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(57) Abstract: The present invention relates generally to the field of compositions containing wholegrain cereals. In particular, the present invention relates to health benefits of wholegrain cereals. One embodiment of the present invention concerns wholegrain cereal for use in the treatment or prevention of a liver disorder, wherein the wholegrain cereal is administered to a subject in an amount of at least 100 g, preferably of at least 160 g per day.



Wholegrain cereal and liver disorders

The present invention relates generally to the field of compositions containing wholegrain cereals. In particular, the present invention relates to health benefits of wholegrain cereals.

- 5 One embodiment of the present invention concerns wholegrain cereal for use in the treatment or prevention of a liver disorder, wherein the wholegrain cereal is administered to a subject in an amount of at least 100 g, preferably of at least 160 g per day.

10 Liver disorders occur widely in the population and are a risk factor for early mortality and more serious liver diseases. For example, fatty liver disease (FLD) exists in approximately 10-24 % of the population, depending on the country, and may have a prevalence of about 75 % among obese people. One German study estimated that having fatty liver disease increased a patients health costs for the Government by 26 % over a 5 year period (Baumeister SE, et al. (2008) Gastroenterology 134: 85-94).

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Fatty liver disease (FLD) is an inflammation of the liver, caused by the build up of fat in the liver cells which can over time lead to decreased liver function and if untreated, cirrhosis (where the liver ceases to function properly). When seen under a microscope slide, fatty liver cells have large apparently empty spaces, which are fat deposits. This fat accumulation ultimately pushes the cell nucleus to the edge of the cell, and the cell ceases to function normally. The reasons for the increase in fat accumulation are widely debated, but are suggested to involve a decrease in function of some cell signalling pathways, general insulin resistance, and/or a lack of the methyl-donor betaine. One of the possible causes of FLD is a metabolic disequilibrium around the one-carbon metabolic cycle. In addition, other metabolic factors may play a roll in the accumulation of lipids (e.g. loss of insulin sensitivity). While a better diet and exercise are the main recommended treatments, there are no specific food recommendations.

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There are two main types of FLD – alcoholic FLD, where subjects regularly consume >20 g/d alcohol, and alcohol is considered to be the main cause of the fat build up; and non-alcoholic FLD (NAFLD), which is not caused by alcohol, and often related to insulin resistance and metabolic syndrome. The extreme form of NAFLD is known as non-alcoholic steatohepatitis (NASH). NASH is considered to be one of the main causes of otherwise undefined cirrhosis

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of the liver (over time, around 20 % of people with NASH will go on to develop cirrhosis of the liver). If treated early enough, the condition is reversible.

NAFLD is often associated with impaired insulin metabolism, obesity and metabolic syndrome. It is also caused by protein malnutrition (rapid weight loss may exacerbate the condition in obese people), and treatment with corticosteroids (e.g. cortisone and prednisone). Alcoholic-FLD is generally associated with chronic alcohol abuse, and in this case the metabolism of alcohol by the enzyme alcohol dehydrogenase also sends a signal for an increase in fatty acid synthesis and a decrease in fatty acid breakdown.

FLD is usually diagnosed using liver function tests. Alcohol consumption over 20 g/d is considered to be the threshold for considering alcohol abuse to be the cause of the FLD condition. Abnormal liver enzyme values (e.g. alanine transaminase; aspartate transaminase, bilirubin and γ -glutamyl transpeptidase), and ultrasound observation of steatosis (abnormal accumulation of fat within cells) are minimally invasive markers of FLD, but definitive diagnosis can only be made with a liver biopsy and histological examination. Current treatments of FLD usually involve weight loss, and may include the administration of metformin and thiazolidinediones. Improvements of diet and exercise habits appear to be the most effective long-term treatments. Some experimental evidence also suggests that supplementation with glycine betaine (betaine) can reverse the progression of both alcoholic and non-alcoholic FLD (Miglio F, et al. (2000) *Arzneim. Forsch./Drug Research* 50: 722-727; Samara K, et al. (2006) *Digestive Diseases and Sciences* 51: 1226-1229).

US2002/0119181A1 discloses nutritional compositions made from conventional foods mixed on-site in a blender for treating patients with hepatic disorders. Thereby, a nutritionally complete, calorically dense mixture comprising a range of vitamin and mineral rich foods is achieved. The purpose of the invention is to address and correct the malnutrition as common in many hepatic diseased patients. However, the document fails to specifically provide a treatment or prevention of the hepatic disorder itself.

US7,078,064B2 relates to compositions comprising antioxidants useful for reducing oxidative stress and lipid peroxidation and thereby treating a.o. chronic liver diseases. In particular, the document relates to compositions supplemented with antioxidants, including certain vitamins prepared from plant materials or produced by synthetic chemistry. The main route for this

formulation is via parental administration, i.e. intra-venous supply of nutrients when the oral route is not possible. The document fails to provide a natural and simple oral food product solution for the treatment or prevention of liver disorders.

- 5 In view of the broad distribution of liver disorders in today's population and substantial healthcare costs it would be desirable to have available a natural composition that would be cheap and available to everyone and could be used to prevent and/or treat liver disorders and/or reverse existing liver damage.
- 10 The present inventors have addressed this need and have found that the subject matter of the independent claim achieves the object described above. The thereon dependant claims further develop the idea of the present invention. In particular, the inventors have found that wholegrain cereal can be used in the treatment or prevention of a liver disease, wherein the wholegrain cereal is administered to a subject in an amount of at least 100 g, preferably of at
- 15 least 160 g per day. The wholegrain cereal could be provided in the form of a composition that can be used, e.g., as food product and would consequently be available to everybody and that can be used to treat or prevent liver disorders.

The term "liver disorder" is to be understood herein as equivalent to the term "liver disease",
20 and vice versa.

The inventors were surprised to see the administration of a wholegrain diet for 7 days results in a continuously increased plasma betaine concentration. It was also shown that the plasma betaine concentration acutely increases after a breakfast wholegrain cereal challenge.

- 25 Example 2 further substantiates the beneficial effect of wholegrain cereal in an animal study. As supplementation with betaine has been linked to improvement in fatty liver, the results show that the compositions of the present invention improve liver stores of betaine, which will help prevent the development and progression of liver diseases. Consequently, one embodiment of the present invention is a composition comprising wholegrain cereal for use in
- 30 the treatment or prevention of liver disorders.

The present invention also related to the use of wholegrain cereal for the preparation of a composition to treat or prevent liver disorders, and particularly where the liver disorder is a non-alcoholic liver disease.

For example, the composition comprises at least about 0.3 g wholegrain cereal/g dry weight. Wholegrain cereal is to be understood for the purposes of the present invention as cereal grains that contain bran and germ as well as the endosperm. Refined cereal contains essentially only the endosperm.

For the purposes of the present invention wholegrain shall be understood as proposed by the guidelines of the United States Food and Drug Administration follows:

“Cereal grains that consist of the intact, ground, cracked or flaked caryopsis, whose principal anatomical components - the starchy endosperm, germ and bran - are present in the same relative proportions as they exist in the intact caryopsis - should be considered a wholegrain... Cereal grains may include amaranth, barley, buckwheat, bulgur, corn (including popcorn), millet, quinoa, rice, rye, oats, sorghum, teff, triticale, wheat, and wild rice (Guidelines of the United States Food and Drug Administration).

Advantageously, wholegrain cereal is an ingredient which is generally well accepted and liked by consumers. It is a natural ingredient that can be incorporated into a large variety of products, and consequently, will be available to everyone in sufficient quantities. Moreover, the composition of the present invention does not require the addition of purified chemical compounds, which may require a careful dosing, and which may have undesirable side effects.

Advantageously, the composition of the present invention may be administered in an amount that corresponds to a consumption of at least about 30g wholegrain cereal per day. Preferably, the amount to be administered corresponds to a consumption of at least about 100g wholegrain cereal per day.

While in principle any amount of wholegrain cereal will have a positive effect on liver health following a dose response curve, good effects are seen if at least 30g wholegrain cereal are consumed per day, and even better effects are achieved by administering at least about 100g wholegrain cereal per day. The preferred embodiment is an administration of a composition that corresponds to a daily consumption of wholegrain cereal of about 160g. Such amounts of wholegrain cereal consumption can be feasibly achieved when for example a part or most of the dietary carbohydrates are based on wholegrain cereals. Excessive consumption of

wholegrain cereal on the other hand is likely not to produce further increasing positive effects on liver health, but rather a saturation effect must be expected. Hence, in one embodiment the composition is to be administered in an amount corresponding to a consumption of between about 30-200g wholegrain cereal per day.

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The wholegrain cereal may be formulated in any kind of composition. However, such wholegrain compositions are generally perceived as pleasant to consume if they contain fibre, carbohydrates, fat and protein in a specific ratio. For example, the composition of the present invention may contain about 3-20 weight-% fibre, about 60-85 weight-% carbohydrates, about 0.5-10 weight-% fat and about 5-15 weight-% protein.

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Optionally, the composition of the present invention may also contain vitamins, minerals, trace elements and other micronutrients, e.g., in accordance with the recommendations of Government bodies such as the USRDA.

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For example, the composition may contain per daily dose one or more of the following micronutrients in the ranges given: 300 to 500 mg calcium, 50 to 100 mg magnesium, 150 to 250 mg phosphorus, 5 to 20 mg iron, 1 to 7 mg zinc, 0.1 to 0.3 mg copper, 50 to 200 µg iodine, 5 to 15 µg selenium, 1000 to 3000 µg beta carotene, 10 to 80 mg Vitamin C, 1 to 2 mg Vitamin B1, 0.5 to 1.5 mg Vitamin B6, 0.5 to 2 mg Vitamin B2, 5 to 18 mg niacin, 0.5 to 2.0 µg Vitamin B12, 100 to 800 µg folic acid, 30 to 70 µg biotin, 1 to 5 µg Vitamin D, 3 to 10 µg Vitamin E.

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The composition may also contain further compounds that are known to have a beneficial effect for the liver.

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For example, for the provision of an immediate effect the composition may further comprise added betaine (N,N,N-trimethylglycine). Betaine may be added as purified betaine or as a composition with a high betaine concentration. This will have the advantage that the added betaine will enforce an immediate beneficial effect for the liver while the wholegrain cereal will ensure that the betaine effect will last for prolonged time periods.

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As wholegrain cereal any kind of cereal or pseudocereal may be used. For example, the cereal may be selected from the group consisting of wheat, rye, triticale, oats, barley, rice, maize, sorghum, millet, quinoa, buckwheat or combinations thereof.

- 5 The wholegrain cereals may be processed, e.g., to improve storage times, the pleasure of consumption, and/or to way wholegrain can be incorporated into a final product.

Hence, the wholegrain cereals may at least in part processed. Typically, wholegrain cereals may be processed by a method selected from the group consisting of enzyme treatments, malting, heating, extrusion, baking, cooking or combinations thereof.

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The wholegrain cereal for use of the present invention relates to a subject, wherein the subject is a human being or an animal, preferably a pet animal, and more preferably a dog or a cat.

