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EFFECT OF ETHANOLIC LEAF EXTRACTS OF *MUCUNA PRURIENS* ON SERUM HORMONAL LEVEL IN ALLOXAN-INDUCED DIABETIC MALE WISTAR RAT.

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Abstract
Objective: This research study was carried out to investigate the effect of ethanolic leaf extract of *Mucuna pruriens* (*M. pruriens*) on serum hormonal level in alloxan-induced diabetic wistar rats.

Methodology: Twenty-five (25) male wistar rats weighing between 200-250g were purchased and acclimatized for two weeks, after which they were divided into 7 groups of 5 rats each and housed in cages. The groups were designated as groups A, B, C, D and E. Groups B-E were induced with diabetes using alloxan. Groups A and B served as control groups and received only distilled water; while groups C – D diabetic that served as test groups received Glucophage, 400mg/kg of *M. pruriens* and 800mg/kg of *M. pruriens* respectively for 21 days via oral route with the aid of oral intubation tube. Blood samples were then collected on day 22 through ocular puncture for hormonal assay. All data were tabulated and statistically analyzed using SPSS version 21.0.

Result: The was significant decrease on serum levels of testosterone, FSH, insulin, and GH in the diabetic group B when compared with the control group A ($P>0.05$), but on administration of variable doses of the ethanolic leaf extract of *M. Pruriens,* there were significant increases on the levels of testosterone, FSH, insulin, and GH in groups D – E when compared with the control group A ($P>0.05$).

Conclusion: The leaf extracts of *M. pruriens* have ameliorative effect on the hormonal levels of alloxan-induced Wistar rats.

Keywords: *Mucuna purines,* testosterone, follicle stimulating hormone (FSH), insulin, growth hormone (GH).

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1.0 INTRODUCTION

The onset of Type I diabetes is known to disrupt the hypothamus-pituitary-gonadal axis (HPG) axis, resulting in impaired spermatogenesis and subsequent subfertility [1]. Disruptions in any part of the HPG axis impair fertility, and lead to infertility [1], thus affecting insulin and leptin levels, GnRH pulses from the hypothalamus, LH and FSH secretion from the pituitary, testosterone secretion from the Leydig cells, and sperm quality. Diabetes mellitus (DM) refers to a chronic, metabolic disease characterized by increased levels of blood glucose that lead to serious damage to the heart, blood vessels, eyes, kidneys, and nerves over time [2]. Its characteristic symptoms include thirst, polyuria, blurring vision and weight loss. Globally, an estimated 422 million adults are living with DM [3], and the number is projected to almost double by 2030 [4]. Approximately 150 million people have diabetes mellitus worldwide, and this number may well double by the 2025 [5].

DM being a life-long condition requires consistent management and tight glyceamic control to reduce the risk of complications development [6]. Reproductive complications such as disruption of male fertility, impotence, retrograde ejaculation and hypogonadism due damage to the beta cells of the pancreas, have been revealed through studies [7; 8]. Studies have also shown that sexual dysfunctions ranging from erectile and testicular dysfunctions, reduced libido, retrograde ejaculation [9], disrupted endocrine control of spermatogenesis [10], impaired sperm DNA integrity [11], reduced sperm count and motility [12] and low serum testosterone [13] are associated with DM. It therefore become pertinent that effort should be intensified through research study in other to develop drug(s) that will not only ameliorate the effect of DM but will also treat the complications associated with DM thus necessitating this research work.

Medicinal plants refers to plants that are being used to attempt to maintain health, to be administered for a specific condition, or both, whether in modern medicine or in traditional medicine [14]. Plants are composed of active component such as anthraquinones, flavonoids, glycosides, saponins and tannins etc which posses medicinal properties that can be harnessed for the treatment of different diseases [15]. *M. pruriens* is good example of such medicinal plants.

Mucuna pruriens is a tropical legume native to Africa and tropical Asia and widely naturalized and cultivated [16]. Its English common names are monkey tamarind, velvet bean, Bengal velvet bean, Florida velvet bean, Mauritius velvet bean, Yokohama velvet bean, cowage, cowitch, lacuna bean, and Lyon bean [16]. The plant is notorious for the extreme itchiness it produces on contact [17], particularly with the young foliage and the seed pods; and also produces many medium-sized red swollen bumps along with the itching. It has agricultural and horticultural value and is used in herbalism.

The seeds of *M pruriens* have been used for the treatment of many dysfunctions in Tibb-e-Unani (Unani Medicine) [18] and also used in Ayurvedic medicine. The plant and its extracts have been long used in tribal communities as a toxin antagonist for various snakebites, and has been studied for its effects against bites by Naja spp. (cobra) [19], Echis (Saw scaled viper), Calloselasma (Malayan Pit viper) and Bangarus (Krait). It has long been used in traditional Ayurvedic Indian medicine in an attempt to treat diseases including Parkinson's disease [20], and has been investigated in low income regions of the world as an alternative treatment for Parkinson's disease due to its high content of L-dopa [20]. The seeds have been recognized for their ability to significantly alleviate neurotoxicity induced by Parkinson's disease [21]. Its dried leaves are sometimes smoked [22].

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Roots, leaves and seeds of the plant are commonly used in the treatment of impotence, snake bite, diabetes, cancer and Parkinsonism [23]. *M pruriens* has also been shown to exhibit neuroprotective effect by increase brain mitochondrial complex-I activity and significantly restoring dopamine and norepinephrine levels in Parkinsonism animal model [24]. Its Seeds possess antioxidant, neuroprotective activities and bioactive substances [25; 26] and improves semen quality [27]. In vitro and in vivo studies on *M. pruriens* extract revealed the presence of substances that exhibit a wide variety of pharmacological effects, including anti-diabetic, anti-inflammatory, neuroprotective and anti-oxidant properties, probably due to the presence of L-dopa, a precursor of the neurotransmitter dopamine [26]. Research studies has also revealed that the main phenolic compound of *Mucuna* seeds is L-dopa (approximately 5%) [28]. Its leaves have mild activity against some bacteria probably due to the presence of phenols and tannins [28].

Hence this study was carried out to investigate the effect of *M. prurines* on the serum hormonal levels of alloxan-induced diabetic wistar rats since no work has been carried out on this.

2.0 MATERIALS AND METHODS

2.1 Animal procurement, care and treatment

Twenty five (25) male wistar rats weighing between 160g to 200g were procured and housed at the Animal house of Anatomy Department, Abia State University, Uturu with wire gauze cages in a well-ventilated area. They were fed with standard commercial pellet diet and water *ad libitum*. There were acclimatized for two weeks before the experiment. Their health statuses were closely monitored before and during the experiment. All procedures were carried out in strict accordance with the Institutional guidelines on the care and use of experimental animals.

2.2 Collection, identification and preparation of plant material

Fresh leaves of *M. pruriens* were harvested from a local settlement in Uturu, Isuikwuato Local Government Area of Abia state. The leaves were properly washed with water to remove sand and other impurities, and were authenticated at the herbarium of the Department of Physiology and Pharmacology, Department of Forestry, College of Natural Resources and Environmental Management, Micheal Okpara University of Agriculture, Umudike. Voucher number was assigned to the identified plant as MOUAU/VPP/17/017. They were air dried and crushed using laboratory blender. Extraction was done using ethanol. The crude ethanol extracts were filtered into a stainless basin with a white cloth and placed in a water bath so as to dry up the ethanol. 250mg of these extracts/kg body weights were dissolved in 10mls of distilled water and administered to the animals.

2.3 Induction of diabetes

The rats were divided into non-diabetic control group and experimental groups. The baseline blood glucose level of the experimental group to be inducted was determined before the induction of diabetes. The rats were allowed to fast over night prior to injection of alloxan and diabetes was induced by intra-peritoneal administration of 150mg of alloxan per kg body weight of rat (150mg/kg body weight). After the induction, the rats were allowed to have free access to the same feed and water. After 72 hours, blood samples obtained through tail tip puncture of the rats were used to confirm diabetes in the rats by testing for hyperglycemia using Glucometer. Diabetes was confirmed at fasting blood glucose levels greater than 200mg/dl [30].

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2.4 Experimental protocol

The animals were grouped into five (5) groups of five (5) rats each. Different doses of the leaf extracts were administered via oral route with the aid of oral intubation tube as shown below:

Group A	(The Control group) distilled water.
Group B	(Diabetic group) distilled water.
Group C	Diabetic + Glucophage
Group D	Diabetic + 400mg/kg of *M. pruriens* leaf extract.
Group E	Diabetic + 800mg/kg of *M. pruriens* leaf extract.

2.5 Sample collection

The extracts were administered for twenty one (21) days. On the 22nd day, blood samples were collected through ocular puncture as described by Hoff and Rlagt, [31] for hormonal assay. The obtained blood samples were spun at 2500pm for 10min using wisperfuge model 1384 centrifuge at 10-25^{0}C, serum samples were collected, refrigerated and assayed for growth hormone, follicle stimulating hormone, testosterone and luteinizing hormone using the microwell enzyme linked immunoassay (ELISA) technique, and analytical grade reagent (Syntron Bioresearch Inc, USA).

2.6 Statistical Analysis

All data were tabulated and statistically analyzed using SPSS version 21.0. Results were expressed as Mean ± standard error of mean (SEM). Comparative analysis amongst groups was done using one-way analysis of variance (ANOVA). A post-hoc analysis using Bonferoni multiple comparative tests was performed to identify significant groups. $P<0.05$ was taken as statistically significant.

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3.0 RESULTS

Result showed a significant decrease ($P>0.05$) in serum testosterone level in groups B (3.21±0.09miu/mL) when compared with the control group A (4.09±0.15miu/mL) and significant increase ($P<0.05$) in serum testosterone level in groups C (3.78±0.07miu/mL), D (3.75±0.06miu/mL) and E (3.86±0.03miu/mL) when compared with the control group A.

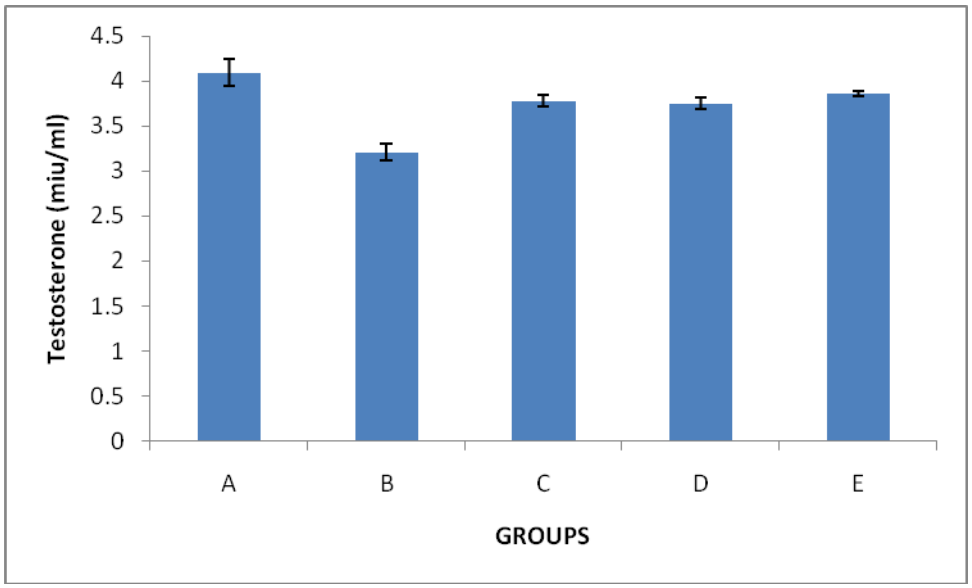

Fig 1: Effect of ethanolic leaf extract of *M. pruriens* on serum the level of testosterone in alloxan-induced male wistar rat.

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Result showed a significant decrease (P>0.05) in serum FSH level in groups B (3.03±0.09miu/mL) when compared with the control group A (4.69±0.25miu/mL), no significant increase serum FSH in group D (3.44±0.13miu/mL) was observed when compared with the control group A. However, test groups C (4.03±0.14miu/mL) and E (3.65±0.07miu/mL) showed significant increase (P<0.05) in serum FSH level when compared with the control group A.

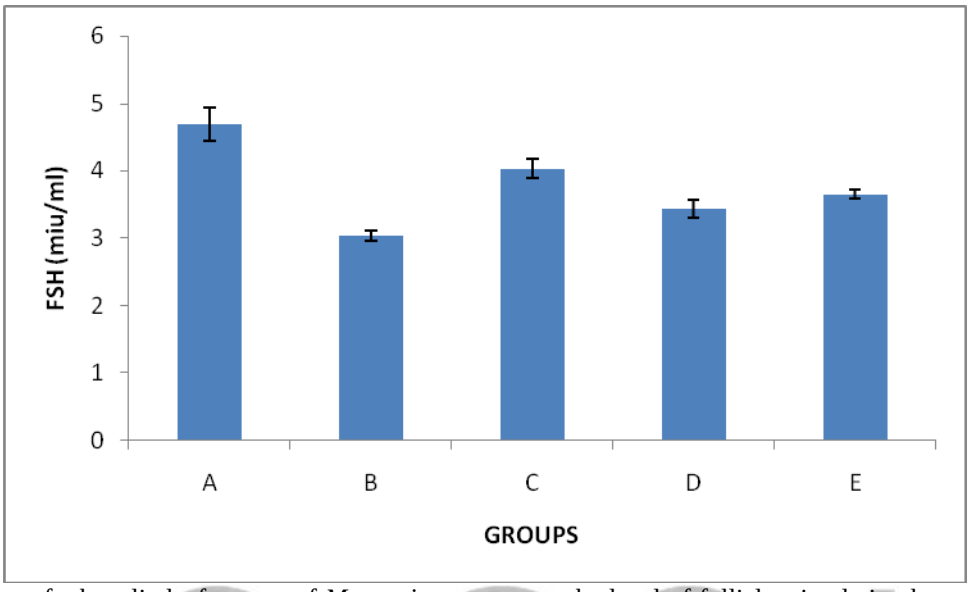

Fig 2: Effect of ethanolic leaf extract of *M. pruriens* on serum the level of follicle stimulating hormone in alloxan-induced male wistar rat.

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Result showed a significant decrease (P>0.05) in serum insulin level in groups B (0.45±0.04miu/mL) when compared with the control group A (1.54±0.07miu/mL), no significant increase in group C (0.89±0.05miu/mL) and group D (0.90±0.04miu/mL) when compared with the control group A. However, test group E showed significant increase (P<0.05) in serum insulin level (1.08±0.06miu/mL) when compared with the control group A.

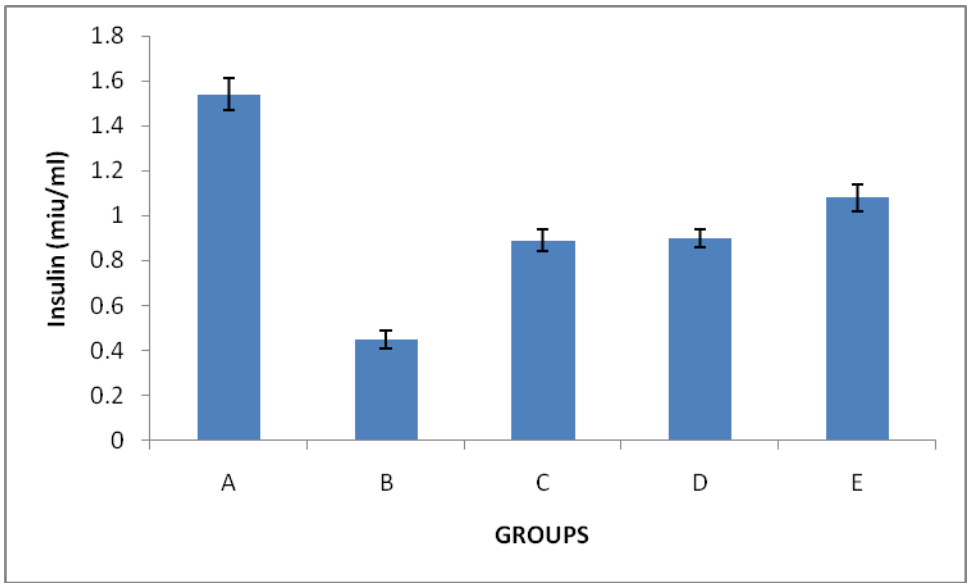

Fig 3: Effect of ethanolic leaf extract of *M. pruriens* on serum the level of insulin in alloxan-induced male wistar rat.

Result showed a significant decrease (P>0.05) in serum growth hormone level in groups B (0.0861±0.006ng/mL) when compared with the control group A (0.114±0.014ng/mL), no significant increase in group C (0.085±0.002miu/mL) and group D (0.080±0.019ng/mL) when compared with the control group A. However, test group E showed significant increase (P<0.05) in serum growth hormone level (0.108±0.007miu/mL) when compared with the control group A.

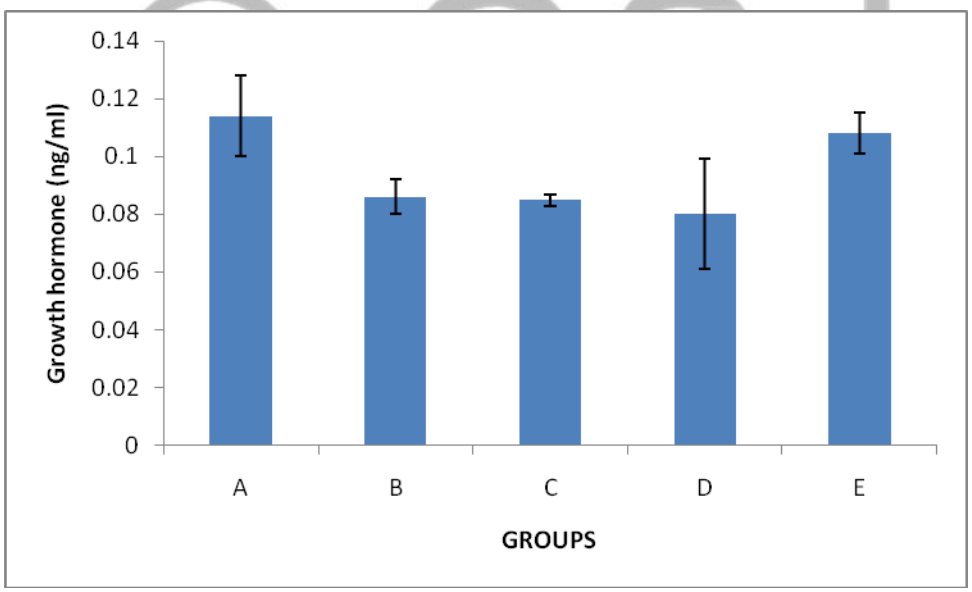

Fig 4: Effect of ethanolic leaf extract of *M. pruriens* on serum the level of growth hormone in alloxan-induced male wistar rat.

4. Discussion

The alloxan induced to the wistar rats selectively kills the insulin-producing beta-cells found in the pancreas thereby inducing diabetes in laboratory animals [32; 33]. This occurs most likely because of selective uptake of the compound due to its structural similarity to glucose as well as the beta-cell's highly efficient uptake mechanism (GLUT2). In addition, it has a high affinity to SH-containing cellular compounds, thus, reduces glutathione content. Also, alloxan inhibits glucokinase, a SH-containing protein essential for insulin secretion induced by glucose [34].

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The significant decrease (P>0.05) in serum testosterone level seen in group B (3.21±0.09miu/mL) when compared with the control group A (4.09±0.15miu/mL) (fig.1) could be due to insulin resistance [35]. Research has shown that men with impaired glucose tolerance had significantly lower levels of total testosterone [36]. Low levels os testosterone suggests that testosterone levels are impaired by Type 1 diabetes in poorly controlled subjects, which could inhibit the process of spermatogenesis [1]. On administration of glucopahage and variable doses of *M. pruriens* to the alloxan-induced diabetic rats, there were increases in the level of testosterone in groups C (0.89±0.05miu/mL) and D (0.90±0.04miu/mL thou not significant when compared with the control group, but was significantly increased (P<0.05) in group E which received higher dose of the leaf extract. no significant increase in group C (0.89±0.05miu/mL) and group D (0.90±0.04miu/mL) when compared with the control group A. this results may be due to the antidiabetic [23] and improvement of semen quality [27] effects of *M. pruriens*. Thus, the leaf extract *M. puriens* has proved to have ameliorative effect on the level of serum testosterone.

In fig.2 above, there was a significant decrease (P>0.05) in serum FSH level in groups B (3.03±0.09miu/mL) when compared with the control group A (4.69±0.25miu/mL). This may be due to disruptions of the HPG axis which impair fertility, and may lead to infertility [1], thus affecting insulin and leptin levels, GnRH pulses from the hypothalamus, LH and FSH secretion from the pituitary, testosterone secretion from the Leydig cells, and sperm quality. There administration of glucophage and variable doses of *M. pruriens* extracts brought ameliorative effect on the level FSH leading to significant increase the level of FSH in group D (3.44±0.13miu/mL), with better effect in groups C (4.03±0.14miu/mL) and E (3.65±0.07miu/mL) which showed significant increase (P<0.05) in serum FSH level when compared with the control group A.

The significant decrease (P>0.05) in serum insulin level in groups B (0.45±0.04miu/mL) when compared with the control group A (1.54±0.07miu/mL) seen in (fig.3) may be due to damage by the induced alloxan which selectively kills the insulin-producing beta-cells found in the pancreas to induce diabetes in laboratory animals [32; 33]. Beta cells (β-cells) of pancreatic islets synthesize and secrete insulin and amylin. They make up 50–70% of the cells in human islets [37]. In type 1 diabetes, beta-cell mass and function are diminished, leading to insufficient insulin secretion and hyperglycemia [38]. Type I diabetes is an autoimmune disorder characterized by a lack of insulin production by the beta cells of the pancreas [39]. This lack of insulin causes a variety of systemic effects such as cardiovascular disease, diabetic neuropathy, and diabetic retinopathy [40; 41; 42; 43]. The administration of a higher dose of M. pruriens in group E (0.108±0.007miu/mL) showed significant increase (P<0.05) in serum growth hormone level when compared with the control group A proving the ameliorative effect of the leaf extracts to the beta-cells of the pancreas..

The significant decrease (P>0.05) in serum growth hormone level in groups B (0.0861±0.006ng/mL) when compared with the control group A (0.114±0.014ng/mL) could be due to insufficient insulin which prevents the body from getting glucose from the blood into the body's cell leading hyperglycemia. When this occurs, the body starts burning fat and muscle for energy causing a reduction in overall weight. This result is in agreement with the result of research work carried out by Ewenighi *et al.,* [44] which reported decreased in body weight in alloxan induced diabetic rats. Junod *et al.,* [45] and Montano *et al.,* [46] also reported significant weight loss after inducing diabetes with streptozotocin on Wistar rats in their respective studies. According to Holly, *et al.*, [47], GH is one of the glucose counter-regulatory hormones, rising in response to hypoglycaemia; and has both intrinsic hyperglycaemic actions and causes insulin

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resistance. On the administration of the leaf extract, there were no significant increase in group C (0.085±0.002miu/mL) and group D (0.080±0.019ng/mL) when compared with the control group A. However, test group E showed significant increase ($P<0.05$) in serum growth hormone level (0.108±0.007miu/mL) when compared with the control group A could be due to ameliorating effect of the leaf extract on the hyperglycemic effect on the alloxan-induced rats thereby increasing the level of GH since GH levels are increased by low glucose levels in the blood [47].

5. Conclusion

This study has shown that ethanolic leaf extract of *M. pruriens* has antidiabetic effect through its ameliorating effect on the serum levels of testoerone, follicle stimulating hormone, insulin and growth hormone on alloxan induced male wistar rats. Thus, this therefore supports the use of *M. pruriens* for the treatment of diabetes mellitus.

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Ethical Approval: Approved by Institutional ethical approval.

REFERENCES.

1. Erica L. S., Samantha S. and Kelle H. M. The effects of type 1 diabtes on the hypothalamic, pituitary and testes axis. *Cell Tissue Res.* 2012 Sep; 349(3): 839–847.
2. World Health Organization. Diabetes. *Global report on diabetes.* 2018; Geneva.
3. World Health Organization. *Global Report on Diabetes*. 2016 Geneva.
4. Wild, S. Roglic, G. Green, A. Sicree, R. King, H. "Global prevalence of diabetes: Estimates for the year 2000 and projections for 2030". *Diabetes Care*, 2004; 27 (5): 1047–53.
5. World Health Organization. Diabetes. *Global report on diabetes.* 2019; Geneva.
6. Wong, N. D. Intensified screening and treatment of the metabolic syndrome for cardiovascular risk reduction. *Preventive Cardiology.* 2005; 8: 47-54.
7. Loeken M. R. A new role for pancreatic insulin in the male reproductive axis. *Diabetes.* 2012; 61: 1667-1668.
8. Schoeller E. L, Albanna G, Frolova A. I. *et al*. Insulin rescues impaired spermatogenesis via the hypothalamic-pituitary-gonadal axis in akita diabetic mice and restores male fertility. *Diabetes.* 2012; 61: 1869-1878.
9. Dunsmuir W. D. and Holmes S. A. The aetiology and management of erectile, ejaculatory, and fertility problems in men with diabetes mellitus. *Diabet Med.* `996;13:700–708
10. Baccetti B, La Marca A, Piomboni P, Capitani S, Bruni E, Petraglia F, De Leo V. Insulin-dependent diabetes in men is associated with hypothalamo- pituitary derangement and with impairment in semen quality. *Hum Reprod.* 2002; 17:2673–2677.
11. Agbaje I. M, Rogers D. A, McVicar C. M, McClure N, Atkinson A. B, Mallidis C, Lewis S. E. Insulin dependent diabetes mellitus: implications for male reproductive function. *Hum Reprod.* 2007; 22:1871–1877.
12. Bhattacharya S. M, Ghosh M, Nandi N. Diabetes mellitus and abnormalities in semen analysis. *J Obstet Gynaecol Res.* 2014; 40:167–171.
13. Verma S, Saxena S. K, Kushwaha J. S, Giri R, Priyadarshi B. P, Singh P. Serum testosterone levels in type 2 diabetes mellitus. *JIACM*.2013; 14:115–118.

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14. Ahn, K. "The worldwide trend of using botanical drugs and strategies for developing global drugs". *BMB Reports.* 2017; *50 (3): 111–116.*
15. Feher, M. and Schmidt, J.M. Property distributions: differences between drugs, natural products, and molecules from combinational chemistry. *Journal of chemical infection and complementary sciences,* 2003; 43: 218-227.
16. Germplasm Resources Information Network (GRIN). *Mucuna pruriens*" Agricultural Research Service (ARS), United States Department of Agriculture (USDA). 2015.
17. Andersen H. H, Elberling J, Arendt-Nielsen L. "Human surrogate models of histaminergic and non-histaminergic itch" (PDF). *Acta Dermato-Venereologica*. 2015; 95 (7): 771–7
18. Amin. K. M. Y., Khan. M. N., Rahman, Hakim Syed Zillur; *et al*. "Sexual function improving effect of Mucuna pruriens in sexually normal male rats". *Fitoterapia*. 1996; 67(1): 53–58.
19. Tan, N. H., Fung, S. Y., Sim, S. M., Marinello, E., Guerranti, R., Aguiyi, J. C. "The protective effect of *Mucuna pruriens* seeds against snake venom poisoning". *Journal of Ethnopharmacology.* 2009; 123 (2): 356–8.
20. Cassani, E., Cilia, R., Laguna, J., Barichella, M., Contin, M., Cereda, E., Isaias, I. U., Sparvoli, F., Akpalu, A., Budu, K. O., Scarpa, M. T., Pezzoli, G. "*Mucuna pruriens* for Parkinson's disease: Low-cost preparation method, laboratory measures and pharmacokinetics profile". *Journal of the Neurological Sciences*. 2016; 365: 175–80.
21. Singh, Surya P.; Gedda, Mallikarjuna R.; Jadhav, Jyoti P.; Patil, Ravishankar R.; Zahra, Walia; Singh, Saumitra S.; Birla, Hareram; Rai, Sachchida N. "*Mucuna pruriens* Protects against MPTP Intoxicated Neuroinflammation in Parkinson's Disease through NF-κB/pAKT Signaling Pathways". *Frontiers in Aging Neuroscience*. 2017; 9: 421.
22. Rätsch, Christian. Enzyklopädie der psychoaktiven Pflanzen. Botanik, Ethnopharmakologie und Anwendungen. Aarau: AT-Verl. 1998; 15.
23. Alo, M.N., Okeh, O.C., Anyim, C., Orji, J.O. The effects of ethanol extract of *Mucuna pruriens* leaves on aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase in albino rats. *Journal Natural Production Plant Resource* 2012; 2 (4), 465-470.
24. Manyam, B. V., Dhanasekaran M., and Hare T. A. Neuroprotective effects of the antiparkinson drug *Mucuna pruriens*. *Phytotherapy Res.*, 2004; 18:706-712.
25. Sharma, M. L., Chandhoke, N., Ray Ghatak, B. J., Jamwal, K. S., Gupta, O. P., and Singh, G. B. Pharmacological screening of Indian medicinal plants. *Indian Journal of Experimental Biology* 1978; 16:228-235.
26. Misra L. and Wagner H. Extraction of bioactive principles from *Mucuna pruriens* seeds. *Indian J. Biochem. Biophys.* 2007; 44:56–60.
27. Daramola, J. O., Abiona, J. O., Smith, O. F., Isah, O. A., Sowande, O. S. Olaniyi, M. O. and Olanite, J. A. Effect of *Mucuna* (*Mucuna pruriens*) on spermiograms of West African dwarf bucks. *Tropical and subtropical Agroecosystems* 2015; 18(2):145-150.
28. Vadivel, V, and Pugalenthi M. Removal of antinutritional/toxic substances and improvement in the protein digestibility of velvet bean seeds during various processing methods. *J. of Food Sci. and Technol.* 2008; 45:242–246.
29. Ogundare, A. O. and Olorunfemi, O. B. Antimicrobial Effecacy of the leaves of *Dioclea reflexa, Mucuna pruriens, Ficus asperifolia* and *Tragia spathulata*. *Research Journal of Microbiology*, 2007; 2:392-396.
30. Adenowo A. F, Ilori M. F, Balogun F. O, Kazeem M. I. Protective effect of ethanol leaf extract of *Carica papaya linn* (*Caricaceae*) in alloxan-induced diabetic rats. *Tropical Journal of Pharmaceutical Research.* 2014; 13(11): 1877-1882.

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31. Hoff J. and Rlagt L. V. Methods of blood collection in the mouse. *Lab animals*. 2000; 29:47-53.
32. Danilova I.G., Sarapultsev P.A., Medvedeva S.U., Gette I.F., Bulavintceva T.S., Sarapultsev A.P. "Morphological Restructuring of Myocardium During the Early Phase of Experimental Diabetes Mellitus". *Anat. Rec*. 2014; 298 (2): 396–407.
33. Loreto D, Elina V. Experimental surgical models in the laboratory rat. Boca Raton: CRC Press. 2009.
34. Szkudelski T. "The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas". *Physiol Res*. 2001; 50 (6): 536–546.
35. Jee-Young O., Elizabeth, B., Nicole, M. W., Deborah, L. W. Endogenous Sex Hormones and the Development of Type 2 Diabetes in Older Men and Women: the Rancho Bernardo Study. *Diabetes Care* 2000; 25 (1) 55-60
36. Goodman-Gruen D. and Barrett-Connor E. Sex differences in the association of endogenous sex hormone levels and glucose tolerance status in older men and women. *Diabetes Care* 2000; 23:912–918.
37. Dolenšek, J., Rupnik, M. S., Stožer, A. "Structural similarities and differences between the human and the mouse pancreas". *Islets.* 2015; 7 (1): e1024405.
38. Chen, C., Cohrs C. M., Stertmann, J., Bozsak, R., Speier, S. "Human beta cell mass and function in diabetes: Recent advances in knowledge and technologies to understand disease pathogenesis". *Molecular Metabolism.* 2017; 6 (9): 943–957.
39. Yoon, J. W. and Jun, H. S. Autoimmune destruction of pancreatic beta cells. *Am J Ther*. 2005; 12:580–591.
40. Sowers, J. R., Epstein, M., Frohlich, E. D. Diabetes, hypertension, and cardiovascular disease: an update. *Hypertension.* 2001; 37:1053–1059
41. Said G. Diabetic neuropathy--a review. *Nat Clin Pract Neurol.* 2007; 3:331–340.
42. Barber A. J. A new view of diabetic retinopathy: a neurodegenerative disease of the eye. *Prog Neuropsychopharmacol Biol Psychiatry.* 2003; 27:283–290.
43. Vinik, A. I., Holland, M. T., Le Beau J. M., Liuzzi, F. J., Stansberry, K. B., *et al.* Diabetic neuropathies. *Diabetes Care.* 1992; 15:1926–1975.
44. Ewenighi C, Dimkpa U, Onyeanusi J, Onoh L, Onoh G, Ezeugwu U. Estimation of Glucose Level and Body Weight in Alloxan Induced Diabetic Rat Treated with Aqueous Extract of Garcinia Kola Seed. *The Ulutas Medical Journal.* 2015; 1(2):26-30.
45. Junod A, Lambert A. E, Stauffacher W, Renold A. E. Diabetogenic action of streptozotocin: relationship of dose to metabolic response. *J Clin Invest*. 1969; (48):2129-2139.
46. Montano M. E, Molpeceres V, Mauriz J. L, Garzo E, Cruz I. B, González P, Barrio J. P. Effect of melatonin supplementation on food and water intake in streptozotocin-diabetic and non-diabetic male Wistar rats. *Nutr Hosp.* 2010; (25):931-938.
47. Holly,J. M., Amiel,S. A., Sandhu, R. R., Rees, L. H., Wass, J. A. The role of growth hormone in diabetes mellitus. *J Endocrinol* 1988; 118(3):353-64.

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