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- (54) **Title:** HCG - NEWER TREATMENT MODALITY FOR TYPE 2 DIABETES MELLITUS (T2DM)

(57) **Abstract:** The present invention discloses a method of treating Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications using hCG which could either be used singly or in combination with anti -diabetic agent(s). The present invention further disclose a pharmaceutical combination of hCG with one or more anti diabetic agents thereof for treating Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications.

**"hCG - Newer Treatment modality for type 2 diabetes mellitus (T2DM)"****The Field Of Invention**

The present invention relates to method of treating Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications using hCG which could either be used singly or in combination with anti -diabetic agent(s). The present invention further relate to a pharmaceutical combination of hCG with one or more anti diabetic agents thereof for treating Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications.

**Background of the Invention**

Type 2 diabetes mellitus (T2DM) is one of the most common forms of chronic disease globally and few societies or ethnic groups are spared. It accounts for about 85% of cases of diabetes in Caucasians and virtually all in certain non - Caucasian ethnic groups. In 2010, it was estimated that 285 million people worldwide have diabetes, of whom 80% live in less developed countries and areas. Among those aged 20 - 79 years, about 6.6% have diabetes globally. The highest number of people with diabetes or the hub of Diabetes would be India by the year 2015.

The number of people with diabetes is expected to reach 438 million by 2030, an increase of 54% compared to predicted figures for 2010. The largest increases will be in countries with rapidly growing economies, such as India and China. With the increasing consumption of high - energy food, increasing adoption of sedentary lifestyles and urbanization, increasing numbers of individuals develop T2DM, and the age of diagnosis is decreasing.

Type 2 diabetes (T2DM) is a heterogeneous disorder, phenotypically, genotypically and pathogenetically. Approximately 10% of patients have a late - onset form of autoimmune diabetes which may represent a hybrid of type 1 and type 2 diabetes; up to another 5% of patients have one of the autosomally dominant inherited forms of maturity - onset

diabetes of youth (MODY); another 1% may have rare genetic mutations involving insulin receptors and elements of the insulin signaling pathway. The remaining 85% of patients have "garden variety" T2DM.

Insulin resistance both precedes and predicts type 2 diabetes mellitus (T2DM). The insulin resistance preceding T2DM is commonly referred to as the metabolic syndrome. Insulin resistance, even when present in individuals without diabetes, is characterized not only by abnormalities in glucose homeostasis, but also by defects in insulin regulation of lipid and uric acid metabolism, vascular function, hemostasis and coagulation.

Hypogonadism is common in clinical practice but is frequently unrecognized and underdiagnosed. There are many pitfalls in making the diagnosis of male hypogonadism. In most studies, it is based only on total testosterone values. Sex hormone-binding globulin (SHBG) levels decrease in patients with obesity and increase with aging. Patients with type 2 diabetes mellitus (T2DM) have even lower SHBG levels in comparison with age- and body mass index (BMI)-matched subjects without diabetes.

Hence, total testosterone levels may not be accurate in the obese diabetic population. Calculated free testosterone (cFT) with use of total testosterone, SHBG, and albumin has been shown to correlate well with free testosterone (FT) measured by equilibrium dialysis and is very useful in patients with altered SHBG levels. Dhindsa et al demonstrated that 33% of men with T2DM had significantly lower levels of FT measured by equilibrium dialysis, the "gold standard" assay for FT estimation.

Kapoor et al, showed that 17% of men with diabetes had overt hypogonadism with total testosterone values less than 2.3 ng/mL, and a further 25% had symptoms of hypogonadism associated with total testosterone levels between 2.3 and 3.5 ng/mL.

A clear demarcation of whether there is a direct cause-effect relationship between testosterone deficiency needs to be evaluated and / defined. However, restoring endogenous gonadal hormone levels have demonstrated an improvement of insulin sensitivity and the components of metabolic syndrome (MetS) [measures of glucose

abnormalities and insulin resistance, obesity with the focus on waist circumference, dyslipidemia, and hypertension].

Traditionally oral antihyperglycemics, has been the mainstay of glycaemic control and preserving beta-cell function. Although understood, testosterone deficiency has a greater role to play on the components of MetS, it would call for a revision of the current treatment algorithms.

Studies published have demonstrated the beneficial effects of subcutaneous administration of human chorionic gonadotropin improved body composition and lipid profile, however insulin sensitivity, endothelial function and blood pressure had not worsened. Current research confirms that, physiological levels of estradiol with a stable androgen pharmacokinetic profile, are critical in improvement of insulin sensitivity.

A considerable body of evidence exists suggesting a link among reduced testosterone plasma levels, Type 2 Diabetes (T2DM), and insulin resistance (IR). Hypogonadal men are at higher risk for T2DM. Testosterone (T) plays a crucial role in maintaining metabolic homeostasis; thus, this hormone may play a vital role in maintaining glycemic control.

Published studies published in peer reviewed journals, reported a positive correlation between serum Testosterone levels and insulin sensitivity, independent of Sex Hormone Binding Globulin(SHBG), in a large cohort of men.

Studies conducted by S.K.Sigh et al.,revealed that males with MetS with or without diabetes had lower serum testosterone than their age matched healthy subjects. Hypogonadism was also common in subjects with MetS. Testosterone therapy to correct hypoandrogenemia improved insulin sensitivity and other biochemical parameters. This emphasizes that hypogonadism in all men with MetS or MetS in all men with hypogonadism needs clinical evaluation and subsequent management.

Further, published observations have indicated that Hypothalamic - Pituitary -Gonadal Axis or low serum testosterone has a clinical relationship with the development of insulin

resistance or may be consequence there upon. Whatever is the situation, attendant hyperandrogenemia may unfavorably influence other parameter of MetS. Restoration of serum T (testosterone) level to near normal ameliorated some of the changes favoring its casual role in development of insulin resistance. Replacement with Testosterone has a potential role in correcting the abnormality of MetS particularly in hypogonadal males.

In various other published studies, exogenous synthetic testosterone has been shown to dramatically suppress gonadotropin release when administered at supra physiological as well as physiological doses. Administration of Testosterone alone has been shown to reduce sperm production in the majority of men to levels acceptable for contraception. However in a study published by Covellio, Matsumo et al. the use of human chorionic gonadotropin to selectively replace Lutenising Hormone (LH) activity within the testis would allow for manipulation of the intratesticular androgenic environment, which could play a vital role in the treatment of Type 2 Diabetes(T2DM).

In view of the aforementioned conventional treatments for the treatment of Type 2 Diabetes(T2DM), the present invention aims at meeting the desired clinical outcomes, in Type 2 Diabetes such as improvement of insulin sensitivity, leptin levels (the markers of adiposity) through an indirect means of improving the serum androgen profile using human chorionic gonadotropin (hCG).

Thus, while testosterone deficiency can be addressed with physiological doses of human chorionic gonadotropin stimulation, it is an object of the invention to provide an effective treatment of Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications using lower dose of hCG which could either be used singly or complementing the main-stay therapy.

Another object of the invention is to provide a pharmaceutical combination of hCG either alone or in combination with other anti-diabetic agents.

**Summary of the invention:**

In accordance with the objectives, one aspect of the present invention disclose a novel treatment modality for type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications using human chorionic gonadotropin (hCG), either alone or in combination with one or more anti-diabetic agent(s).

In an aspect, the method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering hCG preferably in parental form.

In another aspect, the method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering hCG in combination with one or more anti-diabetic agent(s) either sequentially, simultaneously, concurrently, or alternatively.

In another aspect, the anti -diabetic agents are preferably administered orally.

In an aspect, the present invention disclose method for treating physiological testosterone deficient related complications in Type 2 Diabetes Mellitus (T2DM) with associated co-morbid conditions of Metabolic syndrome (Met S).

In another aspect, the present invention provides pharmaceutical combination comprising human chorionic gonadotropin (hCG) with one or more anti-diabetic agent(s) as combination drugs which works synergistically against type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications.

In yet another aspect, the present invention disclose the use of hCG either alone or in

combination with one or more anti-diabetic agent(s) type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications.

**Description of figures:**

**Fig 1** depicts glucose level before diabetes

**Fig 2:** depicts glucose level after diabetes

**Fig 3:** depicts glucose level after treatment

**Fig 4:** depicts comparative levels of glucose in all the Group II, III.

**Fig 5:** depicts comparative levels of glucose in all the Group II, III.

**Fig 6:** depicts initial glucose levels

**Fig 7:** depicts glucose level after 2days diabetes

**Fig 8:** depicts glucose level after 15 days diabetes

**Fig 9:** depicts glucose level after 30days diabetes

**Fig 10:** depicts glucose level after 45 days diabetes

**Fig 11:** depicts glucose level after 60 days diabetes

**Fig 12:** depicts synergistic reduction of glucose levels

**Detailed description of invention**

The invention will now be described in detail in connection with certain preferred and optional embodiments, so that various aspects thereof may be more fully understood and appreciated.

The phrase 'effective amount' means and includes the amount required to treat/alleviate the severity of symptoms associated with the ailments as decided by the persons of ordinary skill in the art.

The phrase 'related metabolic conditions' used herein mean and refer to co -morbid conditions of metabolic syndrome (Met S), insulin sensitivity and physiological testosterone deficient related complications associated with Type 2 diabetes.

The phrase 'after diabetes' used herein mean and refer to the diabetes induced in the animal after the administration of streptozotocin(stz).

The present invention with a view to provide a new treatment modality for type 2 diabetes mellitus (T2DM) also aims at addressing the accompanying co-morbid conditions of metabolic syndrome (Met S), insulin sensitivity and physiological testosterone deficient related complications.

Accordingly, in an embodiment, the present invention is directed to method for treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprising administering to a subject in need thereof an effective amount of human chorionic gonadotropin (hCG) either alone or in combination with one or more anti-diabetic agent(s).

In an embodiment, the method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering hCG in parental form.

In another embodiment, the method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering anti-diabetic agents orally.

In yet another embodiment, the present invention relate to method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprising administering to a subject in need thereof an effective amount of human chorionic gonadotropin (hCG) in combination with one or more anti-diabetic agent(s) either sequentially, simultaneously, concurrently, or alternatively.

The anti-diabetic agent(s) is/are selected from biguanide group such as metformin (met) and /or its organic, inorganic salts; sulfonylureas like tolazamide, glipizide, glimepiride, glibenclamide, gliclazide tolbutamide; Alpha-glucosidase inhibitors like acarbose; members of the glitazone such as rosiglitazone, pioglitazone, a combination of pioglitazone and biguanides; miglitol-a glucosidase inhibitor, postprandial glucose inhibitors such as repaglinide, nateglinide; DPP -4 inhibitors like sitagliptin ( or in combination form), vildagliptin(or in combination form), dutogliptin ,saxagliptin, linagliptin, gemigliptin and alogliptin.

In a preferred embodiment, the present invention to method of treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprising administering to a subject in need thereof an effective amount of human chorionic gonadotropin (hCG) in combination with insulin sensitizer metformin, which has a significant clinical bearing on morbidity and mortality, either sequentially, simultaneously, concurrently, or alternatively.

In an embodiment, the present invention provides a pharmaceutical formulation for treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications, the formulation comprising (hCG) along with suitable pharmaceutical diluents, carriers and/or other excipients. The pharmaceutically acceptable diluents, carriers or excipients suitable to formulate a pharmaceutical composition are those which can be included without affecting the activity and stability of hCG.

In another embodiment, the present invention relate to pharmaceutical combination for treating type 2 diabetes mellitus and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications the formulation comprising human chorionic gonadotropin (hCG) with one or more anti-diabetic agent(s) as combination drugs.

Multiple dosing regimens together, along with large doses, leads to patient non-

compliance, potential side effects & danger of overdosing. Further, the quantity of the compound used in pharmaceutical compositions of the present invention will vary depending upon the body weight of the patient and the mode of administration and can be of any effective amount to achieve the desired therapeutic effect. The present invention provides patient-convenient, cost effective pharmaceutical dosage form to improve the quality of treatment.

In an embodiment, the present invention relate to pharmaceutical dosage of hCG in the range of 50 IU - 15000 IU either alone or in combination with one or more anti-diabetic agent(s). The combination of the present invention may be in the instant release or controlled release form. The hCG used in the said formulation is in highly purified form.

In another embodiment, the combination is in the form of a fixed combination dosage form which provides synergistic effect against type 2 diabetes mellitus and related metabolic diseases i.e the formulation comprises hCG, in the range of 50 IU - 15000 IU, preferably 500IU, and metformin either in the instant release or controlled release form.

The dosage for the anti-diabetic agent(s) used as combination drugs in the present invention with hCG is in the concentration in the range of 500-2500mg, that provides the desired bioactivity of the combination of the present invention.

The pharmaceutical formulation of hCG may be a solid form, a liquid suspension or an injectable composition, preferably as injectables .

The injectable human chronic gonadotrophin (hCG) formulation of the present invention is prepared by a trademarked technology known as Lygotech developed by the applicants themselves.

In an embodiment, the antidiabetic agents are administered, preferably orally when used in combination with hCG

In an embodiment, the present invention provides pharmaceutical formulation comprising

the composition of hCG, formulated as unit dosage in the form of a lyophilized solid which can readily dissolve to form a sterile injectable solution for subcutaneous or intramuscular administration.

The typical excipients include sucrose, lactose, sodium chloride, buffering agents. The solution may be prepared by diluting with water for injection prior to use.

The hCG formulation with anti-diabetic agent (s) of the present invention reduces the number of dosage to be taken, since the subject in need requires to take the formulation once or twice weekly for 12-16 weeks.

In an embodiment, the present invention provides hCG dosage of 500IU as injectables, wherein 1 to 3 injections are administered to the subject for 12-16 weeks.

In an embodiment, the present invention relates to controlled release (CR)-hCG formulation of 500 IU - 10000IU as treatment modality for improvement of said diabetic parameters.

In another embodiment, the pharmaceutical formulation of the present invention comprising hCG ranging from 50 IU - 15000 IU, preferably 500IU is used for treating type 2 diabetes mellitus and associated co -morbid conditions of metabolic syndrome (Met S), insulin sensitivity and physiological testosterone deficient related complications.

In another embodiment, the present invention provides the pharmaceutical combination comprising hCG ranging from 50 IU - 15000 IU, preferably 500IU in combination with anti-diabetic agent(s), in the concentration 500-2000mg that is biocompatible, used for treating type 2 diabetes mellitus and associated co -morbid conditions of metabolic syndrome (Met S), insulin sensitivity and physiological testosterone deficient related complications.

The pharmaceutical formulation of the present invention comprising hCG ranging from 50 IU - 15000 IU, preferably 500IU either alone or in combination with one or more anti-

diabetic agents is used for treating physiological testosterone deficient related complications in Type 2 Diabetes Mellitus (T2DM) with associated co-morbid conditions of Metabolic syndrome (Met S).

Thus the treatment intervention is a value addition to the existing treatment algorithm/s for the management of insulin resistance and (Metabolic Syndrome) MetS in Type 2 Diabetes Mellitus (T2DM). The low doses of hCG maintain baseline levels of ITT (Intratesticular Testosterone) in men with gonadotropin withdrawal from exogenous testosterone administration. Selective replacement of Lutenising Hormone (LH) activity with low-dose hCG, as demonstrated in the study below allows the design of titrating dosages in maintaining intratesticular androgens.

The present invention has the following industrial advantages and applications:

- i. hCG helps in improving insulin resistance, a common phenomenon in Type II Diabetic males. The production of testosterone by hCG improves glycaemic parameters and lipid parameters, which have a significant bearing in terms of morbidity and mortality.
- ii. The novel intervention for T2DM is aimed at addressing the accompanying co-morbid conditions of Met S, Insulin sensitivity and physiological testosterone deficient related complications.
- iii. The intervention is aimed at addressing issues of testosterone deficiency in T2DM either as a single intervention or in combination with existing anti hyperglycemics.
- iv. The intervention is available in a dosage form of 50 IU - 15000 IU hCG.
- v. The intervention plays a physiological role in beta cell function and improvement of endothelial cell function.
- vi. The intervention or treatment strategy has a broad scope in the management of co-morbid conditions associated with T2DM in the ever expanding global diabetic population.

The present invention is further illustrated with the study given below:

**Anti-diabetic activity and related metabolic disorder.**

The pharmaceutical formulation comprising of hCG either alone or in combination with Metformin are tested for its anti-diabetic activity inclusive of blood glucose level, HbA1c (glycosylated haemoglobin-pre and post), lipid profile (cholesterol, HDL, LDL, HDL/LDL ratio), insulin levels, hormones (testosterone and Estradiol) determining the acute and chronic plasma glucose lowering activity of test compound in Male Wistar rats.

**The Animal Model**

Male Wistar Rats weighing 300 - 350 gms were chosen for the study. The number of animals in each group were n=12, to compensate for mortality during the 60 day period.

**Inducing Diabetes**

The Streptozotocin model was chosen as the signs and symptoms exhibited by the animals is similar to Type 2 Diabetes Mellitus, unlike Alloxan which causes complete pancreatic damage, which is similar to Type I Diabetes. The pancreatic dysfunction, was induced using Streptozotocin (STZ) 40 - 60 mg/kg body-weight [reconstituted in citrate buffer].

**Grouping of Experimental Animals**

1. Group I (Control) - Streptozotocin [STZ alone]
2. Group II - 500 IU Highly Purified hCG Injections
3. Group III - 500 IU Highly Purified hCG Injections + Metformin [Biguanide]

**Study Procedure**

The animals once induced with diabetes, exhibited symptoms of polyuria, restlessness and fatigue and the animals were observed for 48 hours and fed a high protein diet.

1. **Group I - Streptozotocin Group:** STZ group was euthanized using carbon dioxide as described in standard laboratory manuals. Blood samples for Blood Glucose Levels, HbA1c, Lipids, Testosterone, Estradiol (E2), and Insulin levels were estimated using ELISA techniques.

2. **Group II - 500 IU Highly Purified hCG Injection Group:** Standard anti-diabetic regimen was initiated with a Sulphonylurea [Gliclazide] and Alpha-Glucosidase Inhibitor [Acarbose] per-orally along with a weekly sub-cutaneous / Intramuscular injection of highly purified hCG [500 IU] which was administered after calibrating the doses as per body-weight.
  
3. **Group III - 500 IU Highly Purified hCG + Biaguanide [Metformin] Group:** Standard anti-diabetic regimen was initiated with a Sulphonylurea [Gliclazide] and Alpha-Glucosidase Inhibitor [Acarbose] per-orally along with a weekly sub-cutaneous / Intramuscular injection of 500 IU Highly Purified hCG which was administered after calibrating the doses as per body-weight.

All the animals were weighed on regular basis to check for either weight gain, or weight loss during the entire study period.

#### **Blood Collection Methodology**

Prior to inducing Diabetes, blood samples were collected by the orbital method in all the group of animals which were mildly anesthetized, prior to collection. Eye drops were instilled to prevent any infection.

All the animals, were euthanized using the Carbon Dioxide Chamber [CO<sub>2</sub>], and blood was collected using the cardiac puncture and both plasma and serum were collected in sufficient quantities for evaluation of the biochemical parameters and was stored at 2-8 degree centigrade, till the tests were performed.

#### **Bio-Chemical Investigations**

Blood Glucose Levels were estimated using an Accucheck [Pre - & Post-Treatment]

HbA1c [Glycosylated Haemoglobin] - Pre- & Post

Lipid Profile [Cholesterol, HDL,LDL,HDL/LDL Ratio]

Insulin Levels

Hormones - Testosterone and Estradiol

All the bio-chemical parameters were estimated using ELISA kits in a NABL Laboratory. Blood sampling was done as per the procedure described above.

**Results:**

**Biochemical parameters: Table 1**

S.no	Parameters	Control	standard(stz )	metformi n	hCG	met+hCG
1	Cholesterol	57.25±14.03	79±12.25	58.28±9.42	46±7.95	56.2±10.9**
2	HDLC	16.87±5.13	18.37±4.5	19.85±5.95	16.28±3.9**	16.2±5.49*
3	LDLC	20.75±9.25	17.5±5.95	23.14±8.19	16.28±3.19*	20.8±5.4**
4	HbA1C	4.02±0.13	3.97±0.09	3.97±0.09	3.99±0.11*	4±0.1**
5	Testosterone	112.95±36.24	76.91±28.04	57.15±43.47	42.66±3.01	43.51±11.55*
6	Estradiol	9.2±0.02	9.8±0.05	9.7±0.02	9.85±0.03	9.3±0.01*
7	Insulin	0.45±0.05	0.05±0.05	0.3±0.05	0.43±0.05	0.60±0.05**

\*Significant compared with control

**Table 2 :** Comparative table of glucose level after treatment with hcg and hCG + metformin with controls.

Sr. no.	Glucose levels	Normal control	Stz control	Metformin	hCG	Met+hCG
1	Before diabetes	86.5±<3.86	105.7±<2.97	86.9±<3.36	97.3±<3.38	106.8±4.93
2	After inducing diabetes	101.1±<3.56	553.6±<17.6	473.8±<22.9	544.5±<11.59	479.9±<29.3
3	After treatment	98.62±<3.4	583±<18.3	308±<17.97	364±<2.09	288.12±<25.56

**Table 3:** Comparative table of glucose level after treatment with hcg and hCG + metformin with controls.

	control	stz control	Metformin	hCG	met+hCG
Before diabetes	86.5	105.7	86.9	97.3	106.8
After diabetes	101.1	553.6	473.8	544.5	479.9
After treatment			388	384	388.12

**Table 4:** Final Values-Body Weights-! Day-Before Diabetes

marking	control	stz control	metformin	hCG	.met+hCG
Initial	386	357	317	336	380
SE[standard error]	13.6	9.9	7.31	12.04	4.71
		2 DAY AFTER DIABETES			
marking	control	stz control	metformin	hCG	met+hCG
2 days after diabetes	369	310	282	297	350
SE	12.6	12.11	4.9	9.77	4.97
			15 DAY		
marking	control	stz control	metformin	hCG	met+hCG
15day			291.5	265	293.88
SE			22.64	14.54	14.11
			30 DAY		
marking	control	stz control	metfor	hCG	met+hCG
30 day			285.5	244.37	271.75
SE			19.74	18.33	16.91
			45 DAY		
marking	control	stz control	metformin	hCG	met+hCG
45 day			297.71	268.75	294
SE			16.94	15.61	15.99
			60 DAY		
marking	control	stz control	metformin	hCG	met+hCG
60 day			267.75	260.5	273.85
SE			16.51	16.26	13.71

Table 5: Comparative table of glucose level after treatment with hcg and hCG + metformin with controls.

marking	control	stz control	metformin	hCG	metformin+hCG
Initial	386	357	317	336	380
2 days after diabetes	369	310	282	297	350
15day			291.5	265	293.88
30 day			285.5	244.37	271.75
45 day			297.71	268.75	294
60 day			267.75	260.5	273.85

From the above tables, it is clear that the Glucose levels which had shown elevated levels initially similar to human T2DM, were well controlled, by treating animals with standard anti-diabetic regimens along with hCG and Metformin [oral] +hCG [injectable].

The reason for variation in glucoses levels appeared in animal models is due to handling of the animals that are under stress. However, in humans as the stress levels are lower; the values with hCG and hCG +Metformin would be higher than Metformin alone.

As regards the weight of the animals, the animals that had lost weight initially a manifestation of T2DM, administration of hCG and hCG along with metformin, did not allow further reduction of weight nor did the animals become obese, once treatment was initiated. Therefore, the administration of hCG either alone or in combination with anti-diabetic drugs proves the synergistic and prolonged effect over the conventional treatments.

**We claim,**

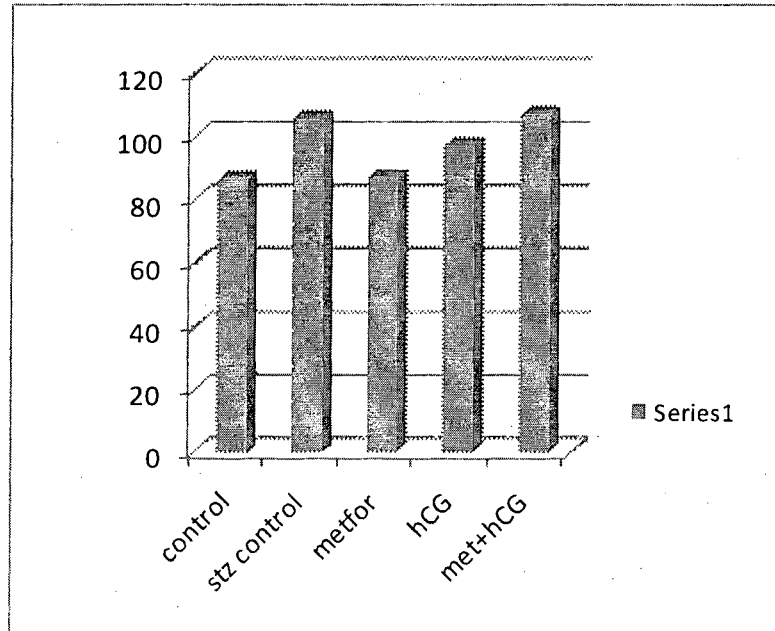
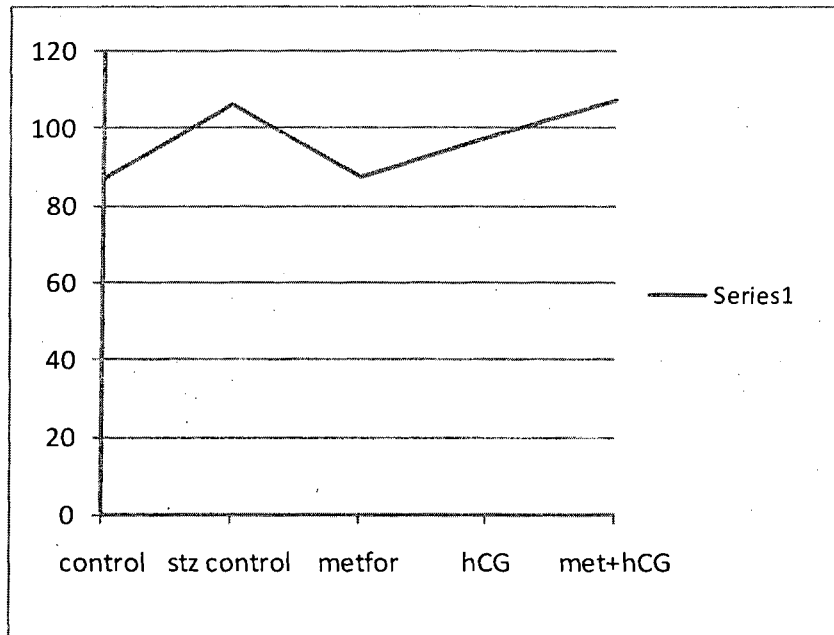
1. A method of treating Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprising administering to a subject in need thereof an effective amount of Human chorionic gonadotropin (hCG) either alone or in combination with one or more anti-diabetic agent(s).
2. The method of treating as claimed in claim 1, wherein said anti-diabetic agent(s) are selected from the group comprising biguanides such as metformin and /or its organic, inorganic salts; sulfonylureas selected from tolazamide, glipizide, glimepiride, glibenclamide, gliclazide tolbutamide; Alpha-glucosidase inhibitors like acarbose; members of the glitazone such as rosiglitazone, pioglitazone, a combination of pioglitazone and biguanides; miglitol-a glucosidase inhibitor, postprandial glucose inhibitors such as repaglinide, nateglinide; DPP -4 inhibitors like sitagliptin ( or in combination form), vildagliptin(or in combination form), dutogliptin ,saxagliptin, linagliptin, gemigliptin and alogliptin
3. The method of treating as claimed in claim 1, wherein said anti-diabetic agent is metformin.
4. The method of treating as claimed in claim 1, wherein treatment of Type 2 Diabetes Mellitus (T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering to a subject in need thereof an effective amount of Human chorionic gonadotropin (hCG) parentally.
5. The method of treating as claimed in claim 1, wherein treatment of Type 2 Diabetes Mellitus (T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications comprises administering to a subject in need thereof an effective amount of Human chorionic gonadotropin (hCG) parentally in

combination with anti-diabetic agents orally, either sequentially, concurrently, simultaneously or alternatively.

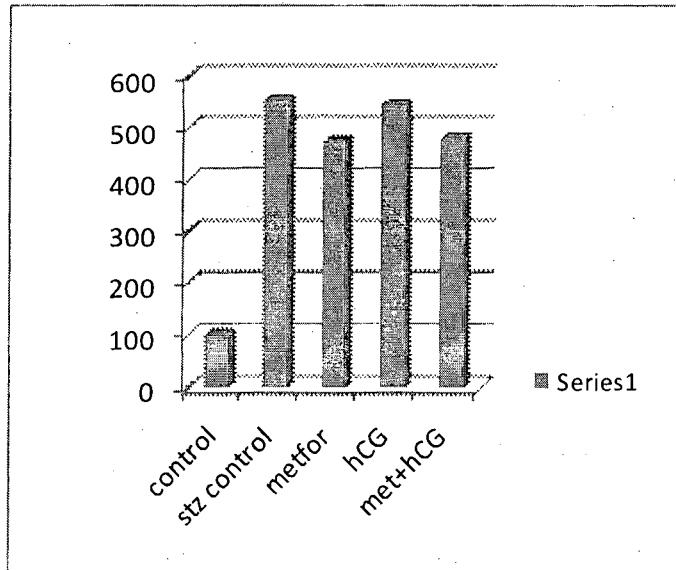
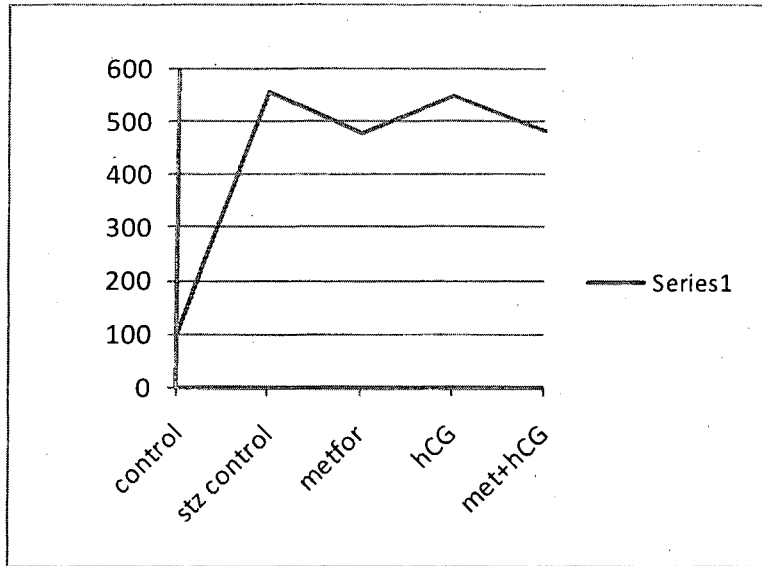
6. The method of treating as claimed in claim 1, wherein dosage of Human chorionic gonadotropin (hCG) is in the range of 50 IU - 15000 IU either alone or in combination with metformin.
7. The method of treating as claimed in claim 5, wherein dosage of hCG is 500 IU either alone or in combination with metformin.
8. A pharmaceutical formulation for the treatment of Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications, wherein said formulation comprises Human chorionic gonadotropin (hCG) along with suitable pharmaceutical diluents and carriers.
9. The pharmaceutical formulation as claimed in claim 8, wherein the said formulation is administered subcutaneously or intramuscularly.
10. A pharmaceutical combination for the treatment of Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), Insulin sensitivity and physiological testosterone deficient related complications, wherein said combination comprises Human chorionic gonadotropin (hCG) in combination with one or more anti-diabetic agent(s) excipients.
11. The pharmaceutical combination as claimed in claim 10, wherein said anti-diabetic agent(s) are selected from the group comprising biguanides such as metformin and /or its organic, inorganic salts; sulfonylureas selected from tolazamide, glipizide, glimepiride, glibenclamide, gliclazide tolbutamide; Alpha-glucosidase inhibitors like acarbose; members of the glitazone such as rosiglitazone, pioglitazone, a combination of pioglitazone and biguanides; miglitol-a glucosidase inhibitor, postprandial glucose inhibitors such as

- repaglinide, nateglinide; DPP -4 inhibitors like sitagliptin ( or in combination form), vildagliptin (or in combination form), dutogliptin ,saxagliptin, linagliptin, gemigliptin and alogliptin.
12. The pharmaceutical formulation as claimed in claim 8, wherein dosage of Human chorionic gonadotropin (hCG) is in the range of 50 IU - 15000 IU.
  13. The pharmaceutical formulation as claimed in claim 12, wherein the dosage of hCG is 500 IU.
  14. The pharmaceutical combination as claimed in claim 10, wherein the dosage of hCG is 500 IU in combination with metformin.
  15. Use of pharmaceutical formulation comprising hCG in highly purified form in association with one or more anti diabetic agent(s) for treatment of Type 2 Diabetes(T2DM) and associated co-morbid conditions of Metabolic syndrome (Met S), insulin sensitivity and physiological testosterone deficient related complications.

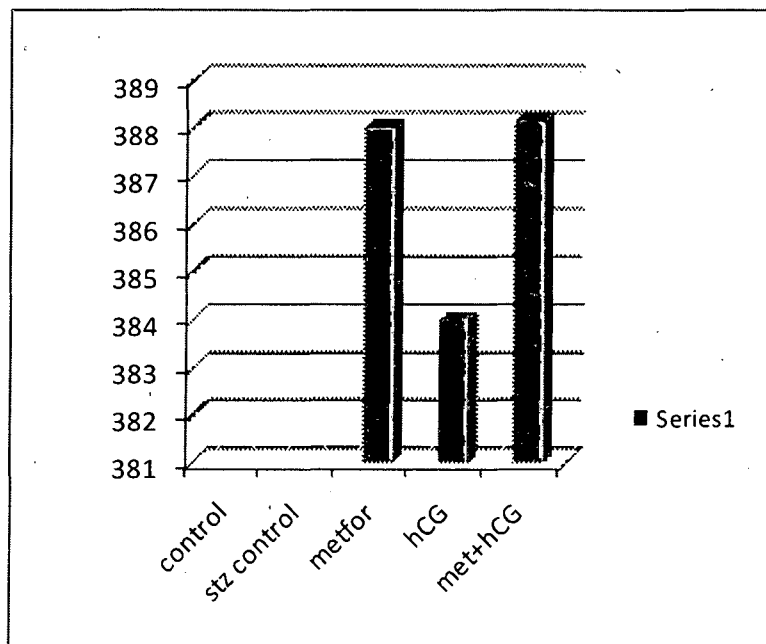
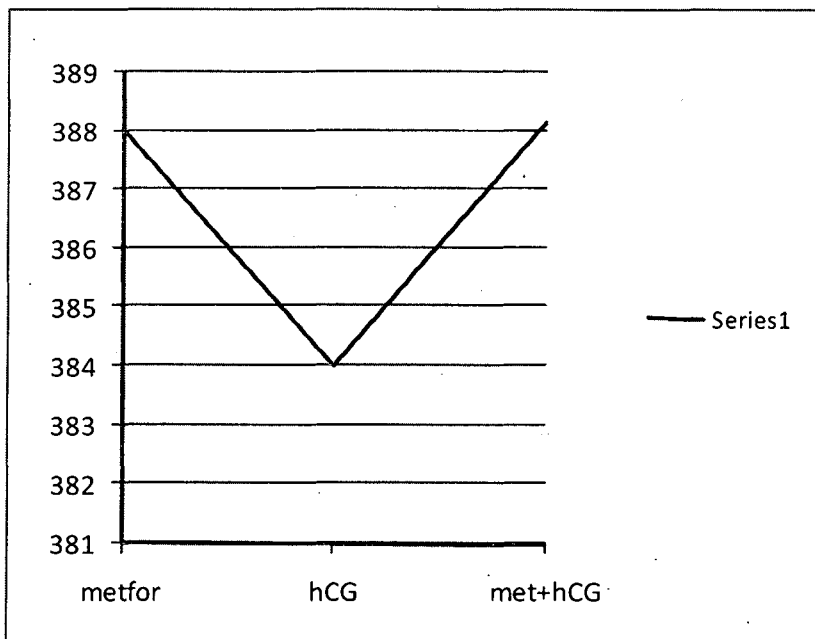
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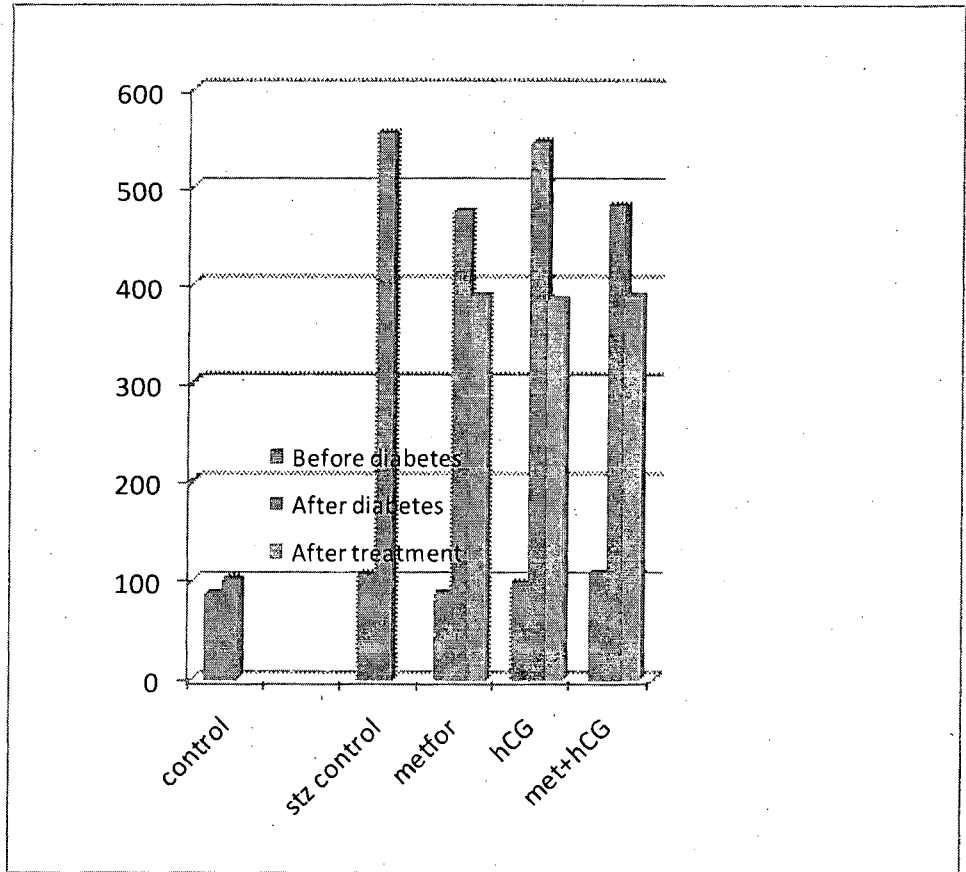
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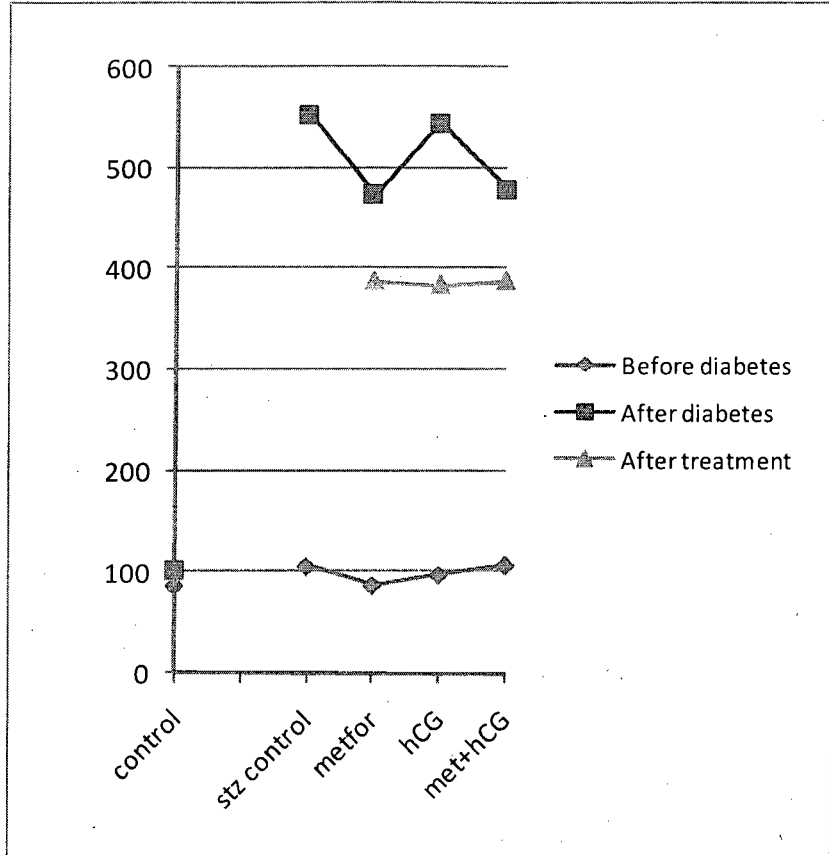
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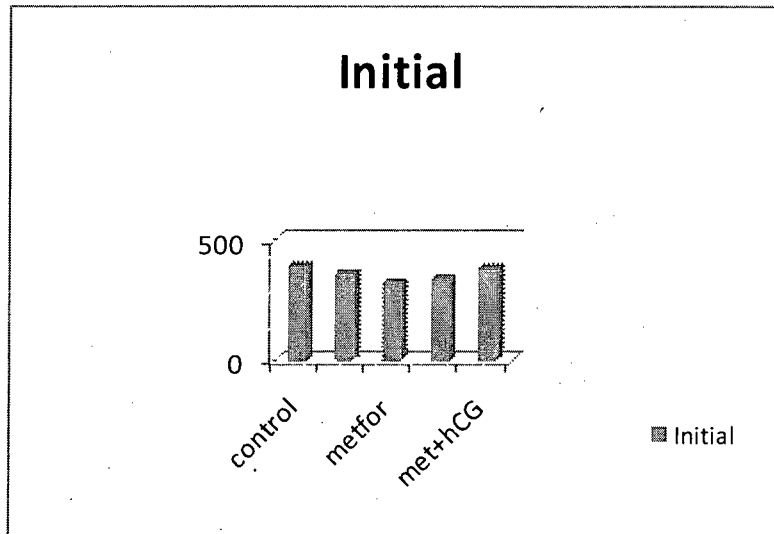
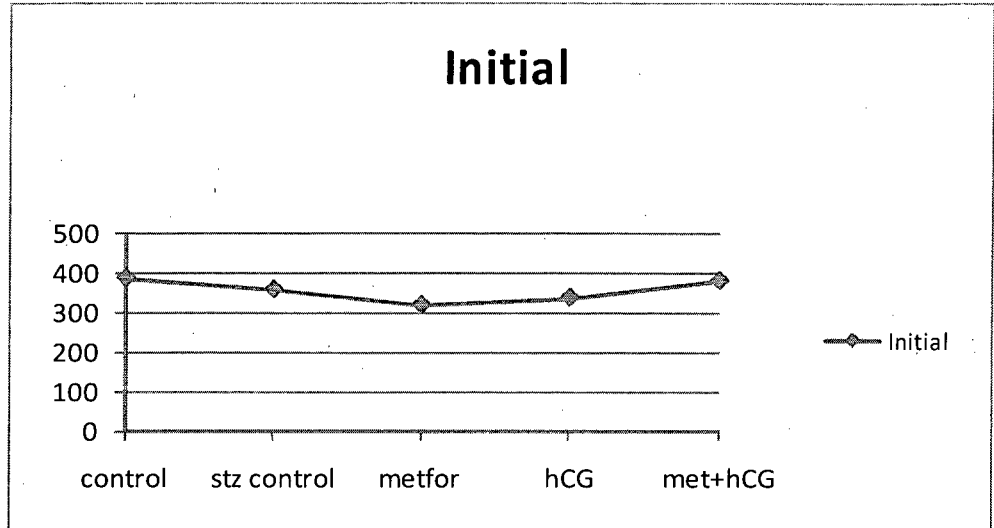
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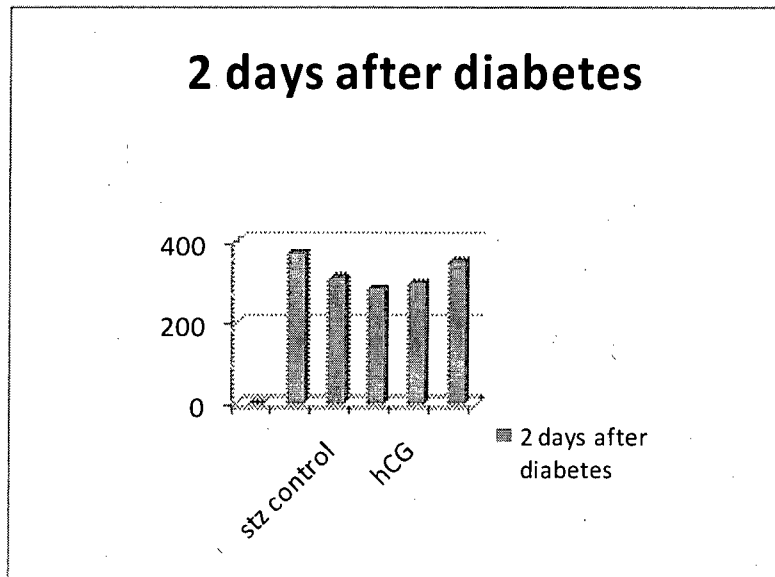
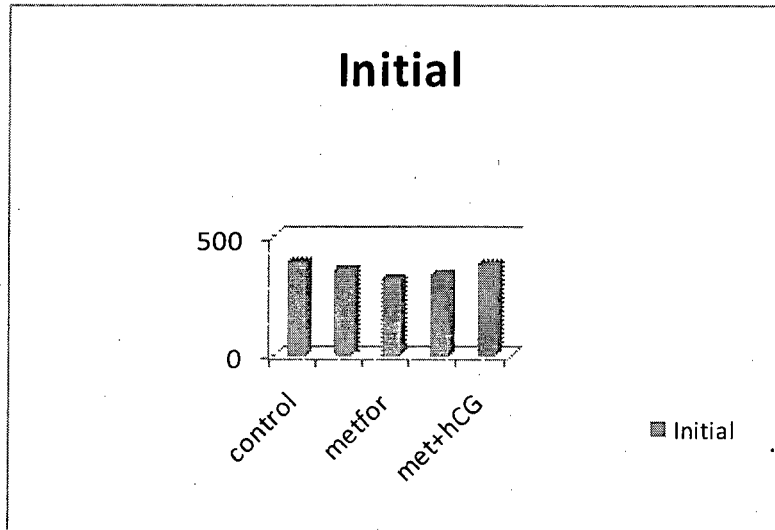
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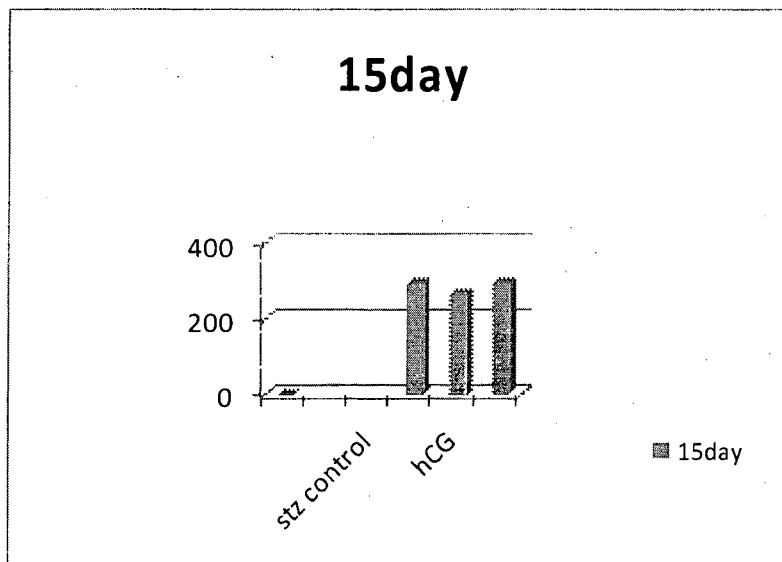
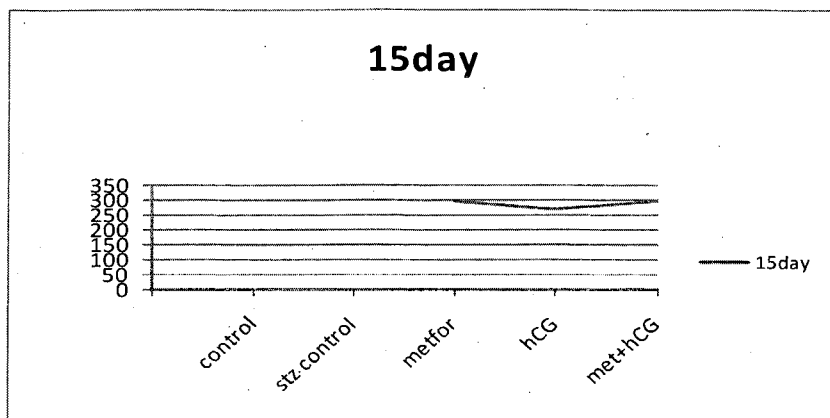
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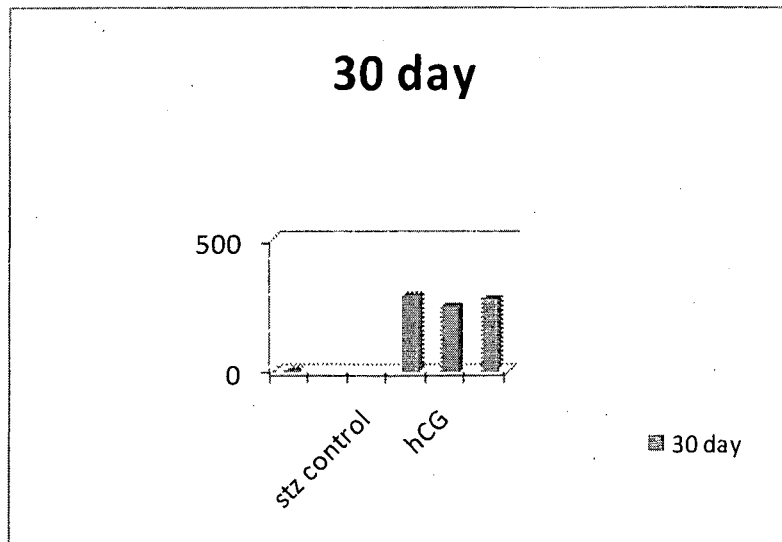
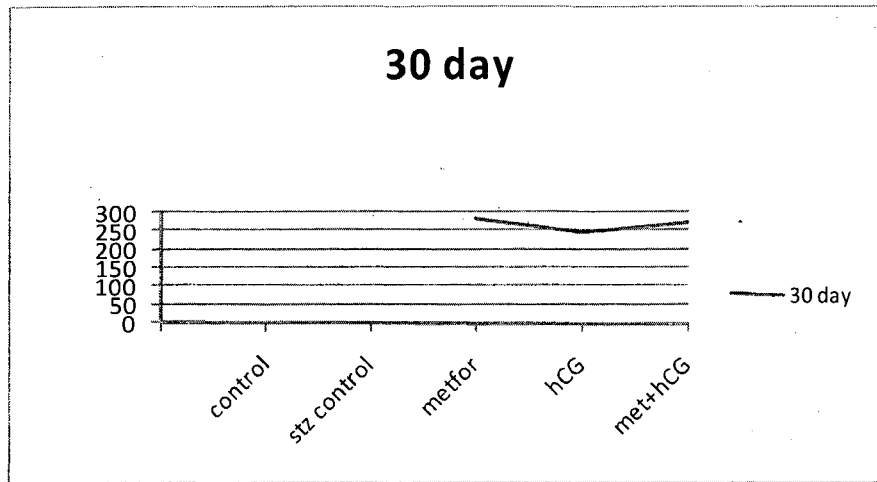
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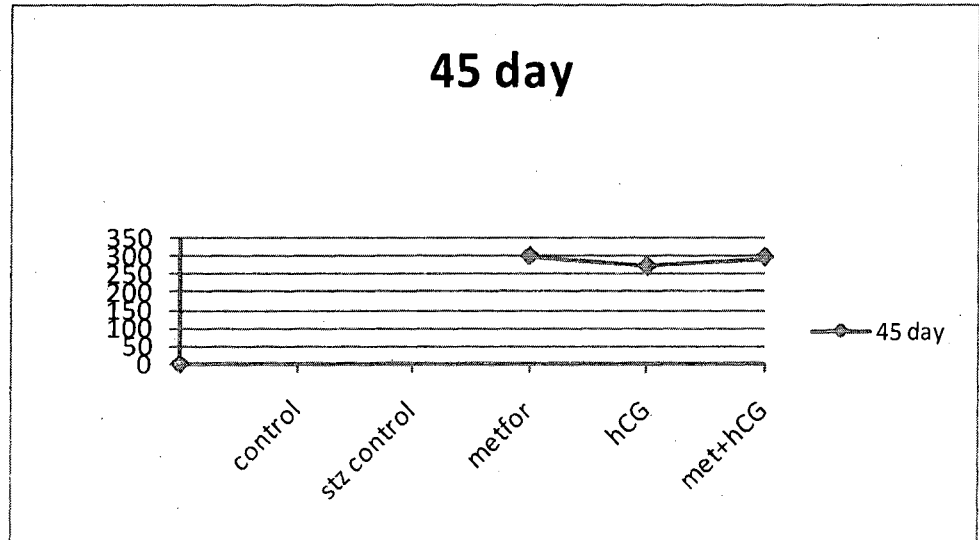
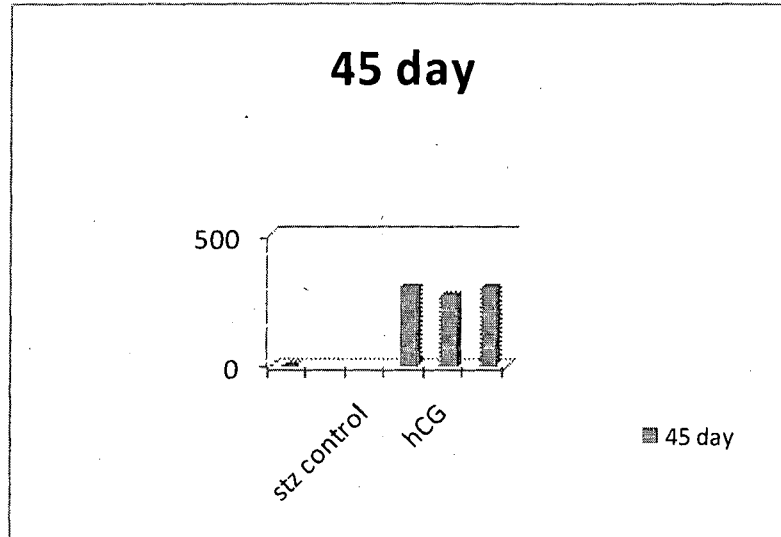
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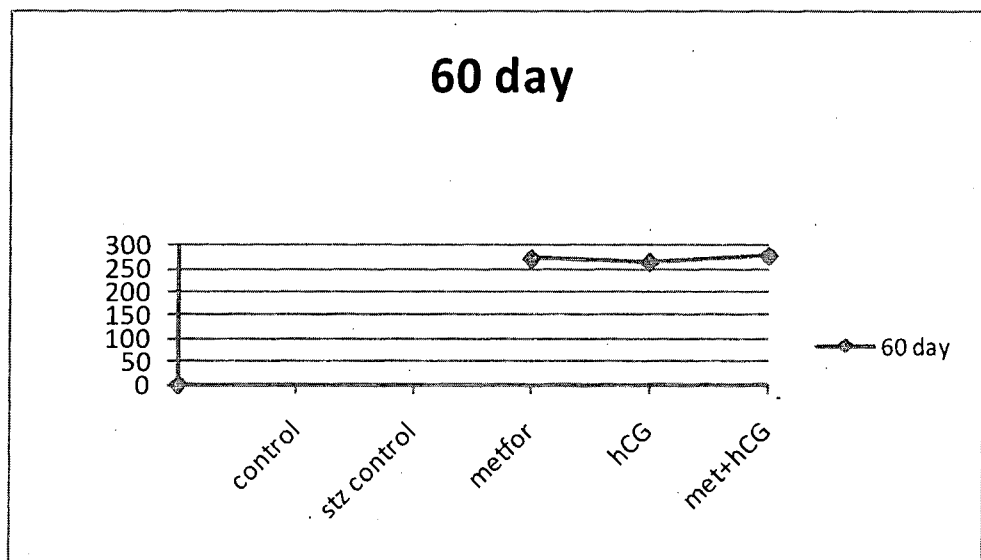
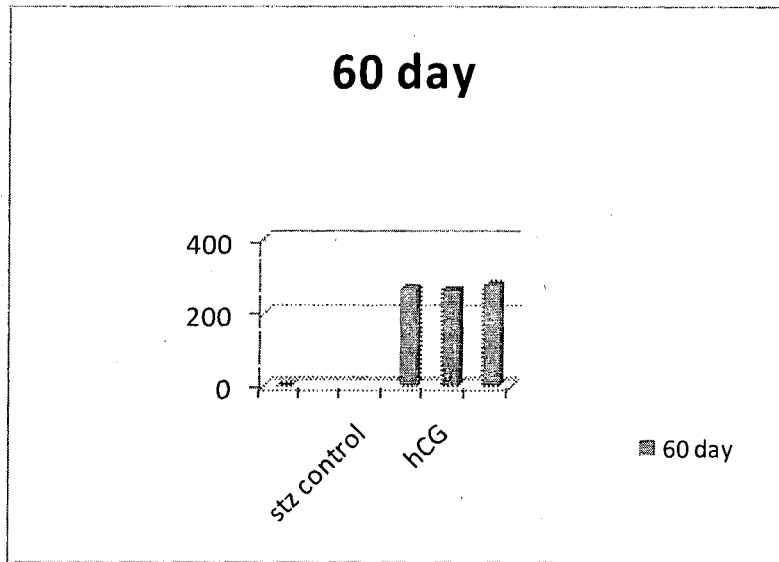
IX/XII



X/XII



XI/XII



XII/XII