- 15 The composition of the present invention may be any kind of consumable product. For example, the composition may be selected from the group consisting of food products, an animal food products, pharmaceutical compositions, nutritional compositions, nutraceuticals, drinks, or food additives.

- 20 Pharmaceutical compositions have the advantage that they are generally administered under the supervision of medical personnel, which allows the determination of exact dosage forms for the subject to be treated.

- 25 Food products, animal food products, nutritional compositions, nutraceuticals, drinks, and food additives have the advantage that they are freely available to everyone and can consequently particularly well be used as preventive measure. Generally, the composition of the present invention can be consumed at any time. However, it was found that the composition is in particular pleasant to consume during or as breakfast. Alternatively, the composition of the present invention may be consumed during or as lunch or breakfast. Without wishing to be bound by theory the inventors believe that this preference is due to the
30 fact that it usually takes longer to digest wholegrain products than products from refined cereals. If this digestion procedure is ongoing through the night, this might be considered problematic for people that have trouble sleeping.

The composition of the invention can also be administered at least twice during the day, whereby one administration may include for example breakfast. Preferably, the composition is administered once in the morning, e.g. during or as breakfast, and once in the after-noon, for example during or as part of the lunch meal. This dieting regimen may particularly be applicable, where consumption of wholegrain cereal of 100g or more per day is envisaged. In this way, the daily consumption can be split into individual servings during the day, which allows to spread the up-take of e.g. carbohydrates and diverse beneficial phytochemicals present in the wholegrain cereal more evenly during the entire day and not to concentrate such consumption to one single meal. Further, the up-take and bioavailability of the effective nutrients from the whole grain will be more evenly spread during the day and hence have a much better and sustainable biological effect to the body of a consumer.

The liver disorder may be selected from the group consisting of fatty liver disease, non-alcoholic steatosis hepatitis, liver cirrhosis, cholestasis or combinations thereof.

The composition of the present invention may be intended for humans or animals. For animals the disorder to be treated or prevented may be fatty liver disease, non-alcoholic steatosis and/or liver cirrhosis and the composition may be formulated as a pet food composition.

Alternatively, the composition may also be formulated as a pet food snack or as a pet food additive.

If intended for humans, the composition of the present invention is particular intended for people at risk of developing a liver disorder, people suffering from liver disorders, and/or people recovering from liver disorders. For example, the composition of the present invention may be to be administered to overweight or obese people.

“Overweight” is defined for an adult human as having a BMI between 25 and 30.

“Obesity” is a condition in which the natural energy reserve, stored in the fatty tissue of animals, in particular humans and other mammals, is increased to a point where it is associated with certain health conditions or increased mortality. “Obese” is defined for an adult human as having a BMI greater than 30.

“Body mass index” or “BMI” means the ratio of weight in kg divided by the height in metres, squared.

Consequently, the composition of the present invention may be to be administered to people with a BMI of above at least 25, preferably above 30.

- 5 Alternatively, the wholegrain cereal for use of the invention may be administered to an over-weight or obese animal, for example an over-weight dog or an over-weight cat.

Since FLD is often caused by increased alcohol consumption, the composition of the present invention may also be to be administered to people with an above average alcohol
10 consumption. For example, the composition may be to be administered to subjects which consume on average at least about 20 g alcohol per day. Of course, the composition of the present invention may also be to be administered to people suffering from alcoholism.

Since the risk of developing liver disorders increases with age, the composition may also be to
15 be administered to people above the age of 40.

Some exemplary embodiments of the present invention are listed below:

In one embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is wheat and the composition is intended for people above the age of 40.

- 20 In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is oats and the composition is intended for people above the age of 50.

In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is maize and the composition is intended for people above the age of 40.

- In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal
25 is rice and the composition is intended for people above the age of 40.

In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is wheat and the composition is intended for people with a BMI of above at least 25.

In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is oats and the composition is intended for people with a BMI of above at least 25.

- 30 In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is maize and the composition is intended for people with a BMI of above at least 25.

In a further embodiment the disorder to be treated or prevented is FLD, the wholegrain cereal is rice and the composition is intended for people with a BMI of above at least 25.

It is clear to those of skill in the art that they can freely combine all features of the present invention disclosed herein without departing from the scope of the invention as disclosed. Further advantages and features of the present invention are apparent from the following example and drawing.

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Figure 1 shows that plasma betaine is increased after one week on a wholegrain cereal diet.

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Figure 2 shows that plasma betaine is increased after a wholegrain breakfast cereal. All points are different from the refined grain diet, and the maximum of 120 minutes is different from all other timepoints measured.

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Figure 3: Image of a section of one rat liver from group 4 (Zucker fa/-, healthy control fed with refined wheat diet) showing periportal fatty changes, mainly microvacuolar with a degree of severity of 2.

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Figure 4: Image of a section of one rat liver from group 1 (Zucker fa/fa rat fed with a refined wheat diet), showing diffuse fat distribution with micro and macrovacuolar (grade 4) and centrilobular hypertrophy (grade 3).

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Figure 5: Image of a section of one rat liver from group 3 (Zucker fa/fa fed with a refined wheat diet with addition of betaine) showing diffuse fat distribution with micro and macrovacuolar (grade 4) and centrilobular hypertrophy (grade 3).

Figure 6: Image of a section of rat liver from group 2 (Zucker fa/fa fed with a whole grain wheat diet) showing less diffuse fat distribution and less severe micro and macrovacuolar (grade 2) and centrilobular hypertrophy (grade 1) compared to rats fed refined wheat or refined wheat plus betaine diets.

Example 1:

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In a randomised cross-over trial, subjects had one of two different energy matched diets: one based on wholegrain cereals, containing Nestlé wholegrain products (e.g. Buitoni pasta and crackers, Lean Cuisine, DON couscous, Shreddies, Shredded Wheat, Uncle Toby's muesli, porridge, barley risotto, rye bread), and the other containing refined grain cereal products (e.g. Cornflakes, Rice Krispies, Nestlé equivalent non-wholegrain cereals). The composition of the

wholegrain cereal foods falls within the following macronutrient specification: 100 – 400 kcal/100 g; 4-15 g protein/100 g; 0.2-30 g fat/100 g and 2-25 g fibre/100 g. The average total daily intake of wholegrains on the wholegrain diet was 150 g/d (dry weight basis). With the exception of fibre, diets were matched for total energy and macronutrient content.

- 5 Eleven female and 6 male healthy subjects aged between 20 and 50 took part in the trial. Subjects followed one of the diets for two weeks, and then followed the other after at least 8 weeks of washout (normal and non-wholegrain diet). On day 8 of each controlled diet period, the subjects undertook a post-prandial challenge, where they consumed a breakfast appropriate for the diet, and then gave blood samples over the next 4 hours.
- 10 Differences between subjects were measured using a repeated measures ANOVA and a paired t-test, and were significant at the $P < 0.05$ level. Plasma betaine concentrations increased after 7 days on the wholegrain diet (Figure 1), and increased acutely after the breakfast cereal challenge (Figure 2).

- As supplementation with betaine has been linked to improvement in fatty liver, the results
- 15 show that the compositions of the present invention improve liver stores of betaine, which will help prevent the development and progression of liver diseases. Other factors in wholegrain cereals may also help to improve insulin sensitivity, which could also help prevent and treat the development of, e.g., FLD.

20 Example 2:

The effect of wholegrain cereal on the reduction of symptoms of fatty liver can be tested in an animal study using for example Zucker rats. Zucker (fa/fa) rats have a defect for the gene that regulates appetite, so spontaneously develop fatty liver because they overeat. One of the main risk factors for fatty liver in humans is obesity brought about by overconsumption of food.

- 25 Zucker (fa/+) rats are genetically similar (littermates), but are heterozygous for this gene defect, and thus serve as normal weight controls.

The study is conducted with four groups, with each group of 10 animals aged 8 weeks at the start of the study and fed with a different diet as follows:

- 1) Zucker (fa/fa) rats fed a refined grain-based diet
- 30 2) Zucker (fa/fa) rats fed a wholegrain-based diet
- 3) Zucker (fa/fa) rats fed a refined grain-based diet, with added glycine betaine to match the diet of Group 2
- 4) Zucker (fa/-) rats fed a refined grain-based diet ('healthy control group')

Diets are based on the AIN-93G diet and matched for total energy, total fibre and percentage of energy coming from fat, carbohydrate and protein. 50 % of the diets by weight come from wheat flour (either refined or wholegrain). This corresponds to a total wholegrain cereal consumption of about 100g to 160g per day for an average sized adult person. All diets are supplemented with standard vitamin and mineral mixes, so that deficiencies of these should not be responsible for any differences observed.

The rats are fed ad libitum, with paired feeding after two weeks if intake between groups is different. In total rats can be fed the different diets for 8 weeks. After 8 weeks, the rats are sacrificed and liver samples taken to assess the development of fatty liver, based on liver histopathology (size of fat deposits and abundance of inflammatory cells).

Example 3:

At present, liver biopsy remains the only reliable way to diagnose NAFLD (non-alcoholic fatty liver disease). It is the gold standard test to assess accurately the different degree of steatosis associated with NAFLD (Kleiner D.E., Brunt E.M., Van Natta M., Behling C., Contos M.J., Cummings O.W., Ferrell L., Liu Y-C. Torbenson M.S., Unalp-Arida A., Yeh M., McCullough A., Sanyal A.J., *Hepatology*, 2005, 1313-1321). In humans, this is carried out using a semi quantitative scale to grade the severity of NAFLD using light microscopy with hematoxylin and eosin (H&E) staining. The same approach is used for grading the severity of NAFLD in rodents, and it is assumed that hepatic pathologies associated with the development of NAFLD in rodents are similar to that in humans.

10 rats in each group were blindly studied by a histopathologist. Histological changes (distribution of lipids, severity grade, incidence and visible fat quantification by image analysis) were recorded during histopathology examination. Severity grades ranges from 1-minimal, 2-slight, 3-moderate and 4-severe. Image analysis was carried out in each liver sample (using Color view III camera at a magnification X 10) and calculated (using Cell Analysis) in area % (expressed as median $\pm$ SEM).

In the first control group (group 4), Zucker fa/- rats fed with a refined wheat diet, the lipids are recorded in the periportal region with a reversible lesion observed as microvacuolar changes (Figure 3). The mean severity observed for peritortal fat was 2.4 for 10 rats. The percentage of lipids measured by image analysis was 26.1 ± 4.9 %.

In the positive control (Zucker fa/fa, group 1), rats fed with a refined wheat diet, the liver showed heavy changes of lipids from normal periportal distribution towards a diffuse fatty changes associated with hepatocellular hypertrophy (Figure 4). The hepatocellular hypertrophy degree of severity was 2.0, whereas the mean severity grade observed for diffuse fat distribution was 3.2 (for 9 rats with one rat having a greater periportal fat change). The percentage of lipids measured by image analysis was 43.2 ± 2.2 %.

The rats from group 3 (Zucker fa/fa fed with refined wheat diet with addition of betaine) showed similar results than rats from group 2 (Figure 5). The liver showed diffuse fat distribution with a degree of severity at 3.8 (for 8 rats). The hepatocellular hypertrophy degree of severity was 1.7. The percentage of lipids measured by image analysis was 55.8 ± 4.7 %.

The rats from group 2 (Zucker fa/fa) fed with whole grain wheat-based diet showed a diffuse fat distribution (Figure 6) with a degree of severity at 3.0 (for only 4 rats). The hepatocellular hypertrophy degree of severity was 1.8. The percentage of lipids measured by image analysis was 36.1 ± 3.3 %.

In conclusion, these results provide evidence that a whole grain diet can lead to several positive alterations of lipids in liver in rats prone to developing NAFLD compared to refined wheat diet. The incidence (number of animals) of having a diffuse fat distribution in liver decreased from 9 to 3 animals between refined and whole grain wheat diet. The percentage of total lipids in liver, measured by image analysis, were significantly decreased ($P=0.043$) between the refined and the whole grain diets. These results also demonstrate that whole grain could reduce the severity of NAFLD compared to a refined wheat diet enriched in betaine. The data presented here demonstrate similar results between refined wheat diet and refined wheat diet with a betaine supplementation, suggesting that betaine did not play the key role in the observed prevention of NAFLD. This is confirmed by the finding that the rats fed whole grain diet had reduced liver fat deposits compared to rats eating refined wheat plus betaine.

Claims :

1. Wholegrain cereal for use in the treatment or prevention of a liver disease, wherein the wholegrain cereal is administered to a subject in an amount of at least 100 g,
5 preferably of at least 160 g per day.
2. The wholegrain cereal for use as of claim 1, wherein the liver disease is a non-alcoholic liver disease.
- 10 3. The wholegrain cereal for use as of claim 1 or 2, wherein the liver disease is selected from the group consisting of non-alcoholic fatty liver disease, non-alcoholic steatosis hepatitis, liver cirrhosis, cholestasis or combinations thereof.
4. The wholegrain cereal for use as of one of the above claims, wherein the cereal is
15 selected from the group consisting of wheat, rye, triticale, oats, barley, rice, maize, sorghum, millet, or combinations thereof.
5. The wholegrain cereal for use as of one of the above claims, wherein the cereal is at least in part processed.
20
6. The wholegrain cereal for use as of claim 5, wherein the cereal is processed by a method selected from the group consisting of enzyme treatments, malting, heating, extrusion, baking, cooking or combinations thereof.
- 25 7. The wholegrain cereal for use as of one of the above claims, wherein the subject is a human being or an animal, preferably a pet animal such as a dog or a cat.
8. The wholegrain cereal for use as of one of the above claims, provided in the form of a human food product, an animal food product, a pharmaceutical composition for a
30 human or an animal, a human nutritional composition, a nutraceutical, a drink or a food additive.

9. The wholegrain cereal for use as of claim 8 to be administered during or as a breakfast.
10. The wholegrain cereal for use as of claim 8 to be administered at least twice a day,
5 preferably at least once in the morning and once during lunch or in the after-noon.
11. The wholegrain cereal for use as of one of the above claims, to be administered to a human subject with a BMI of above at least 25, preferably of above 30.
- 10 12. The wholegrain cereal for use as of one of the claims 1 to 10, to be administered to an over-weight or obese animal.
13. The wholegrain cereal for use as of one of the above claims, wherein the cereal is administered in combination with betaine (N,N,N-trimethylglycine).
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14. The wholegrain cereal for use as of claim 13, wherein the betaine is added to the food product, the pharmaceutical composition, the nutraceutical, the drink or the food additive.

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Figure 1:

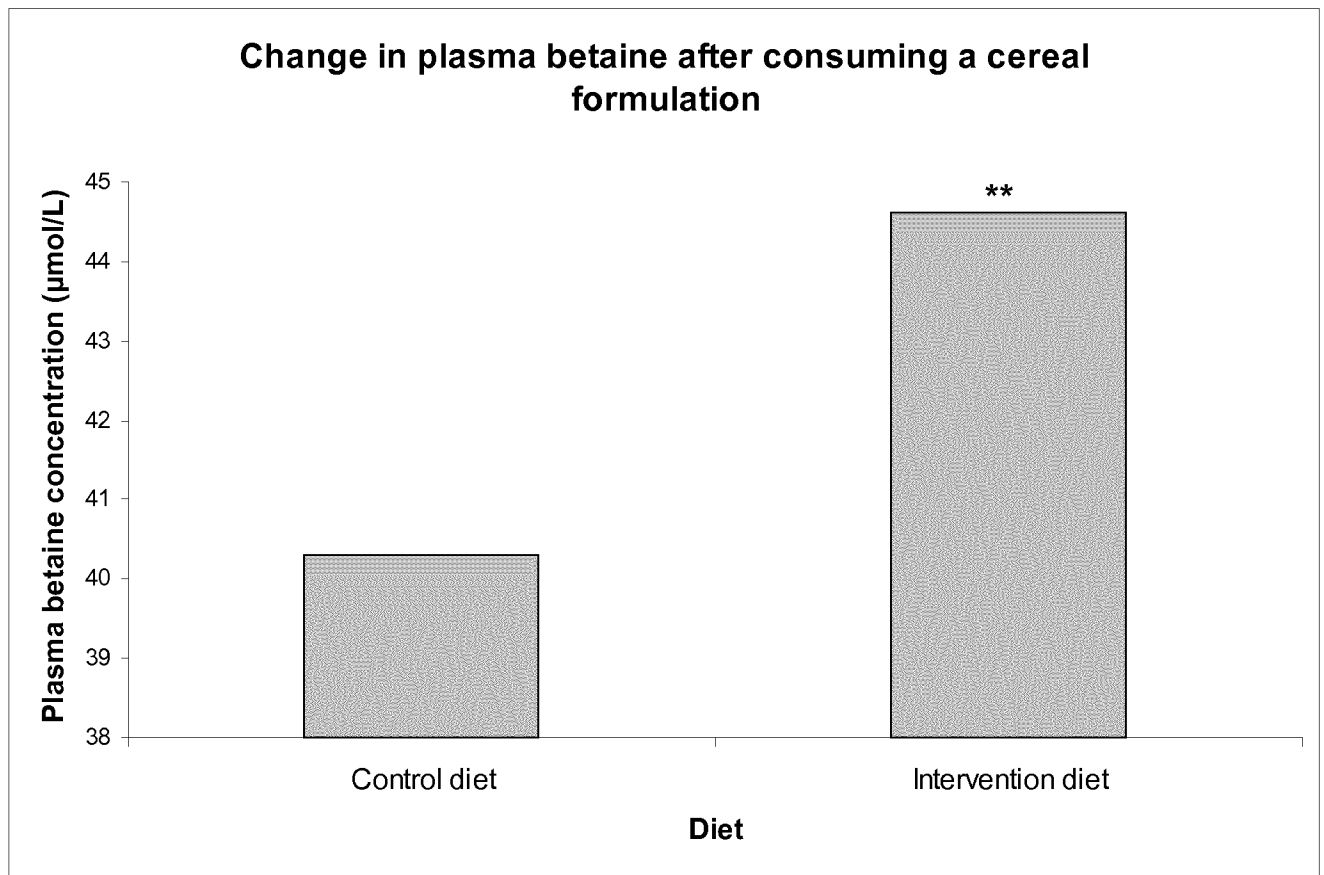


Figure 2:

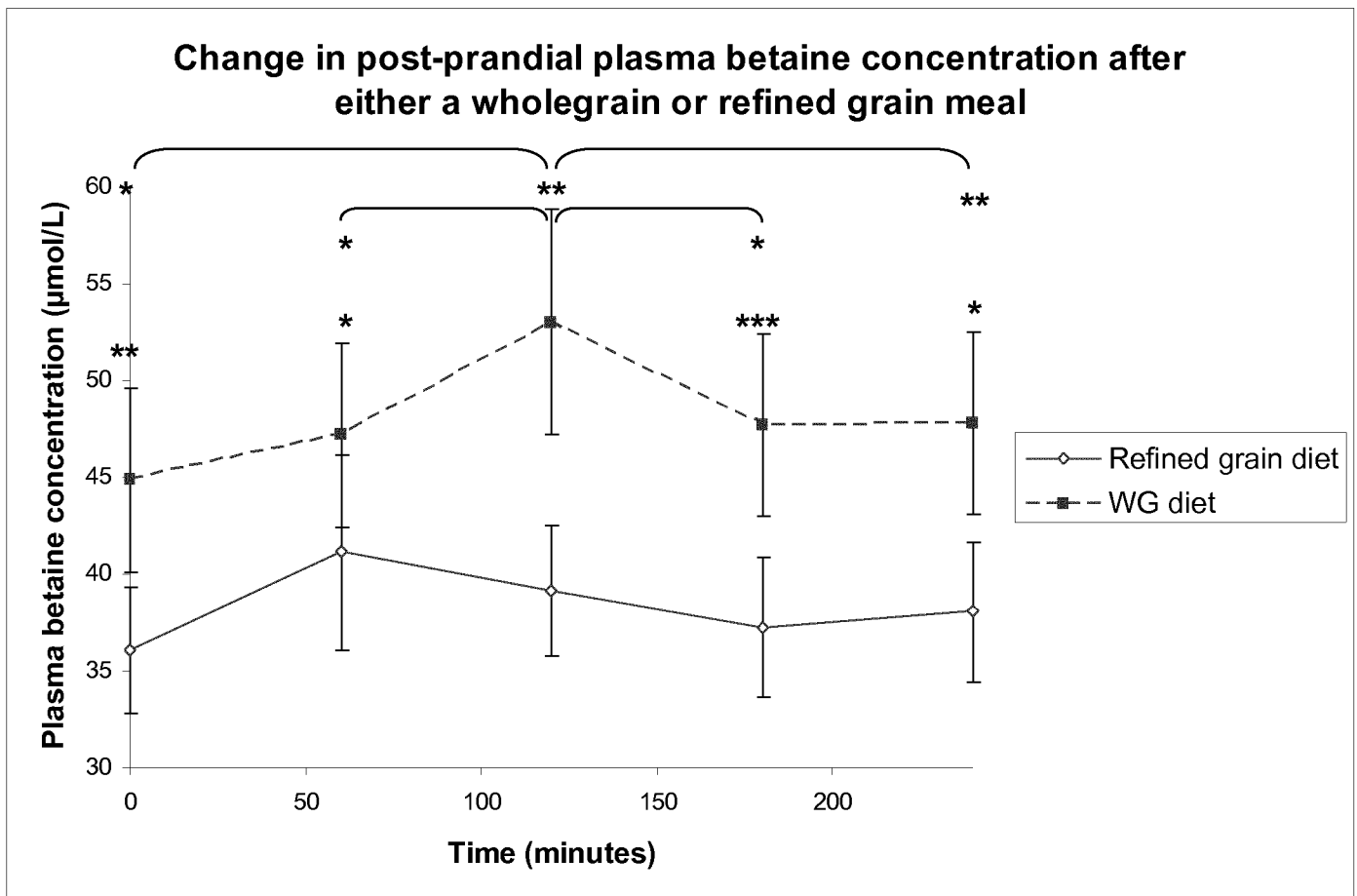


Figure 3:

Image of rat liver from group 4 (Zucker fa/+, healthy control fed with refined wheat diet) showing periportal fatty change, mainly microvacuolar, grade 2.

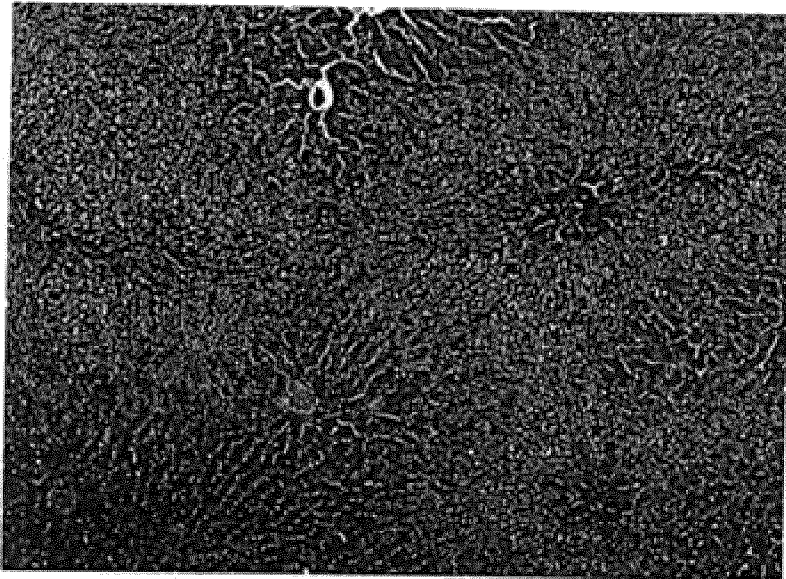


Figure 4:

Image of a section of rat liver from group 1 (Zucker fa/fa rat fed with a refined wheat diet), showing diffuse fat distribution with micro and macrovacuolar (grade 4) and centrilobular hypertrophy (grade 3).

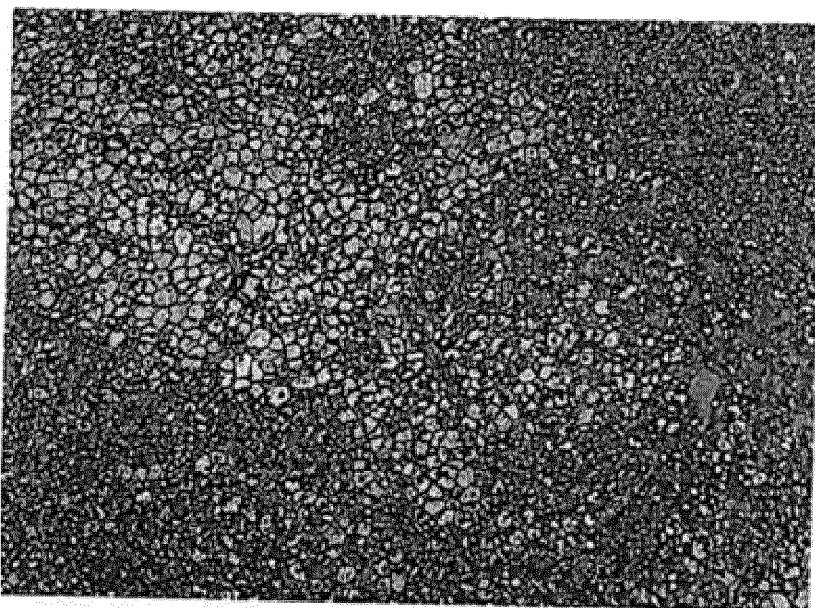


Figure 5:

Image of a section of rat liver from group 3 (Zucker fa/fa fed with a refined wheat diet with addition of betaine) showing diffuse fat distribution with micro and macrovacuolar (grade 4) and centrilobular hypertrophy (grade 3).

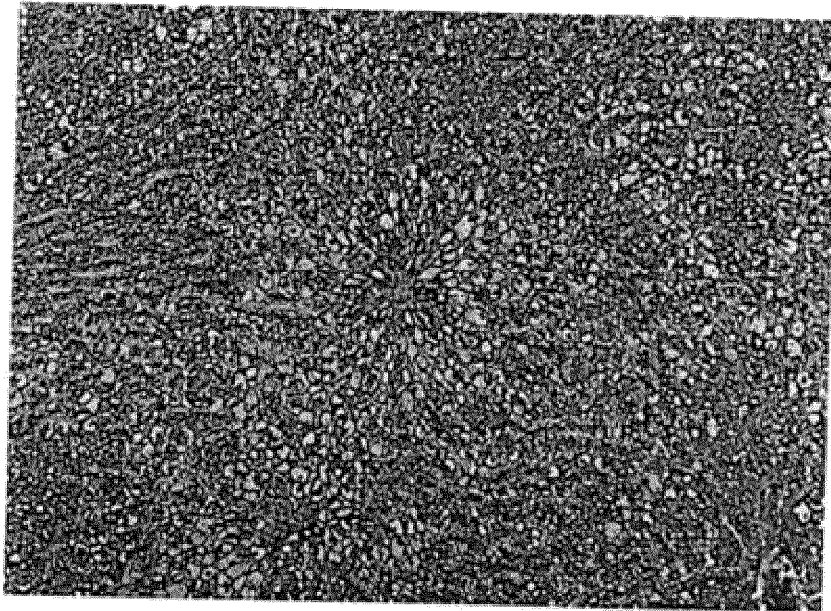
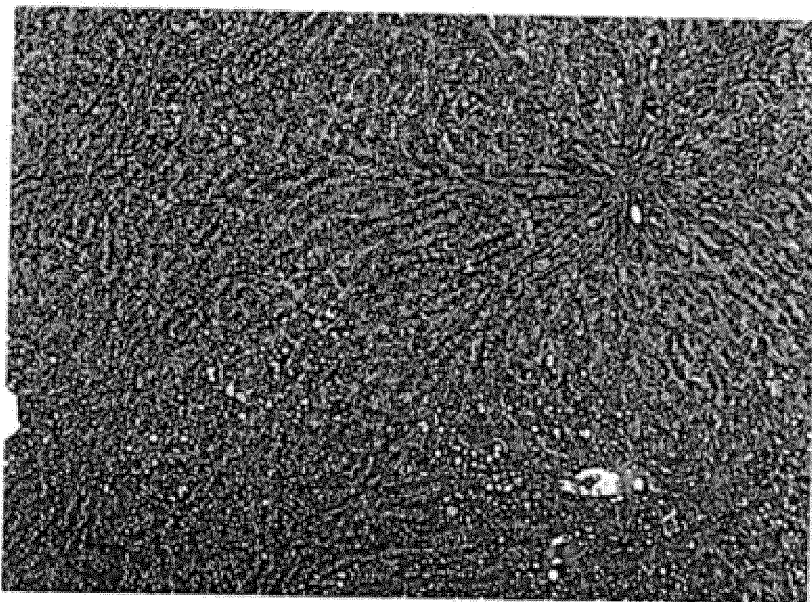


Figure 6:

Image of a section of rat liver from group 2 (Zucker fa/fa fed with a whole grain wheat diet) showing diffuse fat distribution with micro and macrovacuolar (grade 2) and centrilobular hypertrophy (grade 1).



INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2012/061068

A. CLASSIFICATION OF SUBJECT MATTER

INV. A23L1/10 A23L1/164 A23L1/168 A23L1/30 A61K36/899
A61K36/8998 A23K1/14 A23K1/18

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)

A23L A61K A23K

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, WPI Data, FSTA, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	AZADBAKHT ET AL.: "The dietary approaches to stop hypertension eating plan effects C-reactive protein, coagulation abnormalities, and hepatic function tests among type-2 diabetic patients", THE JOURNAL OF NUTRITION, vol. 141, 27 April 2011 (2011-04-27), pages 1083-1088, XP002660434, abstract ----- -/--	1,2,7,8, 11,12,14

☒ Further documents are listed in the continuation of Box C.

☒ 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

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"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

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Date of the actual completion of the international search

5 October 2012

Date of mailing of the international search report

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INTERNATIONAL SEARCH REPORT

International application No

PCT/EP2012/061068

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
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A	<p>anonymous: "What counts as a serving", 2011, XP002684585, Retrieved from the Internet: URL:http://www.wholegrainscouncil.org/whole-grains-101/what-counts-as-a-serving [retrieved on 2012-10-04] the whole document -----</p>	1-14
A	<p>Burros: "'Whole Grain' Labels becoming a little more transparent", 19 April 2006 (2006-04-19), XP002684586, Retrieved from the Internet: URL:http://www.nytimes.com/2006/04/19/dining/19well.html [retrieved on 2012-10-04] the whole document -----</p>	1-14
A	<p>US 2010/189875 A1 (FINOCCHIARO EUGENE TERRY [US]) 29 July 2010 (2010-07-29) the whole document -----</p>	1-14
A	<p>KIM ET AL.: "Saengshik, a formulated health food, prevents liver damage in CC14-induced mice and increases antioxidant activity in elderly women", JOURNAL OF MEDICINAL FOOD, vol. 11, no. 2, 2008, pages 323-330, XP002660431, the whole document -----</p>	1-14
A	<p>KIM ET AL.: "Superfine Saengshik improves liver protection effect compared with fine Saengshik in an animal model", JOURNAL OF FOOD SCIENCE, vol. 74, no. 2, 2009, pages H59-H69, XP002660432, the whole document -----</p>	1-14
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INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2012/061068

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
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A	<p>TANG ET AL.: "Oats supplementation prevents alcohol-induced gut leakiness in rats by preventing alcohol-induced oxidative tissue damage", THE JOURNAL OF PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS, vol. 329, no. 3, 2009, pages 952-958, XP002660435, the whole document</p> <p>-----</p>	1-14
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A	<p>DATABASE WPI Week 200903 Thomson Scientific, London, GB; AN 2009-A67402 XP002660437, & KR 2008 0067085 A (MIN K) 18 July 2008 (2008-07-18) abstract</p> <p>-----</p>	1-14
A	<p>HALLMANS, GÖRAN ET AL: "Tissue concentrations of zinc and iron in rats fed whole grains wheat bread or endosperm wheat bread supplemented with iron", TRACE ELEMENT - ANALYTICAL CHEMISTRY IN MEDECINE AND BIOLOGY, vol. 2, 1983, pages 61-74, XP002660472, the whole document</p> <p>-----</p>	1-14
A	<p>US 2002/119181 A1 (MUSZYNSKA JULIA [US]) 29 August 2002 (2002-08-29) cited in the application paragraph [0022]; claims 1,4,7; figure 16</p> <p>-----</p>	1-14
A	<p>US 2005/123628 A1 (ZABRECKY GEORGE [US]) 9 June 2005 (2005-06-09) cited in the application the whole document</p> <p>-----</p>	1-14
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INTERNATIONAL SEARCH REPORT

International application No

PCT/EP2012/061068

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
	<p>& US 7 078 064 B2 (ZABRECKY GEORGE [US]) 18 July 2006 (2006-07-18) cited in the application -----</p>	

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/EP2012/061068

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