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(72) Inventors; and
(71) Applicants : **WESTPHAL, Sören** [DE/DE]; Fischergasse 6, 89079 Ulm (DE). **FROMME, Tobias** [DE/DE]; Pullinger Hauptstr. 7, 85354 Freising (DE). **KLINGENSPOR, Martin** [DE/DE]; Körnerstr. 7c, 85356 Freising (DE).

(74) Agent: **MEIER, Jürgen**; Vossiuis & Partner, Siebertstraße 3, 81675 München (DE).

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(54) Title: FGF-8 FOR USE IN TREATING DISEASES OR DISORDERS OF ENERGY HOMEOSTASIS

(57) Abstract: The present invention relates to polypeptides for use in treating diseases or disorders of energy homeostasis such as obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or the metabolic syndrome. The invention also relates to polynucleotides encoding said polypeptides for use in treating diseases or disorders of energy homeostasis. Also provided by the present invention are pharmaceutical compositions comprising said polypeptides and polynucleotides for use in treating diseases or disorders of energy homeostasis. Said polypeptides, polynucleotides and pharmaceutical compositions may be administered locally, in particular locally into the visceral adipose tissue. Another aspect of the invention relates to a cosmetic product and the use of said cosmetic product for reducing body weight, in particular for reducing abdominal adipose tissue.

FGF-8 FOR USE IN TREATING DISEASES OR DISORDERS OF ENERGY HOMEOSTASIS

The present invention relates to polypeptides for use in treating diseases or disorders of energy homeostasis such as obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and/or the metabolic syndrome. The invention also relates to polynucleotides encoding said polypeptides for use in treating diseases or disorders of energy homeostasis. Also provided by the present invention are pharmaceutical compositions comprising said polypeptides and/or polynucleotides for use in treating diseases or disorders of energy homeostasis. Said polypeptides, polynucleotides and/or pharmaceutical compositions may be administered locally, in particular locally into the visceral adipose tissue. Another aspect of the invention relates to a cosmetic product and the use of said cosmetic product for reducing body weight, in particular for reducing abdominal adipose tissue.

Obesity is a medical condition in which excess body fat has accumulated to an extent that it has an adverse effect on health, leading to reduced life expectancy and/or increased health problems. For example, obesity is associated with various metabolic disturbances, such as type 2 diabetes, insulin resistance, hyperglycemia, dyslipidemia, high blood pressure and metabolic syndrome. Obesity is most commonly the consequence of a misbalanced energy homeostasis caused by a misbalance of food energy intake and physical activity. However, the misbalanced energy homeostasis leading to obesity can also be the result of genetic susceptibility, endocrine disorders, medications and/or psychiatric illness.

Diseases of energy homeostasis such as obesity and secondary diseases of obesity (like diabetes, insulin resistance, hyperglycemia, dyslipidemia, high blood pressure and metabolic syndrome) are considered a major health issue. For example, obesity has emerged as a global health problem with more than 1.1 billion adults to be classified as overweight or obese (Oh, Curr Top Med Chem (2009), 9: 466-481).

Nevertheless, treatment of diseases of energy homeostasis (such as obesity) is so far not satisfactory.

Mammals have two types of adipose tissue (i.e. fat), brown adipose tissue (BAT, also called brown fat) and white adipose tissue (WAT, also called white fat). The primary function of brown adipose tissue is to produce body heat in mammals without the necessity to shiver. In brown adipose tissue, body heat is produced by signaling the mitochondria to allow protons to run back along the proton gradient without producing ATP (proton leak). This is realized by the uncoupling protein 1 (Ucp1 or thermogenin) which allows re-entry of protons from the intermembrane space into the matrix, thereby bypassing ATP synthase, and thus, uncoupling oxygen consumption from ATP production. This alternative route for protons uncouples oxidative phosphorylation and releases energy as heat.

Brown adipose tissue is highly specialized for non-shivering thermogenesis. For example, as compared to white adipocytes, brown adipocytes have a higher number of mitochondria and these mitochondria have a high concentration of Ucp1 in the inner membrane. The term brown adipocytes refers to all types of thermogenic, UCP1 expressing and/or multilocular cells. These are sometimes categorized into “classical brown” versus “beige” or “brite” and others. The term brown adipocytes is intended to encompass all of this, e.g. “brown adipocytes in white adipose tissue” is synonym to both “beige” and “brite”.

Brown adipocytes are not restricted to uniform, classical BAT depots but are often found interspersed in white adipose tissue (WAT) depots. This second type of brown adipocyte has been termed beige or brite (brown in white) and seems to emerge from a different progenitor cell than classical brown fat cells (reviewed in (Pfeifer & Hoffmann, 2014)). To convert WAT into BAT by means of recruiting brite cells offers a possibility to massively increase the BAT amount accessible to therapeutic activation and at the same time decreases the amount of WAT, thereby replacing an energy-storing organ with an energy-dissipating one. This browning of white fat has been subject to intense research during the last years and several systemic interventions have been identified increasing the number of brite cells in mice, including cold exposure and treatment with β-adrenergic agonists or cardiac

natriuretic peptides (Bordicchia et al., 2012; Fisher et al., 2012; Guerra, Koza, Yamashita, Walsh, & Kozak, 1998; Young, Arch, & Ashwell, 1984).

Brown adipose tissue is especially abundant in newborns and in hibernating mammals. However, this tissue and its ability to combust nutrient energy into heat has recently gained increased attention by the scientific community after the repeated and convincing demonstration that adult healthy humans possess appreciable amounts of metabolically active BAT. It is accepted in the art that physiological or pharmacological activation of BAT thermogenesis is effective in treating some of the most widespread diseases of our time including obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

The thermogenic function of brown adipose tissue (BAT) has been known for more than 50 years and has been subject to intense study. The specific properties of this tissue and its utility in the treatment of human metabolic disease are well accepted in the field and documented in numerous studies. A recent review stated that "*from the literature, it is clear that expansion or increased activity of BAT in rodents is associated with a metabolically healthy phenotype*." (Lidell, (2014) J Int Med 276, 364-377). The presence of BAT is associated with low body mass index, low total adipose tissue content and a lower risk of type 2 diabetes mellitus (Lidell, loc. cit.). Thus, it is accepted in the art that expansion or increased activity of BAT is associated with a healthy phenotype in animals and adult humans. Vice versa, it is accepted that a defect in BAT in animals and adult humans is associated with or is a cause of metabolic disorders like diabetes and obesity (Himms-Hagen (1979) CMA Journal 121, 1361-1364; Rothwell (1979) Nature 281, 31-35). Indeed, recent studies documented the presence of substantial amounts of metabolically active brown adipose tissue in healthy adult humans, while BAT was significantly reduced in overweight or obese adult humans (Saito (2009) Diabetes; Lichtenbelt (2009) NEJM 360, 1500-1508, Virtanen (2009) NEJM 360, 1518-1525).

It is evident that the limiting factor for the therapy of metabolic diseases and disorders is the low amount of brown adipose tissue in adult, especially overweight, humans (Bartelt & Heeren, (2014) *Nat Rev Endocrinol, 10*(1), 24-36; Klingenspor, M., Fromme, T. (2012). Brown adipose tissue. In M. E. Symonds (Ed.), *Adipose Tissue*

Biology (Vol. 414). Heidelberg: Springer; Saito et al., (2009) *Diabetes, 58*(7), 1526-1531; Virtanen et al., (2009) NEJM 360, 1518-1525). Animal experiments have indeed demonstrated that metabolic diseases and disorders like diabetes type I and II, obesity and hyperlipidemia can be treated by increasing the amount of active BAT (e.g. by converting white adipocytes into brown adipocytes); further it is accepted in the art that corresponding results can be obtained in therapy of humans suffering from metabolic diseases and disorders (Cinti (2006) Nutr Metabol Cardiovas Dis 16, 569-574; Bartelt (2011) Nature Medicine doi:10.1038/nm.2297); Gunawardana (2012) Diabetes 61, 674-682; Gunawardana (2014) World J Diabet 15, 420-430; Roman (2014) Translational Research, 1-15;.

The properties of active brown adipose tissue that constitute its potential to counteract specific metabolic diseases are outlined in the following:

In relation to obesity and related disorders it is accepted that brown adipose tissue is characterized by its ability to release chemical energy as heat. Specific heat production is enormous and ranges from approx. 150 to 500 Watt/gram tissue in rodents. Increasing the mass of active brown adipose tissue necessarily leads to increased total energy expenditure and therefore to weight loss, assuming no compensation by increased food intake. The possibility to utilize active brown fat to counter obesity is generally accepted in the field and consequence of fundamental thermodynamics (Himms-Hagen, 1979; Klingenspor, 2012; Rothwell & Stock, 1979).

In relation to diabetes it is known that brown adipose tissue imports and combusts large amounts glucose from the blood stream. In fact, the first unambiguous demonstration of active brown fat in humans relied on a high uptake of labelled glucose and is thus a certain property of human brown fat (van Marken Lichtenbelt et al., 2009; Virtanen et al., 2009). This property can be applied to counter excessive glucose levels that are the primary manifestation of diabetes. In a rodent model, a brown adipose tissue transplant into a white adipose tissue depot even effectively cures diabetes (Gunawardana & Piston, 2012).

The following non-limiting references support the utility of BAT in the therapy of metabolic diseases and disorders, like obesity and diabetes as well as related

disorders, such as insulin-resistance, hyperglycemia and metabolic syndrome. For example, Lidell et al. (2014, loc. cit.) disclosed that "pharmacologic interventions that activate and expand BAT would provide a very attractive means for weight reduction, especially in individuals unable to exercise" and that "data from rodents provide robust indications for amelioration of insulin resistance upon physiological or pharmacological stimulation of BAT." Gunawardana (2014, loc. cit) stated that "increasing the content of endogenous brown adipose tissue is known to combat obesity and type 2 diabetes in both humans and animal models." Roman et al., (2014, loc. cit.) confirmed that "ultimately, the long-term effect of increasing BAT is expected to improve energy expenditure, leading to weight loss and increased insulin sensitivity." "a BAT phenotype of the adipose organ in rodents is important for the prevention of obesity and diabetes. [...] in vivo and in vitro data suggest that the human adipose organ react similarly to the murine adipose organ."

In relation to dyslipidemia, it is known in the art that the excessive metabolic rate of active brown adipose tissue is mainly fueled by lipids which are partially imported from the blood stream. In the active state, brown adipose tissue is considered the master regulator of triglyceride rich lipoprotein clearance and blood lipid abundance (Bartelt et al., 2011). From these established properties it is evident that increased mass of active brown fat can be utilized to lower pathological levels of circulation lipoproteins and lipids. For example, Bartelt et al., (2011, loc. cit. disclose that "BAT activation is able to correct hyperlipidemia". Bartelt & Heeren (2014, loc. cit.) described the following in relation to hyperlipidaemia: „The activation of brown adipose tissue (BAT), the primary organ for heat production, confers beneficial effects on adiposity, insulin resistance and hyperlipidaemia, at least in mice. As the amount of metabolically active BAT seems to be particularly low in patients with obesity or diabetes mellitus who require immediate therapy, new avenues are needed to increase the capacity for adaptive thermogenesis."

Summarizing the above it is evident that an increase in active brown adipose tissue necessarily leads to an improvement in the primary manifestations of a disease or disorder of energy homeostasis, like obesity, diabetes, dyslipidemia, insulin resistance, hyperglycemia and metabolic syndrome.

The amount of human BAT, however, is limited and estimated to account for approximately 0.05-0.1% of body mass as compared to a far more than 10-fold higher specific amount in mice. Thus, to therapeutically use the unique capabilities of BAT in humans, not only acute activators may be required, but also agents that recruit a greater number of brown adipocytes.

Brown adipocytes are not restricted to uniform, classical BAT depots but are often found interspersed in otherwise white adipose tissue depots. This second type of brown adipocytes has been termed beige or brite (brown in white) and seems to emerge from a different progenitor cell than classical brown fat cells. To convert WAT into BAT by means of recruiting brite cells offers a possibility to massively increase the BAT amount accessible to therapeutic activation and at the same time would decrease the amount of WAT, thereby replacing an energy-storing tissue with an energy-dissipating one. This browning of white fat has been subject to intense research during the last years. Meanwhile, several systemic interventions are known which at least to a certain degree increase the number of brite cells in mice. These systemic interventions include cold exposure and treatment with β-adrenergic agonists, cardiac natriuretic peptides or fibroblast growth factor 21 (FGF21). However, these compounds act systemically which bears the risk of severe side effects. Moreover, these compounds mainly target subcutaneous adipose tissue, although particularly the visceral adipose tissue is known to correlate with secondary diseases arising from obesity such as diabetes, metabolic syndrome, cardiovascular disease and premature death (see, e.g., Fisher, Genes & Development (2012) 26, 271-281 and Bordicchia et al. (The Journal of clinical investigation (2012) 122, 1022-36).

Thus, there is a need for medical intervention of diseases or disorders of energy homeostasis.

Accordingly, the technical problem underlying the present invention is the provision of means and methods for the medical intervention of diseases or disorders of energy homeostasis, such as obesity, like central obesity.

This technical problem has been solved by the embodiments provided herein and as described in the claims and illustrated in the appended examples.

Accordingly, the present invention relates to a polypeptide for use in treating a disease or disorder of energy homeostasis,
wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome, and
wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(d) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (c), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(e) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a) and (c).

The function of the above described polypeptide of the invention is the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes. The term “brown adipocytes” as used herein refers to all types of thermogenic, UCP1 expressing and/or multilocular cells. “Brown adipocytes” are in particular defined by expression of uncoupling protein 1 (herein also “UCP1”, “UCP-

1", "ucp-1". "Ucp1" or "Ucp1", i.e. abbreviations as used in the art). The sequence and function of Uncoupling protein 1 (UCP-1) is well documented in the art and disclosed, inter alia, in Aquila et al. (1985) EMBO J 4(9):2369–2376; Bouillaud et al. (1986). J Biol Chem 261(4):1487–1490; Cassard et al., J Cell Biochem. 1990 Jul;43(3):255-64; Nedergaard et al. Biochim Biophys Acta. (2001) Mar 1;1504(1):82-106.Brown adipocytes are sometimes categorized into "classical brown" versus "beige" or "brite" and others. The term brown adipocytes is intended to encompass all of this, e.g. "brown adipocytes in white adipose tissue" is synonym to both "beige" and "brite".

The present invention solves the above identified technical problem since, as documented herein below and in the illustrative appended examples, it was surprisingly found that certain paracrine FGFs (including FGF8b, FGF8f and/or FGF17) induce browning of white adipose tissue.

The appended examples show that paracrine FGF8 and FGF17 of murine origin induced the expression of brown adipocypte specific gene uncoupling protein 1 (Ucp1/UCP1) in white adipocyte cell lines derived from murine inguinal and epididymal adipose tissue depot; Fig. **1A**. Brown adipocypte specific gene uncoupling protein 1 (Ucp1) is a recognized marker of brown adipocytes; see, for example, Sharp LZ, et al., PLOS ONE, 2012 „Human BAT possesses...". In fact, the presence of thermogenic Ucp1 is the decicive hallmark of a functional brown adipocyte.

Murine FGF8 comprises 8 differently spliced transcripts leading to 8 different proteins FGF8a-h. Four of the eight murine FGF8 isoforms are also present in humans, i.e.FGF8a, b, e and f. Human and murine FGF8 isoforms have the following level of identity of amino acid sequences: FGF8a 100%, FGF8b 100%, FGF8e 98,28%, FGF8f 98,36%. Human and murine FGF8 isoforms have the following level of identity of nucleic acid level: FGF8a: 94,96%, FGF8b 95,22%, FGF8e 94,59%, FGF8f 94,83%.

FGF8 isoforms FGF8b and FGF8f are shown to induce Ucp1 mRNA expression in murine adipocytes, while isoforms Fgf8a and FGF8e did not induce Ucp1 mRNA expression; Fig. **1B**. Fgf8b was highly effective in inducing Ucp1 expression in

murine adipocytes of both inguinal and epididymal origin. FGF8f was only effective in inducing Ucp1 expression in adipocytes of epididymal adipocytes. Epididymal fat is viscerally located, while inguinal fat is subcutaneously located. Both paracrine FGFs are accordingly effective in viscerally located fat. In humans, omental fat is the dominant visceral adipose tissue depot and thus functionally comparable to the murine epididymal fat depot. Further, the presence of visceral fat is more dangerous in terms of health risk.

As documented in the appended examples, FGF8b could be shown as capable of increasing Ucp1 mRNA expression level in adipocytes of both inguinal and epididymal origin in a dose-response study; Fig. **2A**. Further, FGF8b increased the expression level of cell death-inducing DNA fragmentation factor alpha like effector A (Cidea) in adipoocytes with a greater effect size in epididymal as compared to inguinal adiopcytes; Fig. **2B**. Cell death-inducing DNA fragmentation factor alpha like effector A (Cidea) is a known marker of brown adipocytes; see, for example, Sharp LZ, et al., PLOS ONE, 2012 „Human BAT possesses...". I

Thus, the appended in vitro experiments show that paracrine FGFs (including FGF8b, FGF8f and/or FGF17, especially FGF8b,) induce the expression level of markers of brown adipocytes (like Ucp1 and Cidea).

Subsequent in vivo experiments as provided herein, document the capacity of FGF8b to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes. FGF8b was surprisingly shown herein to effectively transform epididymal white adipose tissue into brown adipose tissue; Fig. **6**. To this end, pellets that release 100 ng FGF8 per day and placebo pellets were implanted into the epididymal white adipose tissue of adult mice. Brown adipocytes are not restricted to uniform, classical BAT depots but are often found interspersed in white adipose tissue (WAT) depots. This second type of brown adipocyte has been termed beige or brite (brown in white). To convert WAT into BAT by means of recruiting brite cells offers a possibility to massively increase the BAT amount.

After three weeks the mice were sacrificed and the tissue analyzed. By visual inspection, the implantation site of FGF8b pellets and the surrounding tissue was of

brown colour. By contrast, the non-implanted contralateral depot and the implantation site of placebo remained white; Fig. **6A**.The "browning" effect is even more clearly shown in Fig. **6B**. Here, the brown area of the images of Fig. **6A** were digitally converted into white area. The browning effect of FGF8b is evident compared to placebo. Also brown adipocyte markers Ucp1 and Cox7a1 are increased in the depot; Fig. **6C**. Cox7a1 is a marker for BAT; see Beige Adipocytes are a Distinct Type of Thermogenic Fat Cell in Mouse and Human" (Jun Wu et al, Cell (2012).

Thus, the in vivo experiments show that FGF8b is capable of increasing/inducing brown adipose tissue (BAT). Brown adipose tissue also comprises white adipose tissue (WAT) with interspersed brown adipocytes (beige or brite adipocytes). This documented increase/induction may be due to induction of differentiation/ conversion of white adipocytes to brown adipocytes and/or differentiation of white preadipocytes to brown adipocytes or may be due to recruitment of brown adipocytes.

Therefore, the in vitro and in vivo experiments of the present invention demonstrate that paracrine FGFs, (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) have the capacity to increase or induce brown adipose tissue (BAT). Thus, the data in the present application establish a causal link between paracrine FGFs (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) and BAT.

As explained above, it is accepted in the art that an increase in active brown adipose tissue (BAT) necessarily leads to an improvement in the primary manifestations of a disease or disorder of energy homeostasis, like obesity, diabetes, dyslipidemia, insulin resistance, hyperglycemia and/or metabolic syndrome.

Therefore, the present application provides a clear and plausible teaching that paracrine FGFs, (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) can be used in the therapy of diseases or disorders of energy homeostasis, like obesity, diabetes, dyslipidemia, insulin resistance, hyperglycemia and/or metabolic syndrome.

Thus, paracrine FGFs, (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) are used as the active agent(s) (preferably as the therapeutic agent(s)) in accordance with the present invention.

Certain data provided herein do not show a difference in body weight or blood parameters including glucose, plasma lipids and liver enzymes between FGF8b- and placebo-treated animals. These data do, however, not challenge the finding that paracrine FGFs can suitably be used in the therapy of diseases or disorders of energy homeostasis, since the corresponding experiments were not designed to show an effect of FGF8b on body weight or blood parameters. The miniscule amount of recruited brown fat by implantation of a single pellet releasing FGF8b was not designed or expected to affect metabolic parameters in these particular experimental settings. In particular, the experiments were not conducted in disease models, but in healthy, lean mice. Accordingly, these experiments were not designed to detect amelioration of disease parameters (as they are absent to start with) and rather demonstrate the absence of unwanted effects on the healthy phenotype.

In contrast, in particular the in vivo experiments were designed to prove and document the capacity of FGF8b to recruit brown fat. The experiments of this invention successfully confirmed said capacity. Once the capacity to recruit brown fat is established and confirmed in vivo, the therapeutic effect is not only plausible but also credible since the art acknowledges the clear and evident (positive) link between BAT and therapy of disease or disorder of energy homeostasis, like obesity, diabetes, dyslipidemia, insulin resistance, hyperglycemia and/or metabolic syndrome.

The therapeutic effect of paracrine FGFs (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) can be validated in suitable experiments. For example, it is known in the prior art that the amount of BAT which is described as therapeutically effective in the treatment of metabolic diseases, is in the range of 1 % of body mass; see Klingenspor & Fromme (2012). The amount of BAT formed/recruited in the in vivo experiments in the present invention was far below this range. Therefore, a therapeutic effect is envisaged and expected (also in suitable animal models), if the amount of BAT is increased to the level (or above) reported in the prior art. This may, for example, be achieved by increasing the dose of paracrine FGFs, treatment time and pellet number.

FGF8b dose-dependently increases the expression of several markers of brown adipose tissue, including Ucp1. Moreover, the appended examples demonstrate that the paracrine FGFs of the invention (e.g. FGF8b) reprogram both, proliferating and differentiating preadipocytes. Accordingly, the present invention takes advantage of a known mammalian physiological mechanism. In particular, as mentioned above, brown adipose tissue can cause direct energy output in the form of heat which allows mammals to maintain their high body temperature in a cold environment. In this mechanism the uncoupling protein 1 (Ucp1) effects that stored energy is directly converted into heat. This reaction takes place in brown adipose tissue, which is also present in humans. Accordingly, the present invention provides means and methods for replacing an energy-storing organ with an energy-dissipating one, thereby paving the way for reliably reducing fat mass. Moreover, BAT displays a high uptake of both glucose and lipoproteins from the bloodstream. Since Diabetes mellitus has a prevalence of 8.3 % worldwide and affects 382 million individuals in 2013 care providers are in desperate need of innovative therapeutic approaches for treating this condition. Especially diabetes mellitus type 2 is highly associated with obesity and its hallmark is peripheral insulin resistance especially in the obese. Furthermore, a high concentration of circulating free fatty acids in the overweight patients is toxic for insulin-producing beta-cells of the pancreas. Unfortunately there is nearly none – apart from metformin – established therapeutic agent in treating diabetes without increasing body weight. And even metformins impact on reducing body weight is only minor. All other medications, insulin in particular, result in rising body weight. Accordingly, by augmenting the amount of BAT the present invention thus provides means and methods for the reduction of glucose and lipid plasma levels in diabetic and dyslipidemia patients.

In general, brown adipose tissue removes glucose from the blood stream dependent on tissue mass and activation state. At a given sympathetic tone in white adipose tissue, increasing the mass of brown fat increases the amount of removed glucose. Additionally, the sympathetic tone can be increased (i.e. by fasting) or mimicked by acute co-administration of sympathomimetic drugs, such as indirect sympathomimetics (e.g. ephedrine, methylphenidate). In both cases, brown adipose tissue activity can be expected to not only actively remove glucose, but also increase the insulin sensitivity of other tissues (Lidell et al., 2014, loc. cit.).

Different forms of application can be envisioned in the therapy of diabetes:

First, therapy with paracrine FGFs, preferably FGF8b, can be used to improve insulin sensitivity in combination with insulin and/or alternative diabetes drugs like the biguanids metformin, phenformin, buformin, and proguanil, the glitazones like rosiglitazone, pioglitazone, lobeglitazone, troglitazone, netoglitazone, and rivoglitazone, the glucagon-like Peptide-1 (GLP-1)-receptor agonists like exenatide, liraglutide, lixisenatide, albiglutide, dulaglutide, and taspoglutide, the dipeptidyl-Peptidase-4 (DPP-4) inhibitors like sitagliptin, vildagliptin, saxagliptin, linagliptin, anagliptin, teneligliptin, alogliptin, gemigliptin, dutogliptin, berberine, and lupeol, the group of the sulfonyl ureas like carbutamid, tolbutamid, glibenclamid, glibornurid, gliclazid, glipizide, gliquidon, glisoxepid, glycodiazin, and glimepiride, the alpha-glucosidase inhibitor arcabose, and miglitol, and the group of the sodium dependent glucose transporter (SGLT)-2 inhibitors like canagliflozin, empagliflozin, ipragliflozin, and tofogliflozin. The intended outcome of such therapy would be the reduction of previous, combined medication. In less severe cases this may ultimately lead to complete replacement.

Second, by combination of paracrine FGF treatment, preferably FGF8b treatment, with a sympathomimetic drug (such as indirect sympathomimetics (e.g. ephedrine, methylphenidate), patient may be able to regulate glucose disposal specifically to their individual need and thus replace insulin treatment.

The herein provided therapy with paracrine FGFs is suitable for the treatment of both diabetes type 1 and diabetes type 2. Primarly, the therapy effects a lowering/decrease of glucose levels in the blood. Moreover, an increased amount of BAT improves/increases insulin sensitivity which is of primary benefit in the treatment of diabetes type 2. In relation to diabetes type 1 it may be conceivable to use the herein provided therapy with paracrine FGFs in emergency cases, like acute hyperglycemia.

Previously known therapeutic approaches for the treatment of obesity (and secondary diseases arising from obesity) mainly consist of the reduction in caloric

intake (e.g. through diets, pharmacotherapy with appetite suppressants, operational or mechanical measures, inhibition of food adsorption, etc.) and the increase of calorie consumption (e.g. by physical activity). These treatments, however, often do not lead to the desired and long-term weight reduction.

In contrast, with the help of the means and methods provided by the present invention, effective and lasting weight reduction can be achieved. In particular, the amount and duration of administration of the herein provided paracrine FGFs (e.g. FGF8b, FGF8f or FGF17) may be controlled, and thereby the body weight may be reduced up to normal or ideal weight. The ideal human body weight has been a topic of debate forever, is changing over times and is different between cultures. A lot of formulas and theories have been invented, but a broadly accepted answer is still not found. Generally speaking, the ideal weight should be unique for everyone. The major factors that contribute to a person's ideal weight are height, gender, age, body composition, body type, fat distribution and others. From a medical standpoint the ideal weight is the individual body weight without negative influence on health. i. e. a normal height/body mass equation along with regular body mass composition, especially body fat composition. In addition, by constant or repeated administration of the paracrine FGFs of the invention, a lasting weight reduction can be achieved. A method for reliably and sustainable reducing body weight is of high value in the treatment of obesity or secondary diseases of obesity, such as metabolic syndrome which goes along with severe health problems. In addition, using the paracrine FGFs of the present invention for reducing body weight makes it possible to prevent the occurrence of both, obesity (and obesity-related diseases such as the metabolic syndrome), and its short-and long-term health effects (such as atherosclerosis, strokes and heart attacks). Thus, the means and methods provided herein may significantly increase quality of life and life expectancy of overweight or obese individuals.

In the prior art, β_3-adrenergic receptor (also called β_3-adrenoceptor or β_3-adrenoreceptor) agonists have been shown to be effective thermogenic anti-obesity and insulin-sensitizing agents in rodents. However, due to severe side effects and very complex receptor specificity in humans, β_3- adrenergic receptor agonists could not be brought to market (Arch, European Journal of Pharmacology 440 (2002) 99–

107).As shown in the appended examples, the paracrine FGFs provided by the present invention (e.g. FGF8b, FGF8f and/or FGF17) offer the possibility to achieve brown fat recruitment by a different signaling route. In particular, the illustrative appended examples demonstrate that FGF8b effectively induces conversion of white visceral adipose tissue into brown adipose tissue in mice, wherein a panel of blood parameters including glucose, plasma lipids and liver enzymes do not differ between FGF8b and placebo treated animals.

WO 01/00662 speculates that human FGF8 might be used in the therapy of neurological disorders, such as neuropathy associated with diabetes. WO 01/00662 attributes a neuroprotective property to FGF8. By contrast, the present invention demonstrated an increase in BAT by paracrine FGFs. Furthermore, it is contemplated herein that paracrine FGFs can directly lower the blood glucose level. Thus, the paracrine FGFs can, in contrast to WO 01/00662, directly interfere with the primary cause of metabolic diseases, e.g. obesity and related disorders, rather than trying to cure a secondary symptom like neuropathy associated with diabetes. Thus, the mechanism described in WO 01/00662 underlying the therapy of neurological disorders is completely unrelated to that of the therapy provided in the present invention. Furthermore, diabetic neuropathy is a neuron disorder and not diabetes. Rather, diabetic neuropathy is, like diabetic nephropathy, diabetic osteopathy or diabetic retinopathy, a secondary disorder that may be caused by untreated diabetes or diabetes that is associated with average high glucose levels in the blood. These disorders are not regarded as the pathological codition known as diabetes but rather as pathological conditions that may result from diabetes.

WO 2013/138795 describes a technology to deliver antibodies or antibody-like moieties into a cell by complexing them with a polypeptide having positive surface charge (so termed Surf+ peptides). WO 2013/138795 aims at providing a means for delivery antibodies or antibody-like moieties into a cell. FGF is one of the Surf+ peptides disclosed in WO 2013/138795. Thus, FGF is intended to serve as a carrier while the therapeutic effect is conferred by the antibody. By contrast, the paracrine FGFs, particularly FGF8b, are the active agent (preferably therapeutic agent) to be used in accordance with the present invention. Further, it is preferred herein that the herein provided paracrine FGFs are not fused to (an)other protein(s) or peptide(s),

particularly where the protein(s)/peptide(s) confer a further biological activity distinct from that of the paracrine FGF(s).

US 6,037,329 discloses therapeutic compositions comprising FGF-8. However, US 6,037,329 aims at providing a gene therapy, wherein FGF8 is to be used as a carrier for delivery of nucleic acids (the latter are the therapeutic agent in US 6,037,329). By contrast, the paracrine FGFs, particularly FGF8b, are the active (therapeutic) agent to be used in accordance with the present invention.

WO 2013/149258 discloses a treatment of a metabolic syndrome related disorder by administering a compound that increases the activity and/or protein level of follistatin and/or uncoupling protein 1 (UCP-1). WO 2013/149258 proposes many chemical compounds that might be useful to increase the activity and/or protein level of follistatin and/or uncoupling protein 1 (UCP-1). Yet, WO 2013/149258 does neither disclose nor propose a use of paracrine FGFs, like FGF8, in the treatment of a metabolic syndrome related disorder. WO 2013/149258 merely vaguely mentions by reference to Alexandre et al. ((2006) Development) that Follistatin modulators such as FGF8 may be used to modulate fat mobilization. Yet, WO 2013/149258 does not provide any evidence that FGF8 would be a Follistatin activator as required for the therapy of metabolic disorders in WO 2013/149258, let alone any evidence that FGF8 might therapeutically effective in the therapy of a metabolic syndrome related disorder.

WO 2013/149258 does not provide a full citation of the Alexandre et al. paper. It appears that they referred to Alexandre et al. ((2006) Development 133, 2905-2913). Alexandre et al. (loc. cit.) does not support the notion in WO 2013/149258 that FGF8 may be used to modulate fat mobilization. In fact, Alexandre et al. is completely silent on a potential role of FGF8 in the modulation of fat mobilization. Alexandre et al. merely report on positive and negative regulations by FGF8 to midbrain roof plate developmental plasticity in white leghorn chick and Japanese quail embryos. Thus, WO 2013/149258 has incorrectly summarized the content of Alexandre et al.

Further, Alexandre et al. reports that FGF8 might negatively regulate follistatin in the therein dissected brains of embryos. Thus, Alexandre et al. discloses that FGF8 is a

negative regulator, i.e. inhibitor, of follistatin. According to WO 2013/149258 a treatment of a metabolic syndrome related disorder requires however a compound that increases the activity and/or protein level of follistatin. Alexandre et al. discloses that FGF8 is not suitable for said purpose and its teaching is thus not compatible with that of WO 2013/149258. Therefore, Alexandre et al. teaches away from the use of FGF8 in the therapy of metabolic synsdrome related disorders.

The prior art has shown that FGF21 may play a role in the browning of white adipose tissue in adaptive thermogenesis (Fisher, Genes & Development 26 (2012) 271-281). However, amino acid sequences and mechanism of action of members of the FGF family are extremely diverse (Itoh et al. Developmental Dynamics 237:18–27, 2008, Itoh et al. J. Biochem 2011;149(2):121–130). Thus, one cannot assume that other endocrine members of the FGF family might have a similar effect as FGF21, let alone paracrine or intracellular FGFs. FGF21 belongs to the group of endocrine FGFs and therefore exhibits a systemic effect on multiple target tissues. Systemic action bears a high risk of undesired side effects. In contrast to the prior art, the FGFs provided by the present invention (e.g. FGF8b, FGF8f and/or FGF17) are paracrine FGFs. Paracrine FGFs have a protein domain binding to extracellular matrix components and are thereby less mobile and not found in circulation. Their matrix anchor also serves to stabilize interaction with FGF receptors, while endocrine FGFs require an additional cofactor of the klotho family for that purpose. Accordingly, the paracrine FGFs of the present invention (e.g. FGF8b, FGF8f and/or FGF17) act locally on the target tissue they are released into. This local action may for example be accomplished by a local infusion or by pushing an implantable drug depot (e.g. an erodible implant or a minipump) into the target fat depot. Optionally, application of paracrine FGFs may be performed during visceral surgery, e.g. during the implementation of gastric banding or gastric bypass.

Local action is advantageous, since it is accompanied with fewer side effects than systemic action. In addition, the (positive and negative) effects of a local acting agent are better controllable as compared to a systemic agent. Especially for reducing adipose tissue local action is beneficial, as an adipose tissue depot (in a specific region of the body) can be chosen and specifically be targeted by the paracrine FGFs of the present invention (e.g. FGF8b, FGF8f and/or FGF17). As mentioned above,

insertion of an implantable drug depot (e.g. an erodible implant or a minipump), which releases one or several of the paracrine FGFs of the invention, may be performed during visceral surgery, e.g. during the implementation of gastric banding or gastric bypass. A further advantage of local acting agents is that local action combined with a local and continuous administration (e.g. via an erodible implant or a minipump) enables a constant level of the polypeptides of the invention (e.g. FGF8b, FGF8f and/or FGF17) within the target adipose tissue over a desired period of time. Said period may be as long as the desired reduction of adipose tissue has been achieved. For example, said treatment period may be, e.g., at least 1 day, at least 2 days, at least 3 days, at least 4 days, at least 5 days or at least 6 days. Said treatment period may also be at least 1 week, at least 2 weeks, at least 3 weeks or at least 4 weeks. Said treatment period may also be at least 1 month, at least 1.5 months, at least 2 months, at least 2.5 months, at least 3 months, at least 3.5 months, at least 4 months, at least 4.5 months, at least 5 months, at least 5.5 months, at least 0.5 years, at least 1 year, at least 1.5 years, at least 2 years or more. For example, the treatment period may be three weeks.

As mentioned above, the prior art has shown thatβ_3-adrenergic receptor agonists are able to produce brown adipose cells in white adipose tissue. However, these β_3-adrenergic receptor agonists mainly act in subcutaneous adipose depots. It was reported that FGF19 has a positive impact on BAT mass and recruitment state and transgene FGF19 mice seem to exhibit more BAT than wildtype littermates. However, white adipose tissue was not examined for the presence of brown adipocates in this study. With regard to FGF21, Fisher et al. (Genes & Development 26 (2012) 271-281) describe that treatment with this factor increases the appearance of brown-like adipocytes in subcutaneous white adipose tissue.

An advantage of the present invention over the prior art is the fact that the herein provided polypeptides are particularly active in visceral adipose tissue (such as mammal visceral adipose tissue). For example, as documented in the appended illustrative examples, in fully differentiated white adipocyte cell lines, FGF8b increases Ucp1 mRNA abundance 2.1-fold in subcutaneous (inguinal) adipose tissue and 27.4-fold in visceral (epididymal) adipose tissue. In addition, the appended examples further show that in visceral (epididymal) adipose cells, FGF8b induces

Ucp1 mRNA abundance in a dose dependent manner, while in subcutaneous (inguinal) adipose cells only the highest dose increased Ucp1 expression. Moreover, abundance of the brown adipocyte marker *cell death-inducing DNA fragmentation factor alpha like effector A* (Cidea) mRNA was increased by FGF8b treatment similarly to Ucp1 with a greater effect size in visceral (epididymal) as compared to subcutaneous (inguinal) adipocytes. Furthermore, as also shown in the appended examples, FGF8f specifically targets visceral adipocytes (such as mammal adipocytes). Furthermore, as also shown in the appended examples, in vivo treatment with FGF8b effectively recruits brown adipocytes in visceral adipose tissue (such as mammal adipose tissue) and not (or not detectably) in subcutaneous adipose tissue. In a variety of mammalian species, including both mice and men, a difference in characteristics between subcutaneous and visceral adipose tissue is established. Without being bound by theory, it is expected that these differences be causative for the different sensitivity to FGF8b. Accordingly, FGF8b treatment is effective in mammalian visceral adipose tissue, such as human visceral adipose tissue. The nucleic acid and amino acid sequences of the paracrine FGF family and its receptors are very conserved across mammalian species. For instance, human and murine FGF8b are 100 per cent identical on the amino acid level, and 95 per cent on the coding nucleic acid level. The same is true for the respective receptors, e.g. the identity between human and murine FGFR1 is 98 per cent on the amino acid level, and 89 per cent on a nucleotide acid level, respectively. The identity between the sequence of human and murine FGFR4 is 89 per cent on the amino acid level, and 85 per cent on a nucleotid acid level, respectively. In summary, the entire paracrine FGF signaling system is exceptionelly well conserved between mice and humans and it is feasible and evident that also functionality is conserved.

The appended examples further show that adipocytes of subcutaneous (inguinal) origin are only responsive to FGF8b treatment in their fully differentiated state whereas visceral (epididymal) cells respond to early as well as late treatments during adipose differentiation. Thus, the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) are advantageous over the prior art as they specifically target visceral adipose tissue.

Particularly the visceral adipose tissue is known to correlate with secondary diseases arising from obesity. Visceral (or central) obesity is associated with metabolic abnormalities that increase the risk of type 2 diabetes and coronary heart disease (see, e.g., Després, Ann Med. 33 (2001) 534-41). Obese patients with a substantial accumulation of visceral adipose tissue are characterized by higher insulinaemic and glycaemic responses during an oral glucose challenge as well as by a deteriorated plasma lipoprotein-lipid profile compared with individuals with normal body weights or obese subjects with low levels of visceral adipose tissue. Results of the *Quebec Cardiovascular Study* have shown that the cluster of metabolic disturbances observed among subjects with visceral obesity (hyperinsulinaemia, hyperapolipoprotein B and small, dense low-density lipoprotein (LDL) particles) is associated with a 20-fold increase in the risk of coronary heart disease in a sample of middle-aged men followed over 5 years.

As the paracrine FGFs of the present invention (e.g. FGF8b, FGF8f and/or FGF17) reduce adipose tissue specifically in the visceral adipose depots, these polypeptides are particularly useful in the treatment (and/or prevention) of central obesity and secondary diseases of central obesity (such as metabolic syndrome, dyslipidemia, insulin resistance, type 2 diabetes, disturbed glucose tolerance, a pathological fasting blood sugar level, hypertension, coronary heart disease or hyperglycemia). The treatment of obesity in accordance with the present invention encompasses the therapy of secondary diseases of obesity (such as metabolic syndrome, dyslipidemia, insulin resistance, type 2 diabetes, disturbed glucose tolerance, a pathological fasting blood sugar level, hypertension, coronary heart disease or hyperglycemia).The treatment of central obesity in accordance with the present invention encompasses the therapy of secondary diseases of central obesity (such as metabolic syndrome, dyslipidemia, insulin resistance, type 2 diabetes, disturbed glucose tolerance, a pathological fasting blood sugar level, hypertension, coronary heart disease or hyperglycemia). Thus, the present invention paves the way for an efficient and sustainable weight reduction and the reduction of the risk for several secondary diseases of obesity. Furthermore, it gives rise to a method to prevent weight gain beforehand.

A "patient" or "subject" for the purposes of the present invention includes both humans and other animals, particularly mammals, and other organisms. Thus, the methods are applicable to both human therapy and veterinary applications. In the preferred embodiment the patient or subject to be treated is a mammal (like pets, such as cats or dogs), and in the most preferred embodiment the patient is a human patient. However, the present invention also encompasses the medical intervention of metabolic diseases (like obesity dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome) by use of the compounds, means and methods as described herein in a veterinary setting.

Accordingly, the medical therapy of humans/ human patients is preferred herein. Thus, it is preferred herein that paracrine FGFs as defined herein (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) are for use in the treatment of a human patient suffering from or prone to suffering from a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome. Diabetes includes, but is not limited to, diabetes type 1 and type 2, gestational diabetes, MODY (Maturity Onset Diabetes of the Young). Dyslipidemia includes, but is not limited to, LDL increase, HDL decrease, triglycerid increase and genetic forms of dyslipidemia,

Moreover, as mentioned above, BAT displays a high uptake of both glucose and lipoproteins from the bloodstream. This is an important aspect of the present invention as diabetes mellitus (particularly diabetes mellitus type 2) is highly associated with obesity and its hallmark is peripheral insulin resistance especially in the obese and that a high concentration of circulating free fatty acids in the overweight patients is toxic for insulin-producing beta-cells of the pancreas. As mentioned above, there is nearly none – apart from metformin – established therapeutic agent in treating diabetes without increasing body weight. And even metformins impact on reducing body weight is only minor. Accordingly, by augmenting the amount of BAT the present invention thus provides means and methods for the reduction of glucose and lipid plasma levels in diabetic and dyslipidemia patients. Therefore, the paracrine FGFs of the present invention (i.e. FGF8b, FGF8f and/or FGF17) may be used to treat diabetes. Furthermore, the

paracrine FGFs of the present invention (i.e. FGF8b, FGF8f and/or FGF17) may be used to treat dyslipidemia.

Accordingly, the present invention advantageously provides for paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) which are capable of converting disease causing visceral adipose tissue into the beneficial fat-burning brown adipose tissue, which is involved in the prevention of sustained and life threatening hypothermia in the cold. Thus, the invention relates to a paracrine FGF which is useful for the treatment of diseases or disorders of energy homeostasis (e.g. obesity, such as central obesity). This paracrine FGF of the invention may be FGF8b or FGF8f. FGF8, FGF8f and FGF17 are structurally very similar. Therefore, these factors may signal via the same receptor and/or via the same pathway. Preferably, the paracrine FGF of the invention is FGF8b or FGF8f. More preferably, the paracrine FGF of the invention is FGF8b.

The present invention relates in particular to the following items:

1. A polypeptide for use in treating a disease or disorder of energy homeostasis, wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;
   (c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;
   (d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes

and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white visceral adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

2. A polynucleotide for use in treating a disease or disorder of energy homeostasis, wherein the polynucleotide encodes the polypeptide of item 1.

3. A pharmaceutical composition comprising the polypeptide of item 1 and/or the polynucleotide of item 2 for use in treating a disease or disorder of energy homeostasis, further comprising a pharmaceutically acceptable carrier and/or diluent.

4. A method of treating a disease or disorder of energy homeostasis by administering an effective dose of the polypeptide of item 1, the polynucleotide of item 2, or the pharmaceutical composition of item 3, to a subject in need of such treatment.

5. The polypeptide of item 1, the polynucleotide of item 2, the pharmaceutical composition of item 3, or the method of item 4, wherein said polypeptide binds to an FGF receptor.

6. The polypeptide of item 5, the polynucleotide of item 5, the pharmaceutical composition of item 5, or the method of item 5, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3.

7. The polypeptide of any one of items 1, 5 or 6, the polynucleotide of any one of items 2, 5 or 6, the pharmaceutical composition of any one of items 3, 5 or 6, or the method of any one of items 4, 5 or 6, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally.

8. The polypeptide of item 7, the polynucleotide of item 7, the pharmaceutical composition of item 7, or the method of item 7, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally into the visceral adipose tissue.

9. The polypeptide of any one of items 1 and 5 to 8, the polynucleotide of any one of items 2 and 5 to 8, the pharmaceutical composition of any one of items 3 and 5 to 8, or the method of any one of items 4 to 8, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administered into the visceral adipose tissue.

10. The polypeptide of any one of items 1 and 5-9, the polynucleotide of any one of items 2 and 5 to 9, the pharmaceutical composition of any one of items 3 and 5 to 9, or the method of any one of items 4-9, wherein said polypeptide, polynucleotide or pharmaceutical composition is in the form of an erodible implant, an implantable drug release device, a gel for injection or a solution for injection.

11. The polypeptide of any one of items 1 and 5-10, the polynucleotide of any one of items 2 and 5-10, the pharmaceutical composition of any one of items 3 and 5-10, or the method of any one of items 4-10, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administered via a minipump.

12. The polypeptide of any one of items 1 and 5-11, the polynucleotide of any one of items 2 and 5-11, the pharmaceutical composition of any one of items 3 and 5-11, or the method of any one items 4-11, wherein said disease or disorder of energy homeostasis is at least one disease or disorder selected from the

group consisting of obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

13. The polypeptide of item 12, the polynucleotide of item 12, the pharmaceutical composition of item 12, or the method of item 12, wherein said obesity is central obesity.

14. The polypeptide of any one of items 1 and 5-13, the polynucleotide of any one of items 2 and 5-13, the pharmaceutical composition of any one of items 3 and 5-13, or the method of any one of items 4 and 5-13, wherein said polypeptide, polynucleotide or pharmaceutical composition is co-administered with at least one other active agent.

15. The polypeptide of item 14, the polynucleotide of item 14, the pharmaceutical composition of item 14, or the method of item 14, wherein said other active agent is at least one active agent selected from the group consisting of beta-adrenergic agonists (e.g. noradrenalin, isoproterenol, BRL 35135, ICI D7114, CGP-12177A, CL 316243), indirect sympathomimetics (e.g. ephedrine, methylphenidate), atrial natriuretic peptide (e.g. ANP, BNP) and ANP/BNP receptor agonists (e.g. AP-811).

16. The polypeptide of item 15, the polynucleotide of item 15, the pharmaceutical composition of item 15, or the method of item 15, wherein said beta-adrenergic agonist is a beta3-adrenergic agonist.

17 The polypeptide of item 16, the polynucleotide of item 16, the pharmaceutical composition of item 16, or the method of item 16, wherein said beta3-adrenergic agonist is CL 316243.

18. A method for the preparation of a pharmaceutical composition for use in treating a disease or disorder of energy homeostasis, wherein the method comprises the following steps:

(a) contacting the polypeptide of any one of items 1 and 5-17 and/or the polynucleotide of any one of items 2 and 5-17 with a liquid carrier or a solid carrier;

(b) optionally, adjusting the pH and/or the osmolarity of the product obtained in step (a);

(c) optionally, sterilizing the product obtained in step (a) or (b); and

(d) formulating and/or packaging the product obtained in step (a), (b) or (c) as a finished medical product.

19. The method of item 18, wherein said carrier is at least one carrier selected from the group consisting of cellulose, lactose, water, saline, Ringer's solution, dextrose solution, a fixed oil, ethyl oleate and liposomes.

20. A cosmetic product comprising a polypeptide, wherein said polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the

function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

21. Use of the cosmetic product of item 20 for reducing body weight.

22. The cosmetic product of item 20, or the use of item 21, wherein said polypeptide binds to a FGF receptor.

23. The cosmetic product of item 22, or the use of item 22, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1c, FGF receptor 2c and FGF receptor 3c.

24. The cosmetic product of any one of items 20, 22 and 23, or the use of any one of items 21-23, wherein said cosmetic product reduces abdominal adipose tissue.

25. The cosmetic product of any one of items 20 and 22-24, or the use of any one of items 21-24, wherein said cosmetic product is in the form of a cream, salve or gel.

26. The cosmetic product of any one of items 20 and 22-25, or the use of any one of items 21-25, wherein said cosmetic product further comprises a chemical penetration enhancer.

27. The cosmetic product of item 26, or the use of item 26, wherein said chemical penetration enhancer is at least one chemical penetration enhancer selected from the group consisting of ethanol, isopropyl alcohol, decanol, hexanol, lauryl alcohol, myristyl alcohol, octanol, octyl dodecanol, oleyl alcohol, azone, ethyl acetate, octyl salicylate, 2-ethylhexyl 4-(dimethylamino)benzoate, ethyl oleate, glyceryl monoleate, glyceryl monocaprate, glyceryl tricaprylate,

isopropyl myristate, isopropyl palmitate, propylene glycol monolaurate, propylene glycol monocaprylate, 2-(2-ethoxyethoxy)ethanol, lauric acid, linoleic acid, linolenic acid, myristic acid, oleic acid, palmitic acid, stearic acid, isostearic acid, dipropylene glycol, propylene glycol, 1,2-butylene glycol, 1,3-butylene glycol, N-methyl-2-pyrrolidone, 2-pyrrolidone, decylmethyl sulphoxide, dimethyl sulfoxide, sodium lauryl sulphate, alkyl dimethylbenzyl ammonium halides, alkyl trimethyl ammonium halides, alkyl pyridinium halides, 2-(dodecyloxy)ethanol, polyoxyethylen(20)-sorbitan-monooleat, eugenol, d-limonene, menthol, menthone, farnesol and neridol.

As mentioned, FGF8b, FGF8f and FGF17 are paracrine fibroblast growth factors. The nucleic acid and amino acid sequences of FGF8b, FGF8f and FGF17 are known in the art and also disclosed herein. In particular, the nucleic acid and amino acid sequences of FGF8b are disclosed herein as SEQ ID NO: 2 and SEQ ID NO: 16, respectively. The nucleic acid and amino acid sequences of FGF8f are disclosed herein as SEQ ID NO: 4 and SEQ ID NO: 18, respectively. In addition, the nucleic acid and amino acid sequences of FGF17 are disclosed herein as SEQ ID NO: 9 and SEQ ID NO: 23, respectively. The appended examples show that these paracrine FGFs are capable of inducing conversion of white visceral adipocytes to brown adipocytes.

Accordingly, the present invention relates to a paracrine FGF which has the ability to induce differentiation (or conversion) of white adipocytes and/or white preadipocytes to brown adipocytes (e.g. FGF8b, FGF8f and/or FGF17). In particular, the present invention relates to a paracrine FGF which has the ability to induce differentiation (or conversion) of white visceral adipocytes and/or white visceral preadipocytes to brown adipocytes (e.g. FGF8b, FGF8f and/or FGF17). Assays for testing whether a polypeptide has the ability to induce differentiation or conversion of white (visceral) adipocytes and/or preadipocytes to brown adipocytes are employed routinely in the art. For example, the polypeptide to be tested may be contacted with cultivated white adipocytes or white preadipocytes (e.g. cultivated white visceral adipocytes or cultivated white visceral preadipocytes); and (after differentiation of the white adipocytes or of the white preadipocytes) the expression of marker genes of brown adipose tissue (e.g. Ucp1) may be measured. Or, in other words, the potential of the

polypeptide to be tested to induce the expression of brown adipocyte specific genes (e.g. Ucp1) in white visceral adipocyte cell lines may be tested.

For example, an assay for testing the ability to induce differentiation (or conversion) of white adipocytes and/or white preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes as used herein and comprising routine methodology is described as follows: immortalized white visceral adipocyte cell lines may be established from primary stromal-vascular cells isolated from the murine epididymal adipose tissue depot. Treatment of the cells with the polypeptide to be tested may be started after induction of adipocyte differentiation and continued for the entire differentiation period of 6 days. Adipocyte differentiation may be induced by complementing the medium with 250 µM indomethacin, 500 µM isobutylmethylxanthine and 2 µg/ml dexamethasone for 24h after confluence. The concentration to be chosen for the polypeptide to be tested may be based on the biological IC_{50} value determined in fibroblast proliferation assays by the supplier and ranged between 1 and 50 ng/ml.

To characterize whether a polypeptide (e.g. FGF8b, FGF8f and/or FGF17) has the ability to induce differentiation or conversion of either white adipocytes or preadipocytes (e.g. white visceral adipocytes or preadipocytes) to brown adipocytes, a sensitive time window for the conversion during the differentiation process can be identified (1) by treatment during several different time windows (for example, and without limitation, during two days of proliferation, two days of induction or every two day time span during differentiation) and (2) quantification of established BAT markers after full differentiation. Those established BAT markers include Cidea, Cox7a1, Elovl3, Pgc1a, Prdm16, Foxc2, Prb, and above all, Ucp1.

As mentioned above, the invention provides for a polypeptide for use in treating diseases or disorders of energy homeostasis (e.g. obesity). This polypeptide may have a length of 10-1000 amino acids, preferably of 30-800 amino acids, 50-700 amino acids or 70-500 amino acids; more preferably, of 90-300 amino acids, 110-280 amino acids, 150-200 amino acids or 170-270 amino acids; or, most preferably, of 200-230 amino acids. The term “polypeptide” also encompasses fragments and variants of the specific polypeptides provided herein that have the biological

function/activity of the herein described polypeptides. Also the use of short peptides consisting of about 11 or less amino acids is envisaged. These peptides can consist of 10, 9, 8, 7, 6, 5 or 4 amino acids. Also these peptides are polypeptides for use in accordance with the present invention. In one particular aspect of the invention, the polypeptide of the invention has a length of 215 amino acids.

The invention further relates to a polynucleotide for use in treating a disease or disorder of energy homeostasis, wherein the polynucleotide encodes the polypeptide of the invention. The invention also relates to a pharmaceutical composition comprising the polypeptide of the invention and/or the polynucleotide of the invention for use in treating a disease or disorder of energy homeostasis, further comprising a pharmaceutically acceptable carrier and/or diluent. Thus, provided herein is a method of treating a disease or disorder of energy homeostasis by administering an effective dose of the polypeptide of the invention, the polynucleotide of the invention, or the pharmaceutical composition of the invention, to a subject in need of such treatment (e.g. an obese or diabetic or dyslipidemic human being).

The cellular response to FGFs is mediated by FGF receptors (FGFR). A variety of FGFRs is produced from four different genes by differential splicing (FGFR1-4). Thus, one aspect of the invention relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said polypeptide binds to a FGF receptor. Assays for testing whether a given polypeptide binds to a FGF receptor are routinely applied in the art. For example, surface plasmon resonance is commonly used to test whether a particular polypeptide binds to a FGF receptor. Alternatively, binding to a FGF receptor may be assayed by reducing the expression of the FGF receptor in question (e.g. by RNA interference) and analyzing whether the biological effect of the polypeptide to be analyzed (e.g. the conversion of white visceral adipocytes into brown adipocytes) is decreased or lost. A decrease or loss of said biological effect is indicative of a binding of the polypeptide to be analyzed to the FGF receptor in question.

The illustrative appended examples demonstrate that the FGF receptors 1-3 are well detectable in both, subcutaneous (inguinal) and visceral (epididymal) adipose tissue.

Thus, the paracrine FGF of the invention (e.g. FGF8b, FGF8f and/or FGF17) may signal through binding to FGF receptor 1 (FGFR1), FGF receptor 2 (FGFR2) and/or FGF receptor 3 (FGFR3). FGFR1-3 each have the splice forms a, b and c. The murine and human nucleotide and amino acid sequences of FGFR1-4 are shown herein below as SEQ ID NOs:_29 to 36 and 43 to 50. . The nucleotide and amino acid sequences of alternative splice forms of FGFR1-3 are well known in the art and can be retrieved from databases like NCBI or EMBL, for example under the following accession numbers:

	Splice form b	Splice form c
Human		
FGFR1	FGFR1-020 ENST00000397108	*FGFR1-011 ENST00000397103*
FGFR2	FGFR2-201 ENST00000351936	*FGFR2-010 ENST00000457416*
FGFR3	FGFR3-201 ENST00000340107	*FGFR3-203 ENST00000440486*
murine		
Fgfr1	FGFR1-201 ENSMUST00000178276	*FGFR1-202 ENSMUST00000179592*
Fgfr2	FGFR2-011 ENSMUST00000119260	*FGFR2-012 ENSMUST00000117089*
Fgfr3	FGFR3-201 ENSMUST00000114411	*FGFR3-202 ENSMUST00000169212*

Exemplary murine and human nucleotide and amino acid sequences of FGFR3c are shown in SEQ ID NO: 85/86 and 87/88. Exemplary human and murine amino acid sequences of alternative splice forms of FGFR1-3 are shown in SEQ ID NO: 89 to 100. In a preferred aspect of the invention, the paracrine FGFs of the invention (e.g. FGF8b, FGF8f and/or FGF17) achieve browning of white adipose tissue through binding to the FGF receptor e.g. FGFR1c, FGFR2c, FGFR3c and/or FGFR4, particularly to FGFR3c and/or FGFR4. It is known in the art that FGFR1c is strongly expressed in adipose tissue. Thus, the paracrine FGFs of the invention may achieve browning of white adipose tissue through binding to FGFR1c. The appended examples also demonstrate that the FGF receptor 4 is only found in visceral (epididymal) and not in subcutaneous (inguinal) adipose tissue in appreciable

amounts. Moreover, the appended examples show that FGF receptor 4 expression as well as FGF receptor 3c expression correlates with FGF8b sensitivity. These results suggest that the paracrine FGF of the invention (e.g. FGF8b, FGF8f and/or FGF17) accomplish the browning of white adipose tissue through binding to the FGF receptor, particulary FGF receptor 4 or 3c. Thus, the invention further relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3. It is prioritized that the receptor is FGF receptor 1c, 2c or 3c. It is even more prioritized that the FGF receptor is FGF receptor 3c or 4.

The illustrative appended examples demonstrate that local administration of the therein provided paracrine FGFs (e.g. of FGF8b) into the visceral adipose tissue results in a considerable recruitment of brown adipocytes specifically in the visceral adipose tissue depot. Thus, the invention relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally. Local administration encompasses injection or implantation e.g. by using an implantable drug depot.

A further aspect of the invention relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally into the visceral adipose tissue. Thus, according to the invention, it is envisaged that the polypeptide of the invention, polynucleotide of the invention or pharmaceutical composition of the invention is to be administered into the visceral adipose tissue.

The illustrative appended examples show that the herein provided paracrine FGFs (e.g. FGF8b) have the ability to transform white adipose tissue into brown adipose tissue in vivo. More specifically, the appended examples show that the visceral adipose tissue can be effectively and specifically targeted by using an implantable drug depot comprising the herein provided paracrine FGFs (e.g. an implanted “pellet”

comprising FGF8b) which is to be implanted directly into the visceral adipose tissue. Implantable drug depots include erodible implants. An example of an erodible implant is a “pellet” produced by Innovative Research of America, Sarasota, Florida, USA. The catalogue number of a customized “pellet” is Y-999. A further example for an implantable drug depot is an implantable drug release device (such as a minipump) or a single-rod implant. Implantable drug depots (such as a single-rod implant) are commonly known in the art and also used, e.g., for the delivery of contraceptives such as Nexplanon/Implanon. However, the herein provided paracrine FGFs may also be administered within a gel for injection or a solution for injection. For example, a ready-to-use gel for injection or a ready-to-use solution for injection can be used. Thus, the invention provides the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said polypeptide, polynucleotide or pharmaceutical composition is in the form of an erodible implant, an implantable drug release device, a gel for injection or a solution for injection. It is also envisaged in context of the invention that said polypeptide, polynucleotide or pharmaceutical composition is to be administered via a minipump.

Thus, one embodiment of the invention relates to an implantable drug depot for use in administering the paracrine FGFs of the invention (i.e. the polypeptide of the invention, the polynucleotide of the invention or the pharmaceutical composition of the invention) for use in treating obesity and/or secondary diseases of obesity. Accordingly, the invention relates to the use of an implantable drug depot for administering the herein provided paracrine FGFs. The implantable drug depot described herein may be a minipump, such as an osmotic minipump.

The construction and function of a minipump (such as an osmotic minipump) is known in the art and described, e.g., in Theeuwes, Ann Biomed Eng (1976) 4, 343-353 and "http://www.alzet.com". Osmotic minipumps are also designated “miniature pumps” and are miniature, implantable pumps for research and therapy. These minipumps deliver drugs (such as the paracrine FGFs of the invention) continuous and in controlled rates, for durations ranging from one day to several weeks (e.g. six weeks), without the need for external connections or frequent handling. Their

unattended operation eliminates the need for repeated nighttime or weekend dosing by the attending doctor.

The minipump of the invention may be designed as follows. The minipump of the invention may be an osmotic minipump and may consist of a cylindrical reservoir for the fluid (which contains the drug to be delivered, e.g., the paracrine FGF of the inventioin), surrounded by a layer of an osmotic driving agent which, in turn, is encapsulated by a semipermeable membrane. The reservoir material may be chemically inert to most aqueous drug formulations, dilute acids, bases and alcohols. The outer housing of the minipump, which is the membrane, may be highly compatible with tissues when the minipump is implanted in an animal (e.g. a human being). The minipump of the invention may have a total volume of approximately 0.6 ml, and an internal effective volume of approximately 0.2 ml. In the minipump, the active drug (e.g. the paracrine FGF of the invention) may be prepared in a homogenous solution, stable suspension or emulsion and may be contained in the drug reservoir which has a delivery orifice. A collapsible partition such as a diaphragm or bag may separate the drug formulation from the osmotic driving agent that provides the osmotic driving force. The osmotic driving agent may be isolated from the outside environment by a membrane that is permeable to water, but not to the osmotic driving agent. In one embodiment of the invention, the minipump is designed as shown in Fig. 1 of Theeuwes, Ann Biomed Eng (1976) 4, 343-353.

The osmotic minipump described herein operates because of an osmotic pressure difference between a compartment within the minipump, called the salt sleeve, and the tissue environment in which the minipump is implanted. The high osmolality of the salt sleeve causes water to flux into the minipump through a semipermeable membrane which forms the outer surface of the minipump. As the water enters the salt sleeve, it compresses the flexible reservoir, displacing the active agent (e.g. the paracrine FGF of the invention) from the minipump at a controlled, predetermined rate. In one aspect of the invention, the compressed reservoir cannot be refilled and the minipump is designed for single-use only.

The rate of delivery by a minipump of the invention may be controlled by the water permeability of the minipump's outer membrane. Thus, the delivery profile of the

pump is independent of the drug formulation dispensed. Drugs of various molecular configurations, including ionized drugs and macromolecules, can be dispensed continuously in a variety of compatible vehicles at controlled rates. The molecular weight of a compound, or its physical and chemical properties, has no bearing on its rate of delivery by the minipumps of the invention. The volume delivery rate of the minipump of the invention may be fixed at manufacture. The (e.g. osmotic) minipump of the invention may have delivery rates between 0.1 and 10 µl/hr and delivery durations between 1 day and several (e.g. six) weeks. Although the volume delivery rate of the minipump of the invention may be fixed, different dosing rates can be achieved by varying the concentration of agent (e.g. FGF8b, FGF8f and/or FGF17) in the solution or suspension used to fill the minipump reservoir.

The implantable drug depot may also be an erodible implant (e.g. a "pellet" of Innovative Research of America, Sarasota, Florida, USA). Accordingly, this implantable drug depot may be fabricated from a biodegradable matrix. In particular, the implantable drug depot may be made of a matrix fused with an active product (i.e. the herein provided paracrine FGF(s)). For example, the ingredients of the implantable drug depot may comprise a hydrophilic matrix (e.g. cellulose), a surface-active emollient (e.g. cholesterol, phosphates and/or stearates) and filler (e.g. lactose). Thus, the implantable drug depot may be composed of cholesterol, cellulose, lactose, phosphates, stearates and the paracrine FGF(s) provided herein. In accordance with the invention, the implantable drug depot may work by the erosion of the implantable drug depot or by the diffusion of the active product. However, the implantable drug depot described herein may also work by the double process of erosion of the implantable drug depot and diffusion of the active product. The implantable drug depot may locally release 0.1 ng to 1000 mg per day. For example, the implantable drug depot may locally release 1 ng to 500 mg per day, 10 ng to 100 mg per day or 100 ng to 1 mg per day. In one aspect of the invention, the implantable drug depot locally releases 100 ng per day. In context of the invention, that implantable drug depot may locally release the paracrine FGF(s) of the invention (e.g. FGF8b, FGF8f and/or FGF17) for several days. For example, the implantable drug depot may release the paracrine FGF(s) of the invention for at least 1 day, at least 2 days, at least 3 days, at least 4 days, at least 5 days or at least 6 days. The implantable drug depot may also release the paracrine FGF(s) of the invention for at

least 1 week, at least 2 weeks, at least 3 weeks or at least 4 weeks. The implantable drug depot may also release the paracrine FGF(s) of the invention for at least 1 month, at least 1.5 months, at least 2 months, at least 2.5 months, at least 3 months, at least 3.5 months, at least 4 months, at least 4.5 months, at least 5 months, at least 5.5 months, at least 0.5 years, at least 1 year, at least 1.5 years, at least 2 years or more. For example, the implantable drug depot may release the paracrine FGF(s) of the invention for three weeks. An example for an implantable drug depot is, as discussed herein above and herein below, a "pellet" (Innovative Research of America, Sarasota, Florida, USA). Again, dosages/releasing quantities for local release can be determined without further ado by the attending physician. The same holds true for the time frame of the treatment and/or corresponding dosage regimens.

The herein provided paracrine FGFs may also be administered by injecting a gel and/or a solution locally into the target tissue (e.g. into the visceral adipose tissue). Thus, a further embodiment of the invention relates to a gel or solution for use in injecting the polypeptide of the invention, the polynucleotide of the invention of the pharmaceutical composition of the invention locally into the target tissue (e.g. into the visceral adipose tissue) fur use in treating obesity and/or secondary diseases of obesity. Accordingly, one aspect of the invention relates to the use of a gel and/or solution for injecting the polypeptide of the invention, the polynucleotide of the invention or the pharmaceutical composition of the invention into the target tissue (e.g. into the visceral adipose tissue). In accordance with the invention, the gel for injection and/or the solution for injection usually comprise(s) a pharmaceutically acceptable carrier. Examples of non aqueous carriers are propylene glycol, polyethylene glycol, vegetable oils such as olive oil, and injectable organic esters such as ethyl oleate. Aqueous carriers include water, alcoholic/aqueous solutions, emulsions or suspensions, including saline and buffered media. Further carriers are sodium chloride solution, Ringer's dextrose, dextrose and sodium chloride, lactated Ringer's, or fixed oils, fluid and nutrient replenishers, electrolyte replenishers (such as those based on Ringer's dextrose), and the like. Preservatives and other additives may also be present in the herein defined gel for injection and/or solution for injection, such as antimicrobials, anti oxidants, chelating agents, inert gases and the like. The gel for injection may further comprise agents for thickening, such as hyaluronan.

As known in the art, the term "energy homeostasis" relates to any process involved in the balance between energy intake (food consumption) and energy expenditure. In particular, energy homeostasis is the relation between intake of food and output of energy that is positive when the body stores extra food as fats and negative when the body draws on stored fat to provide energy for work. A significant contributor to energy expenditure of mammals is the generation of heat for keeping the body temperature constant as well as muscular and metabolic activities.

Energy turnover in the human body includes the uptake and breakdown of chemical energy delivered by the macronutrients in food (fat, carbohydrate, protein). After resorption across the gut epithelium this chemical energy is either converted to heat and dissipated to the environment, transferred to the environment while conducting external work, excreted as indigestible material with feces and urine or stored mainly as triglycerides in adipose tissues. An adult organism with a healthy energy homeostasis compensates by food consumption the daily amount of energy leaving the body with an equal amount of daily energy uptake into the body . In a balanced state no surplus energy is stored in adipose tissues or elsewhere thus maintaining a stable body mass and body composition (i.e. are not obese). Although in these healthy individuals day-to-day fluctuations in energy stores are detected, these fluctuations are countered over periods of weeks to months by coordinated adjustments of food intake and energy expenditure (see, e.g., Schwartz, Diabetes 52 (2003) 232-238). Unfortunately in modern society, environmental circumstances (thermoneutrality, sedentary lifestyle etc.) and the constant availability of high caloric food counteracts a perfectly balanced energy homeostasis and often lead to a positive energy balance resulting in obesity and secondary diseases of obesity.

In accordance with the invention, a disease or disorder of energy homeostasis is preferably characterized by a positive energy homeostasis (wherein the body is storing energy as fat). Accordingly, the invention relates to the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) for use in treating a disease or disorder of energy homeostasis, wherein said disease or disorder is characterized by a positive energy homeostasis. Such diseases include, e.g., obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

Moreover, BAT displays a high uptake of both glucose and lipoproteins from the bloodstream. This is an important aspect of the present invention as diabetes mellitus (particularly diabetes mellitus type 2) is highly associated with obesity and its hallmark is peripheral insulin resistance especially in the obese. Furthermore, a high concentration of circulating free fatty acids in the overweight patients is toxic for insulin-producing beta-cells of the pancreas. As mentioned above, there is nearly none – apart from metformin – established therapeutic agent in treating diabetes without increasing body weight. And even metformins impact on reducing body weight is only minor. All other medications, insulin in particular, result in rising body weight. Accordingly, by augmenting the amount of BAT the present invention thus provides means and methods for the reduction of glucose and lipid plasma levels in diabetic and dyslipidemia patients. Therefore, the paracrine FGFs of the present invention (i.e. FGF8b, FGF8f and/or FGF17) may be used to treat diabetes.

According to the International Statistical Classification of Diseases and Related Health Problems 10th Revision (ICD-10)-2015-WHO Version for 2015 "diabetes" may be classified as follows:

Diabetes mellitus (E10-E14)

The following fourth-character subdivisions are for use with categories E10-E14:

.0 With coma

Incl.:

Diabetic:

- coma with or without ketoacidosis
- hyperosmolar coma
- hypoglycaemic coma

Hyperglycaemic coma NOS

.1 With ketoacidosis

Incl.:

- Diabetic:
  - acidosis

- without mention of coma

- ketoacidosis

.2† With renal complications

Incl.:

Diabetic nephropathy (N08.3*)

Intracapillary glomerulonephrosis (N08.3*)

Kimmelstiel-Wilson syndrome (N08.3*)

.3† With ophthalmic complications

Incl.:

Diabetic:

- cataract (H28.0*)
- retinopathy (H36.0*)

.4† With neurological complications

Incl.:

Diabetic:

- amyotrophy (G73.0*)
- autonomic neuropathy (G99.0*)
- mononeuropathy (G59.0*)
- polyneuropathy (G63.2*)
  - autonomic (G99.0*)

.5 With peripheral circulatory complications

Incl.:

Diabetic:

- gangrene
- peripheral angiopathy† (I79.2*)
- ulcer

.6 With other specified complications

Incl.:

Diabetic arthropathy† (M14.2*)

Neuropathic diabetic arthropathy† (M14.6*)

.7 With multiple complications

.8 With unspecified complications

.9 Without complications

E10Type 1 diabetes mellitus

Modifier-Hint

[See before E10 for subdivisions]

Incl.:

diabetes (mellitus):

- brittle
- juvenile-onset
- ketosis-prone

Excl.:

diabetes mellitus (in):

- malnutrition-related (E12.-)
- neonatal (P70.2)
- pregnancy, childbirth and the puerperium (O24.-)

glycosuria:

- NOS (R81)
- renal (E74.8)

impaired glucose tolerance (R73.0)

postsurgical hypoinsulinaemia (E89.1)

E11Type 2 diabetes mellitus

Modifier-Hint

[See before E10 for subdivisions]

Incl.:

diabetes (mellitus)(nonobese)(obese):

- adult-onset
- maturity-onset
- nonketotic
- stable

non-insulin-dependent diabetes of the young

Excl.:

diabetes mellitus (in):

- malnutrition-related (E12.-)
- neonatal (P70.2)
- pregnancy, childbirth and the puerperium (O24.-)

glycosuria:

- NOS (R81)
- renal (E74.8)

impaired glucose tolerance (R73.0)

postsurgical hypoinsulinaemia (E89.1)

E12 Malnutrition-related diabetes mellitus

Modifier-Hint

[See before E10 for subdivisions]

Incl.:

malnutrition-related diabetes mellitus:

- type 1
- type 2

Excl.:

diabetes mellitus in pregnancy, childbirth and the puerperium (O24.-)

glycosuria:

- NOS (R81)
- renal (E74.8)

impaired glucose tolerance (R73.0)

neonatal diabetes mellitus (P70.2)

postsurgical hypoinsulinaemia (E89.1)

E13 Other specified diabetes mellitus

Modifier-Hint

[See before E10 for subdivisions]

Excl.:

diabetes mellitus (in):

- malnutrition-related (E12.-)
- neonatal (P70.2)
- pregnancy, childbirth and the puerperium (O24.-)
- type 1 (E10.-)
- type 2 (E11.-)

glycosuria:

- NOS (R81)
- renal (E74.8)

impaired glucose tolerance (R73.0)

postsurgical hypoinsulinaemia (E89.1)

E14 Unspecified diabetes mellitus

Modifier-Hint

[See before E10 for subdivisions]

Incl.:

diabetes NOS

Excl.:

diabetes mellitus (in):

- malnutrition-related (E12.-)
- neonatal (P70.2)
- pregnancy, childbirth and the puerperium (O24.-)

- Type 1 (E10.-)
- Type 2 (E11.-)

glycosuria:

- NOS (R81)
- renal (E74.8)

impaired glucose tolerance (R73.0)
postsurgical hypoinsulinaemia (E89.1)

The treatment of obese diabetes is preferred herein, particularly preferred obese diabetes type 2. The terms "obese diabetes", diabetes associated with obesity", "obesity associated with diabetes" and the like are used interchangeabl herein. In this context "obesity" refers to "overweight" and "obese" as explained and defined herein. In a certain aspect, it is envisaged herein that the treatment of diabetes does not encompass secondary disorders related thereto. For example, it is envisaged that the treatment of diabetes does not encompass the threatment of disorders mentioned in subdivision .2† of the above WHO classification, such as renal complications, like Diabetic nephropathy (N08.3*), Intracapillary glomerulonephrosis (N08.3*) or Kimmelstiel-Wilson syndrome (N08.3*).

Furthermore, the paracrine FGFs of the present invention (i.e. FGF8b, FGF8f and/or FGF17) may be used to treat dyslipidemia.

As mentioned above, obesity is a disorder of energy homeostasis and is defined as an abnormal increase in body fat. Since the eighties of the twentieth century, the incidence of obesity is steadily increasing in countries with a Western lifestyle. For example, more than 30% of the American population is obese. The co-morbidities associated with obesity include dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

As the paracrine FGFs provided herein have the ability to induce conversion of white adipocytes and preadipocytes (e.g. white visceral adipocytes and white visceral preadipocytes) to brown adipocytes, these paracrine FGFs are useful in the

treatment (and/or prevention) of a disease of energy homeostasis such as obesity and secondary diseases of obesity. Thus, one aspect of the invention relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said disease or disorder of energy homeostasis is at least one disease or disorder selected from the group consisting of obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia (such as acute hyperglycemia, e.g. acute hyperglycemia associated with type 1 diabetes) and metabolic syndrome. Said diabetes may be type 2 diabetes. The diabetes may also be type 1 diabetes. The herein provided polypeptide, polynucleotide or method may also be used for treating further secondary disease of obesity such as cardiovascular disease, coronary heart disease, hypertension or stroke.

Obesity and insulin resistance are the core components of the metabolic syndrome which is defined by the World Health Organization (WHO) as the combination of insulin resistance, i.e. type 2 diabetes, impaired glucose tolerance or a pathological fasting blood sugar level, combined with arterial hypertension, dyslipidaemia and obesity (WHO 1998, evidence IV). The metabolic syndrome is also known as the deadly quartet, as it significantly correlates with increased cardiovascular morbidity and mortality (Go, Circulation 125 (2012) 188-97).

With regard to the body mass index (BMI), a measurement obtained by dividing a person's weight in kilograms by the square of the person's height in metres, individuals are considered to be obese when their BMI exceeds 30 kg/m², are considered to be overweight when the BMI lies above 25 and below 30 kg/m², and are considered to be of normal weight when their BMI is above 18.5 and below 25 kg/m². The term “obesity” as used herein encompasses “overweight” and “obese” as defined herein above. Due to the fact that the body composition differs from one human being to the other, the BMI is not necessarily sufficient to predict the individual health risk. Interestingly, close to half the women and more than half the men with normal BMI scores had excessive levels of internal fat deposited around the heart and liver, and streaked through under-used muscles. Therefore, distribution of different fat depots and other tissue masses are crucial.

The total amount of adipose tissue is distributed throughout the human body in a variety of different depots. A common categorization of depots by their localization is the discrimination into subcutaneous (i.e. between skin and abdominal wall) and visceral (i.e. within the abdomial cavity) depots. Subcutaneous adipose tissue is found in all parts of the body and contributes most to total fat mass, typically in the range of 80%.

Common obesity is a state of abdominal fat accumulation. The total amount of abdominal fat can be grouped into subcutaneous (SAT) and visceral (VAT) adipose tissue. The ratio between both is different in men and women. In women, VAT constitutes approx. 25% of SAT (e.g. 3.2 liters SAT; 1,4 l VAT). In men, VAT constitutes approx. 50% (age group under 40 years) to more than 75% (age group above 40 years) of SAT (e.g. 2.7 l SAT; 2.3 l VAT).

During the development of obesity VAT expands more than SAT. Especially VAT mass is associated with negative consequences on human health. Therefore, a human being is considered to be at risk for obesity-related diseases, e.g. dyslipidemia, diabetes, insulin resistance, hyperglycemia and/or the metabolic syndrome, when the individual suffers from increased central fat mass along with or without an above normal BMI.

A formula which takes this into account is the waist to hip ratio defined as the ratio of the circumference of the waist to that of the hips. Waist to hip ratio normal values differ between men and women and are considered to increase with age. The absolute waist circumference (>102 centimetres (40 inch) in men and >88 centimetres (35 inch) in women) and the waist-hip ratio (>0.9 for men and >0.85 for women) are both used as measures of central obesity. Therefore the waist to hip ratio predicts the individual metabolic health risk more accurately.

New imaging techniques, like computed tomography and magnetic resonance imaging, tried to calculate the abdominal fat mass even more precisely. Imaging of the abdomen and a cross-sectional scan of 10-mm thickness centered at the L4-L5 vertebral disc space are obtained with the participant in the supine position with both arms stretched above his/her head. Subsequently, the areas of visceral and

subcutaneous adipose tissue (expressed in square centimeters) are calculated. Visceral fat area (VFA) and subcutaneous fat area (SFA) are summed to obtain the total abdominal fat area (TFA). Individuals with a VFA $\geq$100 cm^2 are considered to be obese and therefore at risk for adverse health conditions.

The latest and most sophisticated attempt is the 3D volumetric and body composition analysis by the Heartlands Hospital, a National Health Service (NHS) Obesity Centre in the United Kingdom. They use a 3D scanner to calculate the Body Volume Index (BVI). It looks at the relationship between mass and volume distribution, i. e. where different body mass is located in the body. It is expected, but not established sufficiently yet, that the BVI will predict the individual health risk for disorders of energy homeostasis most precisely.

As mentioned, the herein provided paracrine FGFs specifically target the visceral adipose tissue which is known to be responsible for several secondary diseases of obesity. Accordingly, the obesity to be treated by the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention may be central obesity. “Central obesity”, “abdominal obesity” or “belly fat” is the accumulation of abdominal fat resulting in an increase in waist size. There is a strong correlation between central obesity and cardiovascular disease. Central obesity, as mentioned above also, is characterized by an excess of visceral fat in the body in which the abdomen protrudes excessively. Central obesity can be diagnosed by taking waist and hip measurements. The absolute waist circumference (>102 centimetres in men and >88 centimetres in women) and the waist-hip ratio (>0.9 for men and >0.85 for women) are both used as measures of central obesity. Another measure of central obesity is the Index of Central Obesity (waist-to-height ratio - WHtR). For persons younger than 40 years, a WHtR ratio of >0.5 (i.e. a waist circumference at least half of the individual's height) is predictive of central obesity (and an increased risk for cardiovascular disease). For persons in the age between 40 and 50 years, a WHtR ratio between 0.5 and 0.6 is indicative of central obesity. For persons older than 50 years, a WHtR of >0.6 is indicative of central obesity. Another diagnosis of obesity is the analysis of intraabdominal fat. The increased amount of fat in this region relates to the higher levels of plasma lipid and lipoproteins. In addition, as described above, central obesity (or abdominal obesity)

can also be diagnosed by using computed tomography and magnetic resonance imaging. In addition or alternatively, central obesity may be diagnosed by using 3D volumetric and body composition analysis as described above.

The appended examples demonstrate that the herein provided paracrine FGFs (e.g. FGF8b) have the ability to increase the total amount of brown adipocytes capable of uncoupled respiration and thus thermogenesis. In particular, the herein provided paracrine FGFs recruit thermogenic potential by inducing the neoformation of brown adipocytes. In accordance with the present invention, the activation of the brown adipose tissue may be accomplished by sympathetic innervation of the surrounding white adipose tissue which releases noradrenalin and stimulates the release of fatty acids of the deposited fat. In addition, the released noradrenalin also activates the newly generated BAT and thereby effectively assists the weight reduction. Sympathetic innervation of white adipose tissue can be activated through energy restriction, e.g. a dietary lifestyle intervention. However, the (newly generated) BAT may also be activated via the administration of pharmaceuticals. Thus, the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) may be co-administered with a pharmaceutical which activates BAT. The co-administration may be a simultaneous, sequential or separate administration. It is prioritized that the administration is sequential.

Thus, the invention relates to the polypeptide of the invention, the polynucleotide of the invention, the pharmaceutical composition of the invention, or the method of the invention, wherein said polypeptide, polynucleotide or pharmaceutical composition is co-administered with at least one other active agent. In accordance with the present invention, said other active agent is at least one active agent selected from the group consisting of beta-adrenergic agonists (e.g. noradrenalin, isoproterenol, BRL 35135, ICI D7114, CGP-12177A, CL 316243), indirect sympathomimetics (e.g. ephedrine, methylphenidate), atrial natriuretic peptide (e.g. ANP, BNP) and ANP/BNP receptor agonists (e.g. AP-811). Said noradrenalin may be native noradrenalin. Said beta-adrenergic agonist may be a beta3-adrenergic agonist and/or said beta3-adrenergic agonist may be CL 316243.

The therapeutic benefit of brown adipose tissue is a function of tissue mass and sympathetic tone (sympathetic catecholamines are activators of brown fat activity). The more brown fat is recruited in white adipose tissue with its specific, given sympathetic tone, the more therapeutic activity is expected. In addition to brown fat recruitment by a paracrine FGF, the sympathetic tone can be increased (e.g. by fasting) or mimicked by co-administration of sympathomimetic drugs. Exemplary sympathomimetic drugs are indirect sympathomimetics (e.g. ephedrine, methylphenidate).

Accordingly, provided herein is a combined preparation of the herein provided FGF (e.g. FGF8b, FGF8f and/or FGF17) and a pharmaceutical which activates BAT for simultaneous, sequential or separate use in therapy. As mentioned above, a pharmaceutical which activates BAT may be a beta-adrenergic agonist, noradrenalin, ephedrine, isoproterenol, methylphenidate, BRL 35135, ICI D7114, CGP-12177A and atrial natriuretic peptide (ANP).

In one aspect of the present invention, the co-administration of the paracrine FGF(s) as provided herein and the other active agent (e.g. an agent for activating brown adipose tissue) may lead to synergistic effects resulting in reduced symptoms and/or faster treatment of a disease or disorder of energy homeostasis (e.g. obesity) as compared to the administration of the inventive paracrine FGF(s) or the other active agent alone.

The term “synergistic effect” is commonly known in the art and used herein to describe a situation where the combined effect of two or more active agents is greater than the sum of the individual active agents. In other words, two or more active agents can interact in a way that the presence of one active agent enhances or magnifies the effect(s) of the second. In particular, synergistic effects may lead to more effective treatment of a disease or disorder of energy homeostasis (e.g. obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and/or metabolic syndrome).

Another embodiment of the invention relates to a method for the preparation of a pharmaceutical composition for use in treating a disease or disorder of energy homeostasis, wherein the method comprises the following steps:

(a) contacting the polypeptide of the invention and/or the polynucleotide of the invention with a liquid carrier or a solid carrier;

(b) optionally, adjusting the pH and/or the osmolarity of the product obtained in step (a);

(c) optionally, sterilizing the product obtained in step (a) or (b); and

(d) formulating and/or packaging the product obtained in step (a), (b) or (c) as a finished medical product.

In this preparation method, said carrier may be at least one carrier selected from the group consisting of cellulose, lactose, water, saline, Ringer's solution, dextrose solution, a fixed oil, ethyl oleate and liposomes. For example, said carrier may be cellulose and/or lactose.

The carrier may contain minor amounts of additives such as substances that enhance isotonicity and/or chemical stability. Such materials are preferably non-toxic to recipients at the dosages and concentrations employed, and may include buffers such as phosphate, citrate, succinate, acetic acid, and other organic acids or their salts; antioxidants such as ascorbic acid; low molecular weight (less than about ten residues) (poly)peptides, e.g., polyarginine or tripeptides; proteins, such as serum albumin, gelatin, or immunoglobulins; hydrophilic polymers such as polyvinylpyrrolidone; amino acids, such as glycine, glutamic acid, aspartic acid, or arginine; monosaccharides, disaccharides, and other carbohydrates including cellulose or its derivatives, glucose, manose, or dextrins; chelating agents such as EDTA; sugar alcohols such as mannitol or sorbitol; counterions such as sodium; and/or nonionic surfactants such as polysorbates, poloxamers, or PEG.

In the preparation method described above, the resulting pharmaceutical composition may be in the form of an erodible implant, a gel, a liquid solution, a pill, a tablet, a capsule, a thin film, a powder, a solid crystal or liposomes. In the herein described method for the preparation of a pharmaceutical composition, the carrier may be at least one carrier selected from the group consisting of cellulose, lactose, water,

saline (e.g. physiological saline), Ringer's solution, dextrose solution, a fixed oil, ethyl oleate and liposomes.

In step (b) of the herein described preparation method, the pH may be adjusted to be the pH of blood (e.g. a pH of 7.35-7.45) or to be the pH of adipose tissue (e.g. a pH of 7.1-7.4. In addition, in step (b) of the preparation method, the osmolarity of the product may be adjusted to be isotonic with blood or adipose tissue. For example, the NaCl content may be adjusted to be isosmotic with blood or adipose tissue. For example, the NaCl content may be adjusted to be 9 g/l NaCl (i.e. 308 mosmol/l).

In step (c) of the above described preparation method, the medical product is sterilized. Methods for sterilization are known in the art. For example, sterilization may be accomplished by, e.g., filtration through sterile filtration membranes (e.g., 0.2 micron membranes). Sterilization may also be accomplished by exposing the medical product to appropriate kind of radiation, id est alpha and beta particles, gamma rays, x-ray, ultraviolet radiation, and neutron radiation. Furthermore, heat, appropriate chemical substances, and plasmas can be used for sterilization.

In step (d) of the herein described preparation method, formulating of the pharmaceutical product is conducted. For example, the product may be shaped into the desired formulation (e.g. into an erodible implant, a gel, a liquid solution, a pill, a tablet, a capsule, a thin film, a powder, a solid crystal or liposomes). Finally, the pharmaceutical product may be packaged. For example, the pharmaceutical product may be placed into a minipump. Alternatively, the pharmaceutical product may be packed in unit or multi-dose containers, for example, sealed ampoules or vials. The method for the preparation of a pharmaceutical composition as described herein results in the production of a finished medical product. Such a product is a product which is ready for administration, sale and distribution. In addition, the package of the pharmaceutical composition may comprise instructions regarding the use of the pharmaceutical composition.

As mentioned above, provided herein are paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) which are capable of converting white visceral adipocytes or preadipocytes to brown adipocytes. FGFs act by binding to and thereby eliciting a response from

FGF receptors The activity of a particular FGF (e.g. FGF8b, FGF8f and/or FGF17) can be mimicked by the activation of the responsive FGF receptor. Accordingly, a small molecule which binds to (and activates) the target receptor of the paracrine FGFs provided herein (e.g. FGF8b, FGF8f and/or FGF17) may be used for treating a disease or disorder of energy homeostasis (e.g. obesity). Thus, one aspect of the present invention relates to a small molecule which simulates binding of the herein provided FGF(s) (e.g. FGF8b, FGF8f and/or FGF17) to its target receptor(s) for use in treating a disease or disorder of energy homeostasis (e.g. obesity).

The illustrative appended examples demonstrate that the FGF receptors 1-3 are well detectable in both, subcutaneous (inguinal) and visceral (epididymal) adipose tissue. The appended examples also show that the FGF receptor 4 is only found in visceral (epididymal) and not in subcutaneous (inguinal) adipose tissue in appreciable amounts and that FGF receptor 4 expression correlates with FGF8b sensitivity. Accordingly, one aspect of the present invention relates to a small molecule which activates at least one FGF receptor selected from the group consisting of FGF receptor 1c, FGF receptor 2c, FGF receptor 3c and FGF receptor 4 for use in treating a disease or disorder of energy homeostasis (e.g. obesity). Preferably, said small molecule activates FGF receptor 4.

As used herein, the term “small molecule” refers to a low molecular weight (<900 Daltons) compound. Small molecules can help to regulate a biological process and have usually a size in the order of 10^{-9} m.

As used herein, the term "polypeptide" relates to a peptide, a protein, or a polypeptide which encompasses amino acid chains of a given length, wherein the amino acid residues are linked by covalent peptide bonds. However, peptidomimetics of such proteins/polypeptides wherein amino acid(s) and/or peptide bond(s) have been replaced by functional analogs as well as polypeptides comprising other than the 20 gene-encoded amino acids (such as selenocysteine) are also encompassed by the invention. Peptides, oligopeptides and proteins may be termed polypeptides. The term polypeptide also refers to, and does not exclude, modifications of the polypeptide, e.g., glycosylation, acetylation, phosphorylation and the like. Such modifications are well described in the art.

The herein provided polypeptides comprise amino acid sequences e.g. the amino acid sequence as shown in SEQ ID NO: 16, or the amino acid sequence comprising amino acids 34-215 of SEQ ID NO: 16, or (a) fragment(s) thereof. As used herein, the term "amino acid" refers to any amino acid known in the art and comprises proteinogenic as well as non-proteinogenic amino acids as known in the art. Proteinogenic amino acids comprise alanine (Ala; A), arginine (Arg; R), asparagine (Asn; N), aspartic acid (Asp; D), cysteine (Cys; C), glutamine (Gln; Q), glutamic acid (Glu; E), glycine (Gly; G), histidine (His; H), isoleucine (Ile; I), leucine (Leu; L), lysine (Lys; K), methionine (Met; M), phenylalanine (Phe; F), proline (Pro; P), serine (Ser; S), threonine (Thr; T), tryptophane (Trp; W), tyrosine (Tyr; Y), valine (Val; V), selenocysteine (Sec; U) and pyrrolysine (Pyl; O). Non-limiting examples for non-proteinogenic amino acids are hydroxyproline, selenomethionine, carnitine, gamma-aminobutyric acid (GABA), lanthionine, dehydroalanine, ornitine, or citrulline. As the skilled person is readily aware of, it is possible that in some cases also non-proteinogenic amino acids may be part of proteins. Amino acids are abbreviated herein by the one-letter code or the three-letter code as commonly used in the art and as also set forth hereinabove.

Without being bound by theory, it is believed that the capacity of paracrine FGFs, preferably FGF8, to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes or to recruit brown adipocytes, may be conferred by the amino acid sequence VTVQSSPNFTQ close to the n-terminal region of the protein. This is explained in more detail below. The corresponding nucleic acid sequence is „GTAACTGTTCAGTCCTCACCTAATTTTACACAG".

The capacity of members of the FGF family to potentially activate UCP-1 mRNA gene expression during the differentiation of different adipocyte cell lines was screened FGF8b and FGF17 were demonstrated to enhance UCP1 mRNA expression in epididymal adipocytes (Figure 1A). It is of note that FGF8 and FGF17 belong to the same subfamily within the FGF gene family, namely the FGF8 subfamily (Itoh et al. Developmental Dynamics 2008). The so called FGF8 subfamily consists of FGF8, FGF17, and FGF18.

FGF8 was demonstrated herein to be the most potent inducer of UCP1. Since the murine FGF8 gene gives rise to 8 differently spliced transcripts leading to 8 different peptides (FGFa-h) in rodents, the ability of the 4 murine isoforms that are also present in humans (FGF8a, b, e, and f) to induce Ucp1 mRNA abundance was assessed (Figure 1B). In adipocytes of inguinal origin, FGF8b was the only isoform with browning potential, while in epididymal adipocytes Fgf8b and Fgf8f were both effective.

Notably, only FGF8b and FGF8f share the amino acid sequence VTVQSSPNFTQ close to the N-terminal region of the protein:

human FGF8 spliceforms

```
human_FGF8a      MGSPRSALSCLLLHLLVLCLQAQ-----------------------------------
human_FGF8b      MGSPRSALSCLLLHLLVLCLQAQ----------------------------VTVQSSPN
human_FGF8f      MGSPRSALSCLLLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQQVTVQSSPN
human_FGF8e      MGSPRSALSCLLLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQ---------
                 **********************

human_FGF8a      ---HVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
human_FGF8b      FTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
human_FGF8f      FTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
human_FGF8e      --QHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
                   ************************************************************

human_FGF8a      TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
human_FGF8b      TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
human_FGF8f      TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
human_FGF8e      TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
                 ************************************************************

human_FGF8a      AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
human_FGF8b      AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
human_FGF8f      AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
human_FGF8e      AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
                 ************************************************************

human_FGF8a      PEPR
human_FGF8b      PEPR
human_FGF8f      PEPR
human_FGF8e      PEPR
                 ****
```

Therefore, these 11 amino acids on exon 1D (nomenclature by MacArthur et al. Development 1995) are likely sufficient to mediate a biological effect. Therefore, it is preferred that the polypeptides for use in accordance with the present invention comprise the amino acid sequence VTVQSSPNFTQ or related sequences like QVTVQSSPNFTQ or QVTVQSSPNFT or fragments thereof that contribute to mediating the biological effect of the polypeptide.

Furthermore, the phenylalanine residue 32 (F32) of this short amino acid peptide has been shown to interact with the hydrophobic groove within the Ig domain III, i.e. the "c" variants of the FGF receptors (Olsen et al. 2013). The "c" spliceforms comprise FGFR1c, FGFR2c, FGFR3c, and FGFR4. Strikingly, FGF17, an endocrine FGF, is also active to induce UCP1 mRNA gene expression (Figure 1A) and also shares F32:

human: FGF8b vs. FGF17

```
human_FGF8b       MGSPRSALS-CLLLHLLVLCLQAQVTVQSSPNFTQHVREQSLVTDQLSRRLIRTYQLYSR
human_FGF17       MGAARLLPNLTLCLQLLILCCQTQGENHPSPNFNQYVRDQGAMTDQLSRRQIREYQLYSR
                  **:.*   .  * *:**:** *:*   :.****.*:**:*. :******* ** ******

human_FGF8b       TSGKHVQVLANKRINAMAEDGDPFAKLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAK
human_FGF17       TSGKHVQVTG-RRISATAEDGNKFAKLIVETDTFGSRVRIKGAESEKYICMNKRGKLIGK
                  ******** . :**.* ****: ****************::***:  ******:****.*

human_FGF8b       SNGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLP
human_FGF17       PSGKSKDCVFTEIVLENNYTAFQNARHEGWFMAFTRQGRPRQASRSRQNQREAHFIKRLY
                  ..**.****************:***::***:*****:****:.*::**:***.**:***

human_FGF8b       RG----HHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR-
human_FGF17       QGQLPFPNHAEKQKQFEFVGSAPTRR----TKRTRRPQPLT
                  :*     : :*:. :***:. .*  *    ::**  *:*
```

FGF8b and FGF17 both exhibit the short amino acid sequence SPNF around F32. This suggests that the potential biological activity of the polypeptides might be conferred by this amino acid sequence. Thus, it is preferred that the polypeptides to be used herein comprise the amino acid sequence SPNF, preferably of from position 29 to position 32 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 33 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). And both FGF8b and FGF17 share the amino acid residue Q at position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or at position 35 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18) Thus, they share the motif "SPNFXQ". Therefore, it is preferred that the polypeptides to be used herein comprise the amino acid sequence SPNFXQ, preferably of from position 29 to position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 35

of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). X may be T or N.

The literature describes that mutation of Phenylalanine F32 to Alanine A32 FGF8bF32A is sufficient to abrogate the organizer activity of FGF8b in the mid-hindbrain development (Olsen et al. 2013). Therefore, FGF8bF32A and FGF17F32A variants can be used to assess whether such variants are no longer capable of inducing differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes or recruiting brown adipocytes.

Abrogation of marker expression, like UCP-1 mRNA expression, by exchange of phenylalanine to alanine would provide evidence for a crucial involvement of F32 in browning of white adipose tissue. Furthermore, fragments like QVTVQSSPNFT, QVTVQSSPNFTQ, VTVQSSPNFTQ, SPNF, SPNFXQ (wherein X may be T or N), or functional fragments thereof, can be used to assess if these short aa peptides are capable of mediating the browning effect. These peptides are also exemplary polypeptides to be used in accordance with the invention.

The following table shows annotations of the positions amino acid residues of the QVTVQSSPNFTQ (and fragments thereof) to exemplary amino acid sequences of FGF8b, FGF8f and FGF17:

Q	V	T	V	Q	S	S	P	N	F	T N	Q	
23	24					29			32		34	FGF8b (SEQ ID NO: 16)
52	53					58			61		63	FGF8f (SEQ ID NO: 18)
						30			33		35	FGF17 (SEQ ID NO: 23)

If polypeptides to be used in accordance with the present invention comprise amino acid sequences like QVTVQSSPNFT, QVTVQSSPNFTQ, VTVQSSPNFTQSPNF, SPNFXQ, SPNF or functional fragments thereof, it is understood that these

sequences (i.e. QVTVQSSPNFT, QVTVQSSPNFTQ, VTVQSSPNFTQ, SPNF, SPNFXQ, or functional fragments thereof) are invariable. In other words, the variation of the amino acid sequence of these polypeptides occurs outside of amino acid sequences like QVTVQSSPNFT, QVTVQSSPNFTQ, VTVQSSPNFTQSPNF, SPNFXQ, SPNF or functional fragments thereof. Variant polypeptides are, for example, polypeptides that are encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined herein (e.g. a nucleic acid molecule having a nucleic acid sequence as depicted in SEQ ID NO: 2, 4 or 9 or a nucleic acid molecule encoding a paracrine FGF, like FGF8b, FGF8f or FGF17, e.g. a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16, 18 or 23. Variant polypeptides are, for example, polypeptides having at least 40% identity to the paracrine FGF polypeptide as defined herein, like FGF8b, FGF8f or FGF17, e.g. a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16, 18 or 23.

Further, different amino acid sequences from the n- and c-terminus of paracrine FGFs (such as FGF8 and/or FGF17) can be added to these fragments. For example, the heparin binding domains of the FGF8 subfamily might be added. The heparin binding domains are of importance because FGF8-subfamily members bind to heparin and are therefore tightly bound to heparansulfate proteoglycans, and subsequently, trapped in the extracellular matrix. This aspect is advantageous in context of the present invention, because this local entrapment of active peptides prevents potential side effects to have systemic consequences.

More than 100 heparin binding proteins have been identified, and the ability to bind heparin itself has, up to now, not been attributed to a specific sequence. Fromm et al. have reported that heparin binding sites frequently contain clusters of basic amino acids, like XBX, XBBX, XBBBX (Fromm Arch Biochem Biophys 1997).

Apart from identifying heparin binding sites, the known splice variants of FGF8, i.e. FGF8a-h can be assessed. As shown in Figure 8C FGF8b-mediated responsiveness correlated with Fgf receptor 4 mRNA abundance in immortalized inguinal as well as epididymal adipocytes. Zhang X et al. analysed receptor activation by FGF8, FGF17, and FGF18 by utilizing BaF3 cell mitogenic assay for b-spliced and c-spliced FGFR1,

2, and-3 als well as FGFR4. They observed a high receptor activation of c-spliced FGFRs and FGFR4 by the members of the FGF8-subfamily (Zhang (2006), J. Biol. Chem. 2006, 281:15694-15700). FGFR4 is considered to belong to the c-spliced FGFR family on a functional level. Als shown in Figure 8 C and 8 D data as provided herein show a similar correlation between the FGF8b-mediated responsiveness FGFR4 and other "c" variants of the FGF receptors, e.g. FGFR3c. Ig domain III "c" variants of the FGF receptor comprise exon 7, 9, and the transmembrane domain of the FGF receptors (Figure 9). It is contemplated herein that the observed effect of FGF8b-mediated browning of white adipocytes is a FGF8-subfamily group effect based on interaction of FGF8-subfamily members with the "c" splice variants of the FGFR 1-3 and FGFR4.

Methods for synthesizing polypeptides are known in the art and comprise, e.g., standard FMOC-synthesis as described in the literature (e.g., solid phase peptide synthesis – "A practical approach" by E. Atherton, R.C. Sheppard, Oxford University press 1989) or liquid phase synthesis, where the peptides are assembled using a mixed strategy by BOC-chemistry and fragment condensation as described in the literature (E. Wünsch, "Synthese von Peptiden" in "Methoden der organischen Chemie" (Houben-Weyl), 15 Ausg. 4, Teil 1 und 2 Thieme, Stuttgart, 1974). Another method for synthesizing the polypeptide(s) of the invention is the generation of transgenic cells which express the polypeptide(s) of the invention. After expression of a desired polypeptide(s) by a transgenic cell, said polypeptide(s) may be purified.

The polypeptides to be used herein may be modified. Especially modifications are contemplated herein that alter the amino acid sequence of the naturally occurring polypeptides so that the polypeptides for use in the present invention are distinct from the naturally occurring polypeptides.

Exemplary modifications are explained below.

Phenylalanine is an aromatic, neutral, and nonpolar amino acid. The interaction domain of FGF8b-mediated UCP1 mRNA inducing effect seems to be dependent on interaction with the hydrophobic groove within in Ig III domain of the FGF receptors. Interaction stability might be enhanced by using alternating amino acids, like other

natural or not natural aromatic amino acids. Natural amino acids of this group are Histidine, Tryptophan, and Tyrosine. Other nonpolar amino acids are Alanine, Cysteine, Glycine, Isoleucine, Leucine, Methionine, Proline, Tryptophan, and Valine. Likewise amino acids with similar characteristics (polar, nonpolar, aromatic, neural, negative, positive) can be used to influence interaction of amino acids, especially amino acids of the QVTVQSSPNFT motif and fragments thereof.

Apart from this strategy, peptide or protein engineering of FGF8-subfamily members might be necessary to develop a molecule suitable for clinical use.

Improvement by using and/or inclusion of disulfide bonds: To yield a biopharmaceutically improved variant of FGF8 it might be useful to stabilize the molecule by disulfide bonds. FGF8f, for example, exhibits various cysteine for this purpose (position 10, 19, 138, and 156). Functionally irrelevant aa could be exchanged by cysteines for artificial disulfide bonds.

Glycosylation may be modified, too. For example, w
Improvement by enhancing stability by preventing n-terminal proteolysis: Proteins are cleaved by ubiquitous and multiple intracellular and extracellular peptidases. High-temperature requirement A1 (Htra1a) serine peptidase 1a is a novel antagonist of FGF signalling via cleavage of FGF8 in the extracelluar domain (Kim et al. Mol Cell Biol 2012). This protease was first identified in bacteria und is characterized by a highly conserved trypsin-like serine protease domain. In order to enhance stability of FGF8b and subsequent strengthen the effect of browning in the herein provided in vivo model Htra1a-mediated cleavage can be addressed by removal of the Htra1a target sequence.

Alternative optimization of the FGF8-mediated effect include introduction of a non-natural amino acid, p-acetylphenylalanine, for the site-specific attachement of polyethylene glycol (PEG), the so called PEGylation method. For example, FGF8 conjugates fused to SUMO, FGF8-CovX-body chimera, and/or fusion of FGF8 to the immunoglobulin Fc can be used to enhance therapeutic effectiveness of the described effect of FGF8-subfamily members to induce browning in white adipose tissue.

As mentioned above, the invention relates to "polynucleotides" for use in treating a disease or disorder of energy homeostasis, wherein the polynucleotides encode the polypeptides of the invention. These polynucleotides may be nucleic acids or nucleic acid analogues such as, e.g., DNA molecules, RNA molecules, oligonucleotide thiophosphates, substituted ribo-oligonucleotides, LNA molecules, PNA molecules, GNA (glycol nucleic acid) molecules, TNA (threose nucleic acid) molecules or morpholino polynucleotides. Furthermore, the term "polynucleotide" is to be construed equivalently with the term "nucleic acid molecule" in context of the present invention and may *inter alia* refer to DNA, RNA, PNA or LNA or hybrids thereof or any modification thereof that is known in the art (see, e.g., US 5,525,711, US 4,711,955, US 5,792,608 or EP 302175 for examples of modifications). Nucleic acid residues comprised by the polynucleotides described and provided herein may be naturally occurring nucleic acid residues or artificially produced nucleic acid residues. Examples for nucleic acid residues are adenine (A), guanine (G), cytosine (C), thymine (T), uracil (U), xanthine (X), and hypoxanthine (HX). As understood by the person of skill in the art, thymine (T) and uracil (U) may be used interchangeably depending on the respective type of polynucleotide. For example, as the skilled person is aware of, a thymine (T) as part of a DNA corresponds to an uracil (U) as part of the corresponding transcribed mRNA. The polynucleotides described and provided herein may be single- or double-stranded, linear or circular, natural or synthetic.

The production of a desired polynucleotide is commonly known in the art. For example, the polynucleotide(s) of the invention may be synthesized by generating transgenic cells which express the polynucleotide(s) of the invention. After replication of (a) desired polynucleotide(s) by a transgenic cell, said polynucleotide(s) may be purified. Various means also exist to artificially accomplish the replication of naturally occurring DNA, or to create artificial gene sequences. For example, polymerase chain reaction (PCR) may be used to replicate a particular polynucleotide. Artificial gene synthesis is the process of synthesizing a gene in vitro without the need for initial template DNA samples. This may be achieved by oligonucleotide synthesis from digital genetic sequences and subsequent annealing of the resultant fragments.

The polynucleotides for use in treating a disease or disorder of energy homeostasis (e.g. obesity) provided herein may be cloned into a vector. Thus, the present invention also relates to a vector for use in treating a disease or disorder of energy homeostasis comprising the polynucleotide as described and provided herein. The term "vector" as used herein particularly refers to plasmids, cosmids, viruses, bacteriophages and other vectors commonly used in genetic engineering. Preferably, these vectors are suitable for the transformation of cells, like fungal cells, cells of microorganisms such as yeast, mammalian cells or prokaryotic cells. In a particularly preferred embodiment, such vectors are suitable for stable transformation of bacterial or eukaryotic cells, for example to express the polynucleotides provided herein.

Generally, expression vectors have been widely described in the literature. As a rule, they may not only contain a selection marker gene and a replication-origin ensuring replication in the host selected, but also a promoter, and in most cases a termination signal for transcription. Between the promoter and the termination signal there is preferably at least one restriction site or a polylinker which enables the insertion of a nucleic acid sequence/molecule desired to be expressed.

Non-limiting examples for the vector into which a polynucleotide for use in treating a disease or disorder of energy homeostasis may be cloned are adenoviral, adeno-associated viral (AAV), lentiviral, HIV-based lentiviral, or nonviral minicircle-vectors.

Furthermore, the herein described polynucleotides and/or vectors may be transduced, transformed or transfected or otherwise introduced into a host cell. Thus, the present invention also relates to a host cell comprising the polynucleotide and/or the vector as described and provided herein. For example, the host cell may be a prokaryotic cell, for example, a bacterial cell. As a non-limiting example, the host cell may also be a mammalian cell. The host cell described herein is intended to be particularly useful for generating the polypeptides provided herein. An overview of examples of different corresponding expression systems to be used for generating the host cell described herein is for instance contained in Methods in Enzymology 153, 1987, 385-516, in Bitter (Methods in Enzymology 153, 1987, 516-544), in Sawers (Applied Microbiology and Biotechnology 46, 1996, 1-9), Billman-Jacobe (Current Opinion in Biotechnology 7, 1996, 500-4), Hockney (Trends in

Biotechnology 12, 1994, 456-463), and in Griffiths, (Methods in Molecular Biology 75, 1997, 427-440). The transformation or genetically engineering of the host cell with a polynucleotide or vector described and provided herein can be carried out by standard methods, as for instance described in Sambrook and Russell, 2001, Molecular Cloning: A Laboratory Manual, CSH Press, Cold Spring Harbor, NY, USA; Methods in Yeast Genetics, A Laboratory Course Manual, Cold Spring Harbor Laboratory Press, 1990.

In context of the present invention, the term "local", in particular "local action" means that the effect of an administered agent is restricted to a particular region of the body and does not have a systemic effect. For example, the effect of a polypeptide of the invention (e.g. FGF8b, FGF8f and/or FGF17) may be restricted to the tissue or organ to which said polypeptide has been administered. Since the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) may be administered into adipose tissue (such as visceral adipose tissue), "local action" of a polypeptide of the invention (i.e. of the paracrine FGFs of the invention) means that said polypeptide acts mainly or specifically on adipose tissue (such as visceral adipose tissue). In line with this, "local action" means that no systemic action can be observed. Furthermore, "local action" of a polypeptide of the invention (e.g. of FGF8b, FGF8f and/or FGF17) means that a particular adipose tissue depot (in a specific region of the body) is specifically targeted by said polypeptide. The adipose tissue depot which may completely or partly be converted into brown fat by the inventive polypeptides may have a particular size. The amount of visceral fat, comprising intraabdominal fat and retroperitoneal fat, in an individual with a BMI of 30 kg/m² may have a size 3 kg (1.4 – 4.9). The amount of subcutanous fat may be 4.7 kg (3.2 – 7.1). The total amount of visceral and subcutaneous fat can be subdivided into specific adipose tissue depots. For example, said adipose tissue may be the mesenteric adipose tissue with a size of approximately 1 kg, the omentum majus with a size of approximately 0.5 kg, the perirenal fat tissue with a size of approximately 0.5 kg, or superficial fat depots. The rate of conversion into brown adipose in a given depot may be 0.1% to 100% of the total amount of visceral or subcutaneous fat, e.g. 0.5% to 80%, 1% to 70%, 5% to 60%, 10% to 50% or 20% to 40% of the total amount of visceral or subcutaneous fat. The polypeptides of the invention (e.g. FGF8b, FGF8f and/or FGF17) act locally since these polypeptides are paracrine factors. Paracrine (signaling) factors are

factors for the cell-cell communication which are produced by cells to induce changes in nearby cells to altering the behavior or differentiation of those cells. Signaling molecules known as paracrine factors diffuse over a relatively short distance (local action), as opposed to endocrine factors (hormones which travel considerably longer distances via the circulatory system) and juxtacrine interactions (autocrine signaling). Cells that produce paracrine factors secrete them into the immediate extracellular environment. The paracrine factors then travel to nearby cells in which the gradient of factor received determines the outcome. The distance a paracrine factor (such as FGF8b, FGF8f and/or FGF17) typically travels is in the range of a few cell diameters, e.g. from 1µM to 1mm.

The terms "white adipose tissue" (WAT, or white fat) and "brown adipose tissue" (BAT, or brown fat) are commonly known in the art and described herein above and below. White adipose tissue is the storage of food energy in the form of fat, brown adipose tissue is used for heat production in defense of body temperature against cold. Brown adipose tissue is characterized by an enormously high energy turnover. In particular brown adipose tissue consumes fat and sugar from the blood and can "burn" large amounts of energy.

Brown adipose tissue accounts for approximately 0.05-0.1% of body mass (i.e. 35-70g in a 70kg man). The performance of brown adipose tissue is in the order of 50 mW/g tissue and (if continuously activated) results in a weight loss of about 4 kg of fat per year. This relatively small theoretical weight loss could be increased by increasing the mass of brown adipose tissue (Klingenspor and Fromme, (2012) M.E. Symonds (ed.), Adipose Tissue Biology, Chapter 3, p. 39-69). Thus, an advantageous effect of the inventive polypeptides (e.g. FGF8b, FGF8f and/or FGF17) is that that these factors have the ability to increase brown adipose tissue in white adipose tissue depots, and thus, can increase weight loss induced through the action of the brown adipose tissue.

The functionality of "brown adipose tissue" is commonly known in the art and described in detail, e.g., in Klingenspor and Fromme (2012) M.E. Symonds (ed.), Adipose Tissue Biology, Chapter 3, p. 39-69. In particular, in mammals, a constant body temperature can only be maintained when the rate of heat dissipation equals

the rate of heat loss. Thermoregulatory heat production mechanisms compensating heat loss are classically categorized as shivering and non-shivering thermogenesis. Non-shivering thermogenesis occurs in brown adipose tissue which represents a unique heater organ.

Brown adipocytes are characterized by an abundance of small lipid droplets (multilocular) in contrast to white adipocytes which typically feature a single large lipid droplet (unilocular). They furthermore contain an unusually high amount of mitochondria which confer the eponymous brown color to the tissue. In mitochondria of brown adipose tissue the proton motive force (PMF) across the inner membrane is dissipated as heat rather than converted to ATP (Klingenspor and Fromme, M.E. Symonds (ed.), Adipose Tissue Biology, Chapter 3, p. 39-69). This tightly regulated process is catalyzed by the uncoupling protein 1 (Ucp1). Non-shivering thermogenesis is elicited by the sympathetic innervation from hypothalamic and brain stem control regions which are activated, e.g., by cold sensation. In a cold environment, up to half of the metabolic rate of rodents can be attributed to non-shivering thermogenesis in brown adipose tissue (Klingenspor and Fromme, M.E. Symonds (ed.), Adipose Tissue Biology, Chapter 3, p. 39-69). Accordingly, brown adipose tissue is significantly involved in the prevention of sustained and life threatening hypothermia in the cold.

Notably, the high thermogenic capacity of brown adipose tissue recruited in the defense of normothermia may also play a role in the regulation of energy balance in the face of hypercaloric nutrition (Klingenspor and Fromme, M.E. Symonds (ed.), Adipose Tissue Biology, Chapter 3, p. 39-69).

The terms “visceral” and “subcutaneous” adipose tissue are commonly known in the art and described, e.g., in Ibrahim, Obesity Reviews 11 (2009) 11-18. In particular, there are differences between adipose tissue present in subcutaneous areas and visceral adipose tissue present in the abdominal cavity. These include anatomical, cellular, molecular, physiological, clinical and prognostic differences. Anatomically, visceral adipose tissue is present mainly in the mesentery and omentum, and drains directly through the portal circulation to the liver. Visceral compared with subcutaneous adipose tissue contains a larger number of inflammatory and immune

cells, lesser preadipocyte differentiating capacity and a greater percentage of large adipocytes. There are more glucocorticoid and androgen receptors in visceral adipose tissue than in subcutaneous adipose tissue. In addition, visceral adipose tissue has a greater capacity to generate free fatty acids and to uptake glucose than subcutaneous adipose tissue and is more sensitive to adrenergic stimulation, while subcutaneous adipose tissue is more avid in absorption of circulating free fatty acids and triglycerides. Importantly, preferential fat storage in visceral adipose tissue is associated with an increased mortality.

Fat present around abdominal viscera in mesentery and omentum, known as visceral fat, is different from that present in subcutaneous areas (subcutaneous fat). The type of fat cells (adipocytes), their endocrine function, lipolytic activity, response to insulin and other hormones differ between subcutaneous adipose tissue (SCAT) and visceral adipose tissue (VAT). Subcutaneous fat accumulation represents the normal physiological buffer for excess energy intake (high caloric diet) with limited energy expenditure (physical inactivity). It acts as a metabolic sink where excess free fatty acids (FFAs) and glycerol are stored as triglycerides (TGs) in adipocytes (Freedland, Nutr. Metab. 1 (2004) 12). When the storage capacity of subcutaneous adipose tissue is exceeded or its ability to generate new adipocytes is impaired because of either genetic predisposition or stresses (physiological and mental stress), fat begins to accumulate in areas outside the subcutaneous tissue (the natural store house for energy) and visceral adipose depots develop (Ibrahim, Obesity Reviews 11 (2009) 11-18). Individuals with upper abdominal, central or android obesity are at a greater risk than those with gluteofemoral, peripheral or gynoid obesity for developing several secondary diseases of obesity such as dyslipidemia, diabetes, insulin resistance, hyperglycemia, metabolic syndrome and premature death. In the following, we will use the terms central and peripheral to distinguish between these fat distribution patterns.

More specifically, central obesity carries have a greater risk of developing diabetes and future cardiovascular events than peripheral obese individuals (Ibrahim, Obesity Reviews 11 (2009) 11-18). In addition, central fat accumulation is associated with tendency to hyperglycaemia, hyperinsulinemia, hypertriglyceridemia, impaired glucose tolerance and increased apolipoproteins B-rich lipoproteins, which are

features of the insulin resistance syndrome (Ibrahim, Obesity Reviews 11 (2009) 11-18). In addition, increased risk of developing diabetes is also greater in individuals with excess visceral adipose tissue (Bjorntorp, Diabetes Metab. 26, Suppl. 3 (2000) 10–12; Lemieux, Diabetes Metab. 20 (1994); Dobbelsteyn, Int. J. Obs. Relat. Metab. Disord. 25 (2001) 652). Furthermore, individuals with high levels of visceral adipose tissue area have higher mean plasma cholesterol and triglyceride levels and lower high-density lipoprotein cholesterol values as compared to peripheral obese individuals (Despres, Metabolism 34 (1985)967–973).

Moreover, an increased body waist circumference is considered to be a prerequisite of the metabolic syndrome. Visceral obesity, like hyperinsulinemia and insulin resistance, not only accompanies but antedates the components of the metabolic syndrome (Lemieux, Diabetes Metab. 20 (1994) 375–393; Bergstrom, Diabetes 39 (1990) 104–111). In addition, elevated arterial blood pressure that is one of the components of the metabolic syndrome was explained by insulin resistance and compensating hyperinsulinemia in central obese individuals (Rocchini, Am. J. Hypertens. 15 (2002) 505–525; Sharma, Curr. Hypertens. Rep. 3 (2001) 152–156; Mc Farlane, J. Clin. Endocrinol. Metabol. 86 (2001) 713–718). Furthermore, central obesity can induce the development of hypertension through increased activity of adipose tissue renin-angiotensin-aldosterone system, sympathetic activation and other mechanisms closely connected with insulin resistance (Ibrahim, Obesity Reviews 11 (2009) 11-18).

Visceral fat quantified as waist size has been identified as an independent risk factor for cardiovascular disease, hypertension and stroke (Dobbelsteyn, Int. J. Obs. Relat. Metab. Disord. 25 (2001) 652; Despres, Int. Congr. Ser. 23 (2003) 27–34). In addition, excess visceral adipose tissue has the potential to cause hypercoagulability because of increased secretion of PAI-1. Importantly, increased waist circumference when accompanied by increased triglycerides leads to increased risk of coronary heart disease (Tanko, Circulation 111 (2005) 1883–1890; Lemieux, Circulation 102 (2000) 179–184).

Central obesity correlates closely with other measures of atherosclerosis such as intima–media thickness (Harris, Obes. Res. 8 (2000) 516–524). Peripheral arterial

disease also has been correlated to visceral adipose tissue, but not to total body fat in elderly subjects (Planas, Int. J. Obes. Relat. Metab. Disord. 25 (2001) 1068–1070). In addition, subjects with abdominal obesity were reported to have greater risk of having an abnormal albumin excretion rate (Mulyadi, Ann. Nutr. Metab. 45 (2000) 6–71) and microalbuminuria signifies enhanced cardiovascular risk. Moreover, hyperinsulinemia, associated with visceral (i.e. central) obesity, is a predictor of coronary artery disease (Fontbonne, Diabetologica 34 (1991) 356–361).

In addition, increase in circulating free fatty acids in abdominal obesity is associated with increase in cardiovascular risk. In addition, elevations in free fatty acid levels have been demonstrated to promote endothelial dysfunction (Sharma, 3 Curr. Hypertens. Rep. 3 (2001) 152–156).

Accordingly, assessment of cardiovascular risk in obese patients from the measurement of body weight may be misleading (Kissebah, Diabet. Rev. 5 (1997) 8–20). Only obese individuals characterized by increased visceral adipose tissue show the complications predictive of type 2 diabetes and cardiovascular disease (Mc Farlane, J. Clin. Endocrinol. Metabol. 86 (2001) 713–718). Women generally display a more favorable risk profile than men, and generally lower level of visceral adipose tissue than men. Interestingly, adjustments for differences in visceral fat between men and women eliminated most of the sex differences in cardiovascular risk factors (Lemieax, Diabetologica 37 (1994) 757–764). Peripheral or gluteofemoral fat distribution seems to be even protective against atherosclerosis (Tanko, Eur. Heart. J. 24 (2003) 1531–1537; Tanko, Circulation 107 (2003) 1626–1631; Lassner, Obes. Res. 9 (2001) 644–646).

It is known that obesity is associated with increased cardiovascular disease mortality (Dagenais, Am. Heart. J. 149 (2005) 54–60; Allison, JAMA 282 (1999) 1530–1538). Cardiovascular disease death rates are directly related to body mass index in both men and women and obesity in adulthood is associated with a striking reduction in life expectancy (Lee, J. Gerontol. A. Biol. Sci. Med. Sci. 56 (2001) 7–19). In addition, in particular abdominal adiposity as measured by waist circumference is a significant predictor of mortality independently of body mass index (Zhang, Arch. Intern. Med. 167 (2007) 886–892).

Advantageously, the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) specifically and effectively target visceral fat which is associated with several severe diseases (such as type 2 diabetes, insulin resistance syndrome, metabolic syndrome, cardiovascular disease, coronary heart disease, hypertension and stroke) and is a strong, independent predictor of all-cause mortality in men (Kuk, Obesity 14 (2006) 336–341).

The anatomical and physiological differences between visceral adipose tissue and subcutaneous adipose tissue help explain the increased metabolic and cardiovascular risks associated with abdominal (i.e. central) obesity.

Anatomical differences between visceral adipose tissue and subcutaneous adipose tissue are known in the art and described, e.g., in Ibrahim, Obesity Reviews 11 (2009) 11-18. In particular, the main areas for subcutaneous fat deposition are the femerogluteal regions. About 80% of all body fat is in the subcutaneous area (Wajchenberg, Endocr. Rev. 21 (2000) 679–738; Arner, J. Endocrinol. 155 (1997) 191–192). Intra-abdominal fat is visceral fat which accounts for up to 10–20% of total fat in men and 5–8% in women (Wajchenberg, Endocr. Rev. 21 (2000) 679–738). The amount of visceral fat increases with age in both genders.

Because of its anatomical position, visceral fat venous blood is drained directly to the liver through the portal vein (Ibrahim, Obesity Reviews 11 (2009) 11-18). This contrasts with subcutaneous fat where venous drainage is through systemic veins. The portal drainage of visceral fat provides direct hepatic access to free fatty acids and adipokines secreted by visceral adipocytes. Adipokines activate hepatic immune mechanisms with production of inflammatory mediators such as C-reactive protein (CRP) (Heinrich, Biochem. J. 265 (1990) 621–636; Mårin, Metabolism 41 (1992) 1241–1248).

There are several physiological and metabolic differences between subcutaneous and visceral adipose tissue (Ibrahim, Obesity Reviews 11 (2009) 11-18). For example, in the obesity state, adipocytes from visceral adipose tissue are more insulin-resistant than subcutaneous adipose tissue adipocytes (Abate, J. Clin. Invest.

96 (1995) 88–98; Frayn, Br. J. Nutr. 83, Suppl. 1 (2000) S71–S77). It is noted that smaller adipocytes tend to be more insulin-sensitive whereas large adipocytes become insulin-resistant (Salans, J. Clin. Invest. 52 (1973) 929–941; Bjorntorp, Diabetes Metab. 26, Suppl. 3 (2000) 10–12). In addition, it has been found that the amount of visceral fat represents an important factor associated with variations in insulin sensitivity (Mårin, Metabolism 41 (1992) 1241–1248; Hisra, Nutrition 19 (2003) 457–466; Kadswaki, Exp. Biol. Med. 228 (2003) 1111–1117).

Insulin resistance prevents glucose and more fat from entering the cell and becoming preferentially oxidized. Subjects with visceral abdominal obesity, when compared with those with peripheral obesity, had lower glucose disposal, glucose oxidation and greater lipid oxidation (Ibrahim, Obesity Reviews 11 (2009) 11-18). Insulin resistance may be one of the most important factors linking abdominal visceral adiposity to cardiovascular risk.

Free fatty acids are known to induce insulin resistance (Ibrahim, Obesity Reviews 11 (2009) 11-18). In the liver, insulin inhibits gluconeogenesis and glycogenolysis and stimulates glycogen formation. It has been shown that the degree of free fatty acid suppression following meal ingestion differs between abdominally and peripherally obese persons. In particular, the release of free fatty acids is greater in the abdominally obese individuals (Freedland, Nutr. Metab. 1 (2004) 12).

In addition, in the healthy state, visceral adipose tissue has higher rate of insulin-stimulated glucose uptake compared with subcutaneous adipose tissue adipocytes. Small adipocytes in subcutaneous adipose tissue have a high avidity for free fatty acid and triglyceride uptake. The new, small, more insulin-sensitive adipocytes act as a sink or powerful 'buffers', avidly absorbing circulating free fatty acids and triglycerides in the postprandial period (Freedland, Nutr. Metab. 1 (2004) 12; Arner, J. Endocrinol. 155 (1997) 191–192). Accordingly, subcutaneous adipose tissue cells may act as a buffer or sink for circulating free fatty acids and triglycerides, but once they reach their capacity they lose their protective benefit and fat begins to accumulate in tissues not suited for lipid storage (Freedland, Nutr. Metab. 1 (2004) 12). Notably, subcutaneous adipose tissue in abdominal wall has higher uptake of triglycerides and larger free fatty acid release per kilograms than femoral fat (Mårin,

Metabolism 41 (1992) 1241–1248; Kadswaki, Exp. Biol. Med. 228 (2003) 1111–1117).

Accordingly, visceral and subcutaneous fat are tissues with anatomical, cellular, molecular and physiological differences. Obviously, these considerable differences are the reason for the fact that especially visceral adipose tissue amount correlates with the risk for several diseases such as dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

As indicated above, the present invention relates to a pharmaceutical composition comprising the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17) for use in treating (and/or preventing) a disease or disorder of energy homeostasis (e.g. obesity), further comprising a pharmaceutically acceptable carrier and/or diluent. In context of the present invention, said "pharmaceutical composition(s)" are medicaments. Such pharmaceutical compositions may be administered to a subject in need of medical intervention of a disease or disorder of energy homeostasis (e.g. obesity [such as central obesity], dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome). Thus, the invention also relates to a method of treating a disease or disorder of energy homeostasis by administering an effective dose of the herein provided paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17), to a subject in need of such treatment. In context of the present invention, a subject is a mammal, e.g., a mouse, rat, hamster, rabbit, guinea pig, ferret, cat, dog, chicken, sheep, bovine species, horse, camel, primate or a human being. It is prioritized that the subject is a pet animal (e.g. a dog, a cat, a rabbit, a hamster, a bird, a horse, a monkey or a camel. It is even more prioritized that the subject is a human being.

The pharmaceutical compositions as described herein may be administered to a subject in need of medical intervention of a disease or disorder of energy homeostasis in an amount of about 1 ng/kg body weight per day to about 100 mg/kg body weight per day. For example, the pharmaceutical composition for use in treating a disease or disorder of energy homeostasis may be administered to the subject in an amount of about 0.01 µg/kg body weight per day to about 50 mg/kg body weight per day, or about 0.05 µg/kg body weight per day to about 50 µg/kg body weight per day, or about 0.1 µg/kg body weight per day to about 10 µg/kg body weight per day,

or about 1 µg/kg body weight per day to about 5 µg/kg body weight per day, or about 2 µg/kg body weight per day to about 3 µg/kg body weight per day. For example, the pharmaceutical composition or the polypeptide of the invention may be administered in an amount of 5 µg/kg body weight per day or in an amount of 2.5 µg/kg body weight per day.

It is envisaged in context of the invention, that an implantable drug depot (e.g. a "pellet" [Innovative Research of America, Sarasota, Florida, USA or a minipump) which locally releases the herein provided pharmaceutical composition, polypeptide and/or polynucleotide, is implanted into the subject in need of such treatment. Said pellet or minipump may locally release the above listed amounts of the herein described pharmaceutical composition, polypeptide and/or polynucleotide. For example, said pellet or minipump may locally release 0.2 µg/kg body weight per day to about 1 µg/kg body weight per day. For example, said pellet or minipump may locally release 0.25 µg/kg body weight per day or 0.5 µg/kg body weight per day. The pharmaceutical composition as disclosed herein as well as releasing quantities for local release from a drug depot will be formulated, dosed and provided in a fashion consistent with good medical practice, taking into account the clinical condition of the individual patient, the site of delivery of the pharmaceutical composition, the method of administration, the scheduling of administration, and other factors known to practitioners/physicians. The "effective amount" of the pharmaceutical composition and/or the releasing quantities for purposes herein described is thus determined by such considerations.

Accordingly, also doses below or above the exemplary ranges described hereinabove are envisioned. The skilled person knows that the effective amount of pharmaceutical compositions administered to an individual will, *inter alia*, depend on the nature of the compound. For example, the dose may be further decreased or increased as subject to therapeutic discretion, in particular if concomitantly certain lipids are applied or if the administered polypeptide is subject to certain chemical modifications. The particular amounts may be determined by conventional tests which are well known to the person skilled in the art. The dosage regimen will be determined by the attending physician and clinical factors. As is well known in the medical arts, dosages for any one patient depends upon many factors, including the

patient's size, body surface area, age, the particular compound to be administered, sex, time and route of administration, general health, and other drugs being administered concurrently.

The polypeptides of the present invention may also be used in combinations of two or more polypeptides provided herein. For example, in context of the present invention, the polypeptides FGF8b, FGF8f and/or FGF17 may be administered in combination (simultaneously, sequentially or separate). Accordingly, the pharmaceutical compositions of the present invention may comprise two or more polypeptides provided herein, optionally also in combination with other compounds described and provided herein. Said other compounds may be compounds which activate brown adipose tissue, such as beta-adrenergic agonists (e.g. beta3-adrenergic agonists such as CL 316243), noradrenalin, ephedrine, isoproterenol, methylphenidate, BRL 35135, ICI D7114, CGP-12177A or atrial natriuretic peptide (ANP)).

It is envisaged in context of the present invention that the herein provided pharmaceutical composition, polypeptide or polynucleotide is administered locally. Accordingly, administration of the pharmaceutical compositions, polypeptides or polynucleotides provided herein may be effected by different ways, e.g., parenterally (e.g. intraviscerally, subcutaneous, transdermally, intramuscularly or intraperitoneally) or as an implantable drug depot (e.g. as an erodible implant made of biodegradable polymers, such as cellulose, polylactate or polyglycolate).

Thus, the pharmaceutical composition described and provided herein may be also administered by sustained-release systems. Further suitable examples of sustained-release compositions include semi-permeable polymer matrices in the form of shaped articles, e.g., films, or mirocapsules. Sustained-release matrices include polylactides (U.S. Pat. No. 3,773,919, EP-A1 58481), copolymers of L-glutamic acid and gamma-ethyl-L-glutamate (Biopolymers, 1983, 22: 547-556), poly (2-hydroxyethyl methacrylate) (J Biomed Mater Res, 1981, 15: 167-277; Langer, Chem Tech, 1982, 12: 98-105), ethylene vinyl acetate (Langer, *loc. cit.*) or poly-D-(-)-3-hydroxybutyric acid (EP-A1 133988). Sustained release of pharmaceutical compositions may also include liposomally entrapped compounds. Liposomes containing the pharmaceutical composition may be prepared by methods known in

the art, such as described in DE 3218121; Proc Natl Acad Sci USA, 1985, 82: 3688-3692; Proc Natl Acad Sci USA 77: 4030-4034, 1980; EP-A1 52322; EP-A1 36676; EP-A1 88046; EP-A1 143949; EP-A1 142641; Japanese Pat. Appl. 83-118008; U.S. Pat. Nos. 4,485,045 and 4,544,545; and EP-A1 102324.

In certain circumstances the herein provided pharmaceutical compositions, polypeptides or polynucleotides may be administered intravenously, intraperitoneal, intramuscular, via inhalation (e.g., intrabronchially) or enterally (e.g. a pill, tablet, [e.g. buccal, sublingual, orally or disintegrating], capsule, thin film, liquid solution suspension, powder, solid crystals or liquid), rectally (e.g., suppository, enema), topically, vaginally, epicutaneously or intranasally. However, it is prioritized in context of the invention that the herein provided pharmaceutical compositions, polypeptides or polynucleotides are administered locally (e.g. locally into the visceral adipose tissue). Thus, the herein provided pharmaceutical compositions, polynucleotides or polypeptides may be administered directly to the target site, e.g., by implantation of an implantable drug depot, biolistic delivery to the target site or by catheter.

As mentioned above, the inventive pharmaceutical compositions comprise a pharmaceutically acceptable carrier and/or diluents. Pharmaceutically acceptable carriers are well known in the art and include, e.g., non-aqueous carriers such as propylene glycol, polyethylene glycol, vegetable oils such as olive oil, and injectable organic esters such as ethyl oleate. Aqueous carriers include water, alcoholic/aqueous solutions, emulsions or suspensions, including saline and buffered media. Parenteral carriers include sodium chloride solution, Ringer's dextrose, dextrose and sodium chloride, lactated Ringer's, or fixed oils. Intravenous carriers include fluid and nutrient replenishers, electrolyte replenishers (such as those based on Ringer's dextrose), and the like. Preservatives and other additives may also be present such as, for example, antimicrobials, anti-oxidants, chelating agents, inert gases and the like. Diluents which are used in pharmaceuticals are inactive ingredients that are added to medicaments in addition to the active drug. Diluents may be used as binders, disintegrants (help the tablet break apart in the digestive system), or flavor enhancers. Some very common diluents include starch, cellulose, cellulose derivatives, lactose and magnesium stearate (a lubricant).

As used herein, the terms "treatment", "treating" and the like also means “preventing” and “ameliorating” of a disease such as a disease or disorder of energy homeostasis (e.g. obesity). These terms are used herein to generally mean obtaining a desired pharmacological and/or physiological effect. The effect may be prophylactic in terms of completely or partially preventing a disease or symptom thereof and/or may be therapeutic in terms of partially or completely curing a disease and/or adverse effect attributed to the disease. The term "treatment/treating" as used herein covers any treatment of a disease in a subject and includes: (a) preventing and ameliorating a disease or disorder of energy homeostasis from occurring in a subject which may be predisposed to the disease; (b) inhibiting a disease or disorder of energy homeostasis, e.g. arresting its development; or (c) relieving a disease or disorder of energy homeostasis, e.g. causing regression of a disease or disorder of energy homeostasis. In accordance with the present invention, the term “prevention“ or “preventing“ of an disease means the disease per se can be hindered of developing or to develop into an even worse situation. Accordingly, it is one aspect of the present invention that the herein described polypeptides can be employed in avoidance of a disease or disorder of energy homeostasis. In accordance with the present invention, the peptides as described herein may be employed before a disease or disorder of energy homeostasis develops.

As described herein, the polypeptides for use in treating a disease or disorder of energy homeostasis as described herein may also be employed in the amelioration and/or treatment of disorders wherein the diseased status has already developed, i.e. in the treatment of an existing disease or disorder of energy homeostasis. Accordingly, the term “treatment/treating” as used herein also relates to medical intervention of an already manifested disorder, like the treatment of an already defined and manifested disease or disorder of energy homeostasis. Thus, the present invention relates to the treatment or prevention of a disease or disorder of energy homeostasis by using the polypeptides as described herein, the polynucleotides as described herein or the pharmaceutical compositions as defined herein.

The terms "paracrine FGF(s) of the (present) invention" "paracrine FGF(s) provided herein", "herein provided paracrine FGF(s)", "inventive paracrine FGF(s)", "paracrine FGF(s) described and provided herein", "herein described and provided paracrine FGF(s)", "paracrine FGF(s) for use in treating a disease or disorder of energy homeostasis" and the like are used interchangeably herein and relate to a polypeptide for use in treating a disease or disorder of energy homeostasis, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As mentioned, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

In accordance with the present invention, the polypeptide of item (e) above may have at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes. Herein, the above defined polypeptide is also termed "polypeptide(s) of the invention" or "polypeptide(s) of the present invention".

In addition, the terms "paracrine FGF(s) of the (present) invention" "paracrine FGF(s) provided herein", "herein provided paracrine FGF(s)", "inventive paracrine FGF(s)", "paracrine FGF(s) described and provided herein", "herein described and provided paracrine FGF(s)", "paracrine FGF(s) as provided herein", "paracrine FGF(s) for use in treating a disease or disorder of energy homeostasis" and the like also relate to the polynucleotide for use in treating a disease or disorder of energy homeostasis, wherein the polynucleotide encodes the polypeptide described above. Herein, this polynucleotide is also termed "polynucleotide(s) of the (present) invention".

The terms "polypeptide(s) of the (present) invention" or "polynucleotide(s) of the (present) invention" relate to paracrine FGFs, in particular FGF8b, FGF8f and/or FGF17, for use in treating a disease or disorder of energy homeostasis (such as obesity). The term "polypeptide(s) of the (present) invention" or "polynucleotide(s) of the (present) invention" also includes (a) functional fragment(s) of the herein described paracrine FGFs (such as functional fragments of FGF8b, FGF8f or FGF17).

As mentioned above, the invention provides for a polypeptide for use in accordance with the present invention, particularly for use in treating diseases or disorders of energy homeostasis (e.g. obesity). This polypeptide may have a length of 10-1000 amino acids, preferably of 30-800 amino acids, 50-700 amino acids or 70-500 amino acids; more preferably, of 90-300 amino acids, 110-280 amino acids, 150-200 amino acids or 170-270 amino acids; or, most preferably, of 200-230 amino acids. The term

"polypeptide" also encompasses fragments and variants of the specific polypeptides provided herein that have the biological function/activity of the herein described polypeptides. Also the use of short peptides consisting of about 11 or less amino acids is envisaged. These peptides can consist of 10, 9, 8, 7, 6, 5 or 4 amino acids. Also these peptides are polypeptides for use in accordance with the present invention. In one particular aspect of the invention, the polypeptide of the invention has a length of 215 amino acids.

It is preferred that the polypeptides for use in accordance with the present invention, particularly for use in treating diseases or disorders of energy homeostasis (e.g. obesity) comprise the amino acid sequence VTVQSSPNFTQ or related sequences like QVTVQSSPNFTQ or QVTVQSSPNFT, preferably of from position 26 to position 37 of the respective amino acid sequence, or fragments thereof that contribute to mediating the biological effect of the polypeptide.

It is preferred that the polypeptides to be used herein comprise the amino acid sequence SPNF, preferably of from position 29 to position 32 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 33 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). And both FGF8b and FGF17 share the amino acid residue Q at position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or at position 35 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). Thus, they share the motif "SPNFXQ". Therefore, it is preferred that the polypeptides to be used herein comprise the amino acid sequence SPNFXQ, preferably of from position 29 to position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 35 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). X may be T or N.

The illustrative appended examples demonstrate that the paracrine FGFs of the invention have the ability to induce differentiation (or conversion) of white (visceral)

adipocytes (and/or preadipocytes) to brown adipocytes also without their signal sequence. Such a protein is, e.g., a polypeptide having the amino acid sequence consisting of the amino acid residues 23-215 of SEQ ID NO: 16. Thus, a functional fragment of the herein provided paracrine FGF(s) include the respective FGF without its signal sequence. However, also other functional fragments of the herein provided paracrine FGF(s) are encompassed by the present invention.

Thus, the invention relates to a polypeptide for use in treating a disease or disorder of energy homeostasis, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16, or a functional fragment thereof, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, or a functional fragment thereof; wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, or a functional fragment thereof; wherein the function comprises the ability to induce

differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As mentioned, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The illustrative appended examples show that the paracrine FGFs of the present invention are active in browning visceral adipose tissue. Moreover, the appended examples show that of 13 fibroblast growth factors, FGF8 strongest induced expression of (the brown adipocyte specific gene) Ucp1 in white adipose cell lines. In these experiments FGF8b was shown to have browning potential in both visceral and subcutaneous adipose tissue. In contrast, FGF8f specifically induces browning of visceral adipose tissue. Therefore, one aspect of the invention relates to FGF8f for use in treating diseases or disorders of energy homeostasis such as (central) obesity.

Thus, one aspect of the invention relates to a polypeptide for use in treating a disease or disorder of energy homeostasis, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 4 or the nucleic acid sequence comprising nucleic acid residues 67-735 in SEQ ID NO: 4;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 18 or an amino acid sequence comprising amino acids 23-244 in SEQ ID NO: 18;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-244 in SEQ ID NO: 18 or having an amino acid sequence as depicted in SEQ ID NO: 18;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

The illustrative appended examples also demonstrate that FGF17 is active in browning visceral adipose tissue. Accordingly, the invention also relates to FGF17 for use in treating a disease or disorder of energy homeostasis.

Thus, one aspect of the invention relates to a polypeptide for use in treating a disease or disorder of energy homeostasis, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 9 or the nucleic acid sequence comprising nucleic acid residues 67-651 in SEQ ID NO: 9;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 23 or an amino acid sequence comprising amino acids 23-216 in SEQ ID NO: 23;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-216 in SEQ ID NO: 23 or having an amino acid sequence as depicted in SEQ ID NO: 23;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand

of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As indicated above, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

Thus, the polypeptide of the invention may be FGF8b, FGF8f or FGF17. Preferably, polypeptide of the invention is FGF8b or FGF8f. More preferably, the polypeptide of the invention is FGF8b.

As used herein, a "functional fragment" of a protein which displays a specific biological activity relates to a fragment of said protein having a sufficient length to display said activity. Accordingly, a functional fragment of a protein showing a specific (e.g. signalling) activity may relate to a polypeptide which corresponds to a fragment of said protein which is still capable of showing said (signalling) activity. For example, a functional fragment of a paracrine FGF as provided herein (e.g. FGF8b) may correspond to the fragment of the paracrine FGF (e.g. of FGF8b) which has the same signalling activity as the paracrine FGF (e.g. as FGF8b). Methods for determining whether a certain fragment of a protein is a functional fragment are known in the art. For example, a test for determining whether a fragment of a paracrine FGF as provided herein (e.g. FGF8b) is functional, (i.e., is still capable of inducing differentiation or conversion of white visceral adipocytes and/or

preadipocytes to brown adipocytes), is for example, treating cultured white adipocytes or preadipocytes with the fragment to be tested and measuring the expression of marker genes of brown adipose tissues (e.g. Ucp1) after differentiation. This method is used herein in the appended examples. Preferably, a functional fragment of a paracrine FGF of the present invention has substantially the same biological activity as the paracrine FGF of the present invention itself. Preferably, the functional fragment is at least 50% more preferably at least 60%, 70%, 80%, 85%, 90%, 95% or 99% of the amino acid sequence of the full length sequences of the paracrine FGF of the invention (e.g. FGF8b).

Without deferring from the gist of the present invention also (a) functional fragment(s) or (a) functional derivative(s) of the herein provided polypeptides or proteins can be used, for example, (functional) fragment(s) or (functional) derivative(s) of the polypeptides as shown in SEQ ID NO. 16, 18 or 23.

Thus, a functional fragment of the above polypeptide(s)/protein(s) provided herein and to be used in accordance with the present invention can be any of the above specific polypeptides as shown in any one of SEQ ID NOs: SEQ ID NO. 16, 18 or 23, respectively, wherein one or more amino acids are deleted.

A (functional) derivative(s) of the above polypeptide(s)/protein(s) provided herein and to be used in accordance with the present invention can be any of the above specific polypeptides as shown in SEQ ID NOs: SEQ ID NO. 16, 18 or 23, respectively, wherein one or more amino acids are inserted, added or substituted.

1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 amino acids can, for example, be deleted, inserted, added or substituted within the amino acid sequence of the polypeptides as shown SEQ ID NOs: 16, 18 or 23.

The term “one or more amino acids deleted” relates to functional fragments of the specific paracrine FGF polypeptides provided herein.

A preferred (functional) fragment of the above mentioned polypeptides provided

herein and to be used in accordance with the present invention consists of from 4 to 15 contiguous amino acids. Accordingly, a (functional) fragment of the above mentioned polypeptides provided herein and to be used in accordance with the present invention preferably consists of 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15, contiguous amino acids.

A (functional) fragment of the above mentioned polypeptides provided herein and to be used in accordance with the present invention preferably consists of from 15 to 25 contiguous amino acids of the amino acid sequence shown in of the polypeptides as shown SEQ ID NOs: 16, 18 or 23.

The fragment or derivative preferably has the same (or essentially the same) biological activity as the full length polypeptide from which it is derived, the full length polypeptide having the amino acid sequence as shown in SEQ ID NO: 16, 18 or 23. In this sense, the fragment or derivative is a "functional" fragment or derivative to be used herein.

The herein provided polypeptide (as shown, for example, in SEQ ID NO: 16, 18 or 23, respectively) may have one or more amino acids deleted, inserted, added and/or substituted provided that the polypeptide maintains essentially the biological activity which is characteristic of the polypeptides from which it is derived.

Preferably, any such deletions, insertions, additions and/or substitutions (in this context particularly substitutions) are conservative, i.e. amino acids are substituted by amino acids having the same or similar characteristics. For example, a hydrophobic amino acid will preferably be substituted by another hydrophobic amino acid and so on.

Preferred fragments to be used herein are QVTVQSSPNFT, QVTVQSSPNFTQ, VTVQSSPNFTQ, SPNF, SPNFXQ (wherein X may be T or N), or functional fragments thereof.

In certain aspects, the present invention relates to the following items:

1. A paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis.

2. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8b.

3. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 1 or 2, wherein the paracrine fibroblast growth factor (FGF) is human FGF8b.

4. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 3, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;
   (c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;
   (d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);
   (e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);
   (f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

5. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8f.

6. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1 or 5, wherein the paracrine fibroblast growth factor (FGF) is human FGF8f.

7. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 and 6, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 4 or the nucleic acid sequence comprising nucleic acid residues 67-735 in SEQ ID NO: 4;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 18 or an amino acid sequence comprising amino acids 23-244 in SEQ ID NO: 18;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-244 in SEQ ID NO: 18 or having an amino acid sequence as depicted in SEQ ID NO: 18;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

8. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF17.

9. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1 or 8, wherein the paracrine fibroblast growth factor (FGF) is human FGF17.

10. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 8 and 9, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 9 or the nucleic acid sequence comprising nucleic acid residues 67-651 in SEQ ID NO: 9;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 23 or an amino acid sequence comprising amino acids 23-216 in SEQ ID NO: 23;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-216 in SEQ ID NO: 23 or having an amino acid sequence as depicted in SEQ ID NO: 23;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

11. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d).

12. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 90% identity to the polypeptide of any one of (a) to (d).

13. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 95% identity to the polypeptide of any one of (a) to (d).

14. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 99% identity to the polypeptide of any one of (a) to (d).

15. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to

induce differentiation or conversion of white adipocytes and/or white preadipocytes to brown adipocytes.

16. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to induce differentiation or conversion of white visceral adipocytes and/or white visceral preadipocytes to brown adipocytes.

17. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence VTVQSSPNFTQ or a fragment thereof.

18. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence VTVQSSPNFTQ from position 24 to position 34 of the amino acid sequence of the polypeptide.

19. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 18, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

20. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence VTVQSSPNFTQ from position 53 to position 63 of the amino acid sequence of the polypeptide.

21. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 20, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

22. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFTQ or a fragment thereof.

23. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 23 to position 34 of the amino acid sequence of the polypeptide.

24. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 23, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

25. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 52 to position 63 of the amino acid sequence of the polypeptide.

26. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 25, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

27. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFT or a fragment thereof.

28. The paracrine fibroblast growth factor (FGF) for use in treating a disease or

disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 23 to position 33 of the amino acid sequence of the polypeptide.

29. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 28, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

30. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 52 to position 62 of the amino acid sequence of the polypeptide.

31. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 30, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

32. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNFXQ or a fragment thereof.

33. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 29 to position 34 of the amino acid sequence of the polypeptide.

34. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 33, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

35. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 58 to position 63 of the amino acid sequence of the polypeptide.

36. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 35, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

37. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 30 to position 35 of the amino acid sequence of the polypeptide.

38. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 37, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

39. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 32 to 38, wherein residue X in the amino acid sequence SPNFXQ is T or N.

40. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNF or a fragment thereof.

41. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 29 to position 32 of the amino acid sequence of the polypeptide.

42. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 41, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

43. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 58 to position 61 of the amino acid sequence of the polypeptide.

44. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 43, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

45. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 30 to position 33 of the amino acid sequence of the polypeptide.

46. The paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 45, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

47. A polynucleotide for use in treating a disease or disorder of energy homeostasis, wherein the polynucleotide encodes the paracrine fibroblast growth factor (FGF) of any one of items 1 to 46.

48. A pharmaceutical composition comprising the paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and/or the polynucleotide of item 47 for use in treating a disease or disorder of energy homeostasis, further comprising a pharmaceutically acceptable carrier and/or diluent.

49. A method of treating a disease or disorder of energy homeostasis by administering an effective dose of the paracrine fibroblast growth factor (FGF) of any one of items 1 to 46, the polynucleotide of item 47, or the pharmaceutical composition of item 48, to a subject in need of such treatment.

50. The method of claim 49, wherein the subject is a human patient.

51. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46, the polynucleotide of item 47, or the pharmaceutical composition of item 48, wherein a human patient suffering from a disease or disorder of energy homeostasis or being prone to suffering from a disease or disorder of energy homeostasis is to be treated.

52. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51, the polynucleotide of item 47 or 51, the pharmaceutical composition of item 48 or 51, or the method of item 49 or 50, wherein said paracrine fibroblast growth factor (FGF) binds to an FGF receptor or is capable of binding to an FGF receptor.

53. The paracrine fibroblast growth factor (FGF) of item 52, the polynucleotide of item 52, the pharmaceutical composition of item 52, or the method of item 52, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3.

54. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 53, the polynucleotide of any one of items 47 and 51 to 53, the pharmaceutical composition of any one of items 48 and 51 to 53, or the method of any one of item 49 to 53, wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is to be administrated locally.

55. The paracrine fibroblast growth factor (FGF) of item 54, the polynucleotide of item 54, the pharmaceutical composition of item 54, or the method of item 54,

wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is to be administrated locally into the visceral adipose tissue.

56. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 55, the polynucleotide of any one of items 47 and 51 to 55, the pharmaceutical composition of any one of items 48 and 51 to 55, or the method of any one of items 49 to 55, wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is to be administered into the visceral adipose tissue.

57. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 56, the polynucleotide of any one of items 47 and 51 to 56, the pharmaceutical composition of any one of items 48 and 51 to 56, or the method of any one of items 49 to 56, wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is in the form of an erodible implant, an implantable drug release device, a gel for injection or a solution for injection.

58. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 57, the polynucleotide of any one of items 48 and 51 to 57, the pharmaceutical composition of any one of items 48 and 51 to 57, or the method of any one of items 49 to 57, wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is to be administered via a minipump.

59. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is at least one disease or disorder selected from the group consisting of obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

60. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is obesity.

61. The paracrine fibroblast growth factor (FGF) of item 60, the polynucleotide of item 60, the pharmaceutical composition of item 60, or the method of item 60, wherein said obesity is central obesity.

62. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is dyslipidemia.

63. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is diabetes.

64. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is insulin resistance.

65. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is hyperglycemia.

66. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 58, the polynucleotide of any one of items 48 and 51 to 58, the pharmaceutical composition of any one of items 49 and 51 to 58, or the method of any one items 49 to 58, wherein said disease or disorder of energy homeostasis is metabolic syndrome.

67. The paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 51 to 66, the polynucleotide of any one of items 47 and 51 to 66, the pharmaceutical composition of any one of items 48 and 51 to 66, or the method of any one of items 49 and 51 to 66, wherein said paracrine fibroblast growth factor (FGF), polynucleotide or pharmaceutical composition is co-administered with at least one other active agent.

68. The paracrine fibroblast growth factor (FGF) of item 67, the polynucleotide of item 67, the pharmaceutical composition of item 67, or the method of item 67, wherein said other active agent is at least one active agent selected from the group consisting of beta-adrenergic agonists (e.g. noradrenalin, isoproterenol, BRL 35135, ICI D7114, CGP-12177A, CL 316243), indirect sympathomimetics (e.g. ephedrine, methylphenidate), atrial natriuretic peptide (e.g. ANP, BNP) and ANP/BNP receptor agonists (e.g. AP-811).

69. The paracrine fibroblast growth factor (FGF) of item 68, the polynucleotide of item 68, the pharmaceutical composition of item 68, or the method of item 68, wherein said beta-adrenergic agonist is a beta3-adrenergic agonist.

70. The paracrine fibroblast growth factor (FGF) of item 69, the polynucleotide of item 69, the pharmaceutical composition of item 69, or the method of item 69, wherein said beta3-adrenergic agonist is CL 316243.

71. A method for the preparation of a pharmaceutical composition for use in treating a disease or disorder of energy homeostasis, wherein the method comprises the following steps:

(a) contacting the paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and/or the polynucleotide of item 47 with a liquid carrier or a solid carrier;

(b) optionally, adjusting the pH and/or the osmolarity of the product obtained in step (a);

(c) optionally, sterilizing the product obtained in step (a) or (b); and

(d) formulating and/or packaging the product obtained in step (a), (b) or (c) as a finished medical product.

72. The method of item 71, wherein said carrier is at least one carrier selected from the group consisting of cellulose, lactose, water, saline, Ringer's solution, dextrose solution, a fixed oil, ethyl oleate and liposomes.

In the present invention and in the appended examples, a link between paracrine FGFs (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) and brown adipose tissue (BAT) has been established. Because the art acknowledges the link between BAT and the treatment of diseases of energy homeostasis (including obesity, diabetes etc.), it is credible and plausible that paracrine FGFs (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) can be used in the therapy of these diseases or disorders. In the same vein, it is evident that the activation of paracrine FGFs will have the same beneficial therapeutic effect in the therapy of these diseases or disorders in subjects in need of such a therapy or treatment. Said subject may be a human subject.

Several mechanisms and pathways exist that can be used to activate paracrine FGFs (including FGF8b, FGF8f and/or FGF17, preferably FGF8b) in accordance with the present invention. In the following exemplary activators of paracrine FGFs are described which can be used in accordance with the invention.

The following table shows activators of FGF signalling:

Pathway	**Activators**
FGF receptors	Strontium ranelate (Caverzasio & Thouverey)

MAPK	Anisomycin (p38 MAP and JNK), PAR C-16 (MEK), t-butylhydroquinone (Erk2)
IP3	Phorbol 12-myristate 13-acetate (PKC), Cell permeant caged IP3
PI3K	740 Y-P (IP3R), sc-3036 (IP3R)

Further activators of paracrine FGFs are shown in the following table:

	substance	IUPAC International Chemical Identifier (InChI), reference or chemical nomenclature/trivial name
Mechanism: Influencing FGF-heparin-binding		
Activators	Sucrose octasulfate	WEPNHBQBLCNOBB-FZJVNAOYSA-N
	Inositol hexasulfate	NBTMNFYXJYCQHQ-UHFFFAOYSA-N
Mechanism: Supply of heparin for activating the FGFR-FGF-Heparin-complex		
activating	administration of heparin (or of a heparin derivative)	Classical heparin; further heparin derivavites, such as certoparin, dalteparin, enoxaparin, nadroparin, danaparoid
Mechanism: Influencing stability of protein conformation		
Stabilizing agent	Alpha-Cyclodextrin and other Cyclodextrinderivatives	HFHDHCJBZVLPGP-RWMJIURBSA-N
Mechanism: Influencing heparanase-mediated degradation of heparan-sulfate proteoglycan (HSPG) of the extracellular matrix		
Heparanase (endo-beta-D-glucuronidase heparanase) inhibitor	PI-88 is a mixture of highly sulfated, monophosphorylated mannose oligosaccharides; Name: MUPARFOSTAT	
Heparanase inhibitor	OGT 2115	2-[4-[[3-(4-Bromophenyl)-1-oxo-2-propenyl]amino]-3-fluorophenyl]-5-benzoxazoleacetic acid

Mechanism: Influencing FGFR activity		
FGFR1		
Activator:	SUN11602	4-[[4-[[2-[(4-Amino-2,3,5,6-tetramethylphenyl)amino]acetyl]methylamino]-1-piperidinyl]methyl]benzamide
FGFR3		
Activator:	Botulinum neurotoxin serotype A (BoNT/A)	
FGFR4		
Activator:	Monoclonal Antibody: 4FA6D3C10	US 2009/0123462 A1

Also heparin sulfate can be used as activator.

In one aspect, the present invention relates to an activator of a paracrine FGF including FGF8b, FGF8f or FGF17 as defined herein for use in treating a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome. In a preferred aspect, the present invention relates to an activator of FGF8b as defined herein for use in treating a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome.

The term "FGF8b" as used herein can refer to a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of

a nucleic acid molecule as defined in (a) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(d) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (c), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white visceral adipocytes and/or preadipocytes to brown adipocytes; and

(e) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), and (c).

The term “FGF8b” also refers to fragments of the polypeptide.

The term “agonist of a paracrine FGF” or “activator of a paracrine FGF” or “enhancer of a paracrine FGF” means in context of the present invention a compound capable of fully or partially stimulating or increasing the physiologic activity and/or expression level of (a) a paracrine FGF. The terms "activator" or “enhancer” are used interchangeably herein.

The term “agonist” can refer to a chemical compound/substance that binds to a receptor and activates the receptor to produce a biological response. Thus, an “agonist of a paracrine FGF” can be a chemical compound/substance that binds to a receptor of paracrine FGF (like FGFR1, FGFR2, FGFR3 or FGFR4) and induces the same or essential the same biological activity as the the paracrine FGF. The term “activator of a paracrine FGF” can encompass “agonist(s) of a paracrine FGF”.

In one aspect, the present invention relates to an agonist of a paracrine FGF including FGF8b, FGF8f or FGF17 as defined herein for use in treating a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome. In a preferred aspect, the present invention relates to an agonist of FGF8b as defined herein for use in treating a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome.

The activator/agonist of paracrine FGF can be administered, for example, locally or or orally. Local administration encompasses injection or implantation e.g. by using an implantable drug depot. As used herein, the term "agonist" or "activator" also encompasses partial agonists or co-agonists/co-activators. In addition thereto, an "activator" of paracrine FGF in the context of the present invention may also be capable of stimulating the function of the paracrine FGF by inducing/enhancing the expression of the nucleic acid molecule encoding for said paracrine FGF. Thus, an activator of a paracrine FGF may lead to an increased expression level of the paracrine FGF (e.g. increased level of paracrine FGF mRNA, paracrine FGF protein); this may be reflected in an increased paracrine FGF activity. In addition thereto, an "activator" of paracrine FGF in the context of the present invention may also be an inhibitor of proteolysis of the paracrine FGF and thus enhancing paracrine FGF function by increasing amounts of effective material. Furthermore, “activators” of paracrine FGF in the context of the invention may be capable of stabilizing paracrine FGF e.g. capable of preventing degradation of paracrine FGF. This increased activity can be measured/detected by the herein described methods.

An activator of the paracrine FGF in the context of the present invention may also encompass transcriptional activators of paracrine FGF expression that are capable of enhancing paracrine FGF function. The term "activator" comprises partial activators. As partial activator the art defines candidate molecules that behave like activator, but that, even at high concentrations, cannot activate a paracrine FGF to the same extent as a full activator. Furthermore, the activator of a paracrine FGF may have an effect on interactions of the paracrine FGF protein(s) with other proteins (thus, for example, having an effect on the activity of complexes involving paracrine FGF protein(s)) or, in general, with its synthesis, e.g. by having an effect on upstream steps of paracrine FGF expression or with signalling pathways in which the paracrine FGF is involved. Depending on the mode of action, such activator may, for example, be denoted “sequestering activator” or “signalling activator”.

Hence, the use of potent activators of a paracrine FGF will lead to an increase of paracrine FGF expression level and/or activity, and thereby increase differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes. In accordance

with the above definition of "activator" also a paracrine FGF itself can be considered as its own activator. For example, overexpression of a paracrine FGF may lead to enhanced paracrine FGF activity, thus increasing paracrine FGF function. Accordingly, it is preferred that a paracrine FGF as defined herein can be used for the treatment of a disease or disorder of energy homeostasis, particularly wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome.

It is envisaged and preferred herein that the activator of a paracrine FGF targets, preferably specifically targets, the paracrine FGF polypeptide (or the nucleic acid encoding same) itself, particularly the biologically active region thereof. The term "targeting" refers in this context to the binding to paracrine FGF polypeptide (and here in particular to the biologically active region thereof) and/or increasing the activity of paracrine FGF, in particular the increase in its capacity/ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes. The increase of the activity of paracrine FGF can also refer, for example, to the interference with/activation of the activity of paracrine FGF to act as a scaffold or as a recruiting platform for interaction partners, in particular forming a complex with its FGFR receptor and/or heparansulfate. The latter member of this FGFR signaling activator complex belongs to the group of the heparine molecules.

As explained above, 11 amino acids on exon 1D of paracrine FGFs to be used in accordance with the present invention are likely sufficient to mediate a biological effect. Therefore, a preferred biologically active region of the polypeptides for use in accordance with the present invention comprises the amino acid sequence VTVQSSPNFTQ or related sequences like QVTVQSSPNFTQ or QVTVQSSPNFT or fragments thereof that contribute to mediating the biological effect of the polypeptide. This biologically active region can be targeted by activators of paracrine FGF.

We have explained above that the phenylalanine residue 32 (F32) of this short amino acid peptide has been shown to interact with the hydrophobic groove within the Ig domain III, i.e. the "c" variants of the FGF receptors (Olsen et al. 2013). The "c" spliceforms comprise FGFR1c, FGFR2c, FGFR3c, and FGFR4. Strikingly, FGF17,

an endocrine FGF, is also active to induce UCP1 mRNA gene expression (Figure 1A) and also shares F32. Thus, it is preferred that the biologically active region that can be targeted by activators of paracrine FGF comprising the amino acid sequence SPNF, preferably of from position 29 to position 32 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 33 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). And both FGF8b and FGF17 share the amino acid residue Q at position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or at position 35 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18) Thus, they share the motif "SPNFXQ". Therefore, it is preferred that the biologically active region that can be targeted by activators of paracrine FGF comprises the amino acid sequence SPNFXQ, preferably of from position 29 to position 34 of the respective amino acid sequence of FGF8b (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 16) or of from position 30 to position 35 of the respective amino acid sequence of FGF17 (exemplary amino acid sequence of FGF8b is shown in SEQ ID NO. 18). X may be T or N..

Further, the biologically active region that can be targeted by activators of paracrine FGF can comprise the heparin binding domains of the FGF8 subfamily. The heparin binding domains are of importance because FGF8-subfamily members bind to heparin molecules and are therefore with regard to the extracellular space tightly bound to heparansulfate proteoglycans, and subsequently, trapped in the extracellular matrix. This aspect is advantageous in context of the present invention, because this local entrapment of active peptides prevents potential side effects to have systemic consequences.

More than 100 heparin binding proteins have been identified, and the ability to bind heparin itself has, up to now, not been attributed to a specific sequence. Fromm et al. have reported that heparin binding sites frequently contain clusters of basic amino acids, like XBX, XBBX, XBBBX (Fromm Arch Biochem Biophys 1997).

Moreover, activators/agonists of paracrine FGF to be used herein can target FGF receptors. As shown in Figure 8C FGF8b-mediated responsiveness correlated with Fgf receptor 4 mRNA abundance in immortalized inguinal as well as epididymal adipocytes. Data as provided herein show a similar correlation between the FGF8b-mediated responsiveness and other "c" variants of the FGF receptors. Ig domain III "c" variants of the FGF receptor comprise exon 7, 9, and the transmembrane domain of the FGF receptors (Figure 9). It is contemplated herein that the observed effect of FGF8b-mediated browning of white adipocytes is a FGF8-subfamily group effect based on interaction of FGF8-subfamily members with the "c" splice variants of the FGFR 1-4. Thus, strengthening this interaction by activators/agonists of paracrine FGF is contemplated herein.

The agonist(s)/activator may be (a) small molecule drug(s), (a) (small) binding molecule, a peptidomemetic, and/or a poly-/monoclonal antibody.

Agonists/activators to be used herein can be (a) small molecule drug(s). The terms "small molecule drug" and "small molecule compound" are used interchangeably herein. (A) small molecule drug(s) to be used herein as agonist/ activators of paracrine FGF can refer to an (organic) low molecular weight (<900 Daltons) compound. Small molecules can help to regulate a biological process and have usually a size in the order of 10^{-9} m. Agonists/activators to be used herein, like small molecules (drugs), can, for example, be identified by screening compound libraries, for example Enamine, Chembridge or Prestwick chemical libraries. Exemplary small molecule drugs to be used herein are provide in the Tables above.

For example, an agonist/activator of paracrine FGF can be a binding molecule(s), such as be (an) aptamer(s) and/or (an) intramer(s).

It is also envisaged in the present invention that peptides, particularly cyclic peptides can be used as agonists/activators of paracrine FGF. Cyclic peptides are polypeptide chains, wherein the amino termini and carboxyl termini, amino termini and side chain, carboxyl termini and side chain, or side chain and side chain are linked with a covalent bond that generates the ring. It is also envisaged herein that biological selection technology, such as phage display is used in order to select peptide ligands

tethered to synthetic molecular structures. These peptide ligands show specificity to target paracrine FGF. In certain aspects of the invention, monomeric monocyclic peptide agonists/activators and dimeric bicyclic peptide agonists/activators of paracrine FGF are used.

The agonist/activators is preferably a selective agonist of paracrine FGF.

Selectivity expresses the biologic fact that at a given compound concentration enzymes (or proteins) are affected to different degrees. In the case of enzymes selective activation can be defined as preferred activation by a compound at a given concentration. Or in other words, an enzyme (or protein) is selectively activated over another enzyme (or protein) when there is a concentration, which results in activation of the first enzyme (or protein) whereas the second enzyme (or protein) is not, or not substantially, affected. To compare compound effects on different enzymes it is crucial to employ similar assay formats, such as the FRET assay, Plus assay, HMT assays, thermoshift assays, biological readouts (of reporter proteins/enzymes, such as Cxcl1/CXCL8), or chemical proteomics.For example, commercially available test kits, like the commercial ELISA kit can be employed (Mouse CXCL1/KC Quantikine ELISA Kit (MKC00B) from R&D Systems.

The agonists/activators to be used herein are preferably specific for paracrine FGF, i.e. the compounds specifically activate paracrine FGF. In other words, the paracrine FGF agonists/activators are preferably selective paracrine FGF agonists/activators.

The term “selective paracrine FGF activator(s)” as used herein refers to (a) paracrine FGF activator(s) as defined herein (in particular (a) small molecule drug(s)) that activates or display(s) increased activity towards paracrine FGF without displaying substantial activity towards another protein or enzyme, in particular another FGF (like FGF1, FGF2, FGF3, FGF4, FGF5, FGF6, FGF7, FGF8a, FGF8d, FGF9, FGF10, FGF11, FGF12, FGF 13, FGF 14, FGF15, FGF16 or FGF18, FGF19, or FGF21) as defined herein above.

Accordingly, a paracrine FGF activator that is selective for a paracrine FGF exhibits a paracrine FGF selectivity of greater than about 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-

fold, 8-fold, 9-fold, 10-fold, 20-fold, 50-fold or greater than about 100-fold with respect to activation of another protein or enzyme (in particular another FGF as defined above).

For example, pan-FGF activators (i.e. compounds that broadly activate substantially any FGF) are not considered herein as selective paracrine FGF activators.

Binding molecules are also envisaged herein as agonists/activators of paracrine FGF. It is envisaged herein that the binding molecule activating paracrine FGF specifically binds to paracrine FGF as defined herein. It is envisaged herein that the aptamers/intramers can specifically target/bind to (functional) fragments or (functional) derivatives of the paracrine FGF proteins as defined herein, for example also to polypeptides having at least 40% or more identity to herein provided paracrine FGF protein(s). Accordingly, the present invention relates to the use of these aptamers/intramers in particular in the therapeutic methods of the present invention.

Activators for use in accordance with the present invention are described and provided herein. Also the use of agonists/activators yet to be generated or known compounds to be tested for their agonizing/activating activity is envisaged in context of the present invention.

Therefore, the present invention provides a method for assessing the activity of a candidate molecule suspected of being an activator of a paracrine FGF as defined and provided herein comprising the steps of:

a) contacting a cell, tissue or a non-human animal comprising a paracrine FGF with said candidate molecule;
b) detecting an increase in activity of said paracrine FGF; and
c) selecting a candidate molecule that increases activity of said paracrine FGF.

An increase of the paracrine FGF receptor activity can indicate the capacity of the selected molecule to activate paracrine FGF.

In a certain aspect, the present invention provides a method for assessing the activity of a candidate molecule suspected of being an agonist of a paracrine FGF as defined and provided herein comprising the steps of:

a) contacting a cell, tissue or a non-human animal comprising a paracrine FGF receptor with said candidate molecule;
b) detecting an increase in activity of said paracrine FGF receptor; and
c) selecting a candidate molecule that increases activity of said paracrine FGF receptor.

An increase of the paracrine FGF receptor activity can indicate the capacity of the selected molecule to agonize a paracrine FGF.

The activity of paracrine FGF can, in particular, be reflected in the increase in the capacity/ability of paracrine FGF to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes. The activity of paracrine FGF can also refer, for example, to the interference with/activation of the activity of paracrine FGF to act as a scaffold or as a recruiting platform for interaction partners, in particular forming a complex with its FGFR receptor and/or heparine .

Such assays are provided herein and described in more detail further below.

Also an increase in the (expression) level can indicate useful agonists of a paracrine FGF. Accordingly, the term “activity” above can comprise and relate to the “expression level” and vice versa.

The present invention relates to a method for assessing the (expression) level of a candidate molecule suspected of being an agonist of a paracrine FGF as defined and provided herein comprising the steps of:

a) contacting a cell, tissue or a non-human animal comprising a paracrine FGF with said candidate molecule;
b) detecting an increase in the (expression) level of said paracrine FGF; and
c) selecting a candidate molecule that increases the (expression) level of said paracrine FGF.

An increase of the paracrine FGF (expression) level can indicate the capacity of the selected molecule to activate paracrine FGF.

It is understood that the detected activity or expression level of a paracrine FGF is compared to a standard or reference value of a paracrine FGF activity. The standard/reference value may be detected in a cell, tissue, or non-human animal as defined herein, which has not been contacted with a potential paracrine FGF agonist/activator or prior to the above contacting step. The increase in the activity or expression level of the paracrine FGF upon contacting with (a) candidate molecule(s) may also be compared to the increase in paracrine FGF activity induced by (a) routinely used reference compound(s). A skilled person is easily in the position to determine/assess whether the activity and/or expression of a paracrine FGF is increased.

In accordance with this invention, in particular the screening or identifying methods described herein, a cell, tissue or non-human animal to be contacted with a candidate molecule comprises paracrine FGF as defined herein. For example said cell, tissue or non-human animal may express a paracrine FGF gene, in particular also (an) additional (copy) copies of a paracrine FGF gene, (a) paracrine FGF mutated gene(s), a recombinant paracrine FGF gene construct and the like. As explained herein, the capability of a candidate molecule to activate/agonize paracrine FGF may, accordingly, be detected by measuring the expression level of such gene products of paracrine FGF or of corresponding gene constructs (e.g. mRNA or protein), wherein a high expression level (compared to a standard or reference value) is indicative for the capability of the candidate molecule to act as activator/agonist.

The term “comprising paracrine FGF” may, for example, relate to to the paracrine FGF gene(s) or proteins known in the art and described herein, but also to a reporter construct which comprises the paracrine FGF (or a functional fragment thereof) and a “reporter”. Exemplary reporters (reporter gene products), which can be used in the screening methods of the invention are luciferase, (green/red) fluorescent protein and variants thereof, EGFP (enhanced green fluorescent protein), RFP (red fluorescent protein, like DsRed or DsRed2), CFP (cyan fluorescent protein), BFP (blue green

fluorescent protein), YFP (yellow fluorescent protein), β-galactosidase or chloramphenicol acetyltransferase. The skilled person is readily in the position to generate and use also other reporters/reporter constructs, which can be employed in accordance with the present invention. The use of fusion proteins containing a paracrine FGF protein (or a functional fragment thereof) and a reporter gene product is also envisaged in the methods of the present invention.

All definitions and explanations provided herein above, inter alia, in relation to "paracrine FGF" (and related compounds), "agonist", "activity" and the like, apply mutatis mutandis in the context of these methods for assessing the activity (or (expression) level) of a candidate molecule suspected of being an activator/agonist of a paracrine FGF.

The following exemplary assays can be used in the determination that a candidate molecule is indeed an agonist/activator of a paracrine FGF to be used in accordance with the present invention:
A candidate molecule can be applied into a culture of proliferating or differentiating white preadipocytes, e.g. immortalized murine white preadipocytes. After full differentiation a quantification of UCP1 mRNA, e.g. by qPCR, is able to demonstrate, whether the candidate molecule led to a recruitment of brown adipocytes, as FGF8b does in the experiments we describe herein.

Furthermore, a candidate molecule can be tested in vivo by introducing it into a white adipose tissue depot, e.g. the epididymal adipose tissue depot, of mice. The candidate molecule may be applied directly or embedded in a releasing matrix or device. After several days or weeks mice are sacrificed, the tissue surrounding the site of application excised and analyzed for expression of UCP1 mRNA, e.g. by qPCR, and/or analyzed histologically for the presence of multilocular cells. These parameters will demonstrate, whether a candidate molecule led to recruitment of brown adipocytes in a similar fashion as FGF8b does in the experiments we describe herein.

Exemplary methods are described in detail in the appended examples.

Antibodies, in particular monoclonal antibodies, that specifically bind to paracrine FGF as defined herein can be used in the herein provided screening assays in order to detect the expression level of a paracrine. For example, such antibodies can be used in techniques like global ChIP-seq, imaging/co-localisations, immunoprecipitation to find new interaction partners by mass-spec, and the like. Such antibodies are valuable research tools.

In certain aspects, the present invention relates to the following items:

1. An activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis.

2. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8b.

3. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 1 or 2, wherein the paracrine fibroblast growth factor (FGF) is human FGF8b.

4. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 3, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;
   (c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as

depicted in SEQ ID NO: 16;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

5. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8f.

6. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1 or 5, wherein the paracrine fibroblast growth factor (FGF) is human FGF8f.

7. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 and 6, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 4 or the nucleic acid sequence comprising nucleic acid residues 67-735 in SEQ ID NO: 4;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 18 or an amino acid sequence comprising amino acids 23-244 in SEQ ID NO: 18;

(c) a polypeptide encoded by a nucleic acid molecule encoding a

polypeptide comprising an amino acid sequence having amino acids 23-244 in SEQ ID NO: 18 or having an amino acid sequence as depicted in SEQ ID NO: 18;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

8. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF17.

9. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to claim 1 or 8, wherein the paracrine fibroblast growth factor (FGF) is human FGF17.

10. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 8 and 9, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 9 or the nucleic acid sequence comprising nucleic acid residues 67-651 in SEQ ID NO: 9;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 23 or an amino acid sequence comprising amino acids 23-216

in SEQ ID NO: 23;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-216 in SEQ ID NO: 23 or having an amino acid sequence as depicted in SEQ ID NO: 23;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

11. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d).

12. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 90% identity to the polypeptide of any one of (a) to (d).

13. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 95% identity to the polypeptide of any one of (a) to (d).

14. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 99% identity to the polypeptide of any one of (a) to (d).

15. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to induce differentiation or conversion of white adipocytes and/or white preadipocytes to brown adipocytes.

16. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to induce differentiation or conversion of white visceral adipocytes and/or white visceral preadipocytes to brown adipocytes.

17. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence VTVQSSPNFTQ or a fragment thereof.

18. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence VTVQSSPNFTQ from position 24 to position 34 of the amino acid sequence of the polypeptide.

19. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 18, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

20. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence VTVQSSPNFTQ from position 53 to position 63 of the amino acid sequence of the polypeptide.

21. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 20, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

22. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFTQ or a fragment thereof.

23. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 23 to position 34 of the amino acid sequence of the polypeptide.

24. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 23, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

25. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 52 to position 63 of the amino acid sequence of the polypeptide.

26. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 25, wherein the

amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

27. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFT or a fragment thereof.

28. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 23 to position 33 of the amino acid sequence of the polypeptide.

29. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 28, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

30. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 52 to position 62 of the amino acid sequence of the polypeptide.

31. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 30, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

32. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNFXQ or a fragment thereof.

33. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 29 to position 34 of the amino acid sequence of the polypeptide.

34. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 33, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

35. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 58 to position 63 of the amino acid sequence of the polypeptide.

36. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 35, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

37. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 30 to position 35 of the amino acid sequence of the polypeptide.

38. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 37, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

39. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 32 to 38, wherein residue X in the amino acid sequence SPNFXQ is T or N.

40. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNF or a fragment thereof.

41. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 29 to position 32 of the amino acid sequence of the polypeptide.

42. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 41, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

43. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 58 to position 61 of the amino acid sequence of the polypeptide.

44. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 43, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

45. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 30 to position 33 of the amino acid sequence of the polypeptide.

46. The activator of paracrine fibroblast growth factor (FGF) for use in treating a disease or disorder of energy homeostasis according to item 45, wherein the

amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

47. A pharmaceutical composition comprising the activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 for use in treating a disease or disorder of energy homeostasis, further comprising a pharmaceutically acceptable carrier and/or diluent.

48. A method of treating a disease or disorder of energy homeostasis by administering an effective dose of the activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46, or the pharmaceutical composition of item 47, to a subject in need of such treatment.

49. The method of claim 48, wherein the subject is a human patient.

50. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46, or the pharmaceutical composition of item 47, wherein a human patient suffering from a disease or disorder of energy homeostasis or being prone to suffering from a a disease or disorder of energy homeostasis is to be treated.

51. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50, the pharmaceutical composition of item 47 or 50, or the method of item 48 or 49, wherein said paracrine fibroblast growth factor (FGF) binds to an FGF receptor or is capable of binding to an FGF receptor.

52. The activator of paracrine fibroblast growth factor (FGF) of item 51, the pharmaceutical composition of item 51, or the method of item 51, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3.

53. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 52, the pharmaceutical composition of any one of items 47 and 50 to 52, or the method of any one of item 48 to 52, wherein said activator of paracrine fibroblast growth factor (FGF) or said pharmaceutical composition is to be administrated orally or locally.

54. The activator of paracrine fibroblast growth factor (FGF) of item 43, the pharmaceutical composition of item 53, or the method of item 53, wherein said activator of paracrine fibroblast growth factor (FGF), or said pharmaceutical composition is to be administrated locally into the visceral adipose tissue.

55. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 54, the pharmaceutical composition of any one of items 47 and 50 to 54, or the method of any one of items 48 to 54, wherein said activator of paracrine fibroblast growth factor (FGF), or said pharmaceutical composition is to be administered into the visceral adipose tissue.

56. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 55, the pharmaceutical composition of any one of items 47 and 50 to 55, or the method of any one of items 48 to 55, wherein said activator of paracrine fibroblast growth factor (FGF), or pharmaceutical composition is in the form of an erodible implant, an implantable drug release device, a gel for injection or a solution for injection.

57. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 56, the pharmaceutical composition of any one of items 47 and 50 to 56, or the method of any one of items 48 to 56, wherein said activator of paracrine fibroblast growth factor (FGF) or said pharmaceutical composition is to be administered via a minipump.

58. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is at least one disease or disorder selected from the group consisting of obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia and metabolic syndrome.

59. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and

50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is obesity.

60. The activator of paracrine fibroblast growth factor (FGF) of item 59, the pharmaceutical composition of item 59, or the method of item 59, wherein said obesity is central obesity.

61. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is dyslipidemia.

62. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is diabetes.

63. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is insulin resistance.

64. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is hyperglycemia.

65. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 57, the pharmaceutical composition of any one of items 48 and 50 to 57, or the method of any one items 48 to 57, wherein said disease or disorder of energy homeostasis is metabolic syndrome.

66. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 65, the pharmaceutical composition of any one of items 47 and

50 to 65, or the method of any one of items 48 and 50 to 65, wherein said activator of paracrine fibroblast growth factor (FGF), pharmaceutical composition is co-administered with at least one other active agent.

67. The activator of paracrine fibroblast growth factor (FGF) of item 66, the pharmaceutical composition of item 66, or the method of item 66, wherein said other active agent is at least one active agent selected from the group consisting of beta-adrenergic agonists (e.g. noradrenalin, isoproterenol, BRL 35135, ICI D7114, CGP-12177A, CL 316243), indirect sympathomimetics (e.g. ephedrine, methylphenidate), atrial natriuretic peptide (e.g. ANP, BNP) and ANP/BNP receptor agonists (e.g. AP-811).

68. The activator of paracrine fibroblast growth factor (FGF) of item 67, the pharmaceutical composition of item 67, or the method of item 68, wherein said beta-adrenergic agonist is a beta3-adrenergic agonist.

69. The activator of paracrine fibroblast growth factor (FGF) of item 68, the pharmaceutical composition of item 68, or the method of item 68, wherein said beta3-adrenergic agonist is CL 316243.

70. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and 50 to 69, or the method of any one of items 48 and 50 to 69, wherein said activator of paracrine fibroblast growth factor (FGF) is capable of influencing FGF-heparin-binding or influences FGF-heparin-binding.

71. The activator of paracrine fibroblast growth factor (FGF) of item 70, the pharmaceutical composition of item 70, or the method of item 70, wherein said activator of paracrine fibroblast growth factor (FGF) is sucrose octasulfate or inositol hexasulfate.

72. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and 50 to 69, or the method of any one of items 48 and 50 to 69, wherein said

activator of paracrine fibroblast growth factor (FGF) is capable of activating the FGFR-FGF-Heparin-complex or activates the FGFR-FGF-Heparin-complex.

73. The activator of paracrine fibroblast growth factor (FGF) of item 72, the pharmaceutical composition of item 72, or the method of item 72, wherein said activator of paracrine fibroblast growth factor (FGF) is heparin or a heparin derivative, such as certoparin, dalteparin, enoxaparin, nadroparin or danaparoid.

74. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and 50 to 69, or the method of any one of items 48 and 50 to 69, wherein said activator of paracrine fibroblast growth factor (FGF) is capable of influencing stability of protein conformation or influences stability of protein conformation.

75. The activator of paracrine fibroblast growth factor (FGF) of item 74, the pharmaceutical composition of item 74, or the method of item 74, wherein said activator of paracrine fibroblast growth factor (FGF) is Alpha-Cyclodextrin or a Cyclodextrin-derivative.

76. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and 50 to 69, or the method of any one of items 48 and 50 to 69, wherein said activator of paracrine fibroblast growth factor (FGF) is capable of influencing heparanase-mediated degradation of heparan-sulfate proteoglycan (HSPG) of the extracellular matrix or influences heparanase-mediated degradation of heparan-sulfate proteoglycan (HSPG) of the extracellular matrix.

77. The activator of paracrine fibroblast growth factor (FGF) of item 76, the pharmaceutical composition of item 76, or the method of item 76, wherein said activator of paracrine fibroblast growth factor (FGF) is PI-88 (a mixture of highly sulfated, monophosphorylated mannose oligosaccharides) or OGT 2115.

78. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and 50 to 69, or the method of any one of items 48 and 50 to 69, wherein said activator of paracrine fibroblast growth factor (FGF) is capable of influencing FGFR activity or influences FGFR activity.

79. The activator of paracrine fibroblast growth factor (FGF) of item 78, the pharmaceutical composition of item 78, or the method of item 78, wherein said FGFR is FGFR1.

80. The activator of paracrine fibroblast growth factor (FGF) of item 79, the pharmaceutical composition of item 79, or the method of item 79, wherein said activator of paracrine fibroblast growth factor (FGF) is SUN11602.

81. The activator of paracrine fibroblast growth factor (FGF) of item 78, the pharmaceutical composition of item 78, or the method of item 78, wherein said FGFR is FGFR3.

82. The activator of paracrine fibroblast growth factor (FGF) of item 81, the pharmaceutical composition of item 81, or the method of item 81, wherein said activator of paracrine fibroblast growth factor (FGF) is Botulinum neurotoxin serotype A (BoNT/A).

83. The activator of paracrine fibroblast growth factor (FGF) of item 78, the pharmaceutical composition of item 78, or the method of item 78, wherein said FGFR is FGFR4.

84. The activator of paracrine fibroblast growth factor (FGF) of item 83, the pharmaceutical composition of item 83, or the method of item 83, wherein said activator of paracrine fibroblast growth factor (FGF) is monoclonal Antibody: 4FA6D3C10.

85. The activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 46 and 50 to 69, the pharmaceutical composition of any one of items 47 and

50 to 69, or the method of any one of items 48 and 50 to 69, wherein said activator of paracrine fibroblast growth factor (FGF) is strontium ranelate, anisomycin, PAF C-16, t-butylhydroquinone, phorbol 12-myristate 13-acetate, cell permeant caged IP3, 740 Y-P, or sc-3036.

86. A method for the preparation of a pharmaceutical composition for use in treating a disease or disorder of energy homeostasis, wherein the method comprises the following steps:
(a) contacting the activator of paracrine fibroblast growth factor (FGF) of any one of items 1 to 85 with a liquid carrier or a solid carrier;
(b) optionally, adjusting the pH and/or the osmolarity of the product obtained in step (a);
(c) optionally, sterilizing the product obtained in step (a) or (b); and
(d) formulating and/or packaging the product obtained in step (a), (b) or (c) as a finished medical product.

87. The method of item 86, wherein said carrier is at least one carrier selected from the group consisting of cellulose, lactose, water, saline, Ringer's solution, dextrose solution, a fixed oil, ethyl oleate and liposomes.

The herein provided paracrine FGFs are able to specifically target visceral fat (i.e. visceral adipose tissue). Visceral fat, also known as organ fat, intra-abdominal fat or belly fat, is located inside the peritoneal cavity and packed in between internal organs and torso. The aesthetic problem arising from an excess of visceral fat is called "pot belly" or "beer belly", in which the abdomen protrudes to an unaesthetic extent. Thus, an excess of visceral fat negatively influences the bodily appearance. Furthermore, a waist to hip ratio (WHR) of 0.7 for women and 0.9 for men has been shown to correlate strongly with fertility and a WHR of less than 0.7 is a significant measure of female attractiveness in Caucasian cultures. Accordingly, the present invention relates to a non-therapeutic cosmetic product comprising the herein provided paracrine FGF (e.g. FGF8b, FGF8f and/or FGF17).

Thus, one embodiment of the present invention relates to a cosmetic product comprising a polypeptide, wherein said polypeptide is selected from the group

consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As mentioned, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The polypeptide of item (e) above may have at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of visceral adipocytes and/or preadipocytes

(e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The illustrative appended examples show that FGF8 is active in browning visceral adipose tissue. Whereas FGF8b was shown to have browning potential in both visceral and subcutaneous adipose tissue, FGF8f specifically induces browning of visceral adipose tissue. Therefore, one aspect of the invention relates to a non-therapeutic cosmetic product comprising FGF8f.

Thus, one aspect of the invention relates to a cosmetic product comprising a polypeptide, wherein said polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 4 or the nucleic acid sequence comprising nucleic acid residues 67-735 in SEQ ID NO: 4;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 18 or an amino acid sequence comprising amino acids 23-244 in SEQ ID NO: 18;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-244 in SEQ ID NO: 18 or having an amino acid sequence as depicted in SEQ ID NO: 18;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As mentioned, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The polypeptide of item (e) above may have at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The illustrative appended examples demonstrate that FGF17 is active in browning visceral adipose tissue. Therefore, one aspect of the invention relates to a non-therapeutic cosmetic product comprising FGF17.

Thus, one aspect of the invention relates to a cosmetic product comprising a polypeptide, wherein said polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 9 or the nucleic acid sequence comprising nucleic acid residues 67-651 in SEQ ID NO: 9;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 23 or an amino acid sequence comprising amino acids 23-216 in SEQ ID NO: 23;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-216 in SEQ ID NO: 23 or having an amino acid sequence as depicted in SEQ ID NO: 23;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a)

to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes; and

(f) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

As mentioned, the function of the above described polypeptide of the invention is ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (in particular white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The polypeptide of item (e) above may have at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The herein provided non-therapeutic cosmetic product can be used to reduce body weight. Thus, one aspect of the invention relates to the use of the cosmetic product of the invention for reducing body weight. Accordingly, the invention provides a method for improving the bodily appearance of a mammal (e.g. a human being) which comprises administering to said mammal the paracrine FGF of the invention (e.g. FGF8b, FGF8f and/or FGF17) in a dose and for a period of time to effectively reduce visceral adipose tissue, and repeating said administration until a cosmetically beneficial loss of body weight has occurred.

In one aspect, the invention relates to the herein provided cosmetic product, or the use of said cosmetic product, wherein said polypeptide binds to a FGF receptor. Said FGF receptor may be at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3. It is preferred that the FGF receptor is FGF receptor 4 (FGFR4) or the FGF receptor c spliceform of FGF receptor 1, 2, and 3.

As mentioned, the cosmetic product provided herein effectively reduces visceral adipose tissue which is visible as “belly fat” or “abdominal fat”. Thus, the invention also relates to the cosmetic product of the invention, or the use of said cosmetic product, wherein said cosmetic product reduces abdominal adipose tissue.

The herein provided cosmetic product may be in the form for oral administration. Thus, the cosmetic product may be in the form of a liquid solution, a pill, a tablet, a capsule or a powder for oral administration. In one example of the invention, the cosmetic product is in the form of a healthy food, e.g. a diet drink. In another aspect of the invention, the cosmetic product is in the form of a cream, salve or gel. In this embodiment the herein provided cosmetic product can be administered on the skin and penetrate the skin in order to effectively target the adipose tissue. Thus, the cosmetic product(s) provided herein may further comprise a chemical penetration enhancer. Chemical penetration enhancers (i.e. skin penetration enhancers) are commonly known to deliver drugs and cosmetics through the skin (see, e.g., Lane, International Journal of Pharmaceutics 447 (2013) 12–21). In accordance with the invention, said chemical penetration enhancer may be at least one chemical penetration enhancer selected from the group consisting of ethanol, isopropyl alcohol, decanol, hexanol, lauryl alcohol, myristyl alcohol, octanol, octyl dodecanol, oleyl alcohol, azone, ethyl acetate, octyl salicylate, 2-ethylhexyl 4-(dimethylamino)benzoate, ethyl oleate, glyceryl monoleate, glyceryl monocaprate, glyceryl tricaprylate, isopropyl myristate, isopropyl palmitate, propylene glycol monolaurate, propylene glycol monocaprylate, 2-(2-ethoxyethoxy)ethanol, lauric acid, linoleic acid, linolenic acid, myristic acid, oleic acid, palmitic acid, stearic acid, isostearic acid, dipropylene glycol, propylene glycol, 1,2-butylene glycol, 1,3-butylene glycol, N-methyl-2-pyrrolidone, 2-pyrrolidone, decylmethyl sulphoxide, dimethyl sulfoxide, sodium lauryl sulphate, alkyl dimethylbenzyl ammonium halides, alkyl trimethyl ammonium halides, alkyl pyridinium halides, 2-(dodecyloxy)ethanol, polyoxyethylen(20)-sorbitan-monooleat, eugenol, d-limonene, menthol, menthone, farnesol and neridol.

In certain aspects, the present invention relates to the following items:

1. A cosmetic product comprising a paracrine fibroblast growth factor (FGF).

2. The cosmetic product according to item 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8b.

3. The cosmetic product according to item 1 or 2, wherein the paracrine fibroblast growth factor (FGF) is human FGF8b.

4. The cosmetic product according to any one of items 1 to 3, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;
   (c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-215 in SEQ ID NO: 16 or having an amino acid sequence as depicted in SEQ ID NO: 16;
   (d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);
   (e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);
   (f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and
   (g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

5. The cosmetic product according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF8f.

6. The cosmetic product according to claim 1 or 5, wherein the paracrine fibroblast growth factor (FGF) is human FGF8f.

7. The cosmetic product according to any one of items 1, 5 and 6, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 4 or the nucleic acid sequence comprising nucleic acid residues 67-735 in SEQ ID NO: 4;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 18 or an amino acid sequence comprising amino acids 23-244 in SEQ ID NO: 18;
   (c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-244 in SEQ ID NO: 18 or having an amino acid sequence as depicted in SEQ ID NO: 18;
   (d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);
   (e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);
   (f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and
   (g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

8. The cosmetic product according to claim 1, wherein the paracrine fibroblast growth factor (FGF) is FGF17.

9. The cosmetic product according to claim 1 or 8, wherein the paracrine fibroblast growth factor (FGF) is human FGF17.

10. The cosmetic product according to any one of items 1, 8 and 9, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide, wherein the polypeptide is selected from the group consisting of:

(a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 9 or the nucleic acid sequence comprising nucleic acid residues 67-651 in SEQ ID NO: 9;

(b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 23 or an amino acid sequence comprising amino acids 23-216 in SEQ ID NO: 23;

(c) a polypeptide encoded by a nucleic acid molecule encoding a polypeptide comprising an amino acid sequence having amino acids 23-216 in SEQ ID NO: 23 or having an amino acid sequence as depicted in SEQ ID NO: 23;

(d) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) or (c);

(e) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (d);

(f) a protein as defined in any one of (a) to (e) wherein one or more amino acids are deleted, inserted, added or substituted; and

(g) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), (c) and (d).

11. The cosmetic product according to any one of items 4, 7 and 10, wherein the

polypeptide is a polypeptide having at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% identity to the polypeptide of any one of (a) to (d).

12. The cosmetic product according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 90% identity to the polypeptide of any one of (a) to (d).

13. The cosmetic product according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 95% identity to the polypeptide of any one of (a) to (d).

14. The cosmetic product according to any one of items 4, 7 and 10, wherein the polypeptide is a polypeptide having at least 99% identity to the polypeptide of any one of (a) to (d).

15. The cosmetic product according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to induce differentiation or conversion of white adipocytes and/or white preadipocytes to brown adipocytes.

16. The cosmetic product according to any one of items 1 to 14, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide that is able to induce differentiation or conversion of white visceral adipocytes and/or white visceral preadipocytes to brown adipocytes.

17. The cosmetic product according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence VTVQSSPNFTQ or a fragment thereof.

18. The cosmetic product according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide

comprising the amino acid sequence VTVQSSPNFTQ from position 24 to position 34 of the amino acid sequence of the polypeptide.

19. The cosmetic product according to item 18, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

20. The cosmetic product according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence VTVQSSPNFTQ from position 53 to position 63 of the amino acid sequence of the polypeptide.

21. The cosmetic product according to item 20, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

22. The cosmetic product according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFTQ or a fragment thereof.

23. The cosmetic product according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 23 to position 34 of the amino acid sequence of the polypeptide.

24. The cosmetic product according to item 23, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

25. The cosmetic product according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFTQ from position 52 to position 63 of the amino acid sequence of the polypeptide.

26. The cosmetic product homeostasis according to item 25, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

27. The cosmetic product according to any one of items 1 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence QVTVQSSPNFT or a fragment thereof.

28. The cosmetic product according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 23 to position 33 of the amino acid sequence of the polypeptide.

29. The cosmetic product according to item 28, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

30. The cosmetic product according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence QVTVQSSPNFT from position 52 to position 62 of the amino acid sequence of the polypeptide.

31. The cosmetic product according to item 30, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

32. The cosmetic product according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNFXQ or a fragment thereof.

33. The cosmetic product according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 29 to position 34 of the amino acid sequence of the polypeptide.

34. The cosmetic product according to item 33, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

35. The cosmetic product according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 58 to position 63 of the amino acid sequence of the polypeptide.

36. The cosmetic product according to item 35, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

37. The cosmetic product according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNFXQ from position 30 to position 35 of the amino acid sequence of the polypeptide.

38. The cosmetic product according to item 37, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

39. The cosmetic product according to any one of items 32 to 38, wherein residue X in the amino acid sequence SPNFXQ is T or N.

40. The cosmetic product according to any one of items 1 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising or consisting of the amino acid sequence SPNF or a fragment thereof.

41. The cosmetic product according to any one of items 1 to 4 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 29 to position 32 of the amino acid sequence of the polypeptide.

42. The cosmetic product according to item 41, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 16 (FGF8b).

43. The cosmetic product according to any one of items 1, 5 to 7 and 11 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 58 to position 61 of

the amino acid sequence of the polypeptide.

44. The cosmetic product according to item 43, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 18 (FGF8f).

45. The cosmetic product according to any one of items 1 and 8 to 16, wherein the paracrine fibroblast growth factor (FGF) is a polypeptide comprising the amino acid sequence SPNF from position 30 to position 33 of the amino acid sequence of the polypeptide.

46. The cosmetic product according to item 45, wherein the amino acid sequence of the polypeptide is shown in SEQ ID NO. 23 (FGF17).

47. Use of the cosmetic product of any one of items 1 to 46 for reducing body weight preferably in a healthy patient, more preferably in a healthy human patient.

48. The cosmetic product of any one of items 1 to 46, or the use of item 47, wherein said paracrine fibroblast growth factor (FGF) binds to a FGF receptor or is capable of binding to an FGF receptor.

49. The cosmetic product of item 48, or the use of item 48, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1c, FGF receptor 2c and FGF receptor 3c.

50. The cosmetic product of any one of items 1 to 46, 48 and 49, or the use of any one of items 47 to 49, wherein said cosmetic product reduces abdominal adipose tissue.

51. The cosmetic product of any one of items 1 to 46, and 48 to 50, or the use of any one of items 47 to 50, wherein said cosmetic product is in the form of a cream, salve or gel.

52. The cosmetic product of any one of items 1 to 46, and 48 to 551, or the use of

any one of items 47 to 51, wherein said cosmetic product further comprises a chemical penetration enhancer.

52. The cosmetic product of item 52, or the use of item 52, wherein said chemical penetration enhancer is at least one chemical penetration enhancer selected from the group consisting of ethanol, isopropyl alcohol, decanol, hexanol, lauryl alcohol, myristyl alcohol, octanol, octyl dodecanol, oleyl alcohol, azone, ethyl acetate, octyl salicylate, 2-ethylhexyl 4-(dimethylamino)benzoate, ethyl oleate, glyceryl monoleate, glyceryl monocaprate, glyceryl tricaprylate, isopropyl myristate, isopropyl palmitate, propylene glycol monolaurate, propylene glycol monocaprylate, 2-(2-ethoxyethoxy)ethanol, lauric acid, linoleic acid, linolenic acid, myristic acid, oleic acid, palmitic acid, stearic acid, isostearic acid, dipropylene glycol, propylene glycol, 1,2-butylene glycol, 1,3-butylene glycol, N-methyl-2-pyrrolidone, 2-pyrrolidone, decylmethyl sulphoxide, dimethyl sulfoxide, sodium lauryl sulphate, alkyl dimethylbenzyl ammonium halides, alkyl trimethyl ammonium halides, alkyl pyridinium halides, 2-(dodecyloxy)ethanol, polyoxyethylen(20)-sorbitan-monooleat, eugenol, d-limonene, menthol, menthone, farnesol and neridol.

In context of the present invention, “homologous” or “percent homology” means that amino acid or nucleotide sequences have identities of at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95% or at least 99% to the sequences shown herein, e.g. those of human FGF8b, FGF8f or FGF17, wherein the higher identity values are preferred upon the lower ones.

In accordance with the present invention, the term "identity/identities" or “percent identity/identities” in the context of two or more nucleic acid or amino acid sequences, refers to two or more sequences or subsequences that are the same, or that have a specified percentage of amino acid residues or nucleotides that are the same (e.g., 40% or 45% identity, preferably, 70-95% identity, more preferably at least 99% identity with the nucleic acid sequences of, e.g., SEQ ID NOs: 2, 4 or 9, or with the amino acid sequences of, e.g., SEQ ID NOs: 16, 18 or 23, and being functional, wherein the function comprises ability to induce differentiation or conversion of white

(e.g. visceral) adipocytes and/or preadipocytes to brown adipocytes), when compared and aligned for maximum correspondence over a window of comparison, or over a designated region as measured using a sequence comparison algorithm as known in the art, or by manual alignment and visual inspection.

Those having skill in the art will know how to determine percent identity between/among sequences using, for example, algorithms such as those based on CLUSTALW computer program (Thompson; 1994; Nucl Acids Res; 2; 4673-4680) or FASTDB (Brutlag; 1990; Comp App Biosci; 6; 237-245), as known in the art.

Also available to those having skill in this art are the BLAST and BLAST 2.0 algorithms (Altschul; 1997; Nucl Acids Res 25; 3389-3402, Altschul; 1993; J Mol Evol; 36; 290-300, Altschul; 1990; J Mol Biol 215; 403-410). The BLASTN program for nucleic acid sequences uses as defaults a word length (W) of 11, an expectation (E) of 10, M=5, N=4, and a comparison of both strands. For amino acid sequences, the BLASTP program uses as defaults a word length (W) of 3, and an expectation (E) of 10. The BLOSUM62 scoring matrix (Henikoff; 1989; PNAS; 89; 10915) uses alignments (B) of 50, expectation (E) of 10, M=5, N=4, and a comparison of both strands.

In addition, the present invention relates to a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code. When used in accordance with the present invention, the term "being degenerate as a result of the genetic code" means that due to the redundancy of the genetic code different nucleotide sequences code for the same amino acid.

In order to determine whether an amino acid residue or nucleotide residue in an amino acid or nucleic acid sequence corresponds to a certain position in the amino acid sequence of, e.g., SEQ ID NO: 16, 18 or 23, or nucleotide sequence of e.g. SEQ ID NOs: 2, 4 or 9, the skilled person can use means and methods well-known in the art, e.g., alignments, either manually or by using computer programs.

In accordance with the present invention, the terms "homology" or "percent homology" or "identical" or "percent identity" or “percentage identity” or “sequence

identity" in the context of two or more nucleic acid sequences refers to two or more sequences or subsequences that are the same, or that have a specified percentage of nucleotides that are the same (at least 40 %, 50 %, 60 %, 70%, 75%, 80%, 85%, most preferably at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97% or 98% identity, most preferably at least 99% identity), when compared and aligned for maximum correspondence over a window of comparison (preferably over the full length), or over a designated region as measured using a sequence comparison algorithm as known in the art, or by manual alignment and visual inspection. Sequences having, for example, 75% to 90% or greater sequence identity may be considered to be substantially identical. Such a definition also applies to the complement of a test sequence. Preferably the described identity exists over a region that is at least about 15 to 25 nucleotides in length, more preferably, over a region that is at least about 50 to 100 nucleotides in length and most preferably over the full length. Those having skill in the art will know how to determine percent identity between/among sequences using, for example, algorithms such as those based on CLUSTALW computer program (Thompson Nucl. Acids Res. 2 (1994), 4673-4680) or FASTDB (Brutlag Comp. App. Biosci. 6 (1990), 237-245), as known in the art.

Although the FASTDB algorithm typically does not consider internal non-matching deletions or additions in sequences, i.e., gaps, in its calculation, this can be corrected manually to avoid an overestimation of the % identity. CLUSTALW, however, does take sequence gaps into account in its identity calculations. Also available to those having skill in this art are the BLAST and BLAST 2.0 algorithms (Altschul, (1997) Nucl. Acids Res. 25:3389-3402; Altschul (1993) J. Mol. Evol. 36:290-300; Altschul (1990) J. Mol. Biol. 215:403-410). The BLASTN program for nucleic acid sequences uses as defaults a word length (W) of 11, an expectation (E) of 10, M=5, N=4, and a comparison of both strands. The BLOSUM62 scoring matrix (Henikoff (1989) PNAS 89:10915) uses alignments (B) of 50, expectation (E) of 10, M=5, N=4, and a comparison of both strands.

For example, BLAST 2.0, which stands for Basic Local Alignment Search Tool BLAST (Altschul; 1997; loc. cit., Altschul; 1993; loc. cit.; Altschul; 1990; loc. cit.), can be used to search for local sequence alignments. BLAST, as discussed above, produces alignments of both nucleotide and amino acid sequences to determine sequence similarity. Because of the local nature of the alignments, BLAST is

especially useful in determining exact matches or in identifying similar sequences. The fundamental unit of BLAST algorithm output is the High-scoring Segment Pair (HSP). An HSP consists of two sequence fragments of arbitrary but equal lengths whose alignment is locally maximal and for which the alignment score meets or exceeds a threshold or cut-off score set by the user. The BLAST approach is to look for HSPs between a query sequence and a database sequence, to evaluate the statistical significance of any matches found, and to report only those matches which satisfy the user-selected threshold of significance. The parameter E establishes the statistically significant threshold for reporting database sequence matches. E is interpreted as the upper bound of the expected frequency of chance occurrence of an HSP (or set of HSPs) within the context of the entire database search. Any database sequence whose match satisfies E is reported in the program output.

Analogous computer techniques using BLAST (Altschul; 1997; loc. cit., Altschul; 1993; loc. cit., Altschul; 1990; loc. cit.) are used to search for identical or related molecules in nucleotide databases such as GenBank or EMBL. This analysis is much faster than multiple membrane-based hybridizations. In addition, the sensitivity of the computer search can be modified to determine whether any particular match is categorized as exact or similar. The basis of the search is the product score which is defined as:

$$\frac{\%\ \text{sequence identity} \times \%\ \text{maximum BLAST score}}{100}$$

and it takes into account both the degree of similarity between two sequences and the length of the sequence match. For example, with a product score of 40, the match will be exact within a 1-2% error; and at 70, the match will be exact. Similar molecules are usually identified by selecting those which show product scores between 15 and 40, although lower scores may identify related molecules. Another example for a program capable of generating sequence alignments is the CLUSTALW computer program (Thompson; 1994; Nucl Acids Res; 2; 4673-4680) or FASTDB (Brutlag; 1990; Comp App Biosci 6; 237-245), as known in the art.

The terms "hybridization", "hybridizes" or “hybridizing” as used herein relate to complementary (antisense) molecules, which specifically interact with/hybridizes to

one or more nucleic acid molecules encoding the herein defined paracrine FGFs (e.g. FGF8b, FGF8f and/or FGF17). Highly mutated complementary constructs, which are not capable of hybridizing to a nucleic acid molecule encoding the paracrine FGF of the invention are not to be employed in the context of the present invention. The person skilled in the art can easily deduce whether a complementary construct specifically hybridizes to sequences encoding the paracrine FGF of the present invention (e.g. FGF8b, FGF8f and/or FGF17). These tests comprise, but are not limited to hybridization assays, RNAse protection assays, Northern Blots, North-western blots, nuclear magnetic resonance and fluorescence binding assays, dot blots, micro- and macroarrays and quantitative PCR. In addition, such a screening may not be restricted to mRNA molecules, but may also include mRNA/protein (RNP) complexes (Hermann; 2000; Angew Chem Int Ed Engl; 39; 1890-1904, DeJong; 2002; Curr Trop Med Chem; 2; 289-302). Furthermore, functional tests including Western blots, immunohistochemistry, immunoprecipitation assays, and bioassays based on responsive promoters are envisaged for testing whether a particular complementary construct is capable of specifically interacting with/hybridizing to the nucleic acid molecule encoding the paracrine FGF of the present invention.

In addition, the terms "hybridization", "hybridizes" or “hybridizing” as used herein further relate to hybridizations under stringent or non-stringent conditions. Said hybridization conditions may be established according to conventional protocols, described, e.g., in Sambrook, Russell "Molecular Cloning, A Laboratory Manual", Cold Spring Harbor Laboratory, N.Y. (2001), Ausubel, "Current Protocols in Molecular Biology", Green Publishing Associates and Wiley Interscience, N.Y. (1989), or Higgins and Hames (Eds.) "Nucleic acid hybridization, a practical approach" IRL Press Oxford, Washington DC, (1985). The setting of conditions is well within the skill of the artisan and can be determined according to protocols described in the art. Thus, the detection of only specifically hybridizing sequences will usually require stringent hybridization and washing conditions such as 0.1 x SSC, 0.1% SDS at 65°C. Non-stringent hybridization conditions for the detection of homologous or not exactly complementary sequences may be set at 6 x SSC, 1% SDS at 65°C. As is well known, the length of the probe and the composition of the nucleic acid to be determined constitute further parameters of the hybridization conditions. Note that variations in the above conditions may be accomplished

through the inclusion and/or substitution of alternate blocking reagents used to suppress background in hybridization experiments. Typical blocking reagents include Denhardt's reagent, BLOTTO, heparin, denatured salmon sperm DNA, and commercially available proprietary formulations. The inclusion of specific blocking reagents may require modification of the hybridization conditions described above, due to problems with compatibility.

Hybridizing nucleic acid molecules also comprise fragments of the above described molecules. Such fragments may comprise complementary nucleic acid sequences of the nucleic acid molecules which code for the paracrine FGF(s) of the present invention (e.g. FGF8b) or functional fragments thereof. Furthermore, also the complementary nucleic acid molecule, the complementary fragments and (allelic) variants of the nucleic acid molecules which hybridize with any of the aforementioned nucleic acid molecules are included in the present invention.

Additionally, a hybridization complex refers to a complex between two nucleic acid sequences by virtue of the formation of hydrogen bonds between complementary G and C bases and between complementary A and T bases; these hydrogen bonds may be further stabilized by base stacking interactions. The two complementary nucleic acid sequences hydrogen bond in an anti-parallel configuration. A hybridization complex may be formed in solution (e.g., Cot or Rot analysis) or between one nucleic acid sequence present in solution and another nucleic acid sequence immobilized on a solid support (e.g., membranes, filters, chips, pins or glass slides to which, e.g., cells have been fixed). In context of the present invention, the terms "complementary" or "complementarity" refers to the natural binding of polynucleotides under permissive salt and temperature conditions by base-pairing. A complementary polynucleotide of a specified nucleotide sequence is an antisense polynucleotide of said specified nucleotide sequence. For example, the sequence "A-G-T" binds to the complementary sequence "T-C-A". Complementarity between two single-stranded molecules may be "partial", in which only some of the nucleic acids bind, or it may be complete when total complementarity exists between single-stranded molecules. The degree of complementarity between nucleic acid strands has significant effects on the efficiency and strength of hybridization between nucleic

acid strands. This is of particular importance in amplification reactions, which depend upon binding between nucleic acids strands.

The term "hybridizing (sequences)" preferably refers to the sequences which display a sequence identity of at least 60%, preferably at least 70%, more preferably at least 80%, even more preferably at least 90%, particularly preferred at least 95%, more particularly preferred at least 96%, even more particularly preferred at least 97% and most preferably at least 98% or up to 100% identity with a complementary nucleic acid sequence of the nucleic acid sequence encoding the paracrine FGF of the invention (e.g. FGF8b, FGF8f and/or FGF17) or a functional fragment thereof and being a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes as described herein. Moreover, the term "hybridizing (sequences)" preferably refers to complementary nucleic acid sequences of the nucleic acid sequences encoding amino acid molecules having a sequence identity of at least 60%, preferably at least 70%, more preferably at least 80%, even more preferably at least 90%, particularly preferred at least 95%, more particularly preferred at least 96%, even more particularly preferred at least 97% and most preferably at least 98% or up to 100% identity with an amino acid sequence of the paracrine FGF of the invention (e.g. SEQ ID NOs: 16, 18 or 23) or a functional fragment thereof and being a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes (e.g. white visceral adipocytes and/or white visceral preadipocytes) to brown adipocytes.

The explanations and definitions given herein above in respect of “homology/identity of nucleic acid sequences” apply, mutatis mutandis, to “amino acid sequences” of the herein provided amino acid sequence of the paracrine FGF of the invention (e.g. SEQ ID NOs: 16, 18 or 23).

The polypeptide to be used in accordance with the present invention may have at least 40, 50, 60 or 70 % identity/similarity to the proteins having the amino acid sequence as, for example, depicted in SEQ ID NO: 16, 18 or 23, respectively. More preferably, the polypeptide has at least 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%

or 98% identity/similarity to the proteins depicted in SEQ ID NO: 16, 18 or 23, respectively, wherein the higher values are preferred. Most preferably, the polypeptide has at least 99% homology to the protein as depicted in SEQ ID NO: 16, 18 or 23.

As used herein, the terms “comprising” and “including” or grammatical variants thereof are to be taken as specifying the stated features, integers, steps or components but do not preclude the addition of one or more additional features, integers, steps, components or groups thereof. This term encompasses the terms “consisting of” and “consisting essentially of.” Thus, the terms “comprising”/“including”/”having” mean that any further component (or likewise features, integers, steps and the like) can/ may be present.

The term “consisting of” means that no further component (or likewise features, integers, steps and the like) is present.

The term “consisting essentially of” or grammatical variants thereof when used herein are to be taken as specifying the stated features, integers, steps or components but do not preclude the addition of one or more additional features, integers, steps, components or groups thereof but only if the additional features, integers, steps, components or groups thereof do not materially alter the basic and novel characteristics of the claimed composition, device or method.

Thus, the term “consisting essentially of” means that specific further components (or likewise features, integers, steps and the like) can be present, namely those not materially affecting the essential characteristics of the composition, device or method. In other words, the term "consisting essentially of" (which can be interchangeably used herein with the term "comprising substantially"), allows the presence of other components in the composition, device or method in addition to the mandatory components (or likewise features, integers, steps and the like), provided that the essential characteristics of the device or method are not materially affected by the presence of other components.

It is preferred herein that the polypeptides to be used in accordance with the present invention consist essentially of, more preferably consist of, the amino acid sequences as defined herein.

The present invention is further described by reference to the following non-limiting figures and example.

The Figures show:

Figure 1.

Screening paracrine FGFs for a browning potential. A, B - Screening of all paracrine FGFs in immortalized white adipocytes.Subcutanous, inguinal and visceral, epididymal white adipocytes were grown to confluence in Dulbecco's modified Eagle's medium with 4.5 g/l glucose, supplemented with 20 nmol/l insulin, 1 nmol/l triiodothyronine, 20% fetal bovine serum, and penicillin/streptomycin. Upon reaching confluence, differentiation was induced with 500 mmol/l isobutylmethylxanthine, 250 mmol/l indomethacine, and 2 mg/ml dexamethasone for 24 hours. Subsequently, members of the paracrine FGF protein family were added to the medium for the following six days. The medium and the FGF indicated were changed every 48 hours. Untreated cells served as a negative control. At the end of the differentiation process total RNA was isolated using TRIzol reagents, RNA quality and concentration was measured by photometric analysis, and 5 µg of total RNA was reverse-transcribed to cDNA. Finally, quantitative analysis of UCP-1 mRNA expression was performed. A bar graph analysis including the SEM of 3 to 6 independent experiments is shown. A: FGF8b and FGF17 lead to increased expression of Ucp1 mRNA expression in epididymal adipocytes as measured by quantitative PCR. B - Four different splicing forms of the Fgf8 gene were compared by quantifying Ucp1 mRNA expression in fully differentiated white adipocytes. UCP-1 mRNA abundance is shown in percent of the FGF8b value. Fgf8b and Fgf8f were able to increase expression in epididymal adipocytes. C - Illustration depicting the different exon usage that characterizes the FGF8 spliceforms a, b, e, and f.

Figure 2.

Dose dependency of Ffg8b action. A, B - Subcutanous, inguinal and visceral, epididymal white adipocytes were grown to confluence in Dulbecco's modified Eagle's medium with 4.5 g/l glucose, supplemented with 20 nmol/l insulin, 1 nmol/l triiodothyronine, 20% fetal bovine serum, and penicillin/streptomycin. Upon reaching confluence, differentiation was induced with 500 mmol/l isobutylmethylxanthine, 250 mmol/l indomethacine, and 2 mg/ml dexamethasone for 24 hours. Subsequently, FGF8b in five indicated concentrations were added to the medium for the following six days. The medium and the FGF8b were changed every 48 hours. Untreated cells served as a negative control. At the end of the differentiation process total RNA was isolated using TRIzol reagents, RNA quality and concentration was measured by photometric analysis, and 5 µg of total RNA was reverse-transcribed to cDNA. Finally, quantitative analysis of UCP-1 mRNA expression was performed. FGF8b induces Ucp1 mRNA abundance in inguinal and epididymal adipocytes. Epididymal fat cells were more dose dependently responsive to Fgf8b treatment as compared to inguinal cells. The latter were responsive to FGF8b treatment above the threshold indicated. B - The absolute increase of several genes typical for brown fat cells. Both Ucp1 and Cidea mRNA were strongly increased by Fgf8b, while Cox7a1 and elovl3 were not significantly altered. In fully differentiated cells, Pgc1a mRNA was found to be lower expresse than in untreated cells.

Figure 3.

Treatment timecourse experiment. A, B – Subcutanous, inguinal and visceral, epididymal white adipocytes were grown to confluence (prolif. denotes proliferation period) in Dulbecco's modified Eagle's medium with 4.5 g/l glucose, supplemented with 20 nmol/l insulin, 1 nmol/l triiodothyronine, 20% fetal bovine serum, and penicillin/streptomycin. Upon reaching confluence, differentiation was induced with 500 mmol/l isobutylmethylxanthine, 250 mmol/l indomethacine, and 2 mg/ml dexamethasone for 24 hours (induction). Subsequently, the cells were cultured for another six days (differentiation). At the end of the differentiation process total RNA was isolated using TRIzol reagents, RNA quality and concentration was measured by photometric analysis, and 5 µg of total RNA was reverse-transcribed to cDNA. Finally, quantitative analysis of UCP-1 mRNA expression was performed. Untreated cells served as a negative control. A bar graph analysis including the SEM of 5 independent experiments is shown. A - Epididymal fat cells were treated with FGF8b

during different time windows as given by black squares indicating one day of treatment each. Treatment during the entire differentiation period led to strongest Ucp1 induction. This effect could be mimicked by only treating the cells during the second day or last day of differentiation indicting sensitive time windows. Treatment during the induction period inhibited Ucp1 induction. B - Inguinal fat cells were treated with FGF8b during different time windows as given by black squares indicating one day of treatment each. Treatment during the last day of differentiation led to the highest increase in Ucp1 mRNA abundance. n.d. denotes not detectable.

Figure 4.

Timecourse of brown adipose tissue (BAT) marker gene expression following a short period of Fgf8b treatment. Visceral, epididymal white adipocytes were grown to confluence in Dulbecco's modified Eagle's medium with 4.5 g/l glucose, supplemented with 20 nmol/l insulin, 1 nmol/l triiodothyronine, 20% fetal bovine serum, and penicillin/streptomycin within 96 hours. During this proliferation period cells were treated with FGF8b for 48 hours during the second half of proliferation. Upon reaching confluence, differentiation was induced with 500 mmol/l isobutylmethylxanthine, 250 mmol/l indomethacine, and 2 mg/ml dexamethasone for 24 hours (induction). Subsequently, the cells were cultured for another six days (differentiation). A subset of culture plates were harvested (1, acute) right after FGF8b treatment, (2, d1 p.i. denotes day one post induction) right after the induction period, (3, d2 p.i. denotes day two post induction) after two days of differentiation, (4, d4 p.i. denotes day four post induction) after four days of differentiation, and (5, d6 p.i. denotes day six post induction) after six days, id est the end of the differentiation period. At these indicated times total RNA was isolated using TRIzol reagents, RNA quality and concentration was measured by photometric analysis, and 5 µg of total RNA was reverse-transcribed to cDNA. Finally, quantitative analysis of UCP-1 mRNA expression was performed. Untreated cells served to normalize the data to the natural course of BAT marker values during differentiation. A graph analysis including the SEM of 5 independent experiments is shown. Ucp1 mRNA abundance increased 6 days after induction for the first time. A similar pattern was observed for the brown fat marker genes Cidea and Cox7a1 increasing at day 4 or 6 after induction. Pgc1a was acutely downregulated by Fgf8b treatment and increased expression above control levels at day 6 after induction. Known or suspected regulators of brown

adipocyte differentiation Prdm16, Foxc2 and Prb displayed characteristic patterns of transient increases during the differentiation process.

Figure 5.

Expression of Fgf receptors Fgfr1, Fgfr2, Fgfr3 and Fgfr4 and cofactors alpha and beta klotho in differentiating epididymal and inguinal adipocytes. Subcutanous, inguinal and visceral, epididymal white adipocytes were grown to confluence in Dulbecco's modified Eagle's medium with 4.5 g/l glucose, supplemented with 20 nmol/l insulin, 1 nmol/l triiodothyronine, 20% fetal bovine serum, and penicillin/streptomycin within 96 hours. Upon reaching confluence, differentiation was induced with 500 mmol/l isobutylmethylxanthine, 250 mmol/l indomethacine, and 2 mg/ml dexamethasone for 24 hours (induction). Subsequently, the cells were cultured for another six days (differentiation). A subset of culture plates were harvested every 24 hours starting after 48 hours in the proliferation phase. Total RNA was isolated using TRIzol reagents, RNA quality and concentration was measured by photometric analysis, and 5 µg of total RNA was reverse-transcribed to cDNA. Finally, quantitative analysis of Fgfr1-4 expression was performed. Analysis of 5 independent experiments is shown. Fgfr1-3 are constantly present during all stages of differentation in both cell lines. Fgfr4 is only little expressed in proliferating cells and induced during the induction phase at day 2 and peaks soon after the change to differentiation medium at day 3. Alpha Klotho is expressed in negligible amounts at all times while beta Klotho continously increased with ongoing differentiation.

Figure 6.

Implantation of Ffg8b releasing pellets into the visceral, epididymal white adipose tissue depot leads to recruitment of brown adipocytes. A - Representative photographs centered on the implantation site of three control pellets (upper panels) and three Fgf8b pellets (lower panels). The epididymal fat depot is discernible in light gray shades surrounded by darker non-adipose tissues. The area of visible browning is marked by a dotted line. B - To highlight "visual browning" in this gray-scale images we digitally removed all pixels with a colour saturation of more than 162 (on a scale of 0 - 255). Here, white areas correspond to brown colour on a background of whitish adipose tissue.C - The mRNA abundance of several brown adipocyte marker

genes in the entire depot. Ucp1 and Cox7a1 mRNA are increased by Fgf8b treatment but not by the placebo pellet.

Figure 7.

Physiological data of animals implanted with drug release pellets. A - The absolute body mass of mice was not altered by Fgf8b. B - Both placebo and Fgf8b implanted mice lost a comparable small amount of body mass during the three weeks of the experimental procedure. C - The masses of the implanted epididymal fat depot and the contralateral non-implanted depot were not different. Interscapular brown adipose tissue was not different in placebo and Fgf8b treated animals. D - The implanted adipose tissue depot did not differ in body mass specific mass between Fgf8b and placebo treated mice. E - Plasma metabolites of carbohydrate and lipid metabolisms were not affected by Fgf8b treatment (GLU - glucose, CHOL - total cholesterol, nHDLc - non-HDL cholesterol, HDL - high density lipoproteins, LDL - low density lipoproteins, VLDL - very low density lipoproteins, TRIG - triglycerides). F - Liver enzymes in the plasma were not altered by Fgf8b treatment.

Figure 8.

Fgfr4 expression correlates with Fgf8b sensitivity. A- Fgfr4 and alpha klotho are present in the epididymal adipose tissue depot but not in the inguinal depot. B - All analyzes receptors/cofactors were present in epididymal adipose tissue, while the amounts of Fgfr4 in inguinal fat is neglecable. C - Comparison of Fgf8b sensitivity and Fgfr4 expression in cultured adipocytes. Filled squares denote the Fgfr4 mRNA abundance at different days of differentiation. Empty squares give the Ucp1 mRNA abundance of fully differentiated adipocytes after Fgf8b treatment at the respective day of differentiation. Both Fgfr4 expression and Fgf8b sensitivity correlate. D – FGFR3 splice variant c mRNA expression in fold of day 1 at the respective day of differentiation.

Figure 9.

The primary structure of the FGF receptors (FGFR). FGFRs each contain three immunoglobulin-like domains (Ig I-III), a single transmembrane domain (TM), and two tyrosine kinase domains (KD1-2). The Ig domains are disulfide-linked and therefore stabilized. Within the Ig III domain are three different exons (exon 7 = A, exon 8 = B,

and exon 9 = C) which encode possible splice alternatives at the N-terminal end of the receptor (Ig III C – exon 7, 9, and 10; Ig III B – exon 7, 8, and 10). The short form of the FGFRs lacks the Ig I domain. There is evidence in the literature that the Ig III C spliceform is epithelial tissue-specific and the Ig III B form is mesenchymal tissue-specific. sp represents the signal peptide; sc represents an acid box. The heparin-binding site (HBS) is marked by a circle in Ig II domain.

Figure 10. Strategy to generate a mouse model with adipocyte specific, inducible expression of FGF8b.

On the exchange vector, a loxP (lox) flanked stop cassette interferes with FGF8b coding sequence ("transgene") expression. It also carries a neomycin (neo) resistance gene. Both cds carry polyadenylation signals. The vector construct is recombined into the Rosa26 target mouse line. The resulting mouseline is crossed with an existing one expressing CreERT2 driven by the adiponectin (adipoq) promotor. Activation of CreERT2 by OHT treatment leads to removal of the stop cassette and FGF8b expression.

Figure 11: Experimental setup for pellet implanted mice.

Mice are fed with purified diets, either low fat control diet (LFD) or a high fat diet (HFD) for 8 weeks in total. Depending on the optimal treatment time "x", the timepoint is chosen to implant pellets releasing FGF8b. After several weeks of treatment oral glucose tolerance tests (oGTT), norepinephrine tests (NE-test), cold tolerance tests are performed and plasma and tissue samples are collected. Body mass, body composition and food intake is determined on a regular basis.

Figure 12: Experimental setup for transgenic mice.

Mice of the C57BL6/N strain are fed with a diet free of genistein followed by a week of OHT treatment to induce FGF8b expression in white adipose tissue.. Afterwards, mice are fed purified diets, either low fat control diet (LFD) or a high fat diet (HFD). Regularly, oral glucose tolerance tests (oGTT) are performed and body mass, body composition and food intake is determined. In one group a norepinephrine tests (NE-test) and a cold tolerance test is performed. In all groups plasma and tissue samples are collected.

Figure 13: FGF signaling.

Canonical FGF signaling involves dimerization of a FGF receptor. Binding of the ligand FGF to the dimer is stabilized either by a klotho cofactor (endocrine FGFs) or by heparan sulfate (paracrine FGFs). FGF receptors trigger three intracellular signaling cascades: the PI3K/AKT, PLC/PKC and MAPK. Non-canonical FGF signaling is mediated by syndecans interacting with heparan sulfate and an FGF to aktivate intracellular PKCα or, alternatively, by an integrin heterodimer interacting with an FGF receptor to activate the intracellular MAPK cascade.

The Example illustrates the invention.

Example 1: Paracrine FGFs induce a brown adipose tissue (BAT) phenotype in white adipose tissue and are therefore useful in therapy of disorders and diseases of energy homeostasis (including obesity, diabetes, dyslipidemia, insulin resistance, hyperglycemia or metabolic syndrome)

Material & Methods

Fibroblast growth factors

We obtained fibroblast growth factors (FGFs) 1, 2 and 9 of murine origin and human FGFs 5 and 16-21 from PeproTech (Hamburg, Germany). The murine FGFs 4, 6, 7, 8b, 10, and 23 and human FGFs 3, 8a, 8e, 8f and 22 were purchased from R&D Systems (Minneapolis, Minnesota, United States of America). If not stated otherwise, the concentration used for screening purposes in cell culture were as follows (ng/ml): FGF1 2.5, FGF2 5.0, FGF3 25.0, FGF4 5.0, FGF5 2.5, FGF6 5.0, FGF7 25.0, FGF8a, b, e, and f 25.0, FGF9 1.0, FGF10 50.0, FGF16 2.5, FGF17 10.0, FGF18 5.0, FGF19 100.0, FGF21 10.0, FGF22 125.0 and FGF23 50.0.

Cell culture

Preadipocytes were isolated from the stromal vascular fraction of subcutaneous inguinal or visceral epididymal white adipose tissue of newborn wild-type mice. Cells were immortalized by infection with a puromycin resistance-conferring retroviral vector encoding the Simian Vacuolating Virus 40 large T antigen (SV40 T-antigen) and selected with puromycin as published previously (J. Klein 2002). Cells were

grown to confluence in Dulbecco's modified Eagle medium (4.5g/l glucose, GE Healthcare Bio-Sciences Corp, Piscataway, New Jersey, USA) supplemented with 20% fetal bovine serum (Life Technologies, Carlsbad, California, USA), 20nM insulin and 1nM T3. Adipocyte differentiation was induced by complementing this medium with 250µM indomethacin, 500µM isobutylmethylxanthine and 2µg/ml dexamethasone for 24h after confluence. Cell culture was continued for up to six more days. Differentiated adipocytes were used between passages 10 and 30.

Quantitative PCR

Total RNA was isolated using the Qiazol reagent (Qiagen, Hilden, Germany). Quality of RNA was tested by photometric analysis and agarose gel electrophoresis. 5µg of total RNA were reverse transcribed using the iScript cDNA Synthesis Kit (Biorad, Hercules, California, USA) in a 20µl reaction. Target mRNAs were amplified in a total volume of 25µl containing iQ SYBR Green Supermix (Biorad, Hercules, California, USA) and 10pmol of each primer using the Mastercycler realplex 2 detection system (Eppendorf, Hamburg, Germany). The mRNA abundance was normalized to the expression of either beta-actin or Hsp90 as housekeeping genes. The following primers were used: Actb AGAGGGAAATCGTGCGTGAC and CAATAGTGATGACCTGGCCGT, Cidea TGCTCTTCTGTATCGCCCAGT and GCCGTGTTAAGGAATCTGCTG, Cox7a1 CCGACAATGACCTCCCAGTA and TGTTTGTCCAAGTCCTCCAA, Elovl3 TCCGCGTTCTCATGTAGGTCT and GGACCTGATGCAACCCTATGA, Foxc2 ACGAGTGCGGATTTGTAACC and CAGTTTGGGGAGGGACCTAT, Hsp90 AGGAGGGTCAAGGAAGTGGT and TTTTTCTTGTCTTTGCCGCT, Otop1 GGACCTGATGCAACCCTATGA and ACCATGCTCTACGTGCTGTG, Ppargc1a GGACGGAAGCAATTTTTCAA and GAGTCTTGGGAAAGGACACG, Prb TAAACATCTCCCAGCGGAGT and ACAACCATGAGCCAGGAGTC, Prdm16 CTGTTAGCTTTGGAGCCGAC and GACGAGGGTCCTGTGATGTT, Ucp1 TCTCTGCCAGGACAGTACCC and AGAAGCCCAATGATGTTCAG, Fgfr1 CCGGATCTACACACACCAGA and CCACCAACTGCTTGAACGTA, Fgfr2 AGGGACACAGGATGGACAAG and AAACACAGAATCGTCCCCTG, Fgfr3 ACCGAGTCTACACCCACCAG and TGAGGATGCGGTCTAAATCC, Fgfr4 TGGAAGCTCTGGACAAGGTC and ATACAACATTGCTGCTCCCC, aklotho GGCTCAACTCTCCCAGTCAG and CGCAAACTAGCCACAAAGGT, bklotho ATGTCCAGGAGGCTCTGAAA and

AGCAAATGGTGCAGTCTGTG, Fgfr3c CTCCTTGTCGGTGGT and ACGGCACGCCCTACG.

Animal experimentation

Pellets 1.5mm in diameter were fabricated from a biodegradable matrix to locally release 100ng FGF8b per day for 21 days (Innovative Research of America, Sarasota, Florida, USA). Matching placebo pellets did not contain FGFs. To implant a pellet into the subcutanous adipose tissue, a mouse of the 129Sv/ev strain was anesthetized, abdominal fur removed and the pellet pushed into the depot through a short cut in the skin. To target the visceral adipose tissue, the pellet was introduced through a small cut each into skin and abdominal wall and placed between lobes of epididymal adipose tissue.

Mice were kept in a specific pathogen free barrier facility at room temperature in a 12:12 hour light:dark cycle with free access to food and water. Three weeks after implantation mice were killed by carbon dioxide exposure and dissected. Blood parameters were analyzed with an automated clinical chemistry analyzer (Piccolo xpress system, Abaxis, Darmstadt, Germany). All animal experiments were performed according to the German animal welfare law (permission no. 55.2-1-54-2532-174-11).

Introduction

Brown adipose tissue (BAT) is an organ equipping mammals with a means of non-shivering thermogenesis. In brown adipocyte mitochondria, uncoupling protein 1 (Ucp1) allows re-entry of protons from the intermembrane space into the matrix bypassing ATP synthase and thus uncoupling oxygen consumption from ATP production. By this mechanism, the energy stored in the form of proton motive force is released as heat (reviewed in (Klingenspor, 2012)).

BAT and its ability to combust nutrient energy into heat has recently gained increased attention after the repeated and convincing demonstration that adult healthy humans possess appreciable amounts of metabolically active BAT (Cypess et al., 2009; van Marken Lichtenbelt et al., 2009; Virtanen et al., 2009). Physiological

or pharmacological activation of BAT thermogenesis may prove effective in treating some of the most widespread diseases of our time including obesity, diabetes and dyslipidemia. The amount of human BAT, however, is limited and estimated to account for approximately 0.05-0.1% of body mass as compared to a far more than 10-fold higher amount in mice (Virtanen et al., 2009). Thus, to therapeutically employ the unique capabilities of BAT, not only acute activators are required, but also agents that recruit a greater number of brown adipocytes.

The term "brown adipocytes" refers to all types of thermogenic, UCP1 expressing and/or multilocular cells. These are sometimes categorized into "classical brown" versus "beige" or "brite" and others. The term "brown adipocytes" is intended to encompass all of this, e.g. "brown adipocytes in white adipose tissue" is synonym to both "beige" and "brite".

Brown adipocytes are not restricted to uniform, classical BAT depots but are often found interspersed in white adipose tissue (WAT) depots. This second type of brown adipocyte has been termed beige or brite (brown in white) and seems to emerge from a different progenitor cell than classical brown fat cells (reviewed in (Pfeifer & Hoffmann, 2014)). To convert WAT into BAT by means of recruiting brite cells offers a possibility to massively increase the BAT amount accessible to therapeutic activation and at the same time decreases the amount of WAT, thereby replacing an energy-storing organ with an energy-dissipating one. This browning of white fat has been subject to intense research during the last years and several systemic interventions have been identified increasing the number of brite cells in mice, including cold exposure and treatment with β-adrenergic agonists or cardiac natriuretic peptides (Bordicchia et al., 2012; Fisher et al., 2012; Guerra, Koza, Yamashita, Walsh, & Kozak, 1998; Young, Arch, & Ashwell, 1984).

FGFs can be grouped by their mechanism of action into intracellular, paracrine and endocrine peptides (reviewed in (Itoh & Ornitz, 2008)). FGF21 belongs to the small group of endocrine FGFs and is therefore able to exhibit systemic effect on multiple target tissues. The largest group is formed by the paracrine FGFs which feature a protein domain binding to extracellular matrix components and are thereby less mobile and not found in circulation. Their matrix anchor also serves to stabilize

interaction with FGF receptors, while endocrine FGFs require an additional cofactor of the klotho family for that purpose. Paracrine FGFs can be expected to act locally on the target tissue they are released into.

Results

It is shown herein that paracrine fibroblast growth factors FGF8 and FGF17 are active in browning visceral adipose tissue.

Screening of paracrine fibroblast growth factors in immortalized white adipocytes

Paracrine fibroblast growth factors are encoded by a gene family of 13 members designated Fgf1-10 and Fgf16-18. We obtained the respective peptides of recombinant murine or human origin to screen their potential to induce the expression of the brown adipocyte specific gene uncoupling protein 1 (Ucp1) in white adipocyte cell lines. We chose two immortalized white adipocyte lines established from primary stromal-vascular cells isolated from the murine inguinal and epididymal adipose tissue depot. Treatment of the cells was started after induction and continued for the entire differentiation period of 6 days. The concentration chosen for each factors was based on the biological IC50 value determined in fibroblast proliferation assays by the supplier and ranged between 1 and 50 ng/ml. Of 13 fibroblast growth factors tested, Fgf8 strongest induced Ucp1 mRNA abundance of both fully differentiated adipocyte cell lines (epididymal: 27.4-fold, inguinal 2.1-fold) (Fig. 1A). The murine FGF8 gene gives rise to 8 differently spliced transcripts leading to 8 different peptide factors Fgfa-f of which we initially tested the major spliceform Fgf8b (Fig. 1C). We compared the ability of those 4 murine isoforms that are also present in humans (Fgf8a, b, e, and f) to induce Ucp1 mRNA abundance (Fig 1B). In adipocytes of inguinal origin, Fgf8b was the only isoform with browning potential, while in epididymal adipocytes Fgf8b and Fgf8f were both effective. For further experiments we chose the most potent spliceform Fgf8b which is the dominant spliceform in mice and humans with a completely identical amino acid sequence in both species.

FGF8b dose dependently induces a brown adipocyte phenotype in white adipocytes

We treated inguinal and epididymal adipocytes with different concentrations of Fgf8b during the entire differentiation phase of 6 days. In both cell lines, the highest concentration of 125 ng/ml proved most effective (Fig 2A). In epididymal cells, Fgf8b induced Ucp1 mRNA abundance in a dose dependent manner, while in inguinal cells only the highest dose increased Ucp1 expression in an above threshold manner.

Abundance of the brown adipocyte marker *cell death-inducing DNA fragmentation factor alpha like effector A* (Cidea) mRNA was increased by Fgf8b treatment similarly to Ucp1 with a greater effect size in epididymal as compared to inguinal adipocytes. Subunit of complex IV 7a1 (Cox7a1) and elongase of very long chain fatty acids 3 (Elovl3) mRNA was not or only slightly increased by Fgf8b treatment. The master regulator of mitochondrial biogenesis, PPAR gamma coactivator 1a (Pgc1a), was strongly downregulated in fully differentiated adipocytes treated with Fgf8b.

FGF8b reprograms both proliferating and differentiating preadipocytes

During differentiation in culture, immortalized (pre-)adipocytes undergo drastic changes in morphology, gene expression signature and function. The sensitivity towards an external stimulus can thus vary between different stages of differentiation. In particular the browning of white adipose tissue has been proposed to either include the transdifferentiation of mature white adipocytes or to be caused by the differentiation of a certain pool of precursor cells. To assess the sensitive time window for FGF8b induced browning, we treated inguinal and epididymal white adipocytes during different days of proliferation, induction and/or differentiation and measured the final Ucp1 mRNA abundance after full differentiation.

Adipocytes of inguinal origin were only responsive to FGF8b treatment in their fully differentiated state (Fig. 3B). Thus, the effect of prolonged treatment during the entire differentiation phase can be attributed to this late, sensitive period. While epididymal cells also respond to a comparably late treatment, they display a second even more sensitive time window during early differentiation directly following induction (Fig. 3A) and a third during proliferation. Interestingly, treatment during the induction phase is

not only ineffective. Including the induction day furthermore abrogates the browning effect of otherwise sensitive treatment regimes.

The sensitivity of proliferating epididymal adipocytes towards FGF8b allowed us to follow the expression of brown adipocyte characteristic transcripts in a timecourse experiment covering the entire differentiation phase. We treated cells for 48 hours prior to induction and took samples immediately afterwards, on day 1 post induction (p.i.), day 2 p.i., day 4 p.i. and after full differentiation at day 6 p.i.. The mRNA abundance of four brown adipocyte marker genes (Ucp1, Cidea, Cox7a1 and Elovl3) was increased in mature adipocytes treated with FGF8b during proliferation with Cidea and Elovl3 already increasing on day 4 p.i. and Ucp1 and Cidea following on day 6 p.i. (Fig. 4). Interestingly, Pgc1a was acutely downregulated by FGF8b treatment in line with our previous data of epididymal cells continuously treated during differentiation (Fig 2B). However, early treatment during proliferation still led to increased Pgc1a mRNA abundance after complete differentiation. The transcription factor Prdm16, presumably implicated in brown adipocyte differentiation, displays a transient increase in abundance directly following induction. The transcription factors Foxc2 and pRb do not acutely respond to FGF8b treatment.

Fgf receptor expression in immortalized adipocytes

The cellular response to FGFs is mediated by FGF receptors (FGFR). A variety of FGFRs is produced from four different genes by differential splicing (FGFR1-4). We quantified mRNA abundance of transcripts of all four genes with primers that do not differentiate between individual spliceforms during every day of adipocyte differentiation in cell culture (Fig. 5). Transcripts of all four genes were present in both cell lines at every timepoint. While FGFR1-3 transcripts did not display marked expression changes during differentiation, FGFR4 was clearly less abundant in proliferating cells and strongly upregulated upon induction and early differentiation. The binding of endocrine FGFs further requires the presence of one of the klotho co-factors, alpha-klotho or beta-klotho. Paracrine FGFs are not considered to require a cofactor for FGFR binding because their affinity for extracellular matrix components stabilizes ligand-receptor interaction. We nevertheless determined transcript abundance of the klotho genes to not overlook a possible, previously non-

appreciated role in FGF8b signaling. Alpha-klotho was hardly detectable at all in any sample and accordingly displayed a very high variability. Beta-klotho strongly increased in abundance during differentiation from very low levels in proliferating cells to strong expression in mature adipocytes.

FGF8b transforms epididymal white adipose tissue into brown adipose tissue in vivo

To investigate the ability of FGF8b to transform white adipose tissue into brown adipose tissue in vivo, we obtained pellets that release 100ng FGF8b per day and placebo pellets. One pellet each was implanted into the epididymal white adipose tissue of adult male 129Sv/ev mice. After three weeks mice were sacrificed and the tissue analyzed. Final body weight was comparable between the FGF8b and the placebo group as well as the change in body weight during the 3 weeks of treatment (Fig. 7). The depot mass of implanted and non-implanted epididymal fat and of interscapular brown adipose tissue was not different between groups. Accordingly, the relationship between depot mass and body mass was not influenced by FGF8b treatment (Fig. 7D). A panel of blood parameters including glucose, plasma lipids and liver enzymes did not differ between FGF8b and placebo treated animals. In summary, we found no evidence of any gross metabolic difference between treatment groups.

By visual inspection, the implantation site of FGF8b pellets and the surrounding adipose tissue was of a brown colour, while both the non-implanted contralateral depot and the implantation site of placebo pellets remained characteristically white (Fig. 6A). The browned area extended approximately 3 mm away from the implanted pellet in line with the effect of an infused paracrine factor. The individual effect size varied considerably with the final position of the pellet within the depot. Implantation of FGF8b pellets into inguinal adipose tissue did not lead to visual browning or increased expression of brown adipocyte marker genes.

We measured mRNA abundance of brown fat marker genes. Ucp1 and Cox7a1 were increased when measuring in a RNA preparation of the complete unilateral depot (Fig 6).

FGF receptor 4 expression correlates with FGF8b sensitivity

The different response of epididymal and inguinal adipose tissue to FGF8b treatment might be explained by a different expression of according receptors. We thus measured mRNA abundance of all FGF receptors and klotho co-receptors in the two depots of untreated male mice. The three receptors Fgf receptor 1-3 and beta-klotho were well detectable in both depots and of comparable abundance (Fig. 8A-B). Appreciable amounts of Fgfr4 and alpha-klotho were only found in epididymal adipose tissue, while only traces were present in inguinal fat.

Since Fgf receptor 4 displayed an expression pattern that may account for the different responsiveness of inguinal and epididymal adipose tissue to FGF8b, we also compared responsiveness of immortalized cultures adipocytes with Fgf4 receptor mRNA abundance. In both inguinal and epididymal cells receptor expression and responsiveness displayed a similar pattern. In inguinal adipocytes the common peak is detected in fully differentiated, mature adipocytes. In epididymal adipocytes both parameters display a maximum during early differentiation.

Discussion

The fibroblast growth factors (FGF) gene family comprises 22 members that are considered key players in proliferation and differentiation of a wide variety of cells and tissues. Most FGFs mediate their biological effects as secreted, extracellular proteins by binding to and activating cell surface tyrosine kinase FGF receptors. The FGF protein family as well as the FGF receptors are highly conserved across species. Three distinct subgroups of FGFs can be defined: endocrine FGFs (19-23), paracrine FGFs (1-12 & 16-18) and intracellular FGFs (11-14).

Members of the endocrine FGFs, especially FGF19 and FGF21, are implicated in energy homeostasis and reported to be activators of brown adipose tissue. In addition, FGF21 has been reported to induce browning in white adipose tissue. In principal, endocrine and paracrine FGFs share the same set of receptors FGFR1-4. Thus,we screened all paracrine FGF proteins for their potential to induce Ucp1 gene expression in white adipocytes.

Interestingly, we identified FGF8 and FGF17 of the paracrine FGF8-like subfamily to be able to strongly induce Ucp1 mRNA expression in subcutaneous, inguinal and visceral, epididymal adipocytes. Of four different FGF8 spliceforms, FGF8b most potently led to UCP1 expression in inguinal and epididymal adipocytes. Thus, further studies were carried out with this paracrine peptide.

FGF8b increased the mRNA expression of Ucp1 and further brown adipocyte marker gernes in epididymal adipocytes. A timecourse treatment during differentiation identified two separate sensitive time windows: epididymal adipocytes responded to FGF8b during two days following induction of differentation, while both inguinal and epididymal adipocytes reacted to acute FGF8b treatment once fully differentiated.
After treatment of epididymal cells during the first day of differentation, several marker genes of mature brown adipocytes were first upregulated during day 4 (Cidea, Elovl3) or day 6 (Ucp1, Cox7a1) of differentiation. The timespan of several days between treatment of early preadipocytes and first marker gene expression in fully differentiated cells clearly indicates a reprogramming of the differentation process and not a direct, acute effect on marker gene transcription. The well-known regulator of early brown adipocyte differentiation, PRDM16, however, was immediately and transiently upregulated upon FGF8b treatment. Possibly, the FGF8b effect is intracellularly mediated by the PRDM16 signaling axis.

We determined mRNA abundance of FGF receptors during differentiation of adipocytes to identify candidate receptors transducing the FGF8b effect. FGFR1-3 were constantly present during all stages in large amounts. FGFR4 was drastically upregulated directly following induction and thus displayed a similar pattern as FGF8b sensitivity of these cells. In murine tissue samples, FGFR4 was clearly present in epididymal adipose tissue while only much lower trace amounts were determined in the inguinal fat depot. Taken together, FGFR4 is a candidate receptor to transduce browning induced by FGF8b.

The efficacy of FGF8b to convert white into brown adipose tissue was determined in vivo. Drug release pellets were designed to locally release 100nmol FGF8b per day and implanted into the epididymal adipose tissue depot. After three weeks, the

adipose tissue surrounding the implanted FGF8b pellet turned visibly brown, while placebo pellets did not display a similar phenomenon. The expression of brown fat marker genes increased in FGF8b treated depots. The small effect size is probably due to a dilution effect of the large amounts of unconverted, white adipose tissue surrounding the affected region. Indeed, the entire depot was used to prepare the RNA sample for gene expressin analysis.

Neither body mass, nor fat mass was affected by pellet implantation. Typical metabolic blood parameters (including glucose, triglyceride and liver enzymes) did not change upon FGF8b pellet implantation. Taken together we find no evidence for any systemic adverse reaction to FGF8b.

In summary, FGF8b is able to locally turn visceral, epididymal white adipocytes into a cell type resembling brown adipocytes in cell culture and in vivo without evidence for a systemic effect.

The herein provided experiments follow a sequence of the following rationale: First, peptides were identified that reprogram white adipocytes to display brown adipocyte characteristics in a cell culture screen. Second, mechanistic details were characterized in a cell culture model. Third, the principal transferability of the cell culture data was confirmed in an animal model. The efficacy of recruiting brown fat cells within white adipose tissue to treat a number of metabolic diseases including obesity, diabetes and dyslipidemia can be confirmed in animal experiments and clinical studies.

Specifically, the lack of changes in parameters of blood chemistry and body composition in the mouse experiments does not argue against a possible therapeutic benefit of such treatment. These experiments were specifically designed and intended to demonstrate that FGF8b is able to recruit brown fat in vivo. The miniscule amount of recruited brown fat by implantation of a single pellet of the employed low dose was not expected to affect metabolic parameters. Conversely, the absence of such alterations even proves the lack of any dramatic side effects.

The in vivo experiments were conducted in lean, healthy mice, not in disease models. Amelioration of disease parameters can thus not be the expected outcome of the experiment (as these were absent to start with).

The amount of brown adipose tissue in a mouse, which has been therapeutically activated to help against metabolic disease in the literature, is in the range of 1% of body mass. To generate additional brown adipose tissue by FGF8b in an amount to notably increase this background, the dose, treatment time and/or pellet number can be increased. In addition, a transgenic mouse model expressing FGF8b in white adipose tissue can be characterized.

The therapeutic benefit of brown adipose tissue is a function of tissue mass and sympathetic tone (sympathetic catecholamines are activators of brown fat activity). The more brown fat is recruited in white adipose tissue with its specific, given sympathetic tone, the more therapeutic activity must be expected. In addition to brown fat recruitment by a paracrine FGF, , the sympathetic tone can be increased (i.e. by fasting) or mimicked by co-administration of sympathomimetic drugs.

In summary, it was not the goal of the mouse experiments provided the above to prove the applicability of brown fat recruitment for the treatment of obesity, diabetes and dyslipidemiaThe in vivo experiment aimed at confirming brown fat recruitment by FGF8b and successfully did so. In view of this demonstrated effect it is credible that paracrine FGFs like FGF8b can be used in the therapy of metabolic diseases, because the art recognizes the link between brown fat recruitment and therapy of metabolic diseases.

The above results can be validated in appropriate animal experiments as follows.

Animal models can be used to determine the physiological consequences of FGF8b generated brown adipose tissue.

For example, pellet treatment can be optimized. In the experiments provided above, pellets were implanted releasing FGF8b into the visceral white adipose tissue of

mice. The pellet design in relation to the parameters peptide amount, release rate and treatment duration can be optimized.

In addition to the pharmacological application of the active agent a mouse model of inducible, white fat specific FGF8b expression can be generated. This model allows a uniform treatment of the entire white adipose tissue for an indefinite time period and will be highly versatile to study the mechanisms underlying brown adipocyte recruitment.

Furthermore, physiological consequences of FGF8b induced brown adipocyte recruitment can be determined. Upon activation, brown adipocytes release chemical energy into heat by oxidizing lipids and glucose, making them an attractive target cell type for the treatment of diseases and disorders of energy homeostasis, including diabetes, obesity and dyslipidemia.

To validate the functionality of FGF8b recruited brown fat cells *in vivo*, the above mouse models can be assessed in respect to their thermogenic capacity, glucose tolereance and resistance towards diet induced obesity. The activity of FGF8b can be increased, for example by different manipulations to stimulate thermogenic activity including treatment with sympathomimetics and activation of the sympathetic nervous system by cold and fasting.

Brown adipocytes recruited by FGF8b need to be activated. The therapeutic benefit of brown adipose tissue is a function of tissue mass and sympathetic tone (sympathetic catecholamines are activators of brown fat activity). The more brown fat is recruited in white adipose tissue with its specific, given sympathetic tone, the more therapeutic activity must be expected. In addition to brown fat recruitment by a paracrine FGF, the sympathetic tone can be increased (e.g. by fasting) or mimicked by co-administration of sympathomimetic drugs.

As a first step, two in vivo model systems will be established. The local release of a paracrine peptide by implanted pellets serves as an ideal model for an therapeutic application in human medicine, while a transgenic mouse model of inducible, white

fat specific FGF8b expression will be highly versatile to further study the mechanisms underlying brown adipocyte recruitment.

Model establishment: Optimization of pellet treatment

FGF8b action is limited to paracrine targets due to an anchor sequence interacting with extracellular matrix components. It can thus be applied locally to a target tissue without affecting other tissues via distribution in the bloodstream. For this purpose, drug release pellets, 3mm in diameter, were implanted into white fat in the above provided experiments. These pellets (Innovative Research of America, www.innovrsrch.com) were produced from a biodegradable carrier matrix and recombinant FGF8b (R&D Systems) to release 100ng peptide per day for 3 weeks. The experiments were performed in a specific pathogen free (SPF) mouse research unit at the TUM.

For further experiments, the required amounts of FGF8b (~1mg) are obtained from the same supplier (R&D Systems) and pellets are produced commercially as outlined below (Innovative Research of America, www.innovrsrch.com).

In this first part, it is aimed to optimize pellet design with respect to dose and treatment duration. Pellets of the known, effective dose (100ng/d) are implanted into the epididymal white adipose tissue of male mice and three different treatment durations of continuous release (1 week, 3 weeks, 6 weeks) are compared. Once the optimal duration is validated, the release rate (10ng/d, 100ng/d and 1µg/d) is varied. Pellets with a respective release rate and depot size of FGF8b can technically be produced according to the manufacturer. All pellet treatments are evaluated in comparison to appropriate placebo pellets.

In the above experiments, mice of the 129Sv/ev-S6 strain were employed. The 129Sv/ev-S6 strain is known for its high number of brown adipocytes in white fat. On the one hand, that may indicate a high sensitivity to browning stimuli like FGF8b. On the other hand, it may decrease the maximal effect size possible. Therefore, in parallel mice of the C57BL6/N mice are used. The extensive phenotyping described herein is limited to the strain proving more susceptible to FGF8b-induced browning.

Color and macroscopic appearance of the implantation sites is documented photographically. To quantify effectiveness of a treatment mRNA abundance of brown adipocyte marker genes (Ucp1, Cidea, Cox7a1, etc.) is determined by quantitative PCR (qPCR) and histological sections are generated.
All RNA and tissue samples generated during this work block are archived for further investigation as outlined below.

Model establishment: Generation of a transgenic mouse model

Release of FGF8b into the visceral, white adipose tissue led to recruitment of brown adipocytes in the experiments provided above. As a complement to local application in the form of pellets (see above), it is aimed to generate a mouse model that intrinsically produces FGF8b in white adipocytes. By this method, the entire white fat can be treated in a uniform manner and for any duration desired. While pharmacological treatment is an ideal model system for a possible therapeutic application, this transgenic mouse line with its envisioned greater effect size is optimal to study the molecular mechanisms behind brown adipocyte recruitment and its metabolic consequences.

A mouse line already established is used, which expresses CreERT2 recombinase under the control of the white fat specific adiponectin promotor. The CreERT2 fusion protein consists of a Cre recombinase and a variant estrogen receptor insensitive to estradiol, but binding to 4-hydroxytamoxifen (OHT). Upon treatment with OHT the fusion protein relocates from the cytosol into the nucleus and allows Cre-mediated recombination. This mouse line is of the C57BL6/N strain.

A further required mouse line of the same strain is generated by a commercial service provider (such as TaconicArtemis). The required mouse line is intended to comprise an expression construct inserted into a defined locus driving FGF8b expression from a ubiquitously active promotor (Fig. 10). This construct is silenced by a stop-cassette flanked by loxP-sites. The stop cassette can be removed by the action of Cre recombinase which will lead to expression of the otherwise silent gene. By interbreeding this FGF8b mouse with the CreERT2 mouse line a mouse model

with white adipocyte specific (adiponectin promotor), inducible (OHT inducible CreER2t) expression of FGF8b is generated.

Activation of CreERT2 is possible by feeding a diet containing OHT (400mg/kg). After a washout phase with a diet devoid of soy genistein for one week, OHT-containing diet is fed ad libitum for a further week. This regime leads to an approximate uptake of 3mg OHT per mouse and day which is effective and well tolerable.

The resulting mouse model is extensively phenotyped.

Molecular characteristics of in vivo recruited brown adipocytes

Both animal models described above (pellet implanted & transgenic) are expected to display FGF8b induced emergence of brown adipocytes in white adipose tissue. The measurements outlined here aim to characterize these cells on the cellular and the molecular level. FGF8b-treated white adipose tissue is compared with other adipose tissue depots: interscapular brown fat exclusively consists of classical brown adipocytes, inguinal white fat contains brite adipocytes and untreated epididymal white fat is considered nearly purly white. This comparison will elucidate whether FGF8b recruited brown adipocytes are rather similar to brown or brite adipocytes or even constitute a different, novel class of adipocyte.

Functional characterization

A remarkable feature of brown as compared to white adipocytes is the excessive respiratory capacity conferred by a large number of mitochondria with dense christae. A change in mitochondrial amount on the level of mitochondrial enzyme activity, specifically citrate synthase of the TCA cycle and complex IV of the respiratory chain is detected. The maximal activity of both solubilized enzymes can be measured under conditions of substrate excess and constitute a surrogate measure for mitochondrial abundance and respiratory capacity. The respective assays are routinely applied.

Furthermore, brown and white adipocyte mitochondria differ in their substrate preference. Glycerol-3-phosphate (G3P) is oxidized preferentially by brown fat mitochondria due to more abundant mitochondrial G3P dehydrogenase (Chaffee, Allen, Cassuto, & Smith, 1964; Gong, Bi, Weintraub, & Reitman, 1998). Sufficient amounts of mitochondria might not be isolated from the small adipose tissue explants. Therefore floating cells from collagen-dissociated tissue can be analyzed. Oxygen consumption of such homogenates are measured in a respirometer (Oroboros O2k Oxygraph) in the absence/presence of different substrates to detect a possible shift in preference between placebo and FGF8b implanted mice.

Transcriptome analysis

Biopsies are subjected to a transcriptome analysis by next generation sequencing (NGS). By comparison of expression patterns to classical brown, white and brite adipocyte insight into the cellular identity of FGF8b generated brown adipocytes is gained. Furthermore, candidate signal transduction pathways activated by FGF8b treatment are identified and tested experimentally.

Importantly, transcriptome analysis will allow identification of both signal transduction and effector genes in an unbiased fashion, i.e. in addition to the current knowledge on downstream FGF receptor signaling (Fig.13). Candidate transcripts are validated by qPCR in independent biological samples. These will include tissue samples of both animal models as well as cultured cells.

For the validation of identified pathways cell culture experiments are performed, in which the respective pathway is activated/inhibited/challenged. The exact experimental design strongly depends on the identified target pathways, but may include pharmacological and biochemical compounds and/or physiological stimuli (starvation etc.). Primary outcome will be Ucp1 mRNA expression in fully differentiated cells.

Metabolic consequences of FGF8b-induced recruitment of brown adipocytes

Both animal models described above (pellet implanted & transgenic) are expected to display FGF8b induced emergence of brown adipocytes in white adipose tissue. The physiological function of brown adipocytes is to release chemical energy from nutrient macromolecules into heat. Increasing the number and/or activity of this cell type can thus be expected to lead to the metabolic consequences listed below.

Maximal non-shivering thermogenic capacity: Brown adipocyte non-shivering thermogenesis is under the control of sympathetic catecholamines. An injection of norepinephrine activates the tissue and maximal thermogenic capacity can be assessed by indirect calorimetry (Meyer et al., 2010). Should the FGF8b recruited brown adipocytes be thermogenic, they will contribute to total capacity.

Glucose homeostasis: Active brown adipocytes take up large amounts of glucose and are therefore discussed as a treatment target for type 2 diabetes (T2D). In several mouse models with an increased number of brite adipocytes in white adipose tissue, an improved glucose tolerance has been described (Armani et al., 2014; Bi et al., 2014). Glucose tolerance tests are performed in both mouse models to assess the plausibility of FGF8b as a candidate T2D treatment option. The technique is established (Bolze et al., 2013).

Blood chemistry: Despite a large glucose uptake, this substrate only amounts to 10% of total energy expenditure in active brown adipocytes while the rest reflects lipid oxidation (Virtanen et al., 2009). Lipids are mobilized from intracellular stores or imported from the blood. This import can lead to massive alterations in the plasma lipoprotein pattern possibly beneficial in dyslipidemic patients (Bartelt et al., 2011). Blood chemical and lipoprotein parameters are determined in the mouse models by an automated clinical chemistry analyzer (Abaxis Piccolo Xpress).

Body mass and composition: Non-shivering thermogenesis causes increased energy expenditure. An altered number or activity of brown adipocytes can thus lead to a shift in energy balance and confer resistance to and/or relieve from diet induced obesity. These properties are assessed in feeding trials with both mouse models (see

experimental setup below). Body composition will be analyzed by nuclear magnetic resonance spectroscopy (Bruker MiniSpec).

Activation of brown adipocytes: The activity of brown adipose tissue is controlled by the sympathetic innervation that releases norepinephrine to activate non-shivering thermogenesis. Brown adipocytes residing ectopically in white adipose tissue depots must be expected to be subject to the sympathetic tone in their respective depot. In white adipose tissue, lipolysis and lipid provision is increased in response to a catabolic state of the organism. Possibly, ectopically recruited brown adipocytes can be stimulated by this route and support fat loss during fasting.

The described metabolic consequences of FGF8b recruited brown adipocytes can be assessed in the following experimental setup (Fig. 11 and 12):

Group 1: Resistance to diet induced obesity

The comparison of control versus treated mice will reveal a possible contribution of FGF8b-recruited brown adipocytes to resistance to diet induced obesity and/or improved glucose homeostasis both in lean and in progressively obese, glucose intolerant mice.

In the pellet implantation mouse model (Fig. 11), mice are continuously fed a low fat diet and implanted with either placebo or FGF8b releasing pellets into the epididymal white adipose tissue following an optimized treatment regime as determined earlier. After maximal expected recruitment of brown adipocytes, mice are switched onto a high fat diet for 8 weeks to induce diet induced obesity. Body mass, body composition, food intake and glucose tolerance is monitored.

In the transgenic mouse model (Fig. 12), after activation of white fat FGF8b expression, the development of body mass, fat mass, food intake and glucose tolerance is monitored for 8 weeks feeding a control low fat diet. Subsequently, mice are fed a high fat diet to induce diet induced obesity.

Both the 129Sv/ev-S6 and the C57BL6/N mouse strain are susceptible to diet-induced obesity under these conditions.

Group 2: Activation by catabolic state

During energy restriction, white adipose tissue lipolysis is activated by norepinephrine released from sympathetic nerve fibers. It is observed whether the increased sympathetic tone in epididymal white fat activates FGF8b-recruited brown adipocytes. In that case, the loss of body mass and fat mass as well as the improvement in glucose tolerance would be increased in FGF8b treated animals.

In the pellet implantation mouse model (Fig. 11), mice are fed a high fat diet ad libitum for 8 weeks to induce obesity. During this phase, pellets (placebo or FGF8b) are implanted into the epididymal white adipose tissue at such a timepoint that maximal recruitment of brown adipocytes can be expected to occur in mice with established obesity. A diet change to low fat diet leads to a catabolic state characterized by a loss in body mass and fat mass.

In the transgenic mouse model (Fig. 12), after activation of white fat FGF8b expression, the development of body mass, fat mass, food intake and glucose tolerance for 8 weeks feeding a high fat diet is monitored. Subsequently, mice are fed a control low fat diet to induce a catabolic state.

Group 3: - Activation by norepinephrine.

Injection of the endogenous activator norepinephrine leads to maximal non-shivering thermogenesis in brown adipocytes. Alternatively, mice can be exposed to a series of decreasing ambient temperatures. In both cases, indirect calorimetry can used to determine maximal cold induced thermogenic capacity (norepinephrine) and cold limit (cold), respectively (Meyer et al., 2010; Nau et al., 2008). The measurement with these parameters will reveal a thermogenic contribution of FGF8b-recruited brown adipocytes.

In the pellet implantation mouse model (Fig. 11), mice are implanted with either placebo or FGF8b releasing pellets into the epididymal white adipose tissue following an optimized treatment regime as determined earlier. After maximal expected recruitment of brown adipocytes, thermogenic capacity is determined by indirect calorimetry following injection of norepinephrine. After a recovery period of several

days, mice are subjected to a series of decreasing ambient temperatures to determine the cold limit.

In the transgenic mouse model (Fig. 12), four weeks after activation of white fat FGF8b expression, thermogenic capacity by norepinephrine injection is assessed. After a recovery period of several days, mice are subjected to a series of decreasing ambient temperatures to determine the cold limit.

In all completed study groups, tissue and plasma samples are collected for further analysis, e.g. to determine expression levels of brown adipocyte marker genes and to determine plasma glucose and lipids. Furthermore, tissue samples will be bioenergetically characterized as outlined above, i.e. citrate synthase activity, complex IV activity and comprehensive respirometry. This sample set allows investigating the persistence of recruited brown adipocytes 8 weeks after maximal recruitment.

Further, the used cell culture model can be refined in order to better characterize the signal transduction cascades responsible.

Immortalized white adipocytes were treated during the entire differentiation phase. To refine this model, it is aimed to shorten treatment to 48 hour time windows during different phases of cell differentiation (i.e. during proliferation, during induction, during different days of differentiation) and determine the most effective treatment scheme. Fresh (non-immortalized) primary white adipocytes from several murine adipose tissue depots are prepared to employ in the following experiment characterizing the responsible signal transduction cascade.

FGF8b exerts its effect by binding to a FGF receptor. Candidate receptors are described herein. They can be validated by analysis of responsive cells and tissues and test their relevance by RNA interference.

FGF receptors are known to couple to a number of different intracellular signal transduction cascades. By pharmacological inhibition the cascade necessary for

brown adipocyte recruitment is pinpointed. The known target genes of such a cascade are searched for possible effector gene products.

When receptor and signal transduction cascade are characterized, possible alternative routes of activation are investigated, e.g. shortened FGF8b versions and small molecules interacting with signaling components.

Fibroblast growth factors and their receptors are highly conserved across mammals and beyond. Metabolically active brown adipocytes are found in both mice and man. Thus, a transferability of the results provided herein to the human system is possible.

This can be validated in cell cultures of primary, human adipocytes of subcutaneous and visceral origin. The cells are treated with FGF8b and further effectors as described herein and identified in accordance with the herein provided teaching. Recruitment of brown adipocytes is measured by the expression of brown fat marker genes.

Respirometric assessment of Ucp1 activity in recruited brown adipocytes

In the experiments provided herein, immortalized epididymal white adipocytes with FGF8b were treated. This treatment led to expression of several brown adipocyte marker genes. The mRNA expression of marker genes alone indicates the presence of functional brown adipocytes. However, the functionality of recruited brown adipocytes can be validated and evaluated by respirometry in an extracellular flux analyzer (XFe96, Seahorse Bioscience).

A measurement protocol for the specific detection of functional uncoupling protein 1 (Ucp1) has been developed by Li, Fromme, Schweizer, Schottl, & Klingenspor, 2014). Briefly, oxygen consumption of fully differentiated adipocytes is determined in the presence of bovine serum albumine to buffer free fatty acids. The fraction of respiration attributable to proton leak is determined by addition of the complex V inhibitor oligomycin. Maximal Ucp1 mediated uncoupling is invoked by adrenergic stimulation with isoproterenol and maximal uncoupled respiration by the chemical uncoupler FCCP. Finally, non-mitochondrial respiration is detected by addition of the

complex III inhibitor antimycin A. From these values, specific Ucp1 activity can be detected and quantified. Data from FGF8b recruited brown adipocytes is compared with both untreated white and brown adipocytes.

Characterization of the signal transduction cascade

Identification of the FGF8b receptor

Fibroblast growth factors (FGFs) exert their biological activity by interacting with FGF receptors. While endocrine FGFs require the presence of the cofactor α- or β-klotho, binding of paracrine FGFs is sufficiently stabilized by interaction with heparan sulfate. FGF receptors are tyrosine kinases coded on four different genes FGFR1-4 giving rise to at least seven different transcripts by differential splicing. The specificity of ligand receptor interaction is not fully resolved and FGF8b can bind to at least four different FGF receptor variants.

Alternatively, FGFs can bind to non-canonical target structures that also transmit information into the cell, either alone or in cooperation with classical FGF receptors. These include abundant cell surface proteins of the syndecan family (4 members) and integrin heterodimers (at least 18 α and 8 β subunits = 144) (Murakami, Elfenbein, & Simons, 2008) (Figure 13).

The receptor(s) that mediate FGF8b signaling implicated in brown adipocyte recruitment are validated and assessed as follows. As a first step, a correlation between receptor expression and responsivity of cells and tissues to FGF8b is assessed.

Correlative approach: time frame experiment

Immortalized, primary white adipocytes isolated from murine epididymal adipose tissue were utilized above to identify FGF8b as a potent activator of BAT. In the setup, cells were treated continuously during the entire phase of differentiation. The time window most sensitive to the browning stimulus is, however, very different for several known causative agents. BMP7, for instance, exerts a maximal effect when applied during proliferation, while rosiglitazone is effective during differentiation (Li, Bolze, Fromme, & Klingenspor, 2014; Tseng et al., 2008).

In a time frame experiment, cells are treated for 2 consecutive days during proliferation, induction phase and differentiation phase. All cells are fully differentiated and RNA prepared. Brown adipocyte marker gene and FGF receptor, syndecan and integrin transcript abundance will be measured by qPCR.

Both immortalized and freshly isolated primary cells of different adipose tissue depots are included. This experiment not only shows, which receptor is expressed in a sensitive timeframe and cell type, but also provides insight into the mechanism of brown adipocyte recruitment: sensitivity of proliferating cells would argue for an early determination process while a late sensitivity would argue for transdifferentiation of already differentiated or committed cells.

Correlative approach: Expression panel

Implanting pellets into epididymal adipose tissue led to recruitment of brown adipocytes, while it did not in inguinal adipose tissue. The expression of FGF receptors, syndecans and integrins in both adipose tissue depots and in the respective immortalized primary cell lines is determined to narrow down the range of FGF receptors conferring this effect.

Candidate validation: RNA interference

A knockdown strategy targeting all seven major FGF receptors and identified candidate syndecans/integrins by RNA interference is developed. Established procedures are used based on the viral transfection of vectors encoding a siRNA expression cassette into preadipocytes or alternatively, chemical transfection of pre-made, commercial siRNAs (Hoffmann et al., 2013).

The available knockdown methods are tested to find optimal strategies for the receptors. The crucial validation test will be whether knockdown of a receptor leads to loss in sensitivity to FGF8b mediated brown adipocyte recruitment as measured by marker gene expression. The receptor(s) directly responsible are pinpointed, thereby providing a target for alternative activation. Furthermore, validation of the receptor will provide candidate intracellular signaling pathways to test in the following experiment.

Elucidation of intracellular signaling cascades

Canonical FGF receptor signaling is triggered by FGF binding and subsequent dimerization of the receptor (Figure 13). Several residues of the dimer are autophosphorylated by its tyrosine kinase activity. These sites form docking domains for interacting proteins that in turn activate downstream signaling cascades. Some of the best studied signal transduction pathways emerge from here: the MAPK pathway (Erk1/2, p38, JNK), the IP3 pathway (PLCγ, IP3/DAG, PKCs) and the PI3K pathway (PI3K, AKT). The less studied non-canonical FGF signaling via syndecans and integrin also includes activation of a MAPK cascade (Murakami et al., 2008).

The signaling pathway essential for the recruitment of brown adipocytes by FGF8b by pharmacological inhibition is validated. All three major pathways can be specifically inhibited at multiple sites by commercially available small molecules (Table 1). Such inhibitors are utilized in cultures of immortalized white adipocytes treated with FGF8b and the mRNA abundance of brown adipocyte marker genes (Ucp1, Cidea, etc.) is measured by qPCR. Conversely, activators of candidate signaling cascades to mimic FGF8b action are employed.

To not overlook a possible unknown route of signal transduction, a pathway analysis with the transcriptome data generated above is conducted. Signaling cascades or groups of target genes coordinately regulated by FGF8b treatment are included into the experiment and targeted with small molecule inhibitors/activators.

The following exemplary compounds may be used.

Table 1 - Activators and inhibitors of FGF signaling

Pathway	**Inhibitors**	**Activators**
FGF receptors	None strictly isoform specific known SU11248 (pan-Receptor-Tyrosine-kinase)	Strontium ranelate (Caverzasio & Thouverey)
MAPK	Sorafenib (Raf), SB203580	Anisomycin (p38 MAP and JNK),

	(pan-p38), SP600125 (pan-JNK), Trametinib (MEK1/2), PD98059 (MEK1), SCH772984 (ERK1/2)	PAR C-16 (MEK), t-butylhydroquinone (Erk2)
IP3	Sotrastaurin (pan-PKC), Bisindolylmaleimide 1 (pan-PKC), Gö6983 (pan-PKC), U-73122 (PLCg)	Phorbol 12-myristate 13-acetate (PKC), Cell permeant caged IP3
PI3K	Wortmannin (PI3K), LY294002 (PI3K)	740 Y-P (IP3R), sc-3036 (IP3R)

Table 2 – Activators and inhibitors of paracrine FGFs

	substance	IUPAC International Chemical Identifier (InChI), reference or chemical nomenclature/trivial name
Mechanism: Influencing FGF-heparin-binding		
Inhibitors	naphthalene-1,3,6-trisulfonate	ZPBSAMLXSQCSOX-UHFFFAOYSA-K
	2-O-Bn sucrose heptasulfate	WO 03/038054A2
	1'-O-Bn sucrose heptasulfate	WO 03/038054A2
	1',2-di-O-Bn sucrose hexasulfate	WO 03/038054A2
	6'-O-hexadecanoyl sucrose hexasulfate	WO 03/038054A2
	2-O-dodecanoylhexasulfate	WO 03/038054A2
	6'-O-hexadecanoyl sucrose hexasulfate	WO 03/038054A2
	4,6-O-isopropylidene sucrose hexasulfate	WO 03/038054A2
Activators	Sucrose octasulfate	WEPNHBQBLCNOBB-FZJVNAOYSA-N
	Inositol hexasulfate	NBTMNFYXJYCQHQ-UHFFFAOYSA-N
Mechanism: Supply of heparin for activating the FGFR-FGF-Heparin-complex		

activating	administration of heparin (or of a heparin derivative)	Classical heparin; further heparin derivavites, such as Certoparin, Dalteparin, Enoxaparin, Nadroparin, Danaparoid
Mechanism: Influencing stability of protein conformation		
Stabilizing agent	Alpha-Cyclodextrin and other Cyclodextrinderivatives	HFHDHCJBZVLPGP-RWMJIURBSA-N
Mechanism: Influencing heparanase-mediated degradation of heparan-sulfate proteoglycan (HSPG) oft eh extracellular matrix		
Heparanase (endo-beta-D-glucuronidase heparanase) inhibitor	PI-88 is a mixture of highly sulfated, monophosphorylated mannose oligosaccharides; Name: MUPARFOSTAT	
Heparanase inhibitor	OGT 2115	2-[4-[[3-(4-Bromophenyl)-1-oxo-2-propenyl]amino]-3-fluorophenyl]-5-benzoxazoleacetic acid
Mechanism: Influencing FGFR activity		
FGFR1		
Activator:	SUN11602	4-[[4-[[2-[(4-Amino-2,3,5,6-tetramethylphenyl)amino]acetyl]methylamino]-1-piperidinyl]methyl]benzamide
Inhibitor:	PD166866	1-[2-Amino-6-(3,5-dimethoxyphenyl)-pyrido[2,3-d]pyrimidin-7-yl]-3-tert-butyl urea
	PD 173074	N-[2-[[4-(Diethylamino)butyl]amino-6-(3,5-dimethoxyphenyl)pyrido[2,3-d]pyrimidin-7-yl]-N′-(1,1-dimethylethyl)urea
FGFR2		
Activator:	none described	
Inhibitor:	Ki23057	2-((2-((4-(4-((4-(tert-butyl)phenyl)amino)phenoxy)-6-methoxyquinolin-7-yl)oxy)ethyl)amino)ethanol
FGFR3		
Activator:	Botulinum neurotoxin serotype A (BoNT/A)	

Inhibitor:	PD 173074	N-[2-[[4-(Diethylamino)butyl]amino-6-(3,5-dimethoxyphenyl)pyrido[2,3-d]pyrimidin-7-yl]-N'-(1,1-dimethylethyl)urea
FGFR4		
Activator:	Monoclonal Antibody: 4FA6D3C10	US 2009/0123462 A1
Inhibitor:	none described	
FGFR (unspecific)		
Activator:	-	
Inhibitor:	FIIN 1 hydrochloride	N-(3-((3-(2,6-dichloro-3,5-dimethoxyphenyl)-7-(4-(diethylamino)butylamino)-2-oxo-3,4-dihydropyrimido[4,5-d]pyrimidin-1(2H)-yl)methyl)phenyl)acrylamide
	PD 161570	N-[6-(2,6-Dichlorophenyl)-2-[[4-(diethylamino)butyl]amino]pyrido[2,3-d]pyrimidin-7-yl]-N'-(1,1-dimethylethyl)urea
	SU 5402	2-[(1,2-Dihydro-2-oxo-3H-indol-3-ylidene)methyl]-4-methyl-1H-pyrrole-3-propanoic acid
	SU 6668	5-[1,2-Dihydro-2-oxo-3H-indol-3-ylidene)methyl]-2,4-dimethyl-1H-pyrrole-3-propanoic acid
Inhibitor FGFR1-3	CH-5183284	CAS#: 1265229-25-1
FGFR downstream (FGFR Kinase) modulation)		
Activator:		
Inhibitor:	AP 24534	3-(2-Imidazo[1,2-b]pyridazin-3-ylethynyl)-4-methyl-N-[4-[(4-methyl-1-piperazinyl)methyl]-3-(trifluoromethyl)phenyl]-benzamide
	BGJ398	3-(2,6-dichloro-3,5-dimethoxyphenyl)-1-(6-((4-(4-ethylpiperazin-1-yl)phenyl)amino)pyrimidin-4-yl)-1-methylurea.

Receptor activation by alternative ligands

FGFs have been successfully modified to peptides as short as 10 amino acids retaining biological activity (Ray, Baird, & Gage, 1997). Shorter variants offer the chance to identify FGF8b-based peptides with increased stability, altered tissue penetration and lower production cost.

The human FGF8 gene gives rise to at least 8 different transcripts, 4 of which are present in the mouse (Fgf8a, b, e and f) (Sunmonu, Li, & Li, 2011). These 4 isoforms were compared in terms of their potency to induce browning in immortalized white adipocytes to narrow down essential regions of the peptide (Figure 1AB). FGF8b and FGF8f treatment is effective in immortalized white adipocytes of epididymal origin, while FGF8b is effective in epididymal as well as inguinal white adipocytes. The only difference between the ineffective FGF8a isoform and FGF8b is the presence of an additional 11 amino acids near the N terminus of FGF8b. Crucial amino acids of a FGF receptor interaction domain are located in precisely this region (Olsen et al. 2013). Therefore, it seems possible that the 11 amino acids on exon 1D might be sufficient to mediate a biological effect. But it remains possible that other parts of the protein might be useful and/or necessary.

This peptide is synthesized commercially and its biological potency to induce Ucp1 mRNA gene expression is tested. Further, the heparan sulfate anchor region is added to preserve paracrine tissue effectiveness *in vivo*. Conversely, the heparan sulfate anchor region is replaced with the klotho-interacting domain of endocrine FGFs to test potential adverse effects of FGF8 treatment *in vivo*. This "endocrinization" of a paracrine FGF has been successfully applied before (Goetz et al., 2012; Suh et al., 2014). Recombinant FGF8b variants are tested in cell cultures of murine epididymal white adipocytes for their potential to recruit brown adipocytes.

FGF8b action on human cells

Both brown adipocytes and the fibroblast growth factor signaling system are conserved in mice and men. The mouse data are transferred to a human system to provide for applied studies employing FGF signaling as a therapeutic means to recruit brown adipocytes in humans.

Primary white adipocytes isolated freshly from surgical biopsies are used. The cells will be subjected to the most effective FGF8b treatment as identified above and mRNA abundance of brown adipocyte marker genes (Ucp1, Cidea etc.) is quantified by qPCR.

Following brown adipocyte recruitment in a human model system , brown adipocyte are characterized and the respective inhibitors and activators identified herein are tested. By this approach it can be confirmed that the same signaling pathways are utilized in both human and mouse cells to induce brown adipocyte recruitment.

Human biopsies

To test the effect of FGF8b on human primary adipocytes clinical material from routine human visceral, trauma or general surgery are gathered; e.g. cholecystomie, hemicolectomy. Written informed consent after intensive counseling is obtained from every participating human being . Relevant individual data will be anonymized and pseudonymized to exclude drawing conclusions back to the involved patient. Patients, from whom significant impact of individual pathology or concomitant disease on study results cannot be excluded, will not be included in this project. Surgery indication is independent from the purpose of this study and no additional material is obtained during these routine operations, an additional risk for patients can be excluded.

Animal experimentation

All animal experimentation as outlined herein is conducted in specific pathogen free (SPF) breeding facilities according to the German Animal Welfare law. Proposals for ethical approvals will be submitted to the Government

The present invention refers to the following nucleotide and amino acid sequences:

The following sequence information is based on the Ensembl.org database using the following accession numbers:

Spliceforms of the human FGF8 gene

Isoform	ENSEMBL transcript no.	ENSEMBL transcript ID
FGF8a	FGF8-003	ENST00000346714
FGF8b	FGF8-001	ENST00000347978
FGF8e	FGF8-004	ENST00000344255
FGF8f	FGF8-002	ENST00000320185

Spliceforms of the murine FGF8 gene

Isoform	ENSEMBL transcript no.	ENSEMBL transcript ID
FGF8a	FGF8-003	ENSMUST00000111927
FGF8b	FGF8-002	ENSMUST00000111928
FGF8e	FGF8-006	ENSMUST00000111925
FGF8f	FGF8-001	ENSMUST00000026240

FGF17 gene

Species	ENSEMBL transcript no.	ENSEMBL transcript ID
human	FGF17-001	ENST00000359441
murine	Fgf17-201	ENSMUST00000022697

FGF15/19 gene (The murine ortholog of human FGF19 is called FGF15.)

Species	ENSEMBL transcript no.	ENSEMBL transcript ID
human	FGF19-001	ENST00000294312
murine	Fgf15-201	ENSMUST00000033389

FGF21 gene

Species	ENSEMBL transcript no.	ENSEMBL transcript ID
human	FGF21-201	ENST00000222157
murine	Fgf21-201	ENSMUST00000033099

For the following FGFR1, 2, 3, and 4 sequences please note: Given are examples for a protein coding main transcript – other transcripts or transcript variants to be

employed in accordance with this invention are deposited in the Ensembl.org database. Principles of alternative slicing of FGFR1, 2, and 3 are explained in Figure 9 and are also known in the art.

FGFR1 gene

Species	ENDEMBL transcript no.	ENSEMBL transcript ID
human	FGFR1-001	ENST00000397091
murine	Fgfr1-001	ENSMUST00000084027

FGFR2 gene

Species	ENDEMBL transcript no.	ENSEMBL transcript ID
human	FGFR2-001	ENST00000358487
murine	Fgfr2-004	ENSMUST00000122054

FGFR3 gene

Species	ENDEMBL transcript no.	ENSEMBL transcript ID
Human	FGFR3-001	ENST00000260795
murine	Fgfr3-002	ENSMUST00000087820

FGFR3 gene transcript variant c

Species	ENDEMBL transcript no.	ENSEMBL transcript ID
Human	FGFR3-203	ENST00000440486
murine	Fgfr3-202	ENSMUST00000169212

FGFR4 gene

Species	ENDEMBL transcript no.	ENSEMBL transcript ID
Human	FGFR4-001	ENST00000292408
murine	Fgfr4-001	ENSMUST00000005452

cDNA sequences sorted by gene/transcript

Code: non-underlined/underlined denoted alternating exons

SEQ ID NO: 1. human FGF8a

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTCCAAGCCCAGCATGTG
AGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAACTCTACAGCCGCACCAGC
GGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAGGACGGCGACCCCTTCGCAAAGCTC
ATCGTGGAGACGGACACCTTTGGAAGCAGAGTTCGAGTCCGAGGAGCCGAGACGGGCCTCTACATCTGCATGAAC
AAGAAGGGGAAGCTGATCGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTCTTCACGGAGATTGTGCTGGAGAAC
AACTACACAGCGCTGCAGAATGCCAAGTACGAGGGCTGGTACATGGCCTTCACCCGCAAGGGCCGGCCCCGCAAG
GGCTCCAAGACGCGGCAGCACCAGCGTGAGGTCCACTTCATGAAGCGGCTGCCCCGGGGCCACCACACCACCGAG
CAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
CCCGAGCCCCGATAG

SEQ ID NO: 2. human FGF8b

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTCCAAGCCCAGGTAACT
GTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTC
ATCCGGACCTACCAACTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCC
ATGGCAGAGGACGGCGACCCCTTCGCAAAGCTCATCGTGGAGACGGACACCTTTGGAAGCAGAGTTCGAGTCCGA
GGAGCCGAGACGGGCCTCTACATCTGCATGAACAAGAAGGGGAAGCTGATCGCCAAGAGCAACGGCAAAGGCAAG
GACTGCGTCTTCACGGAGATTGTGCTGGAGAACAACTACACAGCGCTGCAGAATGCCAAGTACGAGGGCTGGTAC
ATGGCCTTCACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGGCAGCACCAGCGTGAGGTCCACTTCATG
AAGCGGCTGCCCCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACG
CGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCCGAGCCCCGATAG

SEQ ID NO: 3. human FGF8e

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTCCAAGCCCAGGAAGGC
CCGGGCAGGGGCCCTGCGCTGGGCAGGGAGCTCGCTTCCCTGTTCCGGGCTGGCCGGGAGCCCCAGGGTGTCTCC
CAACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAACTCTAC
AGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAGGACGGCGACCCC
TTCGCAAAGCTCATCGTGGAGACGGACACCTTTGGAAGCAGAGTTCGAGTCCGAGGAGCCGAGACGGGCCTCTAC
ATCTGCATGAACAAGAAGGGGAAGCTGATCGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTCTTCACGGAGATT
GTGCTGGAGAACAACTACACAGCGCTGCAGAATGCCAAGTACGAGGGCTGGTACATGGCCTTCACCCGCAAGGGC
CGGCCCCGCAAGGGCTCCAAGACGCGGCAGCACCAGCGTGAGGTCCACTTCATGAAGCGGCTGCCCCGGGGCCAC
CACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAG
AGGACTTGGGCCCCCGAGCCCCGATAG

SEQ ID NO: 4. human FGF8f

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTCCAAGCCCAGGAAGGC
CCGGGCAGGGGCCCTGCGCTGGGCAGGGAGCTCGCTTCCCTGTTCCGGGCTGGCCGGGAGCCCCAGGGTGTCTCC
CAACAGGTAACTGTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTC
AGCCGCCGCCTCATCCGGACCTACCAACTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
CGCATCAACGCCATGGCAGAGGACGGCGACCCCTTCGCAAAGCTCATCGTGGAGACGGACACCTTTGGAAGCAGA
GTTCGAGTCCGAGGAGCCGAGACGGGCCTCTACATCTGCATGAACAAGAAGGGGAAGCTGATCGCCAAGAGCAAC
GGCAAAGGCAAGGACTGCGTCTTCACGGAGATTGTGCTGGAGAACAACTACACAGCGCTGCAGAATGCCAAGTAC
GAGGGCTGGTACATGGCCTTCACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGGCAGCACCAGCGTGAG
GTCCACTTCATGAAGCGGCTGCCCCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTAC
CCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCCGAGCCCCGATAG

SEQ ID NO: 5. murine FGF8a

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTCCAAGCCCAGCATGTG
AGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAGCTCTACAGCCGCACCAGC
GGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAAGACGGAGACCCCTTCGCGAAGCTC
ATTGTGGAGACCGATACTTTTGGAAGCAGAGTCCGAGTTCGCGGCGCAGAGACAGGTCTCTACATCTGCATGAAC
AAGAAGGGGAAGCTAATTGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTATTCACAGAGATCGTGCTGGAGAAC
AACTACACGGCGCTGCAGAACGCCAAGTACGAGGGCTGGTACATGGCCTTTACCCGCAAGGGCCGGCCCCGCAAG
GGCTCCAAGACGCGCCAGCATCAGCGCGAGGTGCACTTCATGAAGCGCCTGCCGCGGGGCCACCACACCACCGAG
CAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
CCGGAGCCCCGATAG

SEQ ID NO: 6. murine FGF8b

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTCCAAGCCCAGGTAACT
GTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTC
ATCCGGACCTACCAGCTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCC
ATGGCAGAAGACGGAGACCCCTTCGCGAAGCTCATTGTGGAGACCGATACTTTTGGAAGCAGAGTCCGAGTTCGC
GGCGCAGAGACAGGTCTCTACATCTGCATGAACAAGAAGGGGAAGCTAATTGCCAAGAGCAACGGCAAAGGCAAG

GACTGCGTATTCACAGAGATCGTGCTGGAGAACAACTACACGGCGCTGCAGAACGCCAAGTACGAGGGCTGGTAC
ATGGCCTTTACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGCCAGCATCAGCGCGAGGTGCACTTCATG
AAGCGCCTGCCGCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACG
CGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCGGAGCCCCGATAG

SEQ ID NO: 7. murine FGF8e

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTCCAAGCCCAGGAAGGC
CCGGGCGGGGGGCCTGCGCTGGGCAGGGAGCCCACTTCCCTGCTCCGAGCTGGCCGGGAGCCCCAGGGTGTTTCC
CAACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAGCTCTAC
AGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAAGACGGAGACCCC
TTCGCGAAGCTCATTGTGGAGACCGATACTTTTGGAAGCAGAGTCCGAGTTCGCGGCGCAGAGACAGGTCTCTAC
ATCTGCATGAACAAGAAGGGGAAGCTAATTGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTATTCACAGAGATC
GTGCTGGAGAACAACTACACGGCGCTGCAGAACGCCAAGTACGAGGGCTGGTACATGGCCTTTACCCGCAAGGGC
CGGCCCCGCAAGGGCTCCAAGACGCGCCAGCATCAGCGCGAGGTGCACTTCATGAAGCGCCTGCCGCGGGGCCAC
CACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAG
AGGACTTGGGCCCCGGAGCCCCGATAG

SEQ ID NO: 8. murine FGF8f

ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTCCAAGCCCAGGAAGGC
CCGGGCGGGGGGCCTGCGCTGGGCAGGGAGCCCACTTCCCTGCTCCGAGCTGGCCGGGAGCCCCAGGGTGTTTCC
CAACAGGTAACTGTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTC
AGCCGCCGCCTCATCCGGACCTACCAGCTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
CGCATCAACGCCATGGCAGAAGACGGAGACCCCTTCGCGAAGCTCATTGTGGAGACCGATACTTTTGGAAGCAGA
GTCCGAGTTCGCGGCGCAGAGACAGGTCTCTACATCTGCATGAACAAGAAGGGGAAGCTAATTGCCAAGAGCAAC
GGCAAAGGCAAGGACTGCGTATTCACAGAGATCGTGCTGGAGAACAACTACACGGCGCTGCAGAACGCCAAGTAC
GAGGGCTGGTACATGGCCTTTACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGCCAGCATCAGCGCGAG
GTGCACTTCATGAAGCGCCTGCCGCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTAC
CCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCGGAGCCCCGATAG

SEQ ID NO: 9. human FGF17

ATGGGAGCCGCCCGCCTGCTGCCCAACCTCACTCTGTGCTTACAGCTGCTGATTCTCTGCTGTCAAACTCAGGGG
GAGAATCACCCGTCTCCTAATTTTAACCAGTACGTGAGGGACCAGGGCGCCATGACCGACCAGCTGAGCAGGCGG
CAGATCCGCGAGTACCAACTCTACAGCAGGACCAGTGGCAAGCACGTGCAGGTCACCGGGCGTCGCATCTCCGCC
ACCGCCGAGGACGGCAACAAGTTTGCCAAGCTCATAGTGGAGACGGACACGTTTGGCAGCCGGGTTCGCATCAAA
GGGGCTGAGAGTGAGAAGTACATCTGTATGAACAAGAGGGGCAAGCTCATCGGGAAGCCCAGCGGGAAGAGCAAA
GACTGCGTGTTCACGGAGATCGTGCTGGAGAACAACTATACGGCCTTCCAGAACGCCCGGCACGAGGGCTGGTTC
ATGGCCTTCACGCGGCAGGGGCGGCCCCGCCAGGCTTCCCGCAGCCGCCAGAACCAGCGCGAGGCCCACTTCATC
AAGCGCCTCTACCAAGGCCAGCTGCCCTTCCCCAACCACGCCGAGAAGCAGAAGCAGTTCGAGTTTGTGGGCTCC
GCCCCCACCCGCCGGACCAAGCGCACACGGCGGCCCCAGCCCCTCACGTAG

SEQ ID NO: 10. murine FGF17

ATGGGAGCCGCCCGCCTGCTGCCTAACCTTACCCTGTGCTTGCAGCTATTGATTCTCTGCTGTCAAACACAGGGG
GAGAATCACCCGTCTCCTAATTTTAACCAGTACGTGAGGGACCAGGGCGCTATGACCGACCAGCTGAGCAGGCGG
CAAATCCGTGAATACCAGCTCTACAGCCGGACCAGTGGCAAGCACGTGCAGGTCACCGGACGTCGCATCTCTGCC
ACCGCAGAGGATGGCAACAAGTTCGCCAAGCTCATCGTGGAGACAGATACATTCGGCAGCAGAGTCCGCATCAAG
GGGGCAGAGAGCGAGAAGTACATCTGTATGAACAAGAGGGGCAAGCTGATTGGGAAGCCGAGCGGGAAGAGCAAA
GACTGCGTGTTCACCGAGATCGTACTGGAGAACAACTACACGGCCTTCCAGAACGCCCGGCACGAGGGCTGGTTC
ATGGCTTTCACTCGGCAGGGCCGGCCACGCCAGGCCTCCCGGAGCCGCCAGAACCAGCGAGAGGCCCACTTCATC
AAGCGCCTCTACCAAGGCCAGCTGCCTTTTCCCAACCACGCTGAAAGGCAGAAGCAGTTCGAATTTGTGGGCTCC
GCCCCCACTCGCAGGACCAAGCGCACTCGGAGGCCCCAGTCCCAAACGTAG

SEQ ID NO: 11. human FGF19

ATGCGGAGCGGGTGTGTGGTGGTCCACGTATGGATCCTGGCCGGCCTCTGGCTGGCCGTGGCCGGGCGCCCCCTC
GCCTTCTCGGACGCGGGGCCCCACGTGCACTACGGCTGGGGCGACCCCATCCGCCTGCGGCACCTGTACACCTCC
GGCCCCCACGGGCTCTCCAGCTGCTTCCTGCGCATCCGTGCCGACGGCGTCGTGGACTGCGCGCGGGGCCAGAGC
GCGCACAGTTTGCTGGAGATCAAGGCAGTCGCTCTGCGGACCGTGGCCATCAAGGGCGTGCACAGCGTGCGGTAC
CTCTGCATGGGCGCCGACGGCAAGATGCAGGGGCTGCTTCAGTACTCGGAGGAAGACTGTGCTTTCGAGGAGGAG
ATCCGCCCAGATGGCTACAATGTGTACCGATCCGAGAAGCACCGCCTCCCGGTCTCCCTGAGCAGTGCCAAACAG
CGGCAGCTGTACAAGAACAGAGGCTTTCTTCCACTCTCTCATTTCCTGCCCATGCTGCCCATGGTCCCAGAGGAG
CCTGAGGACCTCAGGGGCCACTTGGAATCTGACATGTTCTCTTCGCCCCTGGAGACCGACAGCATGGACCCATTT
GGGCTTGTCACCGGACTGGAGGCCGTGAGGAGTCCCAGCTTTGAGAAGTAA

SEQ ID NO: 12. murine FGF15

ATGGCGAGAAAGTGGAACGGGCGTGCGGTGGCCCGAGCCCTGGTCCTGGCCACTCTGTGGCTGGCTGTGTCTGGG
CGTCCCCTGGCTCAGCAATCCCAGTCTGTGTCAGATGAAGATCCACTCTTTCTCTACGGCTGGGGCAAGATTACC
CGCCTGCAGTACCTGTACTCCGCTGGTCCCTATGTCTCCAACTGCTTCCTCCGAATCCGGAGCGACGGCTCTGTG
GACTGCGAGGAGGACCAAAACGAACGAAATTTGTTGGAATTCCGCGCGGTCGCTCTGAAGACGATTGCCATCAAG
GACGTCAGCAGCGTGCGGTACCTCTGCATGAGCGCGGACGGCAAGATATACGGGCTGATTCGCTACTCGGAGGAA
GACTGTACCTTCAGGGAGGAAATGGACTGTTTAGGCTACAACCAGTACAGATCCATGAAGCACCATCTCCATATC
ATCTTCATCCAGGCCAAGCCCAGAGAACAGCTCCAGGACCAGAAACCCTCAAACTTTATCCCCGTGTTTCACCGC
TCCTTCTTTGAAACCGGGGACCAGCTGAGGTCTAAAATGTTCTCCCTGCCCCTGGAGAGTGACAGCATGGATCCG
TTCAGGATGGTGGAGGATGTAGACCACCTAGTGAAGAGTCCCAGCTTCCAGAAATGA

SEQ ID NO: 13. human FGF21

ATGGACTCGGACGAGACCGGGTTCGAGCACTCAGGACTGTGGGTTTCTGTGCTGGCTGGTCTTCTGCTGGGAGCC
TGCCAGGCACACCCCATCCCTGACTCCAGTCCTCTCCTGCAATTCGGGGGCCAAGTCCGGCAGCGGTACCTCTAC
ACAGATGATGCCCAGCAGACAGAAGCCCACCTGGAGATCAGGGAGGATGGGACGGTGGGGGGCGCTGCTGACCAG
AGCCCCGAAAGTCTCCTGCAGCTGAAAGCCTTGAAGCCGGGAGTTATTCAAATCTTGGGAGTCAAGACATCCAGG
TTCCTGTGCCAGCGGCCAGATGGGGCCCTGTATGGATCGCTCCACTTTGACCCTGAGGCCTGCAGCTTCCGGGAG
CTGCTTCTTGAGGACGGATACAATGTTTACCAGTCCGAAGCCCACGGCCTCCCGCTGCACCTGCCAGGGAACAAG
TCCCCACACCGGGACCCTGCACCCCGAGGACCAGCTCGCTTCCTGCCACTACCAGGCCTGCCCCCCGCACTCCCG
GAGCCACCCGGAATCCTGGCCCCCCAGCCCCCCGATGTGGGCTCCTCGGACCCTCTGAGCATGGTGGGACCTTCC
CAGGGCCGAAGCCCCAGCTACGCTTCCTGA

SEQ ID NO: 14. murine FGF21

ATGGAATGGATGAGATCTAGAGTTGGGACCCTGGGACTGTGGGTCCGACTGCTGCTGGCTGTCTTCCTGCTGGGG
GTCTACCAAGCATACCCCATCCCTGACTCCAGCCCCCTCCTCCAGTTTGGGGGTCAAGTCCGGCAGAGGTACCTC
TACACAGATGACGACCAAGACACTGAAGCCCACCTGGAGATCAGGGAGGATGGAACAGTGGTAGGCGCAGCACAC
CGCAGTCCAGAAAGTCTCCTGGAGCTCAAAGCCTTGAAGCCAGGGGTCATTCAAATCCTGGGTGTCAAAGCCTCT
AGGTTTCTTTGCCAACAGCCAGATGGAGCTCTCTATGGATCGCCTCACTTTGATCCTGAGGCCTGCAGCTTCAGA
GAACTGCTGCTGGAGGACGGTTACAATGTGTACCAGTCTGAAGCCCATGGCCTGCCCCTGCGTCTGCCTCAGAAG
GACTCCCCAAACCAGGATGCAACATCCTGGGGACCTGTGCGCTTCCTGCCCATGCCAGGCCTGCTCCACGAGCCC
CAAGACCAAGCAGGATTCCTGCCCCCAGAGCCCCCAGATGTGGGCTCCTCTGACCCCCTGAGCATGGTAGAGCCT
TTACAGGGCCGAAGCCCCAGCTATGCGTCCTGA

SEQ ID NO: 29. human FGFR1

ATGTGGAGCTGGAAGTGCCTCCTCTTCTGGGCTGTGCTGGTCACAGCCACACTCTGCACCGCTAGGCCGTCCCCG
ACCTTGCCTGAACAAGCCCAGCCCTGGGGAGCCCCTGTGGAAGTGGAGTCCTTCCTGGTCCACCCCGGTGACCTG
CTGCAGCTTCGCTGTCGGCTGCGGGACGATGTGCAGAGCATCAACTGGCTGCGGGACGGGGTGCAGCTGGCGGAA
AGCAACCGCACCCGCATCACAGGGGAGGAGGTGGAGGTGCAGGACTCCGTGCCCGCAGACTCCGGCCTCTATGCT
TGCGTAACCAGCAGCCCCTCGGGCAGTGACACCACCTACTTCTCCGTCAATGTTTCAGATGCTCTCCCCTCCTCG
GAGGATGATGATGATGATGACTCCTCTTCAGAGGAGAAAGAAACAGATAACACCAAACCAAACCCCGTAGCT
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TACACCTGCATTGTGGAGAATGAGTACGGCAGCATCAACCACACATACCAGCTGGATGTCGTGGAGCGGTCCCCT
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TCTGCATGGTTGACCGTTCTGGAAGCCCTGGAAGAGAGGCCGGCAGTGATGACCTCGCCCCTGTACCTGGAGATC
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ACCAAGAAGAGTGACTTCCACAGCCAGATGGCTGTGCACAAGCTGGCCAAGAGCATCCCTCTGCGCAGACAGGTA
ACAGTGTCTGCTGACTCCAGTGCATCCATGAACTCTGGGGTTCTTCTGGTTCGGCCATCACGGCTCTCCTCCAGT
GGGACTCCCATGCTAGCAGGGGTCTCTGAGTATGAGCTTCCCGAAGACCCTCGCTGGGAGCTGCCTCGGGACAGA
CTGGTCTTAGGCAAACCCCTGGGAGAGGGCTGCTTTGGGCAGGTGGTGTTGGCAGAGGCTATCGGGCTGGACAAG
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CTGATCTCAGAAATGGAGATGATGAAGATGATCGGGAAGCATAAGAATATCATCAACCTGCTGGGGGCCTGCACG
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TGCGCCTACCAGGTGGCCCGAGGCATGGAGTATCTGGCCTCCAAGAAGTGCATACACCGAGACCTGGCAGCCAGG
AATGTCCTGGTGACAGAGGACAATGTGATGAAGATAGCAGACTTTGGCCTCGCACGGGACATTCACCACATCGAC
TACTATAAAAAGACAACCAACGGCCGACTGCCTGTGAAGTGGATGGCACCCGAGGCATTATTTGACCGGATCTAC
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GAGCTGTACATGATGATGCGGGACTGCTGGCATGCAGTGCCCTCACAGAGACCCACCTTCAAGCAGCTGGTGGAA
GACCTGGACCGCATCGTGGCCTTGACCTCCAACCAGGAGTACCTGGACCTGTCCATGCCCCTGGACCAGTACTCC
CCCAGCTTTCCCGACACCCGGAGCTCTACGTGCTCCTCAGGGGAGGATTCCGTCTTCTCTCATGAGCCGCTGCCC
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ATGTGGGGCTGGAAGTGCCTCCTCTTCTGGGCTGTGCTGGTCACAGCCACTCTCTGCACTGCCAGGCCAGCCCCA
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CTACAGCTTCGCTGTCGGCTTCGCGATGATGTGCAGAGCATCAACTGGCTGCGGGATGGGGTGCAGCTGGTGGAG
AGCAACCGTACCCGCATCACAGGGGAGGAGGTGGAGGTGCGGGACTCCATCCCCGCTGACTCTGGCCTCTACGCT
TGCGTGACCAGCAGCCCCTCTGGCAGCGATACCACCTACTTCTCCGTCAATGTCTCAGATGCACTCCCATCCTCG
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GACAGACTGGTCTTAGGCAAACCACTTGGCGAGGGCTGCTTCGGGCAGGTGGTGTTGGCTGAGGCCATCGGGCTG
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TACCCCGGTGTGCCTGTGGAGGAACTTTTCAAGCTGCTGAAGGAGGGTCATCGAATGGACAAGCCCAGTAACTGT
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TACTCACCCAGCTTTCCCGACACACGGAGCTCCACCTGCTCCTCAGGGGAGGACTCTGTCTTCTCTCATGAGCCG
TTACCTGAGGAGCCCTGTCTGCCTCGACACCCCACCCAGCTTGCCAACAGTGGACTCAAACGGCGCTGA

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ATGGTCAGCTGGGGTCGTTTCATCTGCCTGGTCGTGGTCACCATGGCAACCTTGTCCCTGGCCCGGCCCTCCTTC
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CTCGAACAGTATTCACCTAGTTACCCTGACACAAGAAGTTCTTGTTCTTCAGGAGATGATTCTGTTTTTTCTCCA
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TGTACTGCAGCTAGGACGGTAGACAGTGAAACTTGGTACTTCATGGTGAATGTCACAGATGCCATCTCATCTGGA
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ATGGGCGCCCCTGCCTGCGCCCTCGCGCTCTGCGTGGCCGTGGCCATCGTGGCCGGCGCCTCCTCGGAGTCCTTG
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AGTAACGGGGGACCTCGGACGTGA

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ATGCGGCTGCTGCTGGCCCTGTTGGGGGTCCTGCTGAGTGTGCCTGGGCCTCCAGTCTTGTCCCTGGAGGCCTCT
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TCCTCCAGCGATTCTGTCTTCAGCCACGACCCCCTGCCATTGGGATCCAGCTCCTTCCCCTTCGGGTCTGGGGTG
CAGACATGA

SEQ ID NO: 36. murine FGFR4

ATGTGGCTGCTCTTGGCCCTGTTGAGCATCTTTCAGGGGACACCAGCTTTGTCCCTTGAGGCCTCTGAGGAAATG
GAGCAGGAGCCCTGCCTAGCCCCAATCCTGGAGCAGCAAGAGCAGGTGTTGACGGTGGCCCTGGGGCAGCCTGTG
AGGCTGTGCTGTGGGCGCACCGAGCGTGGTCGTCACTGGTACAAAGAGGGCAGCCGCCTAGCATCTGCTGGGCGA
GTACGGGGTTGGAGAGGCCGCCTGGAGATCGCCAGCTTCCTTCCTGAGGATGCTGGCCGATACCTCTGCCTGGCC
CGTGGCTCCATGACCGTCGTACACAATCTTACGTTGCTTATGGATGACTCCTTAACCTCCATCAGTAATGATGAA
GACCCCAAGACACTCAGCAGCTCCTCGAGTGGTCATGTCTACCCACAGCAAGCACCCTACTGGACACACCCCCAA
CGCATGGAGAAGAAACTGCATGCAGTGCCTGCCGGGAATACTGTCAAATTCCGCTGTCCAGCTGCAGGGAACCCC
ATGCCTACCATCCACTGGCTCAAGGATGGACAGGCCTTCCACGGGGAGAATCGTATTGGAGGCATTCGGCTGCGC
CACCAACACTGGAGCCTGGTGATGGAAAGTGTGGTACCCTCGGACCGTGGCACATACACATGCCTTGTGGAGAAC
TCTCTGGGTAGCATTCGCTACAGCTATCTCCTGGATGTGCTGGAGCGGTCCCCGCACCGGCCCATCCTGCAGGCG
GGGCTCCCAGCCAACACCACAGCTGTGGTTGGCAGCGATGTGGAGCTACTCTGCAAGGTGTACAGCGACGCCCAG
CCCCACATACAGTGGCTGAAACACGTCGTCATCAACGGCAGCAGCTTCGGCGCCGACGGTTTCCCCTACGTACAA
GTCCTGAAGACAACAGACATCAATAGCTCGGAGGTAGAGGTCTTGTATCTGAGGAACGTGTCCGCTGAGGATGCA
GGAGAGTATACCTGTCTGGCGGGCAACTCCATCGGCCTTTCCTACCAGTCAGCGTGGCTCACGGTGCTGCCAGAG
GAAGACCTCACGTGGACAACAGCAACCCCTGAGGCCAGATACACAGATATCATCCTGTATGTATCAGGCTCACTG
GTTCTGCTTGTGCTCCTGCTGCTGGCCGGGGGTGTATCATCGGCAAGTCATCCGTGGCCACTACTCTCGCCAGCCT
GTCACTATACAAAAGCTGTCCCGTTTCCCTTTGGCCCGACAGTTCTCTTTGGAGTCGAGGTCCTCTGGCAAGTCA
AGTTTGTCCCTGGTGCGAGGTGTCCGTCTCTCCTCCAGCGGCCCGCCCTTGCTCACGGGCCTTGTGAATCTAGAC
CTGCCTCTCGATCCGCTTTGGGAATTCCCCCGGGACAGGTTGGTGCTCGGAAAGCCCCTGGGTGAGGGCTGCTTT
GGGCAAGTGGTTCGTGCAGAGGCCTTTGGTATGGATCCCTCCCGGCCCGACCAAACCAGCACCGTGGCTGTGAAG
ATGCTGAAAGACAATGCCTCCGACAAGGATTTGGCAGACCTGGTCTCCGAGATGGAGGTGATGAAGCTAATCGGA
AGACACAAGAACATCATCAACCTGCTGGGTGTCTGCACTCAGGAAGGGCCCCTGTACGTGATTGTGGAATGTGCC
GCCAAGGGAAACCTTCGGGAATTCCTCCGTGCCCGGCGCCCCCCAGGCCCTGATCTCAGCCCTGATGGACCTCGG
AGCAGCGAAGGACCACTCTCCTTCCCGGCCCTAGTCTCCTGTGCCTACCAGGTGGCCCGAGGCATGCAGTATCTG
GAGTCTCGGAAGTGCATCCACCGGGACCTGGCTGCCCGAAATGTGCTGGTGACCGAGGATGATGTGATGAAGATC
GCTGACTTTGGGCTGGCACGTGGTGTCCACCACATTGACTACTATAAGAAAACCAGCAACGGCCGCCTGCCAGTC
AAATGGATGGCTCCAGAGGCATTGTTCGACCGCGTGTACACACACCAGAGTGACGTGTGGTCTTTCGGGATCCTG
CTGTGGGAAATCTTCACCCTCGGGGGCTCCCCATACCCTGGCATTCCGGTGGAGGAGCTCTTCTCACTGCTGCGA
GAGGGGCACAGGATGGAGCGGCCCCCAAACTGCCCCTCAGAGCTGTATGGGCTAATGAGGGAGTGCTGGCACGCA
GCCCCATCTCAGAGGCCTACTTTTAAGCAGCTGGTGGAAGCTCTGGACAAGGTCCTGCTGGCTGTCTCTGAAGAG
TACCTTGACCTCCGCCTGACCTTTGGACCCTTTTCTCCCTCCAATGGGGATGCCAGCAGCACCTGCTCCTCCAGT
GACTCGGTTTTCAGCCACGACCCTTTGCCCCTCGAGCCAAGCCCCTTCCCTTTCTCTGACTCGCAGACGACATGA

SEQ ID NO: 85 human FGFR3 spliceform c

ATGGGCGCCCCTGCCTGCGCCCTCGCGCTCTGCGTGGCCGTGGCCATCGTGGCCGGCGCCTCCTCGGAGTCCTTG
GGGACGGAGCAGCGCGTCGTGGGGCGAGCGGCAGAAGTCCCGGGCCCAGAGCCCGGCCAGCAGGAGCAGTTGGTC
TTCGGCAGCGGGGATGCTGTGGAGCTGAGCTGTCCCCCGCCCGGGGGTGGTCCCATGGGGCCCACTGTCTGGGTC
AAGGATGGCACAGGGCTGGTGCCCTCGGAGCGTGTCCTGGTGGGGCCCCAGCGGCTGCAGGTGCTGAATGCCTCC
CACGAGGACTCCGGGGCCTACAGCTGCCGGCAGCGGCTCACGCAGCGCGTACTGTGCCACTTCAGTGTGCGGGTG
ACAGACGCTCCATCCTCGGGAGATGACGAAGACGGGGAGGACGAGGCTGAGGACACAGGTGTGGACACAGGGGCC
CCTTACTGGACACGGCCCGAGCGGATGGACAAGAAGCTGCTGGCCGTGCCGGCCGCCAACACCGTCCGCTTCCGC
TGCCCAGCCGCTGGCAACCCCACTCCCTCCATCTCCTGGCTGAAGAACGGCAGGGAGTTCCGCGGCGAGCACCGC
ATTGGAGGCATCAAGCTGCGGCATCAGCAGTGGAGCCTGGTCATGGAAAGCGTGGTGCCCTCGGACCGCGGCAAC
TACACCTGCGTCGTGGAGAACAAGTTTGGCAGCATCCGGCAGACGTACACGCTGGACGTGCTGGAGCGCTCCCCG
CACCGGCCCATCCTGCAGGCGGGGCTGCCGGCCAACCAGACGGCGGTGCTGGGCAGCGACGTGGAGTTCCACTGC
AAGGTGTACAGTGACGCACAGCCCCACATCCAGTGGCTCAAGCACGTGGAGGTGAATGGCAGCAAGGTGGGCCCG

GACGGCACACCCTACGTTACCGTGCTCAAGACGGCGGGCGCTAACACCACCGACAAGGAGCTAGAGGTTCTCTCC
TTGCACAACGTCACCTTTGAGGACGCCGGGGAGTACACCTGCCTGGCGGGCAATTCTATTGGGTTTTCTCATCAC
TCTGCGTGGCTGGTGGTGCTGCCAGCCGAGGAGGAGCTGGTGGAGGCTGACGAGGCGGGCAGTGTGTATGCAGGC
ATCCTCAGCTACGGGGTGGGCTTCTTCCTGTTCATCCTGGTGGTGGCGGCTGTGACGCTCTGCCGCCTGCGCAGC
CCCCCCAAGAAAGGCCTGGGCTCCCCCACCGTGCACAAGATCTCCCGCTTCCCGCTCAAGCGACAGGTGTCCCTG
GAGTCCAACGCGTCCATGAGCTCCAACACACCACTGGTGCGCATCGCAAGGCTGTCCTCAGGGGAGGGCCCCACG
CTGGCCAATGTCTCCGAGCTCGAGCTGCCTGCCGACCCCAAATGGGAGCTGTCTCGGGCCCGGCTGACCCTGGGC
AAGCCCCTTGGGGAGGGCTGCTTCGGCCAGGTGGTCATGGCGGAGGCCATCGGCATTGACAAGGACCGGGCCGCC
AAGCCTGTCACCGTAGCCGTGAAGATGCTGAAAGACGATGCCACTGACAAGGACCTGTCGGACCTGGTGTCTGAG
ATGGAGATGATGAAGATGATCGGGAAACACAAAAACATCATCAACCTGCTGGGCGCCTGCACGCAGGGCGGGCCC
CTGTACGTGCTGGTGGAGTACGCGGCCAAGGGTAACCTGCGGGAGTTTCTGCGGGCGCGGCGGCCCCCGGGCCTG
GACTACTCCTTCGACACCTGCAAGCCGCCCGAGGAGCAGCTCACCTTCAAGGACCTGGTGTCCTGTGCCTACCAG
GTGGCCCGGGGCATGGAGTACTTGGCCTCCCAGAAGTGCATCCACAGGGACCTGGCTGCCCGCAATGTGCTGGTG
ACCGAGGACAACGTGATGAAGATCGCAGACTTCGGGCTGGCCCGGGACGTGCACAACCTCGACTACTACAAGAAG
ACGACCAACGGCCGGCTGCCCGTGAAGTGGATGGCGCCTGAGGCCTTGTTTGACCGAGTCTACACTCACCAGAGT
GACGTCTGGTCCTTTGGGGTCCTGCTCTGGGAGATCTTCACGCTGGGGGGCTCCCCGTACCCCGGCATCCCTGTG
GAGGAGCTCTTCAAGCTGCTGAAGGAGGGCCACCGCATGGACAAGCCCGCCAACTGCACACACGACCTGTACATG
ATCATGCGGGAGTGCTGGCATGCCGCGCCCTCCCAGAGGCCCACCTTCAAGCAGCTGGTGGAGGACCTGGACCGT
GTCCTTACCGTGACGTCCACCGACGAGTACCTGGACCTGTCGGCGCCTTTCGAGCAGTACTCCCCGGGTGGCCAG
GACACCCCCAGCTCCAGCTCCTCAGGGGACGACTCCGTGTTTGCCCACGACCTGCTGCCCCCGGCCCCACCCAGC
AGTGGGGGCTCGCGGACGTGA

SEQ ID NO: 86 murine FGFR3 spliceform c

ATGGTAGTCCCGGCCTGCGTGCTAGTGTTCTGCGTGGCGGTCGTGGCTGGAGCTACTTCCGAGCCTCCTGGTCCA
GAGCAGCGAGTTGTGCGGAGAGCGGCAGAGGTTCCAGGGCCTGAACCTAGCCAGCAGGAGCAGGTGGCCTTCGGC
AGTGGGGACACCGTGGAGCTGAGCTGCCATCCTCCTGGAGGTGCCCCCACAGGGCCCACGGTCTGGGCTAAGGAT
GGTACAGGTCTGGTGGCCTCCCACCGCATCCTGGTGGGGCCTCAGAGGCTGCAAGTGCTAAATGCCTCCCACGAA
GATGCAGGGGTCTACAGCTGCCAGCACCGGCTCACTCGGCGTGTGCTGTGCCACTTCAGTGTGCGTGTAACAGAT
GCTCCATCCTCAGGAGATGACGAAGATGGGGAGGACGTGGCTGAAGACACAGGGGCTCCTTATTGGACTCGCCCG
GAGCGAATGGATAAGAAACTGCTGGCTGTGCCAGCCGCAAACACTGTCCGCTTCCGCTGCCCAGCTGCTGGCAAC
CCTACCCCCTCCATCTCCTGGCTGAAGAATGGCAAAGAATTCCGAGGGGAGCATCGCATTGGGGGCATCAAGCTC
CGGCACCAGCAGTGGAGCTTGGTCATGGAAAGTGTGGTACCCTCCGATCGTGGCAACTATACCTGTGTAGTTGAG
AACAAGTTTGGCAGCATCCGGCAGACATACACACTGGATGTGCTGGAGCGCTCCCCACACCGGCCCATCCTGCAG
GCTGGGCTGCCGGCCAACCAGACAGCCATTCTAGGCAGTGACGTGGAGTTCCACTGCAAGGTGTACAGCGATGCA
CAGCCACACATCCAGTGGCTGAAGCACGTGGAAGTGAACGGCAGCAAGGTGGGCCCTGACGGCACGCCCTACGTC
ACTGTACTCAAGACTGCAGGCGCTAACACCACCGACAAGGAGCTAGAGGTTCTGTCCTTGCACAATGTCACCTTT
GAGGACGCGGGGGAGTACACCTGCCTGGCGGGCAATTCTATTGGGTTTTCCCATCACTCTGCGTGGCTGGTGGTG
CTGCCAGCTGAGGAGGAGCTGATGGAAACTGATGAGGCTGGCAGCGTGTACGCAGGCGTCCTCAGCTACGGGGTG
GTCTTCTTCCTCTTCATCCTGGTGGTGGCAGCTGTGATACTCTGCCGCCTGCGCAGTCCCCCAAAGAAGGGCTTG
GGCTCGCCCACCGTGCACAAGGTCTCTCGCTTCCCGCTTAAGCGACAGGTGTCCTTGGAATCTAACTCCTCTATG
AACTCCAACACACCCCTTGTCCGGATTGCCCGGCTGTCCTCAGGAGAAGGTCCTGTTCTGGCCAATGTTTCTGAA
CTTGAGCTGCCTGCTGACCCCAAGTGGGAGCTATCCAGGACCCGGCTGACACTTGGTAAGCCTCTTGGAGAAGGC
TGCTTTGGACAGGTGGTCATGGCAGAAGCTATTGGCATCGACAAGGACCGTACTGCCAAGCCTGTCACCGTGGCC
GTGAAGATGCTGAAAGATGATGCGACTGACAAGGACCTGTCGGACCTGGTATCTGAGATGGAGATGATGAAAATG
ATTGGCAAGCACAAGAACATCATTAACCTGCTGGGGGCGTGCACACAGGGTGGGCCCCTGTATGTGCTGGTGGAG
TACGCAGCCAAGGGCAATCTCCGGGAGTTCCTTCGGGCGCGGCGGCCTCCAGGCATGGACTACTCCTTTGATGCC
TGCAGGCTGCCAGAGGAACAGCTCACCTGCAAGGATCTAGTGTCCTGTGCCTACCAGGTGGCACGGGGCATGGAA
TACTTGGCTTCTCAGAAGTGTATTCACAGAGACTTGGCTGCCAGAAACGTCCTGGTGACCGAGGACAATGTGATG
AAGATTGCGGACTTTGGCCTGGCTCGAGATGTGCACAACCTGGACTACTACAAGAAGACCACAAATGGCCGGCTA
CCTGTGAAGTGGATGGCACCAGAGGCCCTTTTTGACCGAGTCTACACCCACCAGAGTGATGTTTGGTCTTTTGGT
GTCCTCCTCTGGGAGATCTTTACGCTGGGGGGCTCACCGTATCCTGGCATCCCAGTGGAAGAGCTTTTCAAGCTG
TTGAAAGAGGGCCACCGCATGGACAAGCCAGCCAGCTGCACACATGACCTGTACATGATCATGCGGGAATGTTGG
CATGCGGTGCCTTCACAGAGGCCCACCTTCAAGCAGTTGGTAGAGGATTTAGACCGCATCCTCACTGTGACATCA
ACCGACGAGTACTTGGACCTCTCCGTGCCGTTTGAGCAGTACTCGCCAGGTGGCCAGGACACGCCTAGCTCCAGC
TCGTCCGGAGATGACTCGGTGTTCACCCATGACCTGCTACCCCCAGGTCCACCCAGTAACGGGGGACCTCGGACG
TGA

Amino acid sequence sorted by gene/transcript

Code: non-underlined/underlined denoted alternating exons, bold face are aminoacids with intron-spanning codons

SEQ ID NO: 15. human FGF8a

MGSPRSALSC**L**LLHLLVLCLQAQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPF**A**KL
IVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRK
GSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR

SEQ ID NO: 16. human FGF8b

MGSPRSALSC**L**LLHLLVLCLQAQVTVQSSPNFTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINA
MAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWY
MAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR

SEQ ID NO: 17. human FGF8e

MGSPRSALSC**L**LLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQQHVREQSLVTDQLSRRLIRTYQLY
SRTSGKHVQVLANKRINAMAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEI
VLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQ
RTWAPEPR

SEQ ID NO: 18. human FGF8f

MGSPRSALSC**L**LLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQQVTVQSSPNFTQHVREQSLVTDQL
SRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSN
GKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNY
PPFTRSLRGSQRTWAPEPR

SEQ ID NO: 19. murine FGF8a

MGSPRSALSC**L**LLHLLVLCLQAQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPF**A**KL
IVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRK
GSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR

SEQ ID NO: 20. murine FGF8b

MGSPRSALSC**L**LLHLLVLCLQAQVTVQSSPNFTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINA
MAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWY
MAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR

SEQ ID NO: 21. murine FGF8e

MGSPRSALSC**L**LLHLLVLCLQAQEGPGGGPALGREPTSLLRAGREPQGVSQQHVREQSLVTDQLSRRLIRTYQLY
SRTSGKHVQVLANKRINAMAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEI
VLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQ
RTWAPEPR

SEQ ID NO: 22. murine FGF8f

MGSPRSALSC**L**LLHLLVLCLQAQEGPGGGPALGREPTSLLRAGREPQGVSQQVTVQSSPNFTQHVREQSLVTDQL
SRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPF**A**KLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSN

GKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNY
PPFTRSLRGSQRTWAPEPR

SEQ ID NO: 23. human FGF17

MGAARLLPNLTLCLQLLILCCQTQGENHPSPNFNQYVRDQGAMTDQLSRRQIREYQLYSRTSGKHVQVTGRRISA
TAEDGNKFAKLIVETDTFGSRVRIKGAESEKYICMNKRGKLIGKPSGKSKDCVFTEIVLENNYTAFQNARHEGWF
MAFTRQGRPRQASRSRQNQREAHFIKRLYQGQLPFPNHAEKQKQFEFVGSAPTRRTKRTRRPQPLT

SEQ ID NO: 24. murine FGF17

MGAARLLPNLTLCLQLLILCCQTQGENHPSPNFNQYVRDQGAMTDQLSRRQIREYQLYSRTSGKHVQVTGRRISA
TAEDGNKFAKLIVETDTFGSRVRIKGAESEKYICMNKRGKLIGKPSGKSKDCVFTEIVLENNYTAFQNARHEGWF
MAFTRQGRPRQASRSRQNQREAHFIKRLYQGQLPFPNHAERQKQFEFVGSAPTRRTKRTRRPQSQT

SEQ ID NO: 25. human FGF19

MRSGCVVVHVWILAGLWLAVAGRPLAFSDAGPHVHYGWGDPIRLRHLYTSGPHGLSSCFLRIRADGVVDCARGQS
AHSLLEIKAVALRTVAIKGVHSVRYLCMGADGKMQGLLQYSEEDCAFEEEIRPDGYNVYRSEKHRLPVSLSSAKQ
RQLYKNRGFLPLSHFLPMLPMVPEEPEDLRGHLESDMFSSPLETDSMDPFGLVTGLEAVRSPSFEK

SEQ ID NO: 26. murine FGF15

MARKWNGRAVARALVLATLWLAVSGRPLAQQSQSVSDEDPLFLYGWGKITRLQYLYSAGPYVSNCFLRIRSDGSV
DCEEDQNERNLLEFRAVALKTIAIKDVSSVRYLCMSADGKIYGLIRYSEEDCTFREEMDCLGYNQYRSMKHHLHI
IFIQAKPREQLQDQKPSNFIPVFHRSFFETGDQLRSKMFSLPLESDSMDPFRMVEDVDHLVKSPSFQK

SEQ ID NO: 27. human FGF21

MDSDETGFEHSGLWVSVLAGLLLGACQAHPIPDSSPLLQFGGQVRQRYLYTDDAQQTEAHLEIREDGTVGGAADQ
SPESLLQLKALKPGVIQILGVKTSRFLCQRPDGALYGSLHFDPEACSFRELLLEDGYNVYQSEAHGLPLHLPGNK
SPHRDPAPRGPARFLPLPGLPPALPEPPGILAPQPPDVGSSDPLSMVGPSQGRSPSYAS

SEQ ID NO: 28. murine FGF21

MEWMRSRVGTLGLWVRLLLAVFLLGVYQAYPIPDSSPLLQFGGQVRQRYLYTDDDQDTEAHLEIREDGTVVGAAH
RSPESLLELKALKPGVIQILGVKASRFLCQQPDGALYGSPHFDPEACSFRELLLEDGYNVYQSEAHGLPLRLPQK
DSPNQDATSWGPVRFLPMPGLLHEPQDQAGFLPPEPPDVGSSDPLSMVEPLQGRSPSYAS

SEQ ID NO: 43. human FGFR1

MWSWKCLLFWAVLVTATLCTARPSPTLPEQAQPWGAPVEVESFLVHPGDLLQLRCRLRDDVQSINWLRDGVQLAE
SNRTRITGEEVEVQDSVPADSGLYACVTSSPSGSDTTYFSVNVSDALPSSEDDDDDDSSSEEKETDNTKPNPVA
PYWTSPEKMEKKLHAVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWSIIMDSVVPSDKGN
YTCIVENEYGSINHTYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKIGP
DNLPYVQILKTAGVNTTDKEMEVLHLRNVSFEDAGEYTCLAGNSIGLSHHSAWLTVLEALEERPAVMTSPLYLEI
IIYCTGAFLISCMVGSVIVYKMKSGTKKSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLSSS
GTPMLAGVSEYELPEDPRWELPRDRLVLGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDLSD
LISEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLQARRPPGLEYCYNPSHNPEEQLSSKDLVS
CAYQVARGMEYLASKKCIHRDLAARNVLVTEDNVMKIADFGLARDIHHIDYYKKTTNGRLPVKWMAPEALFDRIY
THQSDVWSFGVLLWEIFTLGGSPYPGVPVEELFKLLKEGHRMDKPSNCTNELYMMMRDCWHAVPSQRPTFKQLVE
DLDRIVALTSNQEYLDLSMPLDQYSPSFPDTRSSTCSSGEDSVFSHEPLPEEPCLPRHPAQLANGGLKRR

SEQ ID NO: 44. murine FGFR1

MWGWKCLLFWAVLVTATLCTARPAPTLPEQAQPWGVPVEVESLLVHPGDLLQLRCRLRDDVQSINWLRDGVQLVE
SNRTRITGEEVEVRDSIPADSGLYACVTSSPSGSDTTYFSVNVSDALPSSEDDDDDDSSSEEKETDNTKPNRRP

VAPYWTSPEKMEKKLHAVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWSIIMDSVVPSDK
GNYTCIVENEYGSINHTYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKI
GPDNLPYVQILKTAGVNTTDKEMEVLHLRNVSFEDAGEYTCLAGNSIGLSHHSAWLTVLEALEERPAVMTSPLYL
EIIIYCTGAFLISCMLGSVIIYKMKSGTKKSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLS
SSGTPMLAGVSEYELPEDPRWELPRDRLVLGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDL
SDLISEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLQARRPPGLEYCYNPSHNPEEQLSSKDL
VSCAYQVARGMEYLASKKCIHRDLAARNVLVTEDNVMKIADFGLARDIHHIDYYKKTTNGRLPVKWMAPEALFDR
IYTHQSDVWSFGVLLWEIFTLGGSPYPGVPVEELFKLLKEGHRMDKPSNCTNELYMMMRDCWHAVPSQRPTFKQL
VEDLDRIVALTSNQEYLDLSIPLDQYSPSFPDTRSSTCSSGEDSVFSHEPLPEEPCLPRHPTQLANSGLKRR

SEQ ID NO: 45. human FGFR2

MVSWGRFICLVVVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEVYVAAPGESLEVRCLLKDAAVISWTKD
GVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTASRTVDSETWYFMVNVTDAISSGDDEDDTDGAEDFVSENSNN
KRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPMPTMRWLKNGKEFKQEHRIGGYKVRNQHWSLIMESVVPSD
KGNYTCVVENEYGSINHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVYSDAQPHIQWIKHVEKNGSK
YGPDGLPYLKVLKAAGVNTTDKEIEVLYIRNVTFEDAGEYTCLAGNSIGISFHSAWLTVLPAPGREKEITASPDY
LEIAIYCIGVFLIACMVVTVILCRMKNTTKKPDFSSQPAVHKLTKRIPLRRQVTVSAESSSSMNSNTPLVRITTR
LSSTADTPMLAGVSEYELPEDPKWEFPRDKLTLGKPLGEGCFGQVVMAEAVGIDKDKPKEAVTVAVKMLKDDATE
KDLSDLVSEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLRARRPPGMEYSYDINRVPEEQMTF
KDLVSCTYQLARGMEYLASQKCIHRDLAARNVLVTENNVMKIADFGLARDINNIDYYKKTTNGRLPVKWMAPEAL
FDRVYTHQSDVWSFGVLMWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPANCTNELYMMMRDCWHAVPSQRPTF
KQLVEDLDRILTLTTNEEYLDLSQPLEQYSPSYPDTRSSCSSGDDSVFSPDPMPYEPCLPQYPHINGSVKT

SEQ ID NO: 46. murine FGFR2

MGLPSTWRYGRGPGIGTVTMVSWGRFICLVLVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEAYVVAPGE
SLELQCMLKDAAVISWTKDGVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTAARTVDSETWYFMVNVTDAISSG
DDEDDTDSSEDVVSENRSNQRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPTPTMRWLKNGKEFKQEHRIGG
YKVRNQHWSLIMESVVPSDKGNYTCLVENEYGSINHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVY
SDAQPHIQWIKHVEKNGSKYGPDGLPYLKVLKAAGVNTTDKEIEVLYIRNVTFEDAGEYTCLAGNSIGISFHSAW
LTVLPAPVREKEITASPDYLEIAIYCIGVFLIACMVVTVIFCRMKTTTKKPDFSSQPAVHKLTKRIPLRRQVTVS
AESSSSMNSNTPLVRITTRLSSTADTPMLAGVSEYELPEDPKWEFPRDKLTLGKPLGEGCFGQVVMAEAVGIDKD
KPKEAVTVAVKMLKDDATEKDLSDLVSEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLRARRP
PGMEYSYDINRVPEEQMTFKDLVSCTYQLARGMEYLASQKCIHRDLAARNVLVTENNVMKIADFGLARDINNIDY
YKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLMWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPTNCTNE
LYMMMRDCWHAVPSQRPTFKQLVEDLDRILTLTTNEEYLDLTQPLEQYSPSYPDTRSSCSSGDDSVFSPDPMPYE
PCLPQYPHINGSVKT

SEQ ID NO: 47. human FGFR3

MGAPACALALCVAVAIVAGASSESLGTEQRVVGRAAEVPGPEPGQQEQLVFGSGDAVELSCPPPGGGPMGPTVWV
KDGTGLVPSERVLVGPQRLQVLNASHEDSGAYSCRQRLTQRVLCHFSVRVTDAPSSGDDEDGEDEAEDTGVDTGA
PYWTRPERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGREFRGEHRIGGIKLRHQQWSLVMESVVPSDRGN
YTCVVENKFGSIRQTYTLDVLERSPHRPILQAGLPANQTAVLGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGP
DGTPYVTVLKTAGANTTDKELEVLSLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELVEADEAGSVYAG
ILSYGVGFFLFILVVAAVTLCRLRSPPKKGLGSPTVHKISRFPLKRQVSLESNASMSSNTPLVRIARLSSGEGPT
LANVSELELPADPKWELSRARLTLGKPLGEGCFGQVVMAEAIGIDKDRAAKPVTVAVKMLKDDATDKDLSDLVSE
MEMMKMIGKHKNIINLLGACTQGGPLYVLVEYAAKGNLREFLRARRPPGLDYSFDTCKPPEEQLTFKDLVSCAYQ
VARGMEYLASQKCIHRDLAARNVLVTEDNVMKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQS
DVWSFGVLLWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPANCTHDLYMIMRECWHAAPSQRPTFKQLVEDLDR
VLTVTSTDEYLDLSAPFEQYSPGGQDTPSSSSSGDDSVFAHDLLPPAPPSSGGSRT

SEQ ID NO: 48. murine FGFR3

MVVPACVLVFCVAVVAGATSEPPGPEQRVVRRAAEVPGPEPSQQEQVAFGSGDTVELSCHPPGGAPTGPTVWAKD
GTGLVASHRILVGPQRLQVLNASHEDAGVYSCQHRLTRRVLCHFSVRVTGAPYWTRPERMDKKLLAVPAANTVRF
RCPAAGNPTPSISWLKNGKEFRGEHRIGGIKLRHQQWSLVMESVVPSDRGNYTCVVENKFGSIRQTYTLDVLERS

PHRPILQAGLPANQTAILGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYVTVLKTAGANTTDKELEVL
SLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELMETDEAGSVYAGVLSYGVVFFLFILVVAAVILCRLR
SPPKKGLGSPTVHKVSRFPLKRQVSLESNSSMNSNTPLVRIARLSSGEGPVLANVSELELPADPKWELSRTRLTL
GKPLGEGCFGQVVMAEAIGIDKDRTAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKMIGKHKNIINLLGACTQGG
PLYVLVEYAAKGNLREFLRARRPPGMDYSFDACRLPEEQLTCKDLVSCAYQVARGMEYLASQKCIHRDLAARNVL
VTEDNVMKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLLWEIFTLGGSPYPGIP
VEELFKLLKEGHRMDKPASCTHDLYMIMRECWHAVPSQRPTFKQLVEDLDRILTVTSTDEYLDLSVPFEQYSPGG
QDTPSSSSSGDDSVFTHDLLPPGPPSNGGPRT

SEQ ID NO: 49. human FGFR4

MRLLLALLGVLLSVPGPPVLSLEASEEVELEPCLAPSLEQQEQELTVALGQPVRLCCGRAERGGHWYKEGSRLAP
AGRVRGWRGRLEIASFLPEDAGRYLCLARGSMIVLQNLTLITGDSLTSSNDDEDPKSHRDPSNRHSYPQQAPYWT
HPQRMEKKLHAVPAGNTVKFRCPAAGNPTPTIRWLKDGQAFHGENRIGGIRLRHQHWSLVMESVVPSDRGTYTCL
VENAVGSIRYNYLLDVLERSPHRPILQAGLPANTTAVVGSDVELLCKVYSDAQPHIQWLKHIVINGSSFGADGFP
YVQVLKTADINSSEVEVLYLRNVSAEDAGEYTCLAGNSIGLSYQSAWLTVLPEEDPTWTAAAPEARYTDIILYAS
GSLALAVLLLLAGLYRGQALHGRHPRPPATVQKLSRFPLARQFSLESGSSGKSSSSLVRGVRLSSSGPALLAGLV
SLDLPLDPLWEFPRDRLVLGKPLGEGCFGQVVRAEAFGMDPARPDQASTVAVKMLKDNASDKDLADLVSEMEVMK
LIGRHKNIINLLGVCTQEGPLYVIVECAAKGNLREFLRARRPPGPDLSPDGPRSSEGPLSFPVLVSCAYQVARGM
QYLESRKCIHRDLAARNVLVTEDNVMKIADFGLARGVHHIDYYKKTSNGRLPVKWMAPEALFDRVYTHQSDVWSF
GILLWEIFTLGGSPYPGIPVEELFSLLREGHRMDRPPHCPPELYGLMRECWHAAPSQRPTFKQLVEALDKVLLAV
SEEYLDLRLTFGPYSPSGGDASSTCSSSDSVFSHDPLPLGSSSFPFGSGVQT

SEQ ID NO: 50. murine FGFR4

MWLLLALLSIFQGTPALSLEASEEMEQEPCLAPILEQQEQVLTVALGQPVRLCCGRTERGRHWYKEGSRLASAGR
VRGWRGRLEIASFLPEDAGRYLCLARGSMTVVHNLTLLMDDSLTSISNDEDPKTLSSSSSGHVYPQQAPYWTHPQ
RMEKKLHAVPAGNTVKFRCPAAGNPMPTIHWLKDGQAFHGENRIGGIRLRHQHWSLVMESVVPSDRGTYTCLVEN
SLGSIRYSYLLDVLERSPHRPILQAGLPANTTAVVGSDVELLCKVYSDAQPHIQWLKHVVINGSSFGADGFPYVQ
VLKTTDINSSEVEVLYLRNVSAEDAGEYTCLAGNSIGLSYQSAWLTVLPEEDLTWTTATPEARYTDIILYVSGSL
VLLVLLLLAGVYHRQVIRGHYSRQPVTIQKLSRFPLARQFSLESRSSGKSSLSLVRGVRLSSSGPPLLTGLVNLD
LPLDPLWEFPRDRLVLGKPLGEGCFGQVVRAEAFGMDPSRPDQTSTVAVKMLKDNASDKDLADLVSEMEVMKLIG
RHKNIINLLGVCTQEGPLYVIVECAAKGNLREFLRARRPPGPDLSPDGPRSSEGPLSFPALVSCAYQVARGMQYL
ESRKCIHRDLAARNVLVTEDDVMKIADFGLARGVHHIDYYKKTSNGRLPVKWMAPEALFDRVYTHQSDVWSFGIL
LWEIFTLGGSPYPGIPVEELFSLLREGHRMERPPNCPSELYGLMRECWHAAPSQRPTFKQLVEALDKVLLAVSEE
YLDLRLTFGPFSPSNGDASSTCSSSDSVFSHDPLPLEPSPFPFSDSQTT

SEQ ID NO: 87 human FGFR3 spliceform c

MGAPACALALCVAVAIVAGASSESLGTEQRVVGRAAEVPGPEPGQQEQLVFGSGDAVELSCPPPGGGPMGPTVWV
KDGTGLVPSERVLVGPQRLQVLNASHEDSGAYSCRQRLTQRVLCHFSVRVTDAPSSGDDEDGEDEAEDTGVDTGA
PYWTRPERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGREFRGEHRIGGIKLRHQQWSLVMESVVPSDRGN
YTCVVENKFGSIRQTYTLDVLERSPHRPILQAGLPANQTAVLGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGP
DGTPYVTVLKTAGANTTDKELEVLSLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELVEADEAGSVYAG
ILSYGVGFFLFILVVAAVTLCRLRSPPKKGLGSPTVHKISRFPLKRQVSLESNASMSSNTPLVRIARLSSGEGPT
LANVSELELPADPKWELSRARLTLGKPLGEGCFGQVVMAEAIGIDKDRAAKPVTVAVKMLKDDATDKDLSDLVSE
MEMMKMIGKHKNIINLLGACTQGGPLYVLVEYAAKGNLREFLRARRPPGLDYSFDTCKPPEEQLTFKDLVSCAYQ
VARGMEYLASQKCIHRDLAARNVLVTEDNVMKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQS
DVWSFGVLLWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPANCTHDLYMIMRECWHAAPSQRPTFKQLVEDLDR
VLTVTSTDEYLDLSAPFEQYSPGGQDTPSSSSSGDDSVFAHDLLPPAPPSSGGSRT

SEQ ID NO: 88 murine FGFR3 spliceform c

MVVPACVLVFCVAVVAGATSEPPGPEQRVVRRAAEVPGPEPSQQEQVAFGSGDTVELSCHPPGGAPTGPTVWAKD
GTGLVASHRILVGPQRLQVLNASHEDAGVYSCQHRLTRRVLCHFSVRVTDAPSSGDDEDGEDVAEDTGAPYWTRP
ERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGKEFRGEHRIGGIKLRHQQWSLVMESVVPSDRGNYTCVVE
NKFGSIRQTYTLDVLERSPHRPILQAGLPANQTAILGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYV
TVLKTAGANTTDKELEVLSLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELMETDEAGSVYAGVLSYGV
VFFLFILVVAAVILCRLRSPPKKGLGSPTVHKVSRFPLKRQVSLESNSSMNSNTPLVRIARLSSGEGPVLANVSE

LELPADPKWELSRTRLTLGKPLGEGCFGQVVMAEAIGIDKDRTAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKM
IGKHKNIINLLGACTQGGPLYVLVEYAAKGNLREFLRARRPPGMDYSFDACRLPEEQLTCKDLVSCAYQVARGME
YLASQKCIHRDLAARNVLVTEDNVMKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFG
VLLWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPASCTHDLYMIMRECWHAVPSQRPTFKQLVEDLDRILTVTS
TDEYLDLSVPFEQYSPGGQDTPSSSSSGDDSVFTHDLLPPGPPSNGGPRT

SEQ ID NO: 89 human FGFR1b based on Transcript FGFR1-020 ENST00000397108

MWSWKCLLFWAVLVTATLCTARPSPTLPEQAQPWGAPVEVESFLVHPGDLLQLRCRLRDDVQSINWLRDGVQLAE
SNRTRITGEEVEVQDSVPADSGLYACVTSSPSGSDTTYFSVNVSDALPSSEDDDDDDDSSSEEKETDNTKPNPVA
PYWTSPEKMEKKLHAVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWSIIMDSVVPSDKGN
YTCIVENEYGSINHTYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKIGP
DNLPYVQILKTAGVNTTDKEMEVLHLRNVSFEDAGEYTCLAGNSIGLSHHSAWLTVLEALEERPAVMTSPLYLEI
IIYCTGAFLISCMVGSVIVYKMKSGTKKSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLSSS
GTPMLAGVSEYELPEDPRWELPRDRLVLGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDLSD
LISEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLQARRPPGLEYCYNPSHNPEEQLSSKDLVS
CAYQVARGMEYLASKKCIHRDLAARNVLVTEDNVMKIADFGLARDIHHIDYYKKTTNGRLPVKWMAPEALFDRIY
THQSDVWSFGVLLWEIFTLGGSPYPGVPVEELFKLLKEGHRMDKPSNCTNELYMMMRDCWHAVPSQRPTFKQLVE
DLDRIVALTSNQEYLDLSMPLDQYSPSFPDTRSSTCSSGEDSVFSHEPLPEEPCLPRHPAQLANGGLKRR

SEQ ID NO: 90 human FGFR1c based on transcript FGFR1-011 ENST00000397103

MWSWKCLLFWAVLVTATLCTARPSPTLPEQDALPSSEDDDDDDDSSSEEKETDNTKPNPVAPYWTSPEKMEKKLH
AVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWSIIMDSVVPSDKGNYTCIVENEYGSINH
TYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKIGPDNLPYVQILKHSGI
NSSDAEVLTLFNVTEAQSGEYVCKVSNYIGEANQSAWLTVTRPVAKALEERPAVMTSPLYLEIIIYCTGAFLISC
MVGSVIVYKMKSGTKKSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLSSSGTPMLAGVSEYE
LPEDPRWELPRDRLVLGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDLSDLISEMEMMKMIG
KHKNIINLLGACTQDGPLYVIVEYASKGNLREYLQARRPPGLEYCYNPSHNPEEQLSSKDLVSCAYQVARGMEYL
ASKKCIHRDLAARNVLVTEDNVMKIADFGLARDIHHIDYYKKTTNGRLPVKWMAPEALFDRIYTHQSDVWSFGVL
LWEIFTLGGSPYPGVPVEELFKLLKEGHRMDKPSNCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRIVALTSNQ
EYLDLSMPLDQYSPSFPDTRSSTCSSGEDSVFSHEPLPEEPCLPRHPAQLANGGLKRR

SEQ ID NO: 91 human FGFR2b based on transcript FGFR2-201 ENST00000351936

MVSWGRFICLVVVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEVYVAAPGESLEVRCLLKDAAVISWTKD
GVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTASRTVDSETWYFMVNVTDAISSGDDEDDTDGAEDFVSENSNN
KRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPMPTMRWLKNGKEFKQEHRIGGYKVRNQHWSLIMESVVPSD
KGNYTCVVENEYGSINHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVYSDAQPHIQWIKHVEKNGSK
YGPDGLPYLKVLKVFAAGVNTTDKEIEVLYIRNVTFEDAGEYTCLAGNSIGISFHSAWLTVLPAPGREKEITASP
DYLEIAIYCIGVFLIACMVVTVILCRMKNTTKKPDFSSQPAVHKLTKRIPLRRQVSAESSSSMNSNTPLVRITTR
LSSTADTPMLAGVSEYELPEDPKWEFPRDKLTLGKPLGEGCFGQVVMAEAVGIDKDKPKEAVTVAVKMLKDDATE
KDLSDLVSEMEMMKMIGKHKNIINLLGACTQDGPLYVIVEYASKGNLREYLRARRPPGMEYSYDINRVPEEQMTF
KDLVSCTYQLARGMEYLASQKCIHRDLAARNVLVTENNVMKIADFGLARDINNIDYYKKTTNGRLPVKWMAPEAL
FDRVYTHQSDVWSFGVLMWEIFTLGGSPYPGIPVEELFKLLKEGHRMDKPANCTNELYMMMRDCWHAVPSQRPTF
KQLVEDLDRILTLTTNEEYLDLSQPLEQYSPSYPDTRSSCSSGDDSVFSPDPMPYEPCLPQYPHINGSVKT

SEQ ID NO: 92 human FGFR2c based on transcript FGFR2-010 ENST00000457416

MVSWGRFICLVVVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEVYVAAPGESLEVRCLLKDAAVISWTKD
GVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTASRTVDSETWYFMVNVTDAISSGDDEDDTDGAEDFVSENSNN
KRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPMPTMRWLKNGKEFKQEHRIGGYKVRNQHWSLIMESVVPSD
KGNYTCVVENEYGSI
NHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVYSDAQPHIQWIKHVEKNGSKYGPDGLPYLKVLKHS
GINSSNAEVLALFNVTEADAGEYICKVSNYIGQANQSAWLTVLPKQQAPGREKEITASPDYLEIAIYCIGVFLIA
CMVVTVILCRMKNTTKKPDFSSQPAVHKLTKRIPLRRQVTVSAESSSSMNSNTPLVRITTRLSSTADTPMLAGVS
EYELPEDPKWEFPRD
KLTLGKPLGEGCFGQVVMAEAVGIDKDKPKEAVTVAVKMLKDDATEKDLSDLVSEMEMMKMIGKHKNIINLLGAC
TQDGPLYVIVEYASKGNLREYLRARRPPGMEYSYDINRVPEEQMTFKDLVSCTYQLARGMEYLASQKCIHRDLAA
RNVLVTENNVMKIADFGLARDINNIDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLMWEIFTLGGSPY
PGIPVEELFKLLKEG

HRMDKPANCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRILTLTTNEEYLDLSQPLEQYSPSYPDTRSSCSSGD
DSVFSPDPMPYEPCLPQYPHINGSVKT

SEQ ID NO: 93 human FGFR3b based on transcript FGFR3-201 ENST00000340107

MGAPACALALCVAVAIVAGASSESLGTEQRVVGRAAEVPGPEPGQQEQLVFGSGDAVELSCPPPGGGPMGPTVWV
KDGTGLVPSERVLVGPQRLQVLNASHEDSGAYSCRQRLTQRVLCHFSVRVTDAPSSGDDEDGEDEAEDTGVDTGA
PYWTRPERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGREFRGEHRIGGIKLRHQQWSLVMESVVPSDRGN
YTCVVENKFGSIRQT
YTLDVLERSPHRPILQAGLPANQTAVLGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYVTVLKSWISE
SVEADVRLRLANVSERDGGEYLCRATNFIGVAEKAFWLSVHGPRAAEEELVEADEAGSVYAGILSYGVGFFLFIL
VVAAVTLCRLRSPPKKGLGSPTVHKISRFPLKRQVSLESNASMSSNTPLVRIARLSSGEGPTLANVSELELPADP
KWELSRARLTLGKPL
GEGCFGQVVMAEAIGIDKDRAAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKMIGKHKNIINLLGACTQGGPLYV
LVEYAAKGNLREFLRARRPPGLDYSFDTCKPPEEQLTFKDLVSCAYQVARGMEYLASQKCIHRDLAARNVLVTED
NVMKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLLWEIFTLGGSPYPGIPVEEL
FKLLKEGHRMDKPAN
CTHDLYMIMRECWHAAPSQRPTFKQLVEDLDRVLTVTSTDEYLDLSAPFEQYSPGGQDTPSSSSSGDDSVFAHDL
LPPAPPSSGGSRT

SEQ ID NO: 94 human FGFR3c based on transcript FGFR3-203: ENST00000440486

MGAPACALALCVAVAIVAGASSESLGTEQRVVGRAAEVPGPEPGQQEQLVFGSGDAVELSCPPPGGGPMGPTVWV
KDGTGLVPSERVLVGPQRLQVLNASHEDSGAYSCRQRLTQRVLCHFSVRVTDAPSSGDDEDGEDEAEDTGVDTGA
PYWTRPERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGREFRGEHRIGGIKLRHQQWSLVMESVVPSDRGN
YTCVVENKFGSIRQT
YTLDVLERSPHRPILQAGLPANQTAVLGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYVTVLKTAGAN
TTDKELEVLSLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELVEADEAGSVYAGILSYGVGFFLFILVV
AAVTLCRLRSPPKKGLGSPTVHKISRFPLKRQVSLESNASMSSNTPLVRIARLSSGEGPTLANVSELELPADPKW
ELSRARLTLGKPLGE
GCFGQVVMAEAIGIDKDRAAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKMIGKHKNIINLLGACTQGGPLYVLV
EYAAKGNLREFLRARRPPGLDYSFDTCKPPEEQLTFKDLVSCAYQVARGMEYLASQKCIHRDLAARNVLVTEDNV
MKIADFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLLWEIFTLGGSPYPGIPVEELFK
LLKEGHRMDKPANCT
HDLYMIMRECWHAAPSQRPTFKQLVEDLDRVLTVTSTDEYLDLSAPFEQYSPGGQDTPSSSSSGDDSVFAHDLLP
PAPPSSGGSRT

SEQ ID NO: 95 murine FGFR1b based on transcript FGFR1-201 ENSMUST00000178276

MWGWKCLLFWAVLVTATLCTARPAPTLPEQDALPSSEDDDDDDDSSSEEKETDNTKPNPVAPYWTSPEKMEKKLH
AVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWSIIMDSVVPSDKGNYTCIVENEYGSINH
TYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKIGPDNLPYVQILKHSGI
NSSDAEVLTLFNVTE
AQSGEYVCKVSNYIGEANQSAWLTVTRPVAKALEERPAVMTSPLYLEIIIYCTGAFLISCMLGSVIIYKMKSGTK
KSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLSSSGTPMLAGVSEYELPEDPRWELPRDRLV
LGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDLSDLISEMEMMKMIGKHKNIINLLGACTQD
GPLYVIVEYASKGNL
REYLQARRPPGLEYCYNPSHNPEEQLSSKDLVSCAYQVARGMEYLASKKCIHRDLAARNVLVTEDNVMKIADFGL
ARDIHHIDYYKKTTNGRLPVKWMAPEALFDRIYTHQSDVWSFGVLLWEIFTLGGSPYPGVPVEELFKLLKEGHRM
DKPSNCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRIVALTSNQEYLDLSIPLDQYSPSFPDTRSSTCSSGEDS
VFSHEPLPEEPCLPR
HPTQLANSGLKRR

SEQ ID NO: 96 murine FGFR1c based on transcript FGFR1-202 ENSMUST00000179592

MWGWKCLLFWAVLVTATLCTARPAPTLPEQVGSSSWPLWVAAAAQPWGVPVEVESLLVHPGDLLQLRCRLRDDVQ
SINWLRDGVQLVESNRTRITGEEVEVRDSIPADSGLYACVTSSPSGSDTTYFSVNVSDALPSSEDDDDDDSSSE
EKETDNTKPNPVAPYWTSPEKMEKKLHAVPAAKTVKFKCPSSGTPNPTLRWLKNGKEFKPDHRIGGYKVRYATWS

IIMDSVVPSDKGNYT
CIVENEYGSINHTYQLDVVERSPHRPILQAGLPANKTVALGSNVEFMCKVYSDPQPHIQWLKHIEVNGSKIGPDN
LPYVQILKTAGVNTTDKEMEVLHLRNVSFEDAGEYTCLAGNSIGLSHHSAWLTVLEALEERPAVMTSPLYLEIII
YCTGAFLISCMLGSVIIYKMKSGTKKSDFHSQMAVHKLAKSIPLRRQVTVSADSSASMNSGVLLVRPSRLSSSGT
PMLAGVSEYELPEDP
RWELPRDRLVLGKPLGEGCFGQVVLAEAIGLDKDKPNRVTKVAVKMLKSDATEKDLSDLISEMEMMKMIGKHKNI
INLLGACTQDGPLYVIVEYASKGNLREYLQARRPPGLEYCYNPSHNPEEQLSSKDLVSCAYQVARGMEYLASKKC
IHRDLAARNVLVTEDNVMKIADFGLARDIHHIDYYKKTTNGRLPVKWMAPEALFDRIYTHQSDVWSFGVLLWEIF
TLGGSPYPGVPVEEL
FKLLKEGHRMDKPSNCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRIVALTSNQEYLDLSIPLDQYSPSFPDTR
SSTCSSGEDSVFSHEPLPEEPCLPRHPTQLANSGLKRR

SEQ ID NO: 97 murine FGFR2b based on transcript FGFR2-011 ENSMUST00000119260

MVSWGRFICLVLVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEAYVVAPGESLELQCMLKDAAVISWTKD
GVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTAARTVDSETWYFMVNVTDAISSGDDEDDTDSSEDVVSENRSN
QRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPTPTMRWLKNGKEFKQEHRIGGYKVRNQHWSLIMESVVPSD
KGNYTCLVENEYGSI
NHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVYSDAQPHIQWIKHVEKNGSKYGPDGLPYLKVLKAA
GVNTTDKEIEVLYIRNVTFEDAGEYTCLAGNSIGISFHSAWLTVLPAPVREKEITASPDYLEIAIYCIGVFLIAC
MVVTVIFCRMKTTTKKPDFSSQPAVHKLTKRIPLRRQVSAESSSSMNSNTPLVRITTRLSSTADTPMLAGVSEYE
LPEDPKWEFPRDKLT
LGKPLGEGCFGQVVMAEAVGIDKDKPKEAVTVAVKMLKDDATEKDLSDLVSEMEMMKMIGKHKNIINLLGACTQD
GPLYVIVEYASKGNLREYLRARRPPGMEYSYDINRVPEEQMTFKDLVSCTYQLARGMEYLASQKCIHRDLAARNV
LVTENNVMKIADFGLARDINNIDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLMWEIFTLGGSPYPGI
PVEELFKLLKEGHRM
DKPTNCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRILTLTTNEEYLDLTQPLEQYSPSYPDTRSSCSSGDDSV
FSPDPMPYEPCLPQYPHINGSVKT

SEQ ID NO: 98 murine FGFR2c based on transcript FGFR2-012 ENSMUST00000117089

MVSWGRFICLVLVTMATLSLARPSFSLVEDTTLEPEEPPTKYQISQPEAYVVAPGESLELQCMLKDAAVISWTKD
GVHLGPNNRTVLIGEYLQIKGATPRDSGLYACTAARTVDSETWYFMVNVTDAISSGDDEDDTDSSEDVVSENRSN
QRAPYWTNTEKMEKRLHAVPAANTVKFRCPAGGNPTPTMRWLKNGKEFKQEHRIGGYKVRNQHWSLIMESVVPSD
KGNYTCLVENEYGSI
NHTYHLDVVERSPHRPILQAGLPANASTVVGGDVEFVCKVYSDAQPHIQWIKHVEKNGSKYGPDGLPYLKVLKHS
GINSSNAEVLALFNVTEMDAGEYICKVSNYIGQANQSAWLTVLPKQQAPVREKEITASPDYLEIAIYCIGVFLIA
CMVVTVIFCRMKTTTKKPDFSSQPAVHKLTKRIPLRRQVSAESSSSMNSNTPLVRITTRLSSTADTPMLAGVSEY
ELPEDPKWEFPRDKL
TLGKPLGEGCFGQVVMAEAVGIDKDKPKEAVTVAVKMLKDDATEKDLSDLVSEMEMMKMIGKHKNIINLLGACTQ
DGPLYVIVEYASKGNLREYLRARRPPGMEYSYDINRVPEEQMTFKDLVSCTYQLARGMEYLASQKCIHRDLAARN
VLVTENNVMKIADFGLARDINNIDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLMWEIFTLGGSPYPG
IPVEELFKLLKEGHR
MDKPTNCTNELYMMMRDCWHAVPSQRPTFKQLVEDLDRILTLTTNEEYLDLTQPLEQYSPSYPDTRSSCSSGDDS
VFSPDPMPYEPCLPQYPHINGSVKT

SEQ ID NO: 99 murine FGFR3b based on transcipt FGFR3-201 ENSMUST00000114411

MVVPACVLVFCVAVVAGATSEPPGPEQRVVRRAAEVPGPEPSQQEQVAFGSGDTVELSCHPPGGAPTGPTVWAKD
GTGLVASHRILVGPQRLQVLNASHEDAGVYSCQHRLTRRVLCHFSVRVTDAPSSGDDEDGEDVAEDTGAPYWTRP
ERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGKEFRGEHRIGGIKLRHQQWSLVMESVVPSDRGNYTCVVE
NKFGSIRQTYTLDVL
ERSPHRPILQAGLPANQTAILGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYVTVLKSWISENVEADA
RLRLANVSERDGGEYLCRATNFIGVAEKAFWLRVHGPQAAEEELMETDEAGSVYAGVLSYGVVFFLFILVVAAVI
LCRLRSPPKKGLGSPTVHKVSRFPLKRQVSLESNSSMNSNTPLVRIARLSSGEGPVLANVSELELPADPKWELSR
TRLTLGKPLGEGCFG

QVVMAEAIGIDKDRTAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKMIGKHKNIINLLGACTQGGPLYVLVEYAA
KGNLREFLRARRPPGMDYSFDACRLPEEQLTCKDLVSCAYQVARGMEYLASQKCIHRDLAARNVLVTEDNVMKIA
DFGLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLLWEIFTLGGSPYPGIPVEELFKLLKE
GHRMDKPASCTHDLY
MIMRECWHAVPSQRPTFKQLVEDLDRILTVTSTDEYLDLSVPFEQYSPGGQDTPSSSSSGDDSVFTHDLLPPGPP
SNGGPRT

SEQ ID NO: 100 murine FGFR3c based on transcript FGFR3-202 ENSMUST00000169212

MVVPACVLVFCVAVVAGATSEPPGPEQRVVRRAAEVPGPEPSQQEQVAFGSGDTVELSCHPPGGAPTGPTVWAKD
GTGLVASHRILVGPQRLQVLNASHEDAGVYSCQHRLTRRVLCHFSVRVTDAPSSGDDEDGEDVAEDTGAPYWTRP
ERMDKKLLAVPAANTVRFRCPAAGNPTPSISWLKNGKEFRGEHRIGGIKLRHQQWSLVMESVVPSDRGNYTCVVE
NKFGSIRQTYTLDVL
ERSPHRPILQAGLPANQTAILGSDVEFHCKVYSDAQPHIQWLKHVEVNGSKVGPDGTPYVTVLKTAGANTTDKEL
EVLSLHNVTFEDAGEYTCLAGNSIGFSHHSAWLVVLPAEEELMETDEAGSVYAGVLSYGVVFFLFILVVAAVILC
RLRSPPKKGLGSPTVHKVSRFPLKRQVSLESNSSMNSNTPLVRIARLSSGEGPVLANVSELELPADPKWELSRTR
LTLGKPLGEGCFGQV
VMAEAIGIDKDRTAKPVTVAVKMLKDDATDKDLSDLVSEMEMMKMIGKHKNIINLLGACTQGGPLYVLVEYAAKG
NLREFLRARRPPGMDYSFDACRLPEEQLTCKDLVSCAYQVARGMEYLASQKCIHRDLAARNVLVTEDNVMKIADF
GLARDVHNLDYYKKTTNGRLPVKWMAPEALFDRVYTHQSDVWSFGVLLWEIFTLGGSPYPGIPVEELFKLLKEGH
RMDKPASCTHDLYMI
MRECWHAVPSQRPTFKQLVEDLDRILTVTSTDEYLDLSVPFEQYSPGGQDTPSSSSSGDDSVFTHDLLPPGPPSN
GGPRT

Primer sequences:

Actb AGAGGGAAATCGTGCGTGAC (SEQ ID NO: 51),

CAATAGTGATGACCTGGCCGT (SEQ ID NO: 52);

Cidea TGCTCTTCTGTATCGCCCAGT (SEQ ID NO: 53),

GCCGTGTTAAGGAATCTGCTG (SEQ ID NO: 54);

Cox7a1 CCGACAATGACCTCCCAGTA (SEQ ID NO: 55),

TGTTTGTCCAAGTCCTCCAA (SEQ ID NO: 56);

Elovl3 TCCGCGTTCTCATGTAGGTCT (SEQ ID NO: 57),

GGACCTGATGCAACCCTATGA (SEQ ID NO: 58);

Foxc2 ACGAGTGCGGATTTGTAACC (SEQ ID NO: 59),

CAGTTTGGGGAGGGACCTAT (SEQ ID NO: 60);

Hsp90 AGGAGGGTCAAGGAAGTGGT (SEQ ID NO: 61),

TTTTTCTTGTCTTTGCCGCT (SEQ ID NO: 62);

Otop1 GGACCTGATGCAACCCTATGA (SEQ ID NO: 63),

ACCATGCTCTACGTGCTGTG (SEQ ID NO: 64);

Ppargc1a GGACGGAAGCAATTTTTCAA (SEQ ID NO: 65),

GAGTCTTGGGAAAGGACACG (SEQ ID NO: 66);

Prb TAAACATCTCCCAGCGGAGT (SEQ ID NO: 67),
ACAACCATGAGCCAGGAGTC (SEQ ID NO: 68);
Prdm16 CTGTTAGCTTTGGAGCCGAC (SEQ ID NO: 69),
GACGAGGGTCCTGTGATGTT (SEQ ID NO: 70);
Ucp1 TCTCTGCCAGGACAGTACCC (SEQ ID NO: 71),
AGAAGCCCAATGATGTTCAG (SEQ ID NO: 72);
Fgfr1 CCGGATCTACACACACCAGA (SEQ ID NO: 73),
CCACCAACTGCTTGAACGTA (SEQ ID NO: 74);
Fgfr2 AGGGACACAGGATGGACAAG (SEQ ID NO: 75),
AAACACAGAATCGTCCCCTG (SEQ ID NO: 76);
Fgfr3 ACCGAGTCTACACCCACCAG (SEQ ID NO: 77),
TGAGGATGCGGTCTAAATCC (SEQ ID NO: 78);
Fgfr4 TGGAAGCTCTGGACAAGGTC (SEQ ID NO: 79),
ATACAACATTGCTGCTCCCC (SEQ ID NO: 80);
aklotho GGCTCAACTCTCCCAGTCAG (SEQ ID NO: 81),
CGCAAACTAGCCACAAAGGT (SEQ ID NO: 82);
bklotho ATGTCCAGGAGGCTCTGAAA (SEQ ID NO. 83),
AGCAAATGGTGCAGTCTGTG (SEQ ID NO: 84)
fgfr3c rev CTCCTTGTCGGTGGT (SEQ ID NO: 101)and
fgfr3c fwd ACGGCACGCCCT ACG (SEQ ID NO: 102).

Human and murine FGF8 isoforms have the following level of identity of amino acid sequences: FGF8a 100%, FGF8b 100%, FGF8e 98,28%, FGF8f 98,36%. Human and murine FGF8 isoforms have the following level of identity of nucleic acid level: FGF8a: 94,96%, FGF8b 95,22%, FGF8e 94,59%, FGF8f 94,83%.

aa-Human vs. Mouse FGF8a

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8a         100.00  100.00
     2: m8a         100.00  100.00
CLUSTAL O(1.2.1) multiple sequence alignment

h8a       MGSPRSALSCLLLHLLVLCLQAQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANK
m8a       MGSPRSALSCLLLHLLVLCLQAQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANK
          ************************************************************

h8a       RINAMAEDGDPFAKLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTE
m8a       RINAMAEDGDPFAKLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTE
          ************************************************************
```

```
h8a       IVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRF
m8a       IVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRF
          ************************************************************

h8a       EFLNYPPFTRSLRGSQRTWAPEPR
m8a       EFLNYPPFTRSLRGSQRTWAPEPR
          ************************
```

aa-Human vs. Mouse FGF8b

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8b         100.00  100.00
     2: m8b         100.00  100.00
CLUSTAL O(1.2.1) multiple sequence alignment

h8b       MGSPRSALSCLLLHLLVLCLQAQVTVQSSPNFTQHVREQSLVTDQLSRRLIRTYQLYSRT
m8b       MGSPRSALSCLLLHLLVLCLQAQVTVQSSPNFTQHVREQSLVTDQLSRRLIRTYQLYSRT
          ************************************************************

h8b       SGKHVQVLANKRINAMAEDGDPFAKLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKS
m8b       SGKHVQVLANKRINAMAEDGDPFAKLIVETDTFGSRVRVRGAETGLYICMNKKGKLIAKS
          ************************************************************

h8b       NGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPR
m8b       NGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKGSKTRQHQREVHFMKRLPR
          ************************************************************

h8b       GHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR
m8b       GHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR
          ***********************************
```

aa-Human vs. Mouse FGF8e

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8e         100.00   98.28
     2: m8e          98.28  100.00


CLUSTAL O(1.2.1) multiple sequence alignment

h8e       MGSPRSALSCLLLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQQHVREQSLV
m8e       MGSPRSALSCLLLHLLVLCLQAQEGPGGGPALGREPTSLLRAGREPQGVSQQHVREQSLV
          *************************** ******* :**:********************

h8e       TDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETDTFGSRVRVRGA
m8e       TDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETDTFGSRVRVRGA
          ************************************************************

h8e       ETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKG
m8e       ETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYMAFTRKGRPRKG
          ************************************************************

h8e       SKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR
m8e       SKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWAPEPR
          ******************************************************
```

aa-Human vs. Mouse FGF8f

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8f         100.00   98.36
     2: m8f          98.36  100.00
```

```
CLUSTAL O(1.2.1) multiple sequence alignment

h8f       MGSPRSALSCLLLHLLVLCLQAQEGPGRGPALGRELASLFRAGREPQGVSQQVTVQSSPN
m8f       MGSPRSALSCLLLHLLVLCLQAQEGPGGGPALGREPTSLLRAGREPQGVSQQVTVQSSPN
          ************************** ******* :**:********************

h8f       FTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
m8f       FTQHVREQSLVTDQLSRRLIRTYQLYSRTSGKHVQVLANKRINAMAEDGDPFAKLIVETD
          ************************************************************

h8f       TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
m8f       TFGSRVRVRGAETGLYICMNKKGKLIAKSNGKGKDCVFTEIVLENNYTALQNAKYEGWYM
          ************************************************************

h8f       AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
m8f       AFTRKGRPRKGSKTRQHQREVHFMKRLPRGHHTTEQSLRFEFLNYPPFTRSLRGSQRTWA
          ************************************************************

h8f       PEPR
m8f       PEPR
          ****
```

ns-Human vs. Mouse FGF8a

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8a         100.00   94.96
     2: m8a          94.96  100.00

CLUSTAL O(1.2.1) multiple sequence alignment

h8a       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTC
m8a       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTC
          ************************************************* *********

h8a       CAAGCCCAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATC
m8a       CAAGCCCAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATC
          ************************************************************

h8a       CGGACCTACCAACTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
m8a       CGGACCTACCAGCTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
          *********** ************************************************

h8a       CGCATCAACGCCATGGCAGAGGACGGCGACCCCTTCGCAAAGCTCATCGTGGAGACGGAC
m8a       CGCATCAACGCCATGGCAGAAGACGGAGACCCCTTCGCGAAGCTCATTGTGGAGACCGAT
          ******************* ***** *********** ******** ******** **

h8a       ACCTTTGGAAGCAGAGTTCGAGTCCGAGGAGCCGAGACGGGCCTCTACATCTGCATGAAC
m8a       ACTTTTGGAAGCAGAGTCCGAGTTCGCGGCGCAGAGACAGGTCTCTACATCTGCATGAAC
          ** ************** ***** ** ** ** ***** ** *****************

h8a       AAGAAGGGGAAGCTGATCGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTCTTCACGGAG
m8a       AAGAAGGGGAAGCTAATTGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTATTCACAGAG
          ************** ** ********************************* ***** ***

h8a       ATTGTGCTGGAGAACAACTACACAGCGCTGCAGAATGCCAAGTACGAGGGCTGGTACATG
m8a       ATCGTGCTGGAGAACAACTACACGGCGCTGCAGAACGCCAAGTACGAGGGCTGGTACATG
          ** ******************* *********** ************************

h8a       GCCTTCACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGGCAGCACCAGCGTGAG
m8a       GCCTTTACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGCCAGCATCAGCGCGAG
```

```
          ***** ************************************ ***** ***** ***

h8a       GTCCACTTCATGAAGCGGCTGCCCCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTC
m8a       GTGCACTTCATGAAGCGCCTGCCGCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTC
          ** ************** ***** ************************************

h8a       GAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
m8a       GAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
          ************************************************************

h8a       CCCGAGCCCCGATAG
m8a       CCGGAGCCCCGATAG
          ** ************
```

ns-Human vs. Mouse FGF8b

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8b          100.00   95.22
     2: m8b           95.22  100.00

CLUSTAL O(1.2.1) multiple sequence alignment


h8b       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTC
m8b       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTC
          ************************************************** *********

h8b       CAAGCCCAGGTAACTGTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGC
m8b       CAAGCCCAGGTAACTGTTCAGTCCTCACCTAATTTTACACAGCATGTGAGGGAGCAGAGC
          ************************************************************

h8b       CTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAACTCTACAGCCGCACC
m8b       CTGGTGACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAGCTCTACAGCCGCACC
          ******************************************** ***************

h8b       AGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAGGACGGC
m8b       AGCGGGAAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAAGACGGA
          ************************************************** *****

h8b       GACCCCTTCGCAAAGCTCATCGTGGAGACGGACACCTTTGGAAGCAGAGTTCGAGTCCGA
m8b       GACCCCTTCGCGAAGCTCATTGTGGAGACCGATACTTTTGGAAGCAGAGTCCGAGTTCGC
          *********** ******** ******** ** ** ************** ***** **

h8b       GGAGCCGAGACGGGCCTCTACATCTGCATGAACAAGAAGGGGAAGCTGATCGCCAAGAGC
m8b       GGCGCAGAGACAGGTCTCTACATCTGCATGAACAAGAAGGGGAAGCTAATTGCCAAGAGC
          ** ** ***** ** ********************************* ** *********

h8b       AACGGCAAAGGCAAGGACTGCGTCTTCACGGAGATTGTGCTGGAGAACAACTACACAGCG
m8b       AACGGCAAAGGCAAGGACTGCGTATTCACAGAGATCGTGCTGGAGAACAACTACACGGCG
          ********************** ***** ***** ******************** ***

h8b       CTGCAGAATGCCAAGTACGAGGGCTGGTACATGGCCTTCACCCGCAAGGGCCGGCCCCGC
m8b       CTGCAGAACGCCAAGTACGAGGGCTGGTACATGGCCTTTACCCGCAAGGGCCGGCCCCGC
          ******** ***************************** ********************

h8b       AAGGGCTCCAAGACGCGGCAGCACCAGCGTGAGGTCCACTTCATGAAGCGGCTGCCCCGG
m8b       AAGGGCTCCAAGACGCGCCAGCATCAGCGCGAGGTGCACTTCATGAAGCGCCTGCCGCGG
          ***************** ***** ***** ***** ************** ***** ***

h8b       GGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACG
```

```
m8b       GGCCACCACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACG
          ************************************************************

h8b       CGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCCGAGCCCCGATAG
m8b       CGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCCCCGGAGCCCCGATAG
          ********************************** ************
```

ns-Human vs. Mouse FGF8e

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8e         100.00   94.59
     2: m8e          94.59  100.00


CLUSTAL O(1.2.1) multiple sequence alignment

h8e       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTC
m8e       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTC
          ************************************************** *********

h8e       CAAGCCCAGGAAGGCCCGGGCAGGGGCCCTGCGCTGGGCAGGGAGCTCGCTTCCCTGTTC
m8e       CAAGCCCAGGAAGGCCCGGGCGGGGGGCCTGCGCTGGGCAGGGAGCCCACTTCCCTGCTC
          ********************** **** ******************* * ******** **

h8e       CGGGCTGGCCGGGAGCCCCAGGGTGTCTCCCAACAGCATGTGAGGGAGCAGAGCCTGGTG
m8e       CGAGCTGGCCGGGAGCCCCAGGGTGTTTCCCAACAGCATGTGAGGGAGCAGAGCCTGGTG
          ** ************************ ***********************************

h8e       ACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAACTCTACAGCCGCACCAGCGGG
m8e       ACGGATCAGCTCAGCCGCCGCCTCATCCGGACCTACCAGCTCTACAGCCGCACCAGCGGG
          ************************************* **********************

h8e       AAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAGGACGGCGACCCC
m8e       AAGCACGTGCAGGTCCTGGCCAACAAGCGCATCAACGCCATGGCAGAAGACGGAGACCCC
          ********************************************** ***** ******

h8e       TTCGCAAAGCTCATCGTGGAGACGGACACCTTTGGAAGCAGAGTTCGAGTCCGAGGAGCC
m8e       TTCGCGAAGCTCATTGTGGAGACCGATACTTTTGGAAGCAGAGTCCGAGTTCGCGGCGCA
          ***** ******** ******** ** ** ************** ***** ** ** **

h8e       GAGACGGGCCTCTACATCTGCATGAACAAGAAGGGGAAGCTGATCGCCAAGAGCAACGGC
m8e       GAGACAGGTCTCTACATCTGCATGAACAAGAAGGGGAAGCTAATTGCCAAGAGCAACGGC
          ***** ** ******************************** ** ***************

h8e       AAAGGCAAGGACTGCGTCTTCACGGAGATTGTGCTGGAGAACAACTACACAGCGCTGCAG
m8e       AAAGGCAAGGACTGCGTATTCACAGAGATCGTGCTGGAGAACAACTACACGGCGCTGCAG
          ***************** ***** ***** ******************** *********

h8e       AATGCCAAGTACGAGGGCTGGTACATGGCCTTCACCCGCAAGGGCCGGCCCCGCAAGGGC
m8e       AACGCCAAGTACGAGGGCTGGTACATGGCCTTTACCCGCAAGGGCCGGCCCCGCAAGGGC
          ** ***************************** ****************************

h8e       TCCAAGACGCGGCAGCACCAGCGTGAGGTCCACTTCATGAAGCGGCTGCCCCGGGGCCAC
m8e       TCCAAGACGCGCCAGCATCAGCGCGAGGTGCACTTCATGAAGCGCCTGCCGCGGGGCCAC
          *********** ***** ***** ***** ************** ***** *********

h8e       CACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGC
m8e       CACACCACCGAGCAGAGCCTGCGCTTCGAGTTCCTCAACTACCCGCCCTTCACGCGCAGC
          ************************************************************

h8e       CTGCGCGGCAGCCAGAGGACTTGGGCCCCCGAGCCCCGATAG
```

```
m8e       CTGCGCGGCAGCCAGAGGACTTGGGCCCCGGAGCCCCGATAG
          **************************** ************
```

ns-Human vs. Mouse FGF8f

```
Percent Identity  Matrix - created by Clustal2.1
     1: h8f         100.00   94.83
     2: m8f          94.83  100.00

CLUSTAL O(1.2.1) multiple sequence alignment

h8f       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTCCTCTGCCTC
m8f       ATGGGCAGCCCCCGCTCCGCGCTGAGCTGCCTGCTGTTGCACTTGCTGGTTCTCTGCCTC
          ************************************************** *********

h8f       CAAGCCCAGGAAGGCCCGGGCAGGGGCCCTGCGCTGGGCAGGGAGCTCGCTTCCCTGTTC
m8f       CAAGCCCAGGAAGGCCCGGGCGGGGGGCCTGCGCTGGGCAGGGAGCCCACTTCCCTGCTC
          ********************* **** ****************** * ******** **

h8f       CGGGCTGGCCGGGAGCCCCAGGGTGTCTCCCAACAGGTAACTGTTCAGTCCTCACCTAAT
m8f       CGAGCTGGCCGGGAGCCCCAGGGTGTTTCCCAACAGGTAACTGTTCAGTCCTCACCTAAT
          ** ********************** **********************************

h8f       TTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATC
m8f       TTTACACAGCATGTGAGGGAGCAGAGCCTGGTGACGGATCAGCTCAGCCGCCGCCTCATC
          ************************************************************

h8f       CGGACCTACCAACTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
m8f       CGGACCTACCAGCTCTACAGCCGCACCAGCGGGAAGCACGTGCAGGTCCTGGCCAACAAG
          *********** ************************************************

h8f       CGCATCAACGCCATGGCAGAGGACGGCGACCCCTTCGCAAAGCTCATCGTGGAGACGGAC
m8f       CGCATCAACGCCATGGCAGAAGACGGAGACCCCTTCGCGAAGCTCATTGTGGAGACCGAT
          ******************** ***** *********** ******** ******** **

h8f       ACCTTTGGAAGCAGAGTTCGAGTCCGAGGAGCCGAGACGGGCCTCTACATCTGCATGAAC
m8f       ACTTTTGGAAGCAGAGTCCGAGTTCGCGGCGCAGAGACAGGTCTCTACATCTGCATGAAC
          ** ************** ***** ** ** ** ***** ** ******************

h8f       AAGAAGGGGAAGCTGATCGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTCTTCACGGAG
m8f       AAGAAGGGGAAGCTAATTGCCAAGAGCAACGGCAAAGGCAAGGACTGCGTATTCACAGAG
          ************** ** ********************************* ***** ***

h8f       ATTGTGCTGGAGAACAACTACACAGCGCTGCAGAATGCCAAGTACGAGGGCTGGTACATG
m8f       ATCGTGCTGGAGAACAACTACACGGCGCTGCAGAACGCCAAGTACGAGGGCTGGTACATG
          ** ******************** *********** ************************

h8f       GCCTTCACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGGCAGCACCAGCGTGAG
m8f       GCCTTTACCCGCAAGGGCCGGCCCCGCAAGGGCTCCAAGACGCGCCAGCATCAGCGCGAG
          ***** ************************************** ***** ***** ***

h8f       GTCCACTTCATGAAGCGGCTGCCCCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTC
m8f       GTGCACTTCATGAAGCGCCTGCCGCGGGGCCACCACACCACCGAGCAGAGCCTGCGCTTC
          ** ************** ***** ************************************

h8f       GAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
m8f       GAGTTCCTCAACTACCCGCCCTTCACGCGCAGCCTGCGCGGCAGCCAGAGGACTTGGGCC
          ************************************************************

h8f       CCCGAGCCCCGATAG
m8f       CCGGAGCCCCGATAG
```

References cited herein

Bartelt, A., Bruns, O. T., Reimer, R., Hohenberg, H., Ittrich, H., Peldschus, K., . . . Heeren, J. (2011). Brown adipose tissue activity controls triglyceride clearance. *Nat Med, 17*(2), 200-205. doi: 10.1038/nm.2297

Bartelt, A., & Heeren, J. (2014). Adipose tissue browning and metabolic health. *Nat Rev Endocrinol, 10*(1), 24-36. doi: 10.1038/nrendo.2013.204

Cinti, S. (2006). The role of brown adipose tissue in human obesity. *Nutr Metab Cardiovasc Dis, 16*(8), 569-574. doi: 10.1016/j.numecd.2006.07.009

Gunawardana, S. C. (2014). Benefits of healthy adipose tissue in the treatment of diabetes. *World J Diabetes, 5*(4), 420-430. doi: 10.4239/wjd.v5.i4.420

Gunawardana, S. C., & Piston, D. W. (2012). Reversal of type 1 diabetes in mice by brown adipose tissue transplant. *Diabetes, 61*(3), 674-682. doi: 10.2337/db11-0510

Himms-Hagen, J. (1979). Obesity may be due to a malfunctioning of brown fat. *Can Med Assoc J, 121*(10), 1361-1364.

Klingenspor, M., Fromme, T. (2012). Brown adipose tissue. In M. E. Symonds (Ed.), *Adipose Tissue Biology* (Vol. 414). Heidelberg: Springer.

Lidell, M. E., Betz, M. J., & Enerback, S. (2014). Brown adipose tissue and its therapeutic potential. *J Intern Med, 276*(4), 364-377. doi: 10.1111/joim.12255

Roman, S., Agil, A., Peran, M., Alvaro-Galue, E., Ruiz-Ojeda, F. J., Fernandez-Vazquez, G., & Marchal, J. A. (2014). Brown adipose tissue and novel therapeutic approaches to treat metabolic disorders. *Transl Res*. doi: 10.1016/j.trsl.2014.11.002

Rothwell, N. J., & Stock, M. J. (1979). A role for brown adipose tissue in diet-induced thermogenesis. *Nature, 281*(5726), 31-35.

Saito, M., Okamatsu-Ogura, Y., Matsushita, M., Watanabe, K., Yoneshiro, T., Nio-Kobayashi, J., . . . Tsujisaki, M. (2009). High incidence of metabolically active brown adipose tissue in healthy adult humans: effects of cold exposure and adiposity. *Diabetes, 58*(7), 1526-1531. doi: 10.2337/db09-0530

van Marken Lichtenbelt, W. D., Vanhommerig, J. W., Smulders, N. M., Drossaerts, J. M., Kemerink, G. J., Bouvy, N. D., . . . Teule, G. J. (2009). Cold-activated brown adipose tissue in healthy men. *N Engl J Med, 360*(15), 1500-1508. doi: 10.1056/NEJMoa0808718

Virtanen, K. A., Lidell, M. E., Orava, J., Heglind, M., Westergren, R., Niemi, T., . . . Nuutila, P. (2009). Functional brown adipose tissue in healthy adults. *N Engl J Med, 360*(15), 1518-1525. doi: 10.1056/NEJMoa0808949

List of references

Armani, A., Cinti, F., Marzolla, V., Morgan, J., Cranston, G. A., Antelmi, A., . . . Caprio, M. (2014). Mineralocorticoid receptor antagonism induces browning of white adipose tissue through impairment of autophagy and prevents adipocyte dysfunction in high-fat-diet-fed mice. FASEB J, 28(8), 3745-3757. doi: 10.1096/fj.13-245415

Bartelt, A., Bruns, O. T., Reimer, R., Hohenberg, H., Ittrich, H., Peldschus, K., . . . Heeren, J. (2011). Brown adipose tissue activity controls triglyceride clearance. Nat Med, 17(2), 200-205. doi: 10.1038/nm.2297

Bi, P., Shan, T., Liu, W., Yue, F., Yang, X., Liang, X. R., . . . Kuang, S. (2014). Inhibition of Notch signaling promotes browning of white adipose tissue and ameliorates obesity. Nat Med, 20(8), 911-918. doi: 10.1038/nm.3615

Bolze, F., Rink, N., Brumm, H., Kuhn, R., Mocek, S., Schwarz, A. E., . . . Klingenspor, M. (2013). Characterization of the melanocortin-4-receptor nonsense mutation W16X in vitro and in vivo. Pharmacogenomics J, 13(1), 80-93. doi: 10.1038/tpj.2011.43

Bordicchia, M., Liu, D., Amri, E. Z., Ailhaud, G., Dessi-Fulgheri, P., Zhang, C., . . . Collins, S. (2012). Cardiac natriuretic peptides act via p38 MAPK to induce the brown fat thermogenic program in mouse and human adipocytes. J Clin Invest, 122(3), 1022-1036. doi: 10.1172/JCI59701

Caverzasio, J., & Thouverey, C. (2011). Activation of FGF receptors is a new mechanism by which strontium ranelate induces osteoblastic cell growth. Cell Physiol Biochem, 27(3-4), 243-250. doi: 10.1159/000327950

Chaffee, R. R., Allen, J. R., Cassuto, Y., & Smith, R. E. (1964). Biochemistry of Brown Fat and Liver of Cold-Acclimated Hamsters. Am J Physiol, 207, 1211-1214.

Cypess, A. M., Lehman, S., Williams, G., Tal, I., Rodman, D., Goldfine, A. B., . . . Kahn, C. R. (2009). Identification and importance of brown adipose tissue in adult humans. N Engl J Med, 360(15), 1509-1517. doi: 10.1056/NEJMoa0810780

Fisher, F. M., Kleiner, S., Douris, N., Fox, E. C., Mepani, R. J., Verdeguer, F., . . . Spiegelman, B. M. (2012). FGF21 regulates PGC-1alpha and browning of white

adipose tissues in adaptive thermogenesis. Genes Dev, 26(3), 271-281. doi: 10.1101/gad.177857.111

Goetz, R., Ohnishi, M., Kir, S., Kurosu, H., Wang, L., Pastor, J., . . . Mohammadi, M. (2012). Conversion of a paracrine fibroblast growth factor into an endocrine fibroblast growth factor. J Biol Chem, 287(34), 29134-29146. doi: 10.1074/jbc.M112.342980

Gong, D. W., Bi, S., Weintraub, B. D., & Reitman, M. (1998). Rat mitochondrial glycerol-3-phosphate dehydrogenase gene: multiple promoters, high levels in brown adipose tissue, and tissue-specific regulation by thyroid hormone. DNA Cell Biol, 17(3), 301-309.

Guerra, C., Koza, R. A., Yamashita, H., Walsh, K., & Kozak, L. P. (1998). Emergence of brown adipocytes in white fat in mice is under genetic control. Effects on body weight and adiposity. J Clin Invest, 102(2), 412-420. doi: 10.1172/JCI3155

Hanssen (2014) Diabetologia doi 10.1007/ss0125-014-3465-8

Hoffmann, C., Zimmermann, A., Hinney, A., Volckmar, A. L., Jarrett, H. W., Fromme, T., & Klingenspor, M. (2013). A novel SP1/SP3 dependent intronic enhancer governing transcription of the UCP3 gene in brown adipocytes. PLoS One, 8(12), e83426. doi: 10.1371/journal.pone.0083426

Itoh, N., & Ornitz, D. M. (2008). Functional evolutionary history of the mouse Fgf gene family. Dev Dyn, 237(1), 18-27. doi: 10.1002/dvdy.21388

Klein, J., Westphal, S., Kraus, D., Meier, B., Perwitz, N., Ott, V., . . . Klein, H. H. (2004). Metformin inhibits leptin secretion via a mitogen-activated protein kinase signalling pathway in brown adipocytes. J Endocrinol, 183(2), 299-307. doi: 10.1677/joe.1.05646

Klingenspor, M., Fromme, T. (2012). Brown adipose tissue. In M. E. Symonds (Ed.), Adipose Tissue Biology (Vol. 414). Heidelberg: Springer.

Kuchler, S., Perwitz, N., Schick, R. R., Klein, J., & Westphal, S. (2010). Arginine-vasopressin directly promotes a thermogenic and pro-inflammatory adipokine expression profile in brown adipocytes. Regul Pept, 164(2-3), 126-132. doi: 10.1016/j.regpep.2010.05.016

Li, Y., Bolze, F., Fromme, T., & Klingenspor, M. (2014). Intrinsic differences in BRITE adipogenesis of primary adipocytes from two different mouse strains. Biochim Biophys Acta, 1841(9), 1345-1352. doi: 10.1016/j.bbalip.2014.06.003

Li, Y., Fromme, T., Schweizer, S., Schottl, T., & Klingenspor, M. (2014). Taking control over intracellular fatty acid levels is essential for the analysis of thermogenic

function in cultured primary brown and brite/beige adipocytes. EMBO Rep. doi: 10.15252/embr.201438775

Meyer, C. W., Willershauser, M., Jastroch, M., Rourke, B. C., Fromme, T., Oelkrug, R., . . . Klingenspor, M. (2010). Adaptive thermogenesis and thermal conductance in wild-type and UCP1-KO mice. Am J Physiol Regul Integr Comp Physiol, 299(5), R1396-1406. doi: 10.1152/ajpregu.00021.2009

Murakami, M., Elfenbein, A., & Simons, M. (2008). Non-canonical fibroblast growth factor signalling in angiogenesis. Cardiovasc Res, 78(2), 223-231. doi: 10.1093/cvr/cvm086

Nau, K., Fromme, T., Meyer, C. W., von Praun, C., Heldmaier, G., & Klingenspor, M. (2008). Brown adipose tissue specific lack of uncoupling protein 3 is associated with impaired cold tolerance and reduced transcript levels of metabolic genes. J Comp Physiol B, 178(3), 269-277. doi: 10.1007/s00360-007-0219-7

Pfeifer, A., & Hoffmann, L. S. (2014). Brown, Beige, and White: The New Color Code of Fat and Its Pharmacological Implications. Annu Rev Pharmacol Toxicol. doi: 10.1146/annurev-pharmtox-010814-124346

Ray, J., Baird, A., & Gage, F. H. (1997). A 10-amino acid sequence of fibroblast growth factor 2 is sufficient for its mitogenic activity on neural progenitor cells. Proc Natl Acad Sci U S A, 94(13), 7047-7052.

Seyfarth, K., Poschmann, G., Rozman, J., Fromme, T., Rink, N., Hofmann, A., Wurst, W., Stühler, K., Klingenspor, M. (2014). The development of diet-induced obesity and associated metabolic impairments in Dj-1 deficient mice. Journal of Nutritional Biochemistry, in press.

Suh, J. M., Jonker, J. W., Ahmadian, M., Goetz, R., Lackey, D., Osborn, O., . . . Evans, R. M. (2014). Endocrinization of FGF1 produces a neomorphic and potent insulin sensitizer. Nature. doi: 10.1038/nature13540

Sunmonu, N. A., Li, K., & Li, J. Y. (2011). Numerous isoforms of Fgf8 reflect its multiple roles in the developing brain. J Cell Physiol, 226(7), 1722-1726. doi: 10.1002/jcp.22587

Tseng, Y. H., Kokkotou, E., Schulz, T. J., Huang, T. L., Winnay, J. N., Taniguchi, C. M., . . . Kahn, C. R. (2008). New role of bone morphogenetic protein 7 in brown adipogenesis and energy expenditure. Nature, 454(7207), 1000-1004. doi: 10.1038/nature07221

van Marken Lichtenbelt, W. D., Vanhommerig, J. W., Smulders, N. M., Drossaerts, J. M., Kemerink, G. J., Bouvy, N. D., . . . Teule, G. J. (2009). Cold-activated brown adipose tissue in healthy men. N Engl J Med, 360(15), 1500-1508. doi: 10.1056/NEJMoa0808718

Virtanen, K. A., Lidell, M. E., Orava, J., Heglind, M., Westergren, R., Niemi, T., . . . Nuutila, P. (2009). Functional brown adipose tissue in healthy adults. N Engl J Med, 360(15), 1518-1525. doi: 10.1056/NEJMoa0808949

Wagner, I. V., Perwitz, N., Drenckhan, M., Lehnert, H., & Klein, J. (2011). Cannabinoid type 1 receptor mediates depot-specific effects on differentiation, inflammation and oxidative metabolism in inguinal and epididymal white adipocytes. Nutr Diabetes, 1, e16. doi: 10.1038/nutd.2011.12

Westphal, S., Perwitz, N., Iwen, K. A., Kraus, D., Schick, R., Fasshauer, M., & Klein, J. (2008). Expression of ATRAP in adipocytes and negative regulation by beta-adrenergic stimulation of JAK/STAT. Horm Metab Res, 40(3), 165-171. doi: 10.1055/s-2007-1022547

Wree, A., Mayer, A., Westphal, S., Beilfuss, A., Canbay, A., Schick, R. R., . . . Vaupel, P. (2012). Adipokine expression in brown and white adipocytes in response to hypoxia. J Endocrinol Invest, 35(5), 522-527. doi: 10.3275/7964

Young, P., Arch, J. R., & Ashwell, M. (1984). Brown adipose tissue in the parametrial fat pad of the mouse. FEBS Lett, 167(1), 10-14.

All references cited herein are fully incorporated by reference. Having now fully described the invention, it will be understood by a person skilled in the art that the invention may be practiced within a wide and equivalent range of conditions, parameters and the like, without affecting the spirit or scope of the invention or any embodiment thereof.

CLAIMS

1. A polypeptide for use in treating a disease or disorder of energy homeostasis, wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome, and
wherein the polypeptide is selected from the group consisting of:
   (a) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule having the nucleic acid sequence as depicted in SEQ ID NO: 2 or the nucleic acid sequence comprising nucleic acid residues 67-648 in SEQ ID NO: 2;
   (b) a polypeptide comprising an amino acid sequence as depicted in SEQ ID NO: 16 or an amino acid sequence comprising amino acids 23-215 in SEQ ID NO: 16;
   (c) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule hybridizing under stringent conditions to the complementary strand of a nucleic acid molecule as defined in (a) and encoding a functional polypeptide, wherein the function comprises the ability to induce differentiation or conversion of white adipocytes and/or preadipocytes to brown adipocytes;
   (d) a polypeptide having at least 40% identity to the polypeptide of any one of (a) to (c), whereby said polypeptide is functional, wherein the function comprises the ability to induce differentiation or conversion of white visceral adipocytes and/or preadipocytes to brown adipocytes; and
   (e) a polypeptide comprising an amino acid sequence encoded by a nucleic acid molecule being degenerate as a result of the genetic code to the nucleotide sequence of a nucleic acid molecule as defined in (a), and (c).

2. A polynucleotide for use in treating a disease or disorder of energy

homeostasis,

wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome, and

wherein the polynucleotide encodes the polypeptide of claim 1.

3. A pharmaceutical composition comprising the polypeptide of claim 1 and/or the polynucleotide of claim 2 for use in treating a disease or disorder of energy homeostasis, further comprising a pharmaceutically acceptable carrier and/or diluent, wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome.

4. A method of treating a disease or disorder of energy homeostasis by administering an effective dose of the polypeptide of claim 1, the polynucleotide of claim 2, or the pharmaceutical composition of claim 3, to a subject in need of such treatment, wherein said disease or disorder is obesity, dyslipidemia, diabetes, insulin resistance, hyperglycemia or metabolic syndrome.

5. The polypeptide for use according to claim 1, the polynucleotide for use according to claim 2, the pharmaceutical composition for use according to claim 3, or the method of claim 4, wherein said polypeptide binds to an FGF receptor.

6. The polypeptide for use according to claim 5, the polynucleotide for use according to claim 5, the pharmaceutical composition for use according to claim 5, or the method of claim 5, wherein said FGF receptor is at least one FGF receptor selected from the group consisting of FGF receptor 4, FGF receptor 1, FGF receptor 2 and FGF receptor 3.

7. The polypeptide for use according to any one of claims 1, 5 or 6, the polynucleotide for use according to any one of claims 2, 5 or 6, the pharmaceutical composition for use according to any one of claims 3, 5 or 6, or the method of any one of claims 4, 5 or 6, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally.

8. The polypeptide for use according to claim 7, the polynucleotide for use according to claim 7, the pharmaceutical composition for use according to claim 7, or the method of claim 7, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administrated locally into the visceral adipose tissue.

9. The polypeptide for use according to any one of claims 1 and 5 to 8, the polynucleotide for use according to any one of claims 2 and 5 to 8, the pharmaceutical composition for use according to any one of claims 3 and 5 to 8, or the method of any one of claims 4 to 8, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administered into the visceral adipose tissue.

10. The polypeptide for use according to any one of claims 1 and 5-9, the polynucleotide for use according to any one of claims 2 and 5 to 9, the pharmaceutical composition for use according to any one of claims 3 and 5 to 9, or the method of any one of claims 4-9, wherein said polypeptide, polynucleotide or pharmaceutical composition is in the form of an erodible implant, an implantable drug release device, a gel for injection or a solution for injection.

11. The polypeptide for use according to any one of claims 1 and 5-10, the polynucleotide for use according to any one of claims 2 and 5-10, the pharmaceutical composition for use according to any one of claims 3 and 5-10, or the method of any one of claims 4-10, wherein said polypeptide, polynucleotide or pharmaceutical composition is to be administered via a minipump.

12. The polypeptide for use according to any one of claims 1 and 5 to 11, the polynucleotide for use according to any one of claims 2 and 5 to 11, the pharmaceutical composition for use according to any one of claims 3 and 5 to 11, or the method of any one of claims 4 to 11, wherein said obesity is central obesity.

13. The polypeptide for use according to any one of claims 1 and 5 to 12, the polynucleotide for use according to any one of claims 2 and 5 to 12, the pharmaceutical composition for use according to any one of claims 3 and 5 to 12, or the method of any one of claims 4 and 5 to 12, wherein said polypeptide, polynucleotide or pharmaceutical composition is co-administered with at least one other active agent.

14. The polypeptide for use according to claim 13, the polynucleotide for use according to claim 13, the pharmaceutical composition for use according to claim 13, or the method of claim 13, wherein said other active agent is at least one active agent selected from the group consisting of beta-adrenergic agonists (e.g. noradrenalin, isoproterenol, BRL 35135, ICI D7114, CGP-12177A, CL 316243), indirect sympathomimetics (e.g. ephedrine, methylphenidate), atrial natriuretic peptide (e.g. ANP, BNP) and ANP/BNP receptor agonists (e.g. AP-811).

Figure 1.

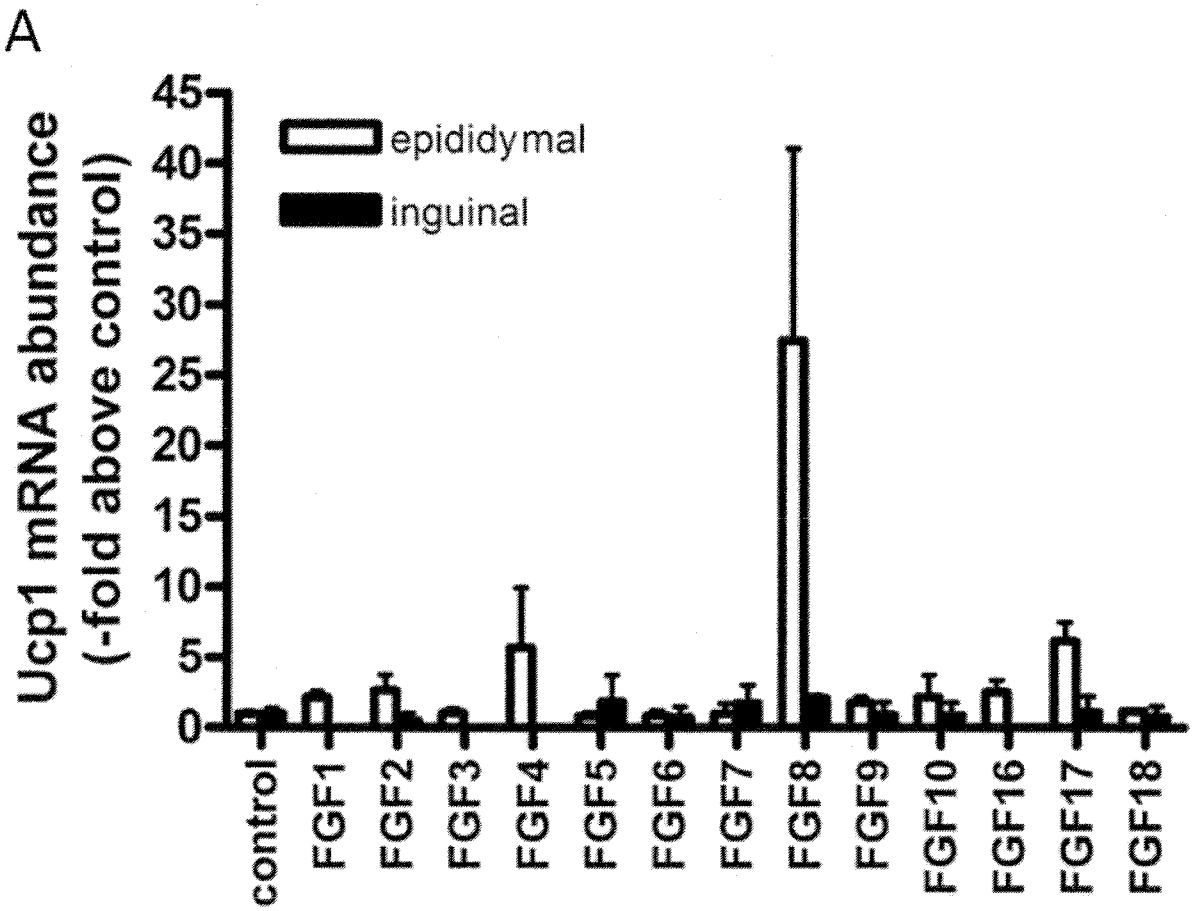
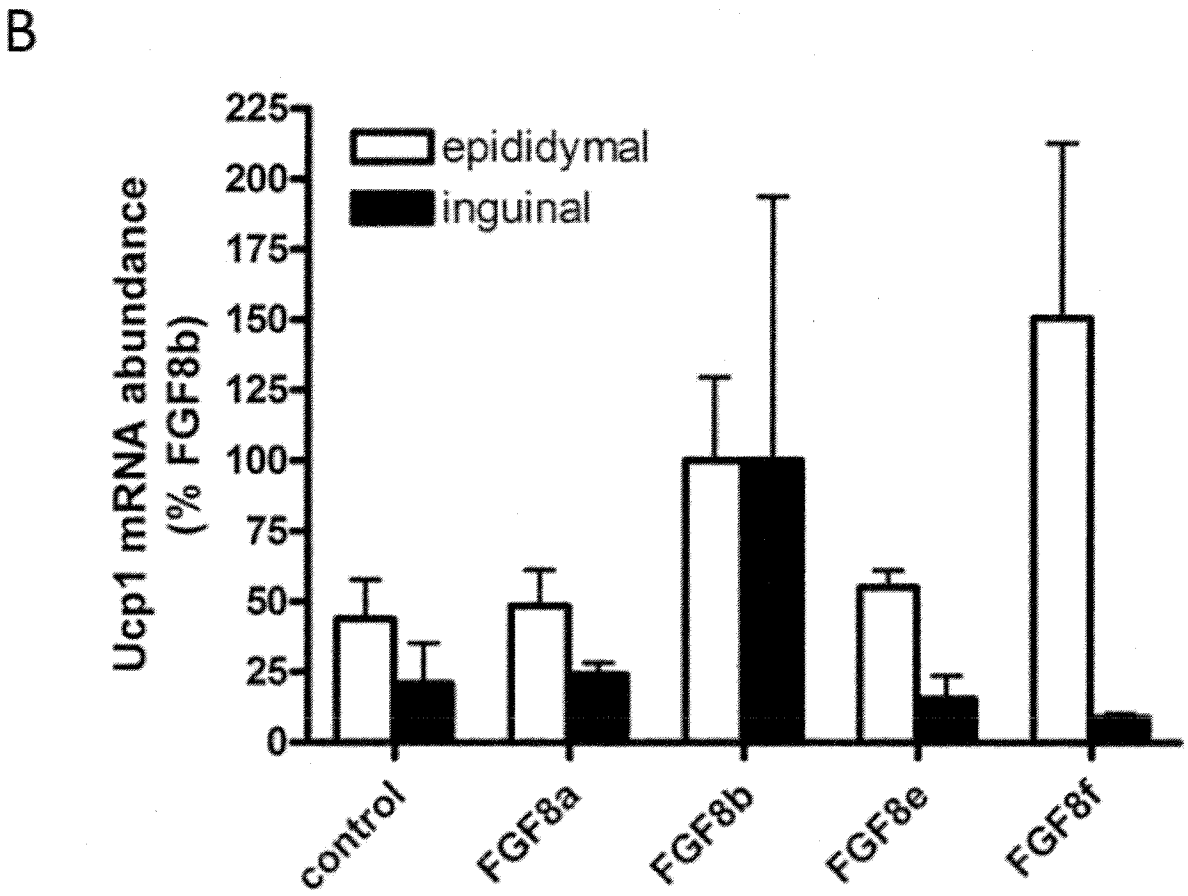

A
Ucp1 mRNA abundance
(-fold above control)
45
40
35
30
25
20
15
10
5
0
epididymal
inguinal
control
FGF1
FGF2
FGF3
FGF4
FGF5
FGF6
FGF7
FGF8
FGF9
FGF10
FGF16
FGF17
FGF18

Figure 1 (cont.).



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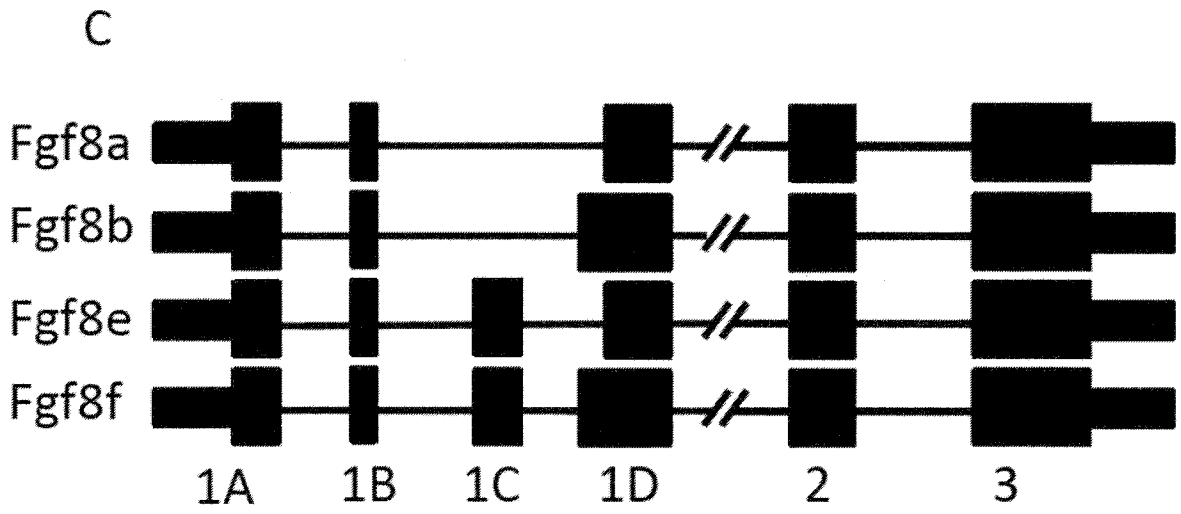


Figure 2.

A

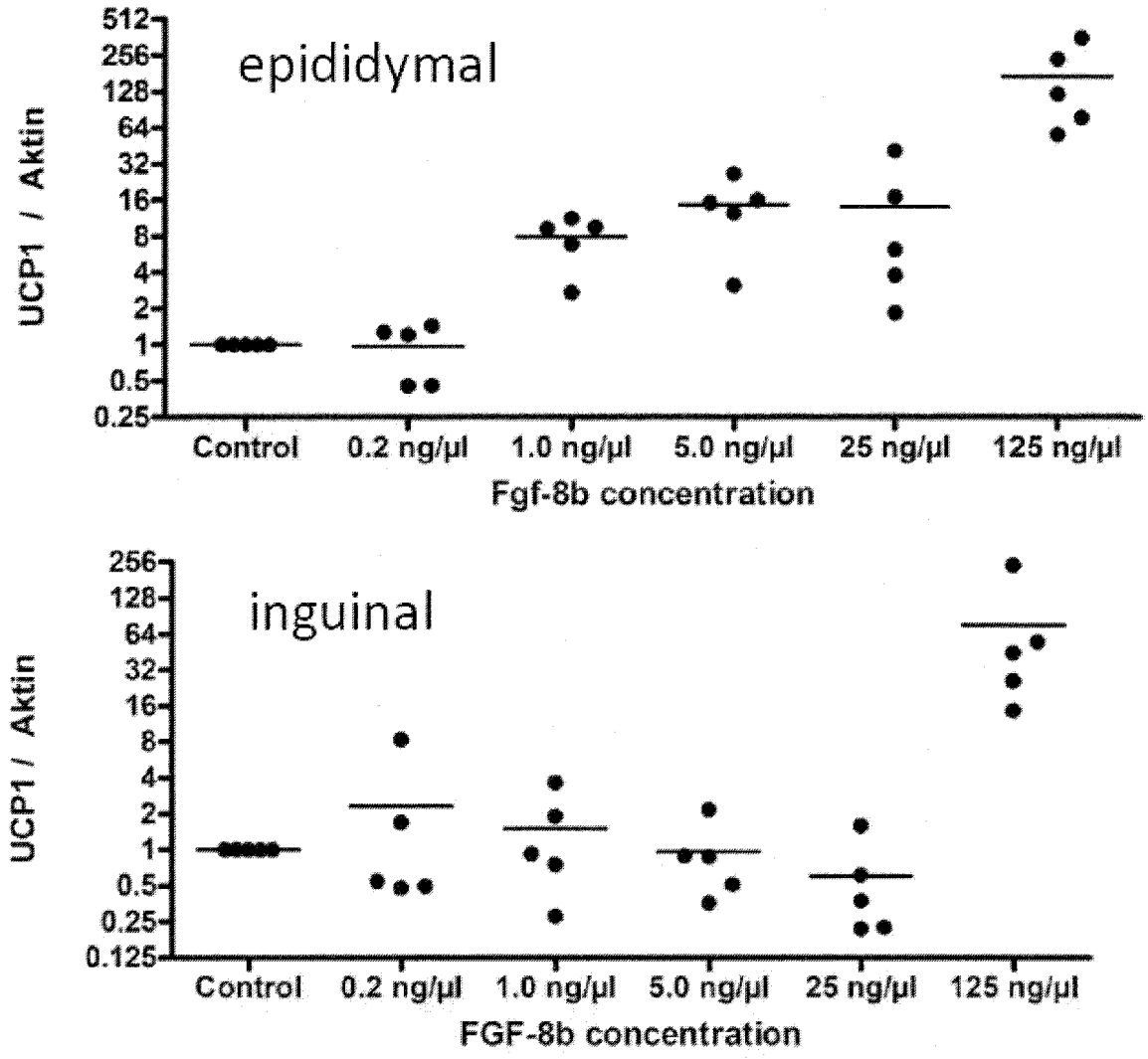

epididymal
UCP1 / Aktin
512
256
128
64
32
16
8
4
2
1
0.5
0.25
Control
0.2 ng/µl
1.0 ng/µl
5.0 ng/µl
25 ng/µl
125 ng/µl
Fgf-8b concentration
inguinal
UCP1 / Aktin
256
128
64
32
16
8
4
2
1
0.5
0.25
0.125
Control
0.2 ng/µl
1.0 ng/µl
5.0 ng/µl
25 ng/µl
125 ng/µl
FGF-8b concentration

Figure 2 (cont.).

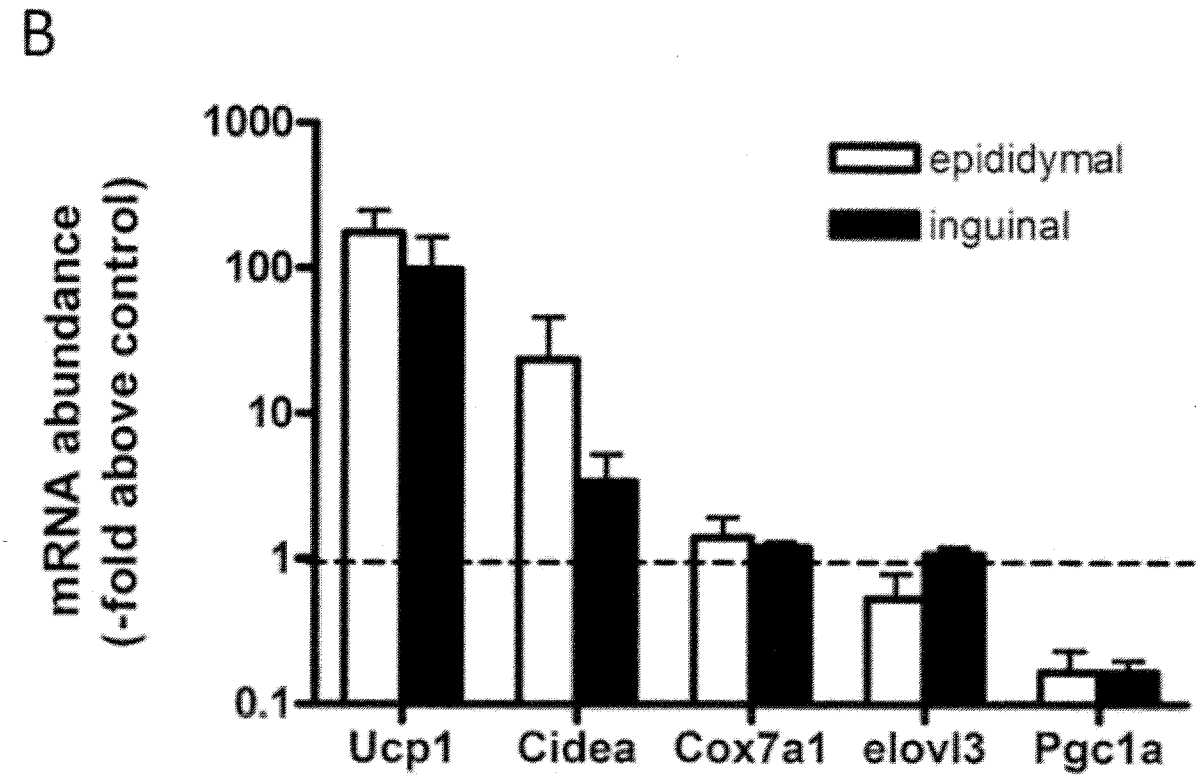


Figure 3.

A

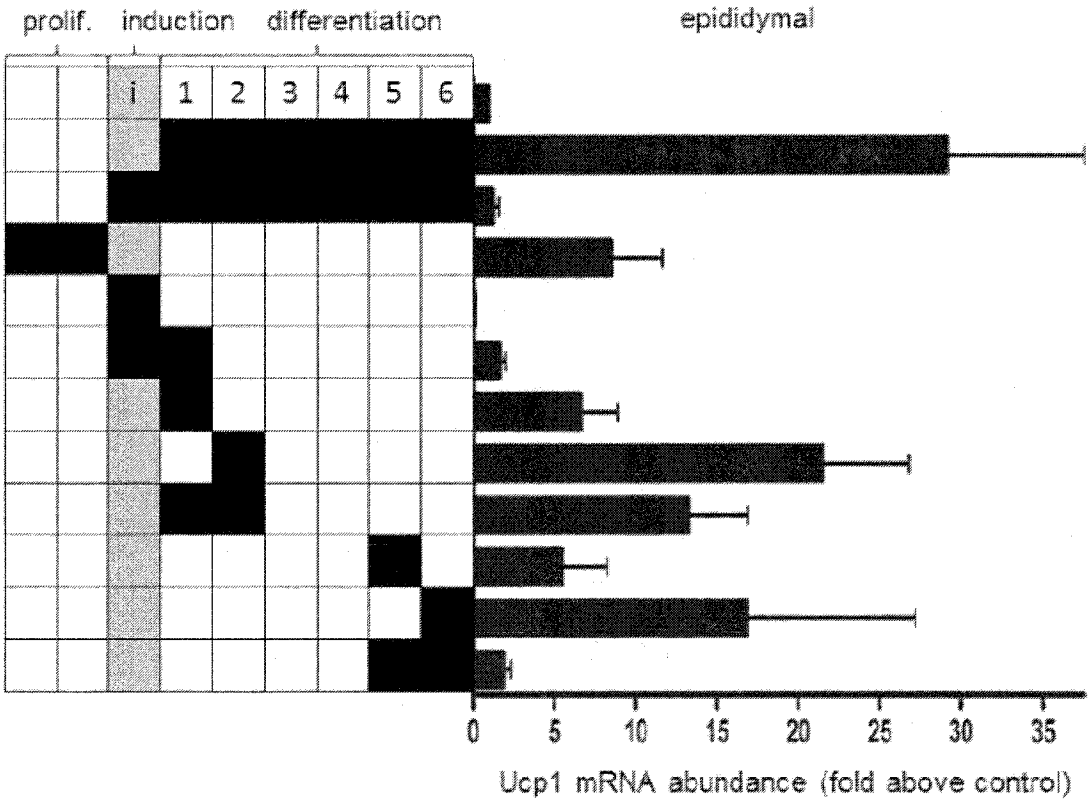
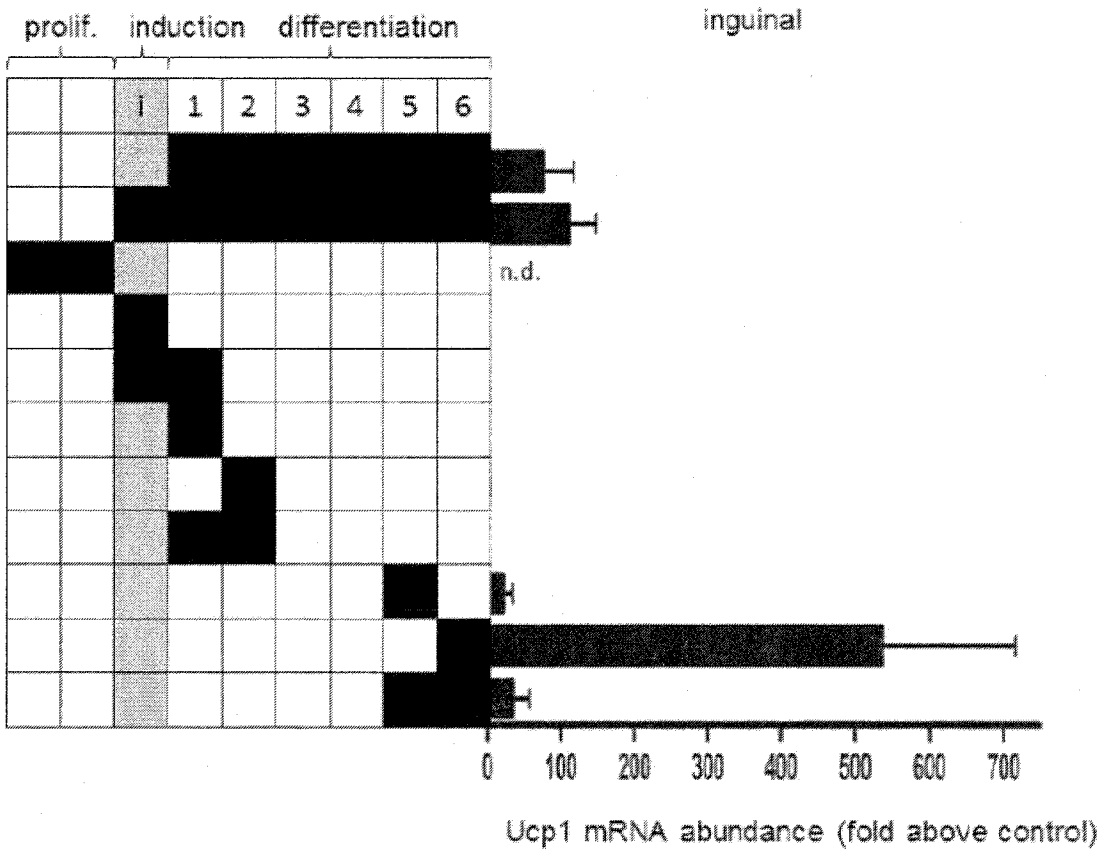

prolif.
induction
differentiation
epididymal
i
1
2
3
4
5
6
0
5
10
15
20
25
30
35
Ucp1 mRNA abundance (fold above control)

Figure 3 (cont.).

B



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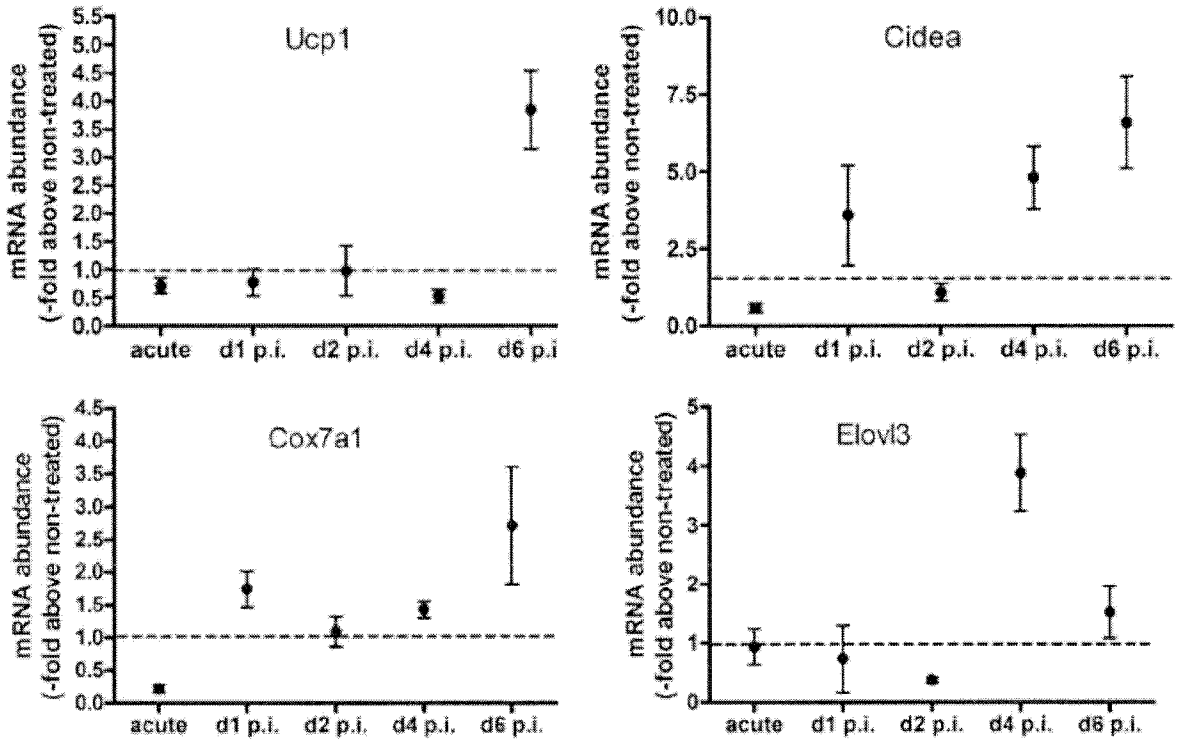
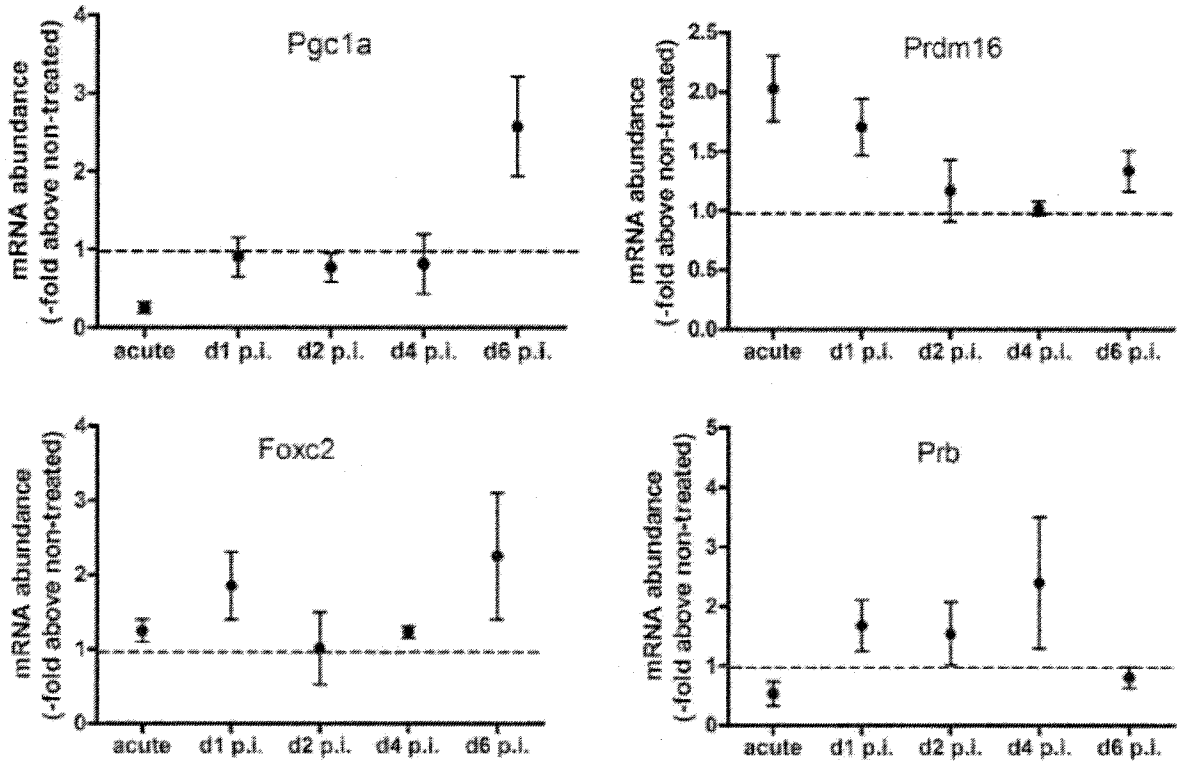

Ucp1
Cidea
Cox7a1
Elovl3
mRNA abundance
(-fold above non-treated)
acute
d1 p.i.
d2 p.i.
d4 p.i.
d6 p.i

Figure 4 (cont.).



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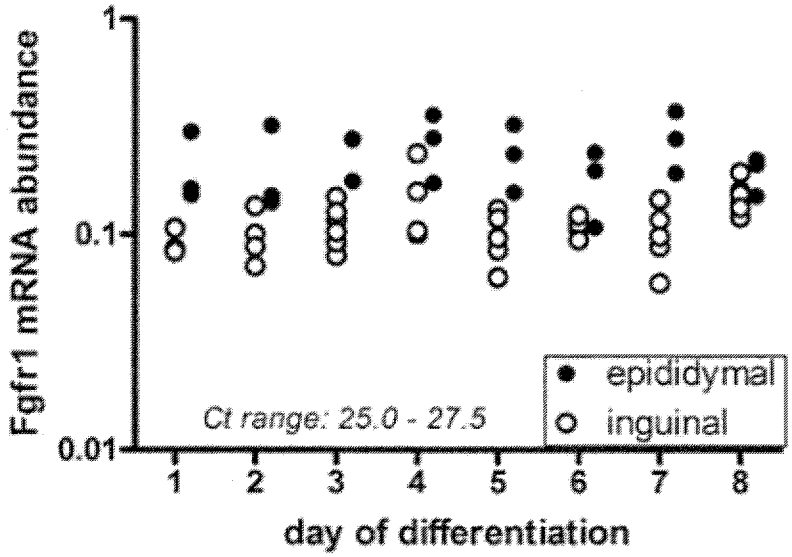

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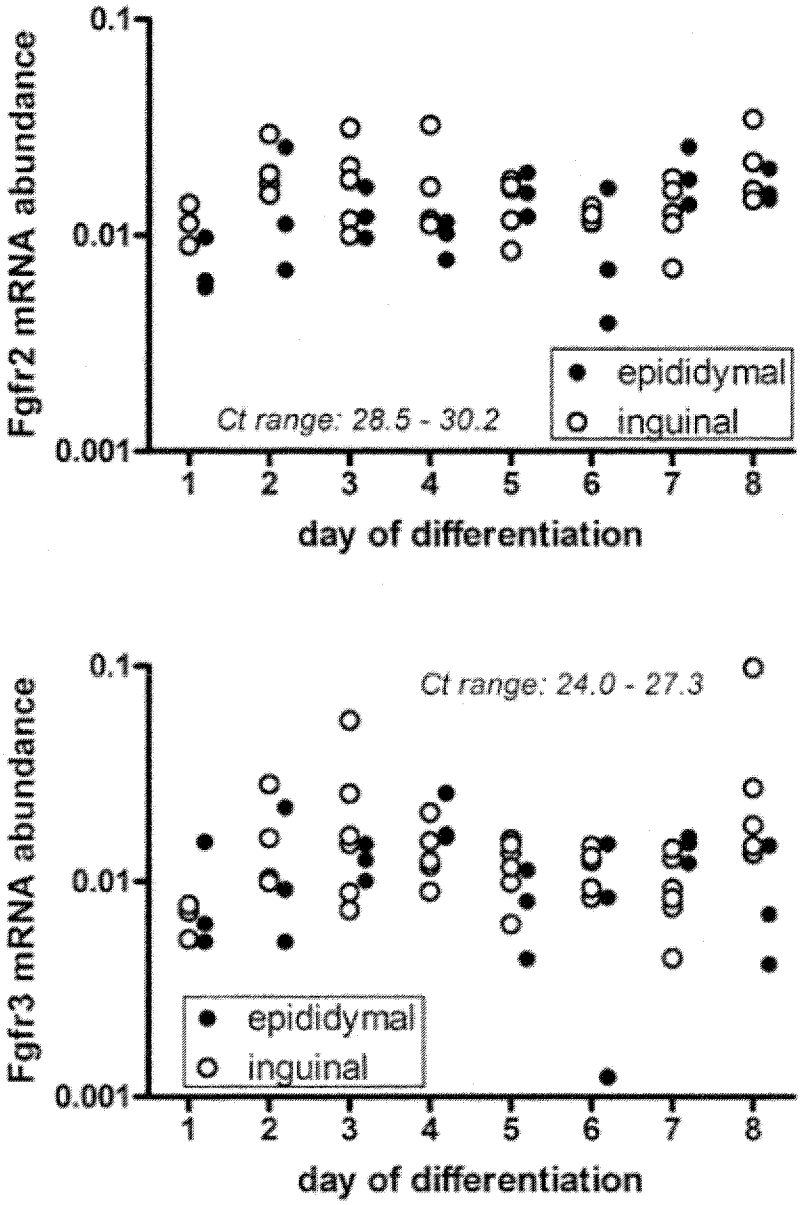

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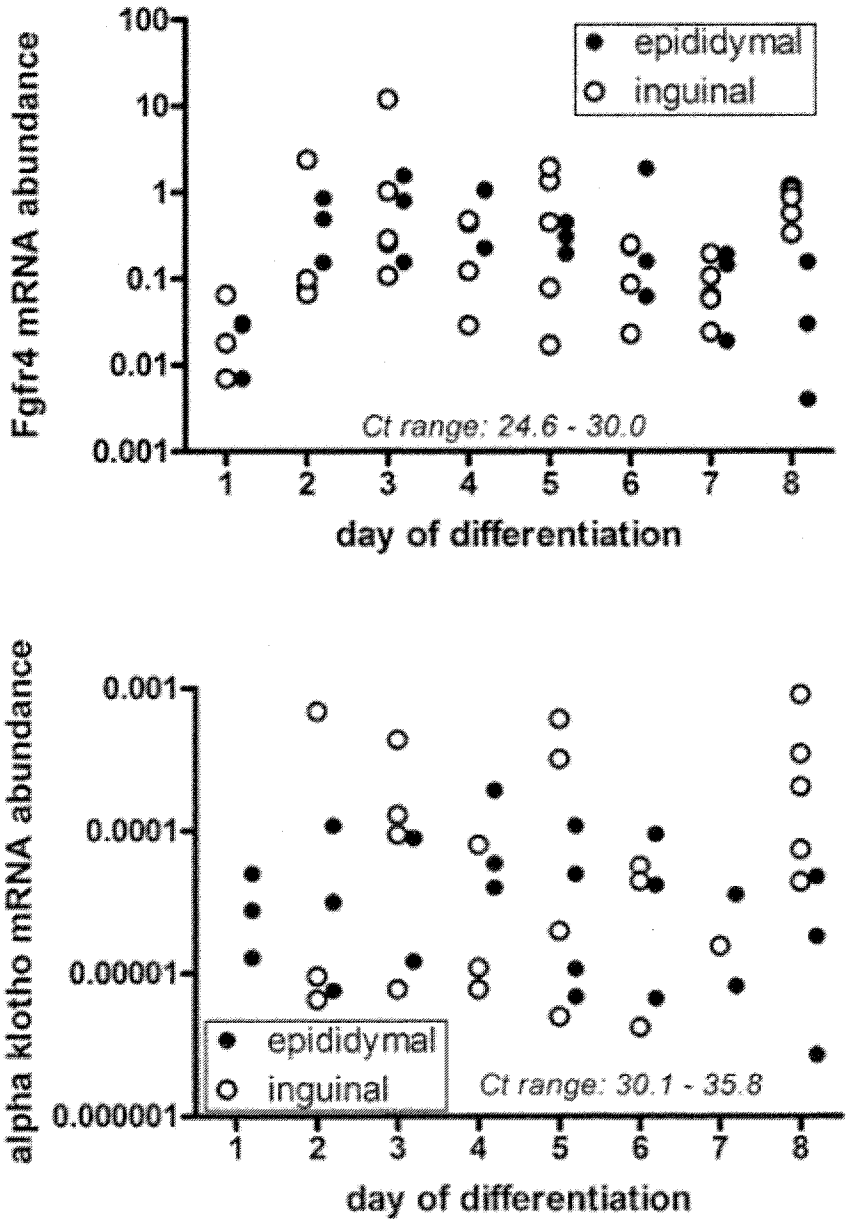

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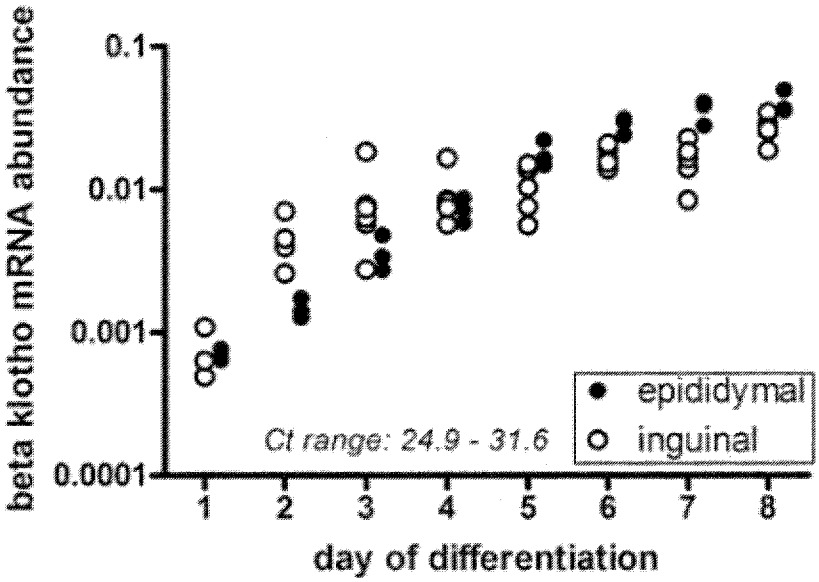

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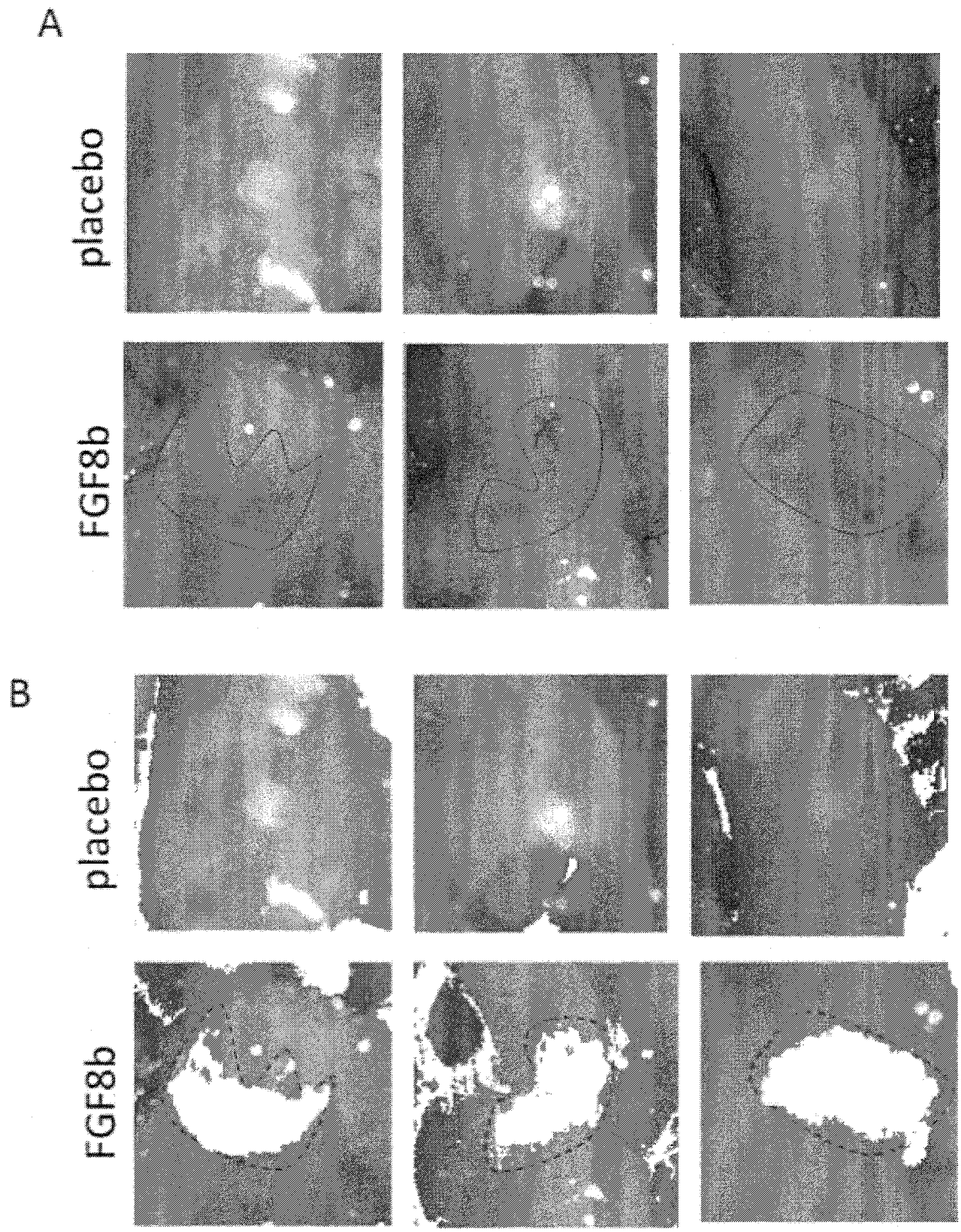


Figure 6 (cont.).

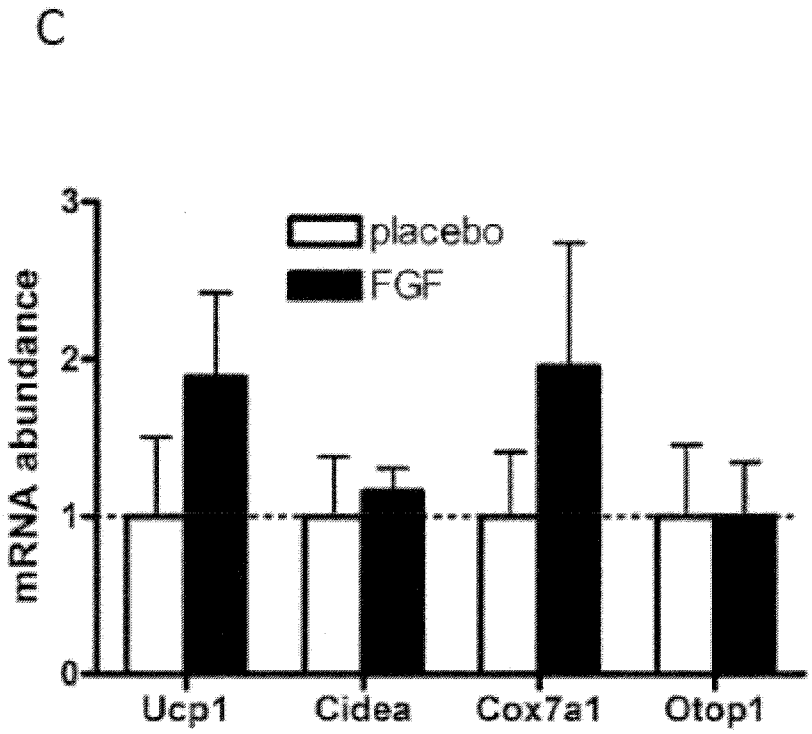
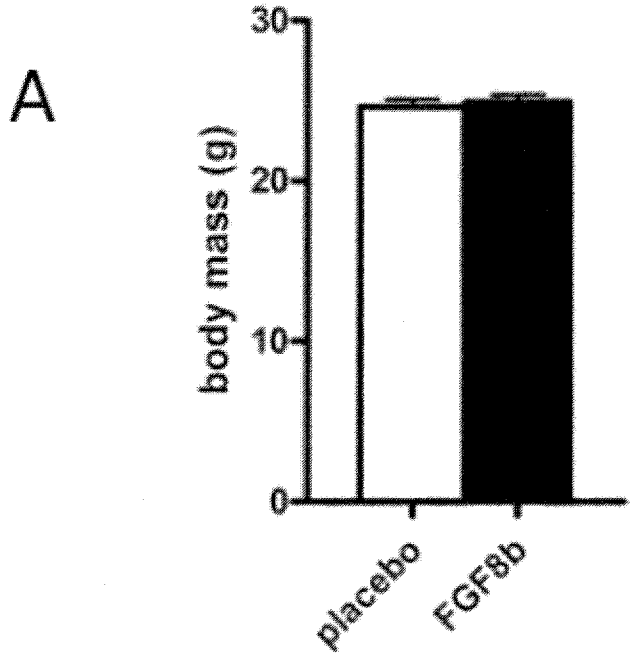


Figure 7.



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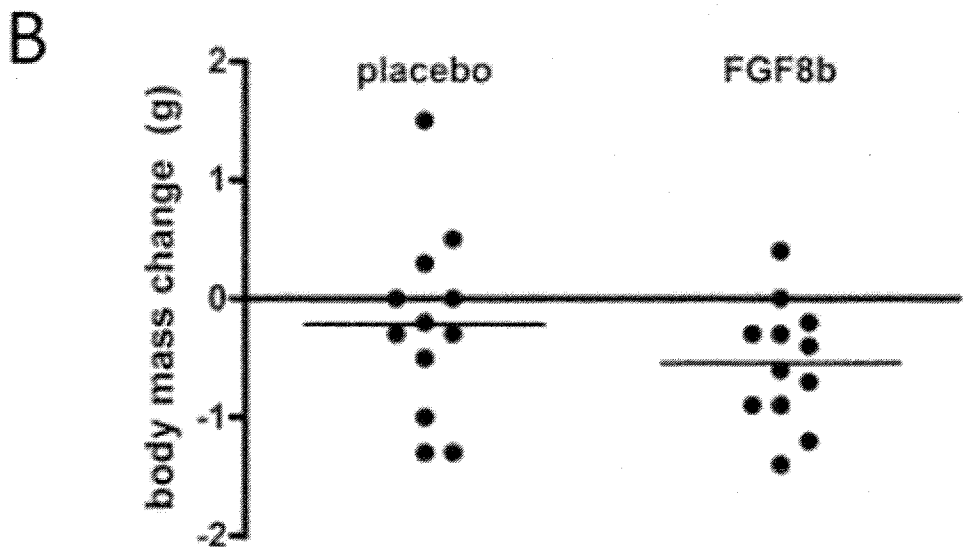

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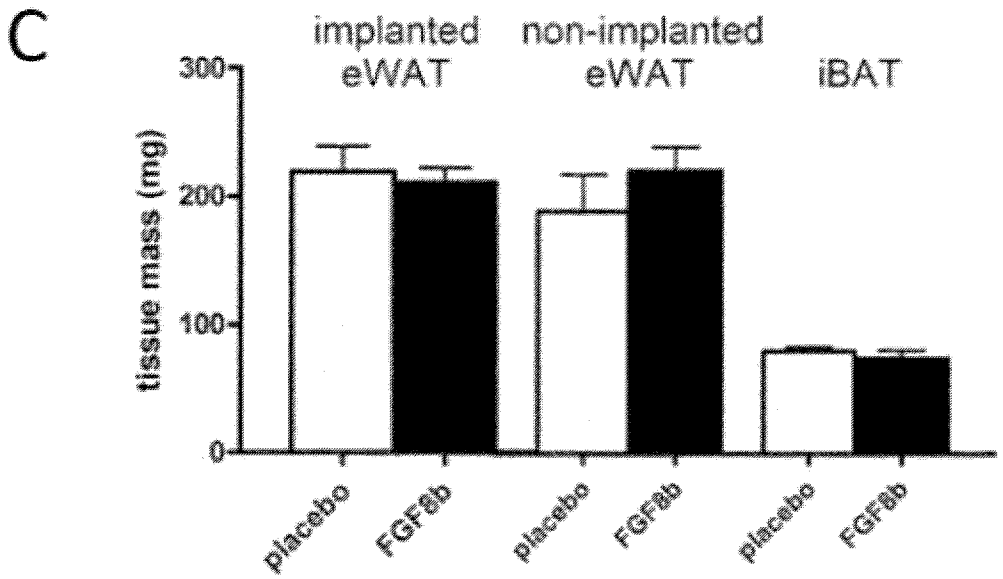

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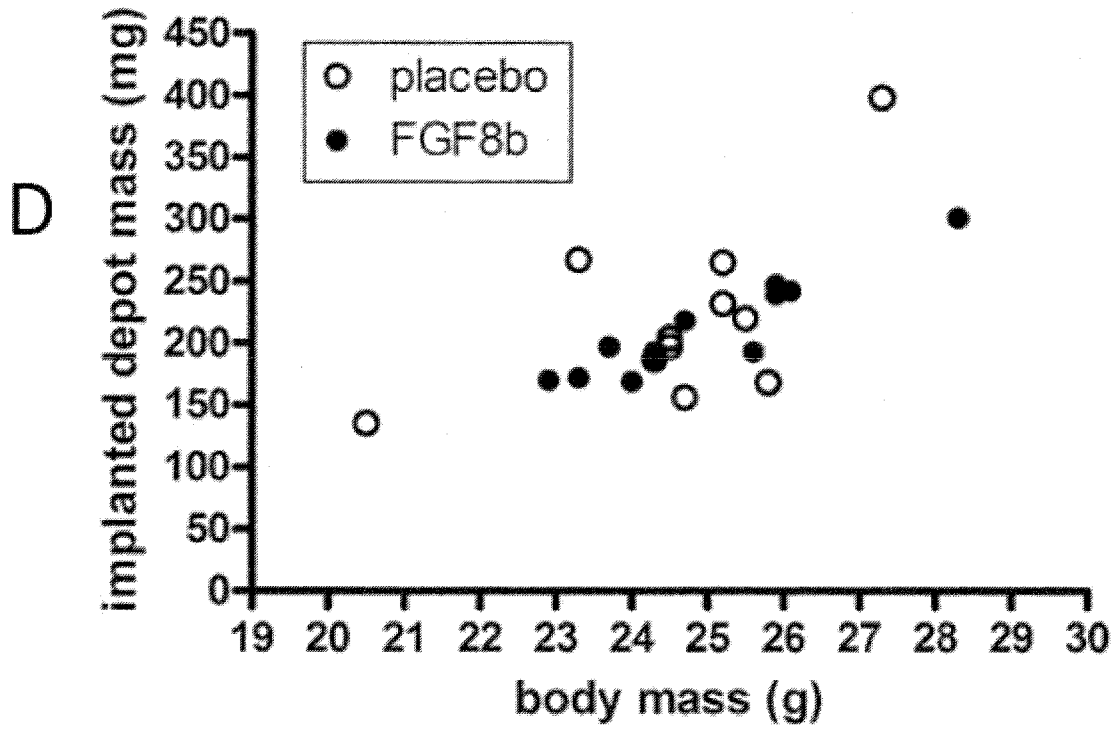

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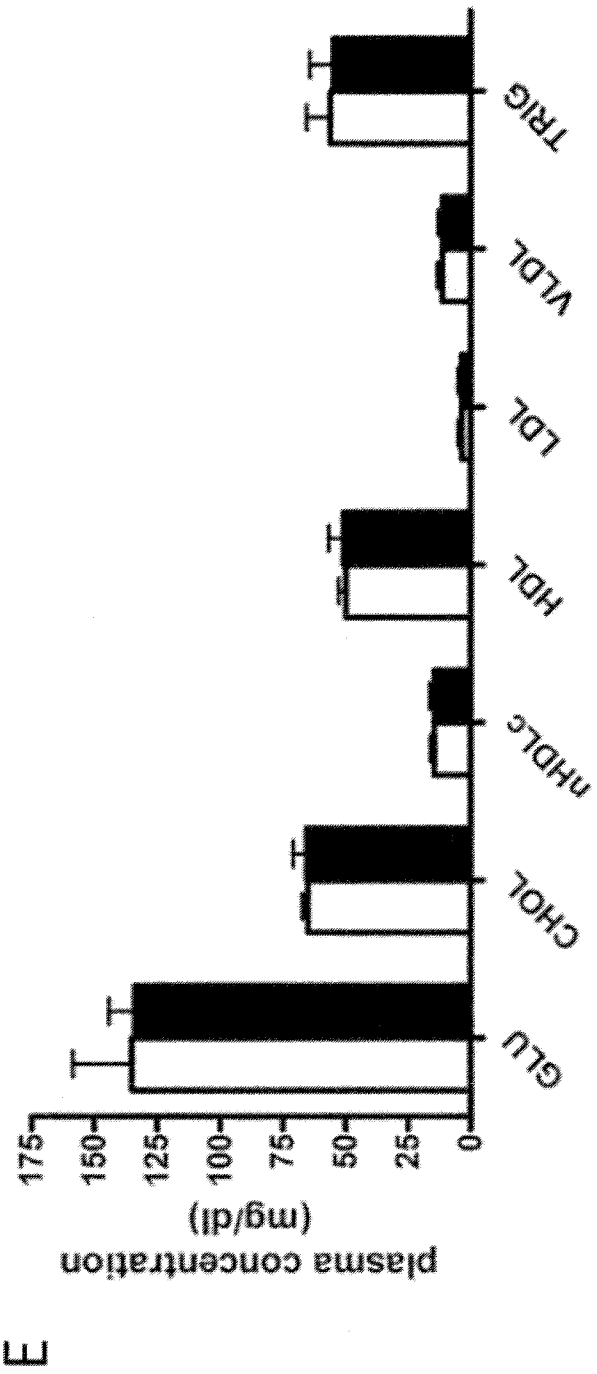
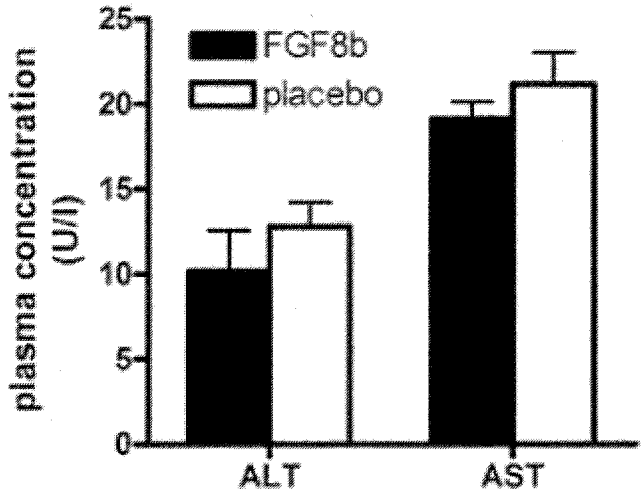


E

Figure 7 (cont.).

F



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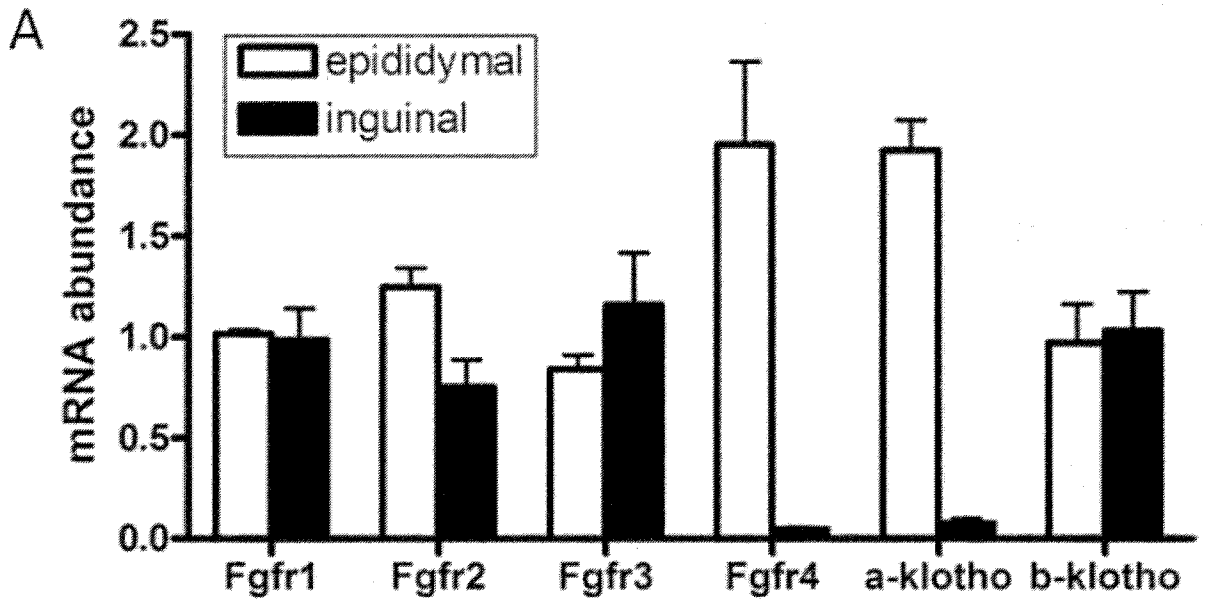

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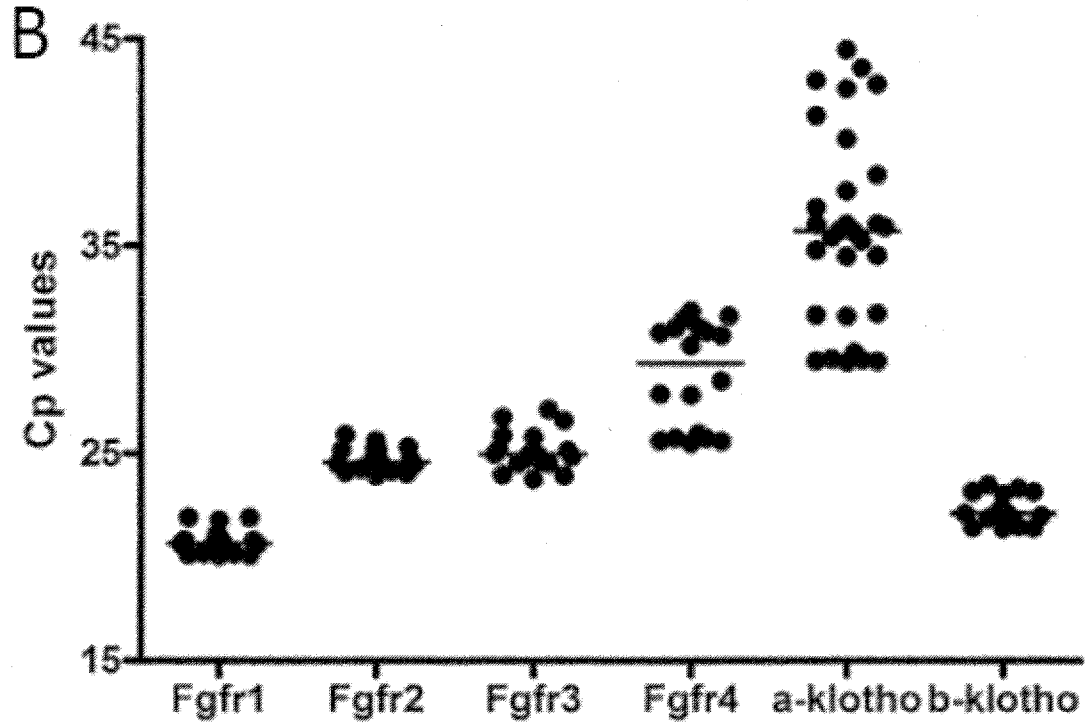

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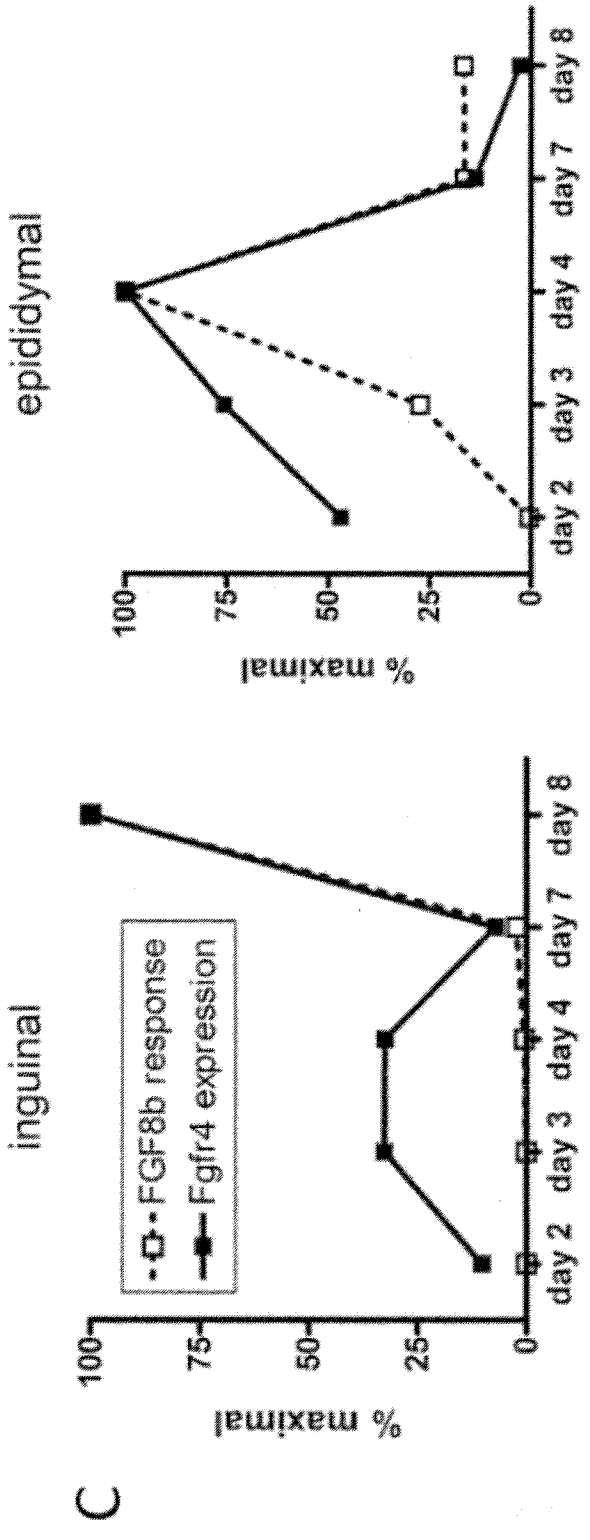

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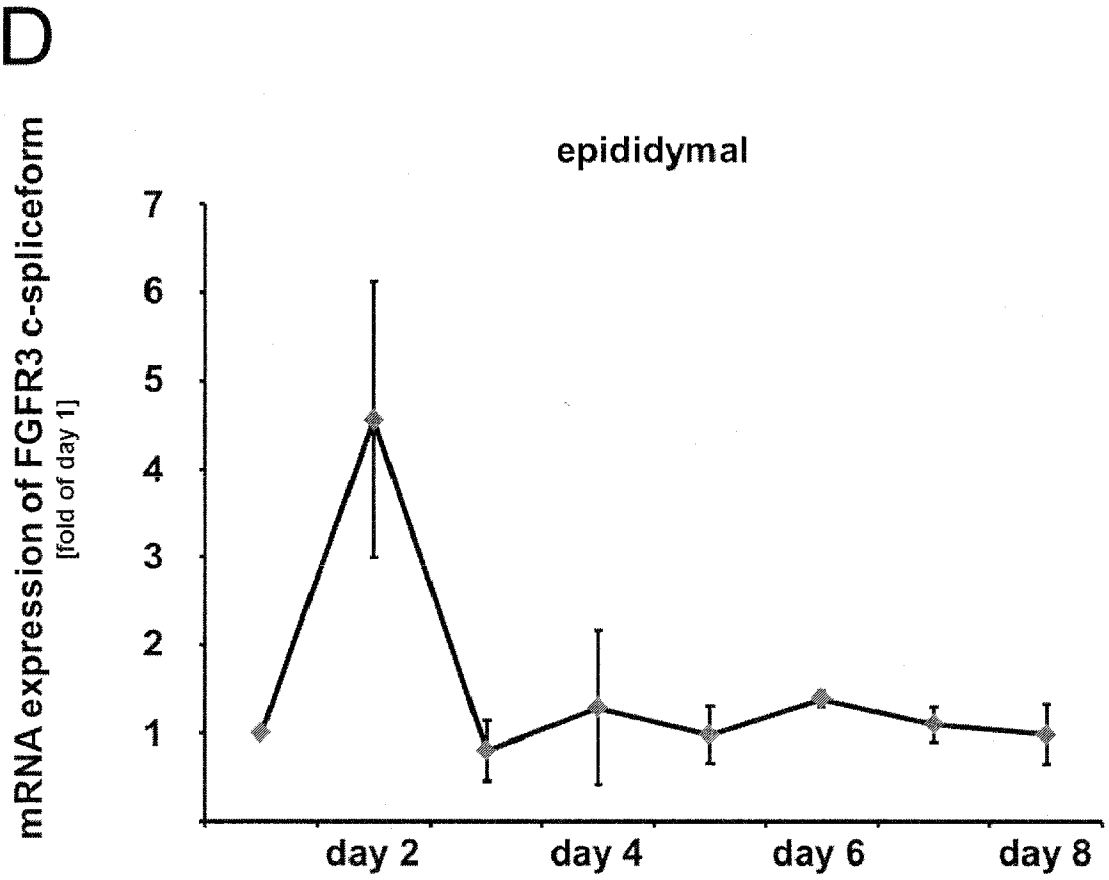

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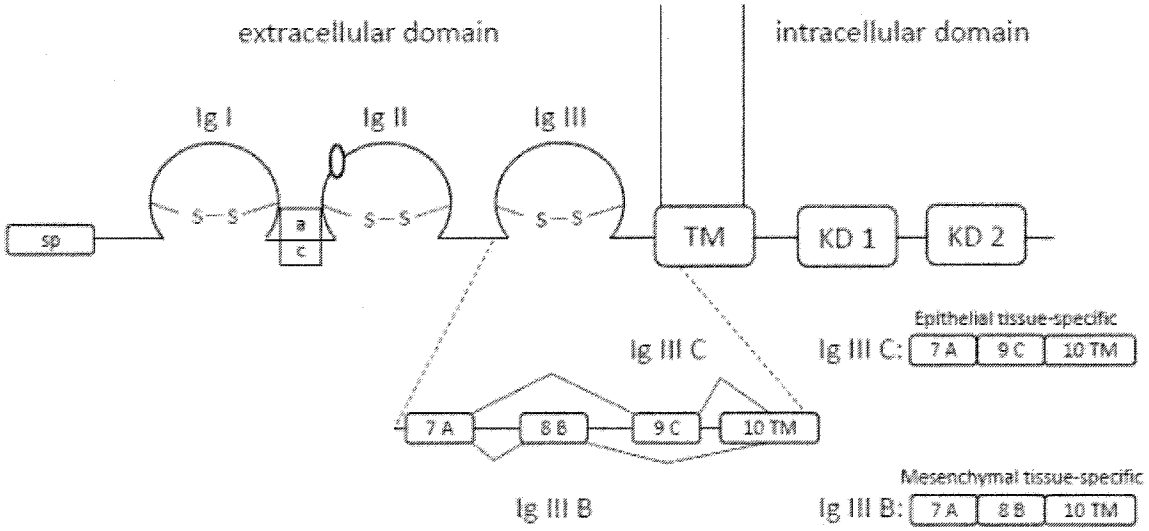


Figure 10.

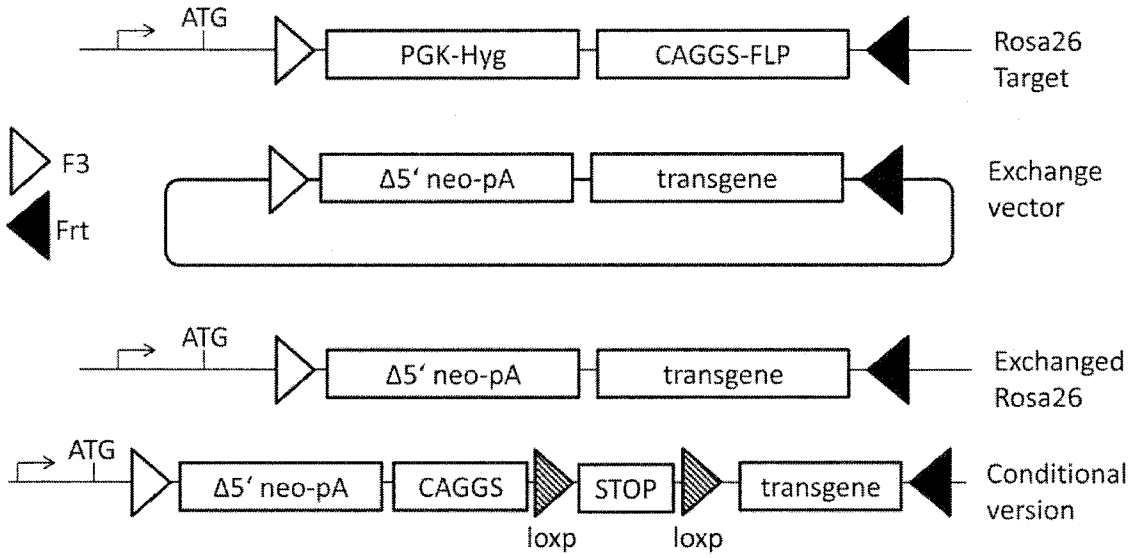


Figure 11.

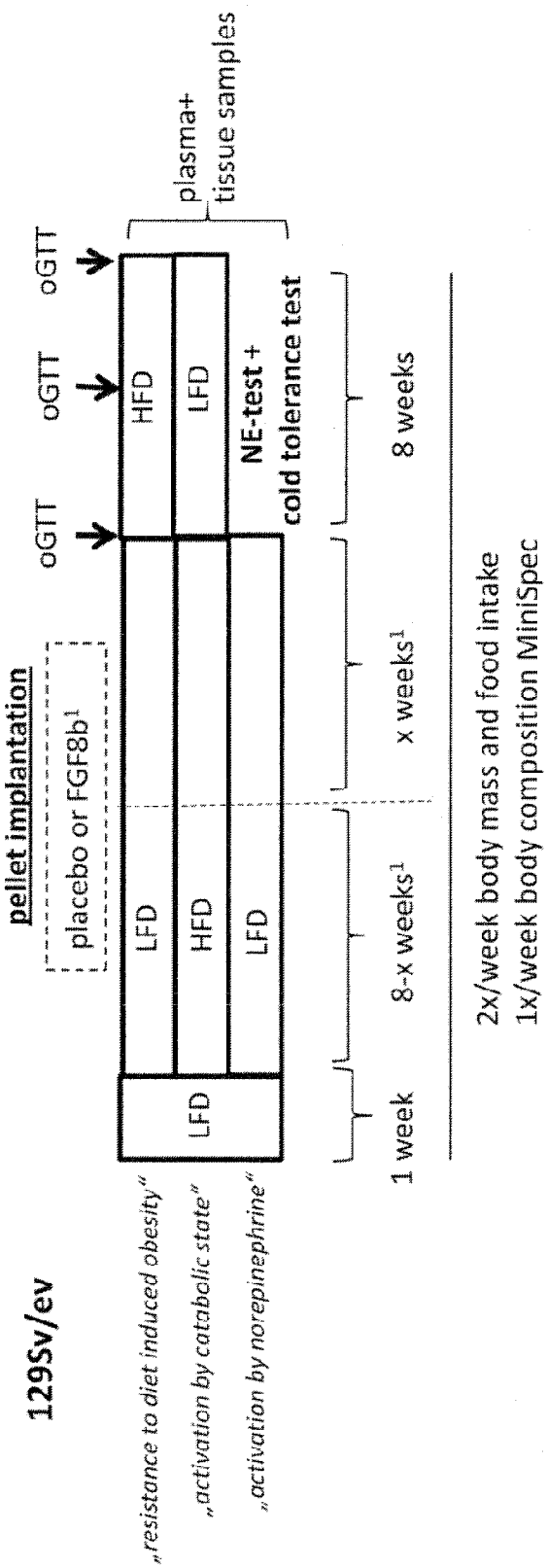

129Sv/ev
„resistance to diet induced obesity"
„activation by catabolic state"
„activation by norepinephrine"
pellet implantation
placebo or FGF8b1
LFD
HFD
LFD
oGTT
oGTT
oGTT
HFD
LFD
NE-test +
cold tolerance test
plasma+
tissue samples
1 week
8-x weeks1
x weeks1
8 weeks
2x/week body mass and food intake
1x/week body composition MiniSpec

Figure 12.

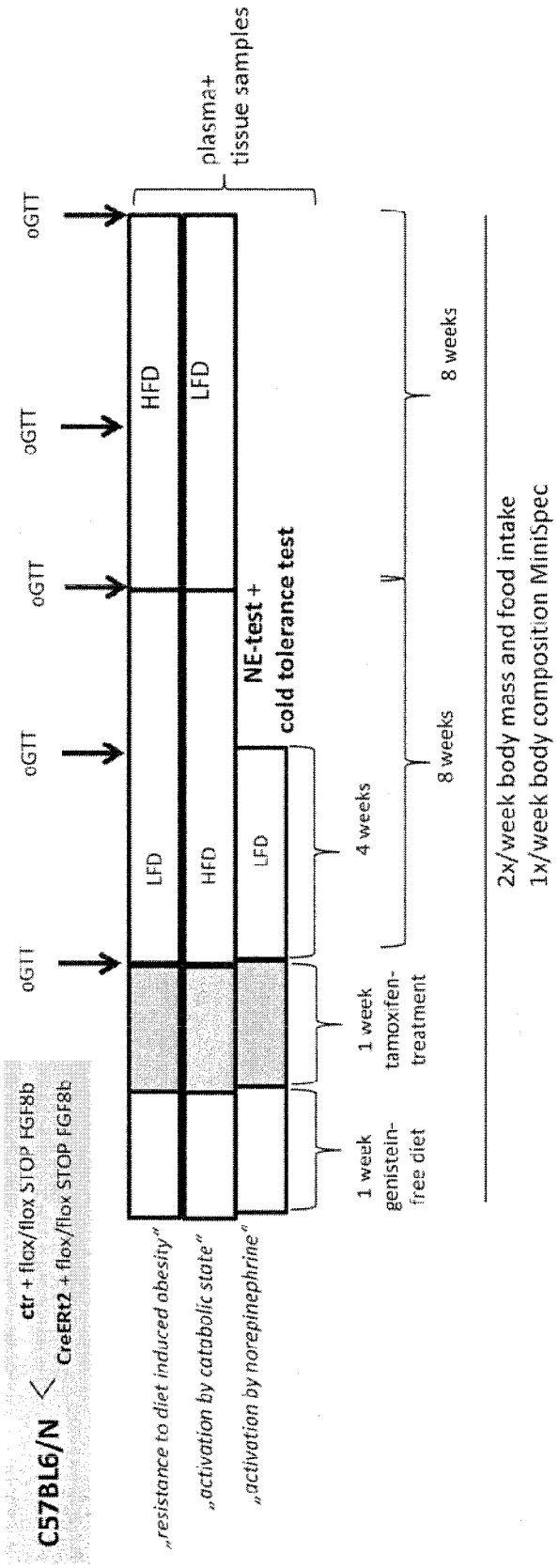
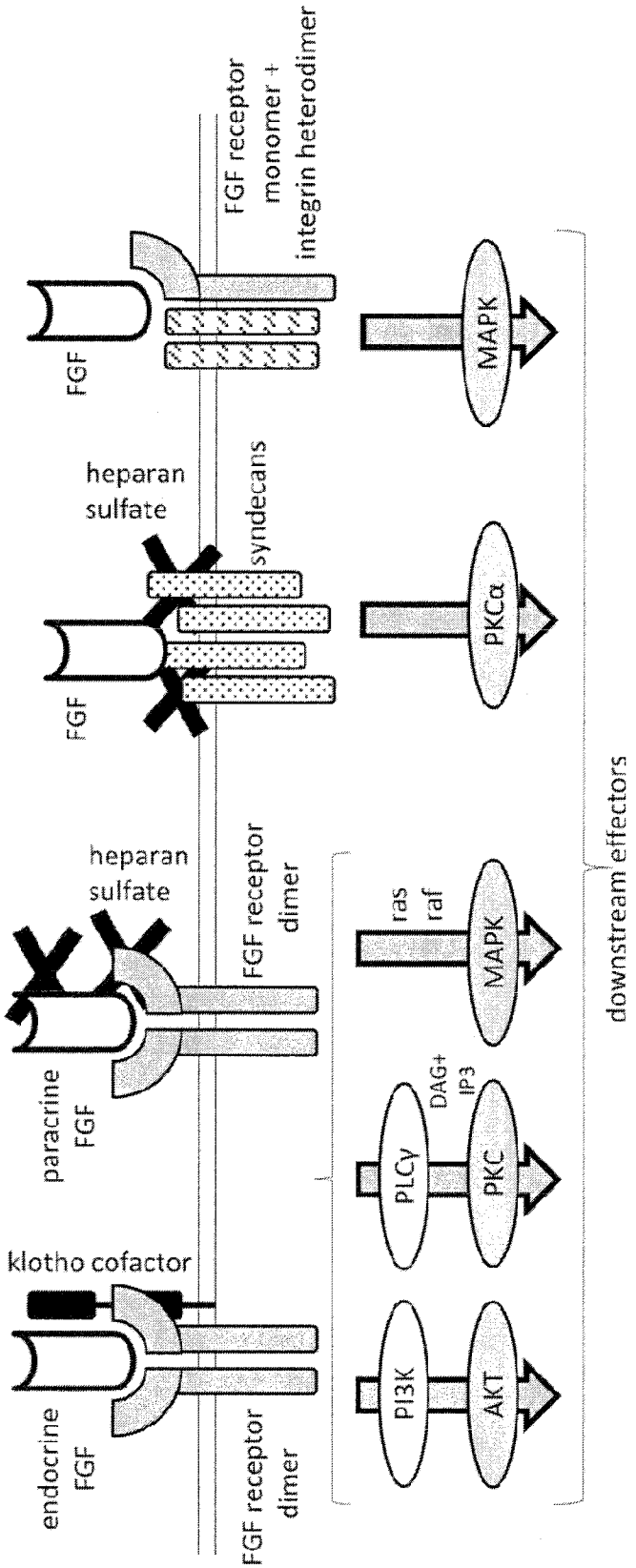

C57BL6/N
ctr + flox/flox STOP FGF8b
CreERt2 + flox/flox STOP FGF8b
oGTT
LFD
HFD
LFD
1 week genistein-free diet
1 week tamoxifen-treatment
4 weeks
8 weeks
8 weeks
NE-test + cold tolerance test
plasma+ tissue samples
„resistance to diet induced obesity"
„activation by catabolic state"
„activation by norepinephrine"
2x/week body mass and food intake
1x/week body composition MiniSpec

Figure 13.


FGF receptor monomer + integrin heterodimer
FGF
MAPK
heparan sulfate
syndecans
FGF
PKCα
heparan sulfate
FGF receptor dimer
ras raf
MAPK
paracrine FGF
DAG+ IP3
PLCγ
PKC
klotho cofactor
endocrine FGF
PI3K
AKT
FGF receptor dimer
downstream effectors

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2015/053165

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K38/18 A61P3/00 A61P3/08 A61P3/10 A61P3/06
A61K8/64
ADD.
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K A61P

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO 01/00662 A2 (LILLY CO ELI [US]; SINGH JAI PAL [US]; WAGLE ASAVARI PRASAD [US]) 4 January 2001 (2001-01-04) claims 1, 8, 9, 10, 11, 12 sequences 1, 2 the whole document -----	1-14
Y	WO 2013/138795 A1 (PERMEON BIOLOG INC [US]; VOGAN ERIK M [US]; FRANZUSOFF ALEX [US]; EDWA) 19 September 2013 (2013-09-19) the whole document sequence 22 claim 78 pages 39-41; table 1 page 92, lines 9-10 -----	1-14
	-/--	

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
29 April 2015	26/05/2015

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Fayos, Cécile

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C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 6 037 329 A (BAIRD J ANDREW [US] ET AL) 14 March 2000 (2000-03-14) the whole document claim 29	1-14
Y	WO 2013/149258 A2 (CHARLES DREW UNIVERSITY OF MEDICINE AND SCIENCE [US]; SINGH RAJAN CHAR) 3 October 2013 (2013-10-03) the whole document page 42, lines 29-32 claims 1-55	1-14
A	WO 2013/006486 A2 (NGM BIOPHARMACEUTICALS INC [US]; LING LEI [US]; LINDHOUT DARRIN A [US]) 10 January 2013 (2013-01-10) the whole document	1-14
A	EP 2 359 843 A1 (SANOFI SA [FR]) 24 August 2011 (2011-08-24) the whole document	1-14
A	BEENKEN ANDREW ET AL: "The FGF family: biology, pathophysiology and therapy.", NATURE REVIEWS. DRUG DISCOVERY MAR 2009, vol. 8, no. 3, March 2009 (2009-03), pages 235-253, XP002624720, ISSN: 1474-1784 the whole document	1-14
A	KHARITONENKOV A ET AL: "FGF-21 as a novel metabolic regulator", JOURNAL OF CLINICAL INVESTIGATION, AMERICAN SOCIETY FOR CLINICAL INVESTIGATION, US, vol. 115, no. 6, 1 June 2005 (2005-06-01), pages 1627-1635, XP002362553, ISSN: 0021-9738, DOI: 10.1172/JCI23606 the whole document	1-14

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/EP2015/053165

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 0100662 A2	04-01-2001	AU 5866600 A	31-01-2001
		WO 0100662 A2	04-01-2001
WO 2013138795 A1	19-09-2013	AU 2013231851 A1	11-09-2014
		CA 2867188 A1	19-09-2013
		EP 2825561 A1	21-01-2015
		WO 2013138795 A1	19-09-2013
US 6037329 A	14-03-2000	US 6037329 A	14-03-2000
		US 6503886 B1	07-01-2003
		US 2003143217 A1	31-07-2003
WO 2013149258 A2	03-10-2013	EP 2875359 A2	27-05-2015
		WO 2013149258 A2	03-10-2013
WO 2013006486 A2	10-01-2013	AU 2012279237 A1	16-05-2013
		CA 2835607 A1	10-01-2013
		CN 103649127 A	19-03-2014
		CO 6940374 A2	09-05-2014
		EP 2726511 A2	07-05-2014
		HK 1198297 A1	27-03-2015
		JP 2014527402 A	16-10-2014
		KR 20140039322 A	01-04-2014
		PE 17272014 A1	26-11-2014
		US 2013023474 A1	24-01-2013
		WO 2013006486 A2	10-01-2013
EP 2359843 A1	24-08-2011	AR 079939 A1	29-02-2012
		AU 2011208650 A1	09-08-2012
		CA 2787131 A1	28-07-2011
		CL 2012002031 A1	14-12-2012
		CN 102753192 A	24-10-2012
		CO 6592082 A2	02-01-2013
		DK 2525807 T3	09-02-2015
		EP 2359843 A1	24-08-2011
		EP 2460527 A1	06-06-2012
		EP 2525807 A1	28-11-2012
		EP 2754449 A1	16-07-2014
		ES 2529612 T3	23-02-2015
		HR P20150029 T1	27-03-2015
		JP 2013518035 A	20-05-2013
		KR 20120123445 A	08-11-2012
		MA 33931 B1	02-01-2013
		NZ 601306 A	29-11-2013
		PT 2525807 E	04-02-2015
		RU 2012135686 A	27-02-2014
		SG 182652 A1	30-08-2012
		SI 2525807 T1	27-02-2015
		TW 201141511 A	01-12-2011
		US 2013203651 A1	08-08-2013
		UY 33194 A	31-08-2011
		WO 2011089203 A1	28-07-2011

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP2015/053165

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:

   a. [ ] forming part of the international application as filed:

      [ ] in the form of an Annex C/ST.25 text file.

      [ ] on paper or in the form of an image file.

   b. [X] furnished together with the international application under PCT Rule 13*ter*.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.

   c. [X] furnished subsequent to the international filing date for the purposes of international search only:

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      [ ] on paper or in the form of an image file (Rule 13*ter*.1(b) and Administrative Instructions, Section 713).

2. [X] In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments: