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Review Article

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Fertility potentials of five medicinal plants in the treatment of infertility caused by polycystic ovarian syndrome in female wistar albino rats

Abstract

Polycystic ovarian syndrome cause infertility in females within childbearing age, Patient with Polycystic ovarian syndrome (PCOS) go through the stress of taking serial medication for years including various medical procedures which are quite traumatic and painful. PCOS was induce in the experimental animals by administering 1mg/kg per body weight of letrozole for twenty one days. After 21days oral administration of letrozole to the experimental animal Blood samples were collected for Glucose and Lipid profile. Serum Progesterone, Estradiol, Testosterone, Luteinizing and Follicle stimulating hormones were measured with enzyme – linked immunosorbent Assay Kit. After the confirmation of the establishment of PCOS, the following plant extracts were administered at 300mg/kg and 100mg/kg per body weight for 15days to different groups of animals in separate cages: *Trigonella foenum graecum*, *Sesamum indicum, Glycine max*, *Glycyrrhiza glabra* and *Lepidium meyenii*. The result shows *T. feonum graecum* and *G. glabra* effectively reduced insulin resistance by reducing the glucose level from 7.60mmol/L in the PCOS group to 5.50mmol/L in *T. feonum graecum* and 5.23mmol/L in G glabra. Increased in lipid profile caused by polycycstic ovarian syndrome as seen in the PCOS group was reduced effectively by *S. indicum* and *G. glabra.* More so, effective control of increased androgen level and hirsitism was shown by *T. feonum graenum* and *L. meyenii* by reducing the testosterone level from 34.4ng/ml in the PCOS group to 5.33ng/ml in *T. feonum graecum* and 7.10ng/ml in *L meyenii*. Ability to increase follicle stimulating hormone was shown in *S indicum, T graecum*, *G max*, and *G glabra*, whereas the ability to ovulate, go through a successful gestation period, and deliver live babies was seen in *S indicum* and *T greacum*, this could be attributed to the high flavoniod (phytoestrogens) seen in the two plant extracts. The best of all the plants extracts is the extract of *T graenum* because of the continuous fertility recorded in this group and also their none toxic effect on the vital body organs.

Keywords: polycystic ovarian syndrome, infertility, testosterone, follicle stimulating hormone, luteinizing hormone, progesterone

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Introduction

Female fertility is the ability of a female to conceive and bear a biological child after copulation with the opposite sex. The time it takes to get pregnant varies in all females. Some females have the ability to get pregnant having a regular unprotected sex within one month, some within one year and others up to three years. However, some life style such as smoking, too much of alcohol, excess intake of caffeine, the use of illicit drugs and weight gain can affect fertility.

Apart from the lifestyle of an individual there are many other causes of infertility in females, these includes, polycystic ovarian syndrome, blockage of the fallopian tube, ovulation problem, endometrioses, production of non-viable eggs, age, premature ovarian insufficiency, sexual transmitted diseases, sterility, fibroid, cancer of the cervix, implantation failure, structural problems of the reproductive system.[1] PCOS is a major area of concern as it affects – reproductive age women causing infertility.[2,3] Moreso, research outcome on global infertility suggests about 48 million of couples worldwide suffer for infertility and about 186 million individuals live with infertility globally.[1] Smeltzer et al.,[4] reported that 5% –10% of women in childbearing age have PCOS. Females with PCOS have imbalance of sex hormones, increase in level of testosterone {male hormone}, this prevents the development and release of mature eggs thereby causing infertility in females.[5] Women with PCOS have insulin resistance, this results in too much sugar in the body, this in turn force the body to produce more insulin and too much of insulin in the body causes increase in testosterone level which affects ovulation and fertility.[5] There are some complications of PCOS such as irregular menstruation, which affect fertility, in that it makes one not to be able to predict ovulation day, also imbalance hormones prevent endometrium lining for strong implantation of the embryo which in turn causes infertility. Other complications of PCOS include pregnancy induced diabetes and pregnancy induced high blood pressure, both result to premature delivery and sometimes maternal and child mortality. Polycystic Ovarian Syndrome leads to increased secretion of androgen hormones which impaired the growth and releases of mature follicle and also result to the formation of cyst around the ovary. Clinical presentations of PCOS include infertility, irregular menstration, hirsitisim, obesity, increase in glucose level, increased in lipid profile, breathing difficulty while sleeping.

Diagnosis of PCOS include; hormonal essay, pelvic examination for the presence of any inflammation, ultra sound scan to check for cyst in the ovaries, routine blood pressure check, glucose and lipid level and screening for depression and anxiety. Treatment of PCOS in modern medicine includes use of progesterone supplement before and after conception, these medication are; Clomid, Metformin, Gonadotropins, Spironolactone, electrolysis and laparoscopic ovarian drilling.

Modern medicine can be effective however it takes a longer time for conception to occur and also it has no lasting effect, very expensive and do not provide cure. This lead to an increase interest in herbal medicine that is less expensive and without side effect and above all curative. Moreso, the cost of diagnosis and treatment of PCOS is alarming. Ricardo et al 2005 reported that the total cost of evaluating and providing care for women within reproductive age PCOS women in the United States is 4.36billion, out of which diagnosis is approximately 2% and the rest for treatment.

Material and methods

Materials purchased: Five hormonal Assay kit were used in this research. Rat Progesterone Elisa kit with the code: ER1255 and Batch No;R1255f123 j, Rat Estradiol Elisa Kit with code no: ER1507 and Batch No: R1507F123 j, Rat Testosterone Kit with Code No: ER1462 and Batch No: R1462F123 J, Rat Luteinizing hormone with Code No: ER1123 and Batch No: R1123f123 j, Rat Follicle stimulating hormone with Code No: ER0960 and Batch No: R0960F123 J. The five kits were purchase from Fine Test in Wuhan, China.

The microplate reader that was used to read the absorbent of each sample during hormonal analysis is a product of Kayto RT-2100C.

Letrozole, metformin and clomidphene were purchase from Andy pharmacy a recognized supplier in Kaduna State of Nigeria.

Experimental animals: Twenty (20) adult male and sixty (60) female rats were obtained from the Institute of Trypanosomiasis Research Kaduna, Kaduna state. The experimental rats were caged in a standard cage, allowed to acclimatize with animal house conditions for two weeks and were also allowed free access to food and water. Ethical clearance approval for the research was granted from Nigerian Defence Academy Ethical committee.

Quantitative phytochemical screening: After the extraction of the plant extract using ethanol, small quantity of the plant extract were then used for phytochemical analysis. Phytochemical screening for cardiac glycosides, saponis, glycosides, Alkaloids, Amino Acid, Starch, Reducing sugar, Phenol, volatile oil, tannins, flavoniods and steroids will be carried out using the method of.[6] This analysis were carried out in Chemistry lab, NigerianDefence Academy, Kaduna State Nigeria.

Administration of letrozole to induce polycystic ovarian syndrome on the rat

The animals were grouped into twelve(12) groups, each containing five rats. Most importantly, each group was placed in a separate cage. Eleven (11) out of the 12 groups of animals were administered letrozole orally, the only group not administered letrozole is the normal control group. The letrozole was administered for 21days at 1mg/kg per body weight.

Observation of clinical signs after administrations of letrozole

The rats were observed every morning for the 21days of administration of letrozole, for any change in feeding habit, any signs of malaise, any stooling any changes on their skin, loss of appetite, etc.

Confirmation of inducement of polycystic ovarian syndrome

After administering letrozole for 21days, ten rats were randomly selected and transported to vetenary clinic, School of Vetenary Medicine, Ahmadu Bello University Zaria, Kaduna state Nigeria were histology of the ovaries was carried out to confirm the inducement of PCOS on them, Hormonal Assay was also carried out on the rats. Serum and Blood samples of the rats were also collected and taken to Human Anatomy Department and University Teaching Hospital Medical labouratory, Ahmadu Bello University Zaria for screening of blood glucose level, lipid profile, and all these analysis is to ascertain the establishment of polycystic ovarian syndrome before the commencement of treatment.

Experimental design

The experimental animals were grouped into 12 groups, each group were caged separately to avoid any major experimental errors.

GROUP A_1; The animals in this group were administered letrozole for twenty one days followed by *Trigonella feonum graecum* ethanolic extract at 300mg/kg for fifteen days.

GROUP A2: The animals in this group were administered letrozole for twenty one days and *Trigonella feonum graecum* ethanolic extract at 100mg/kg for fifteen days.

GROUP B1: The animals in this group were administered letrozole for twenty one days followed by *Glycine max* extract at 300mg/kg for fifteen days.

Group B2: The animals in this group were administered letrozole for twenty one days followed by *Glycine max* ethanolic extract at 100mg/kg for fifteen days.

Group C1: The animals in this group were administered letrozole for twenty one days followed by *Sesamum indicum* ethanol extract at 300mg/kg for fifteen days.

Group C2: The animals in this group were administered letrozole for twenty one days followed by *Sesamum indicum* ethanol extract at 100mg/kg for fifteen days.

Group D1: The animals in this group were administered letrozole for twenty one days followed by *Glycyrrhiza glabra* ethanol extract at 300mg/kg for fifteen days.

GROUP D2: The animals in this group were administered letrozole for twenty one days followed by *Glycyrrhiza glabra* ethanol extract at 100mg/kg for fifteen days.

Group E1: The animals in this group were administered letrozole for twenty one days followed by *lepidium meyenii* at 300mg/kg for fifteen days.

Group E2: The animals in this group were administered letrozole for twenty one days followed by *Lepidium meyenii* at 100mg/kg for fifteen days

Group PCOS: This group was administered letrozole for twenty one day only without any treatment.

Administration of plant extract to treat the syndrome

After the administration of letrozole, the weights of the experimental animals were taken and an identity was given to them using picric acid and that is what was used to calculate the dose of extract to be administered to each of the experimental animals. The extract administration was done for fifthteen (15) days and all treatment were via oral route.

Dose Calculation for each rats was done by using this formula

$$\text{Dose} = \frac{\text{Test dose X Weight of rat Vol(ml)}}{\text{1000 X Conc of extract}}$$

The concentration chosen in this research is 200mg/kg

Vol(ml): volume of the extract to be aspirated with the syringe and administered

Dose(mg/kg); Dose of the plant extract that is due to the rat which could either be 300mg/kg or 100mg/kg

Weight of Rat: weight of the particular rat that will receive the plant extract.

1000: This is the converting factor from kilogram to gram

Conc of extract: This is the chosen concentration of the extract which is 200mg/kg

Observation of clinical signs after administration of plant extract

During and after administration of the plant extract on the experimental animals, they were observed for any signs of malaise, stooling, lack of appetite, isolation, any changes on their skin etc.,

Biochemical analysis: All collected blood samples for biochemical analysis, for Glucose, insulin, cholesterone, triglyceride and HDL cholesterone level were taken to school of vetenary medicine, Ahmadu Bello university Teaching hospital Zaria were enzymes – linked immune sorbent assay based kit, imported from Wuhan China were used to carry out the analysis. All analysis was carried out using carefully the manufactures instructions and guidelines.

Procedure for hormonal assay: Hormonal Assay for Estrogen, Progesterone, Luteinizing, Follicle stimulating hormone, testosterone and prolactin level was measured using enzymes – linked immune sorbent assay based kit.The pre coated plate were wash twice, before the wells were added, the position of the standard, samples and control wells were labeled appropriately, then 50ul of standard or sample were then added to the appropriate wells, after which 50ul of Biotin labeled Antibody was then immediately added into each wells, the plate was then gently tap to ensure thorough mixture before incubating for 45mins at 37^0C. After 45mins incubation the content of the wells were aspirated with micro pippete and washed three times. Then 100ul of Streptavidin conjugate (SABC) was then added into each wells and incubated for 30mins at 37^0C. After the second incubation of 30mins, the content of the wells were aspirated with micro pippete and washed 5 times. After washing the wells 5 times, 90ul of TMB substrate solution was then added to each well and then incubated for 10-20 mins at 37^0C.The experiment is brought to an end, by the addition of 50ul of stop solution. After which the plate is then placed in a microplate reader to read at 450nm immediately and the solubility level of each calculated accurately.

Observation of signs of pregnancy and delivery: The two female rats left in each cage (each group) were then exposed to a male rat for mating to occur. After two weeks of mating the animals were observed for signs of pregnancy. Three to four weeks of mating the animals were then observed for possible delivery.

Results

The result of the phytochemical screening of plant show the presence of Saponins, Cardiac glycosides and Alkaliods across all the plant extract at various concentration. Anthroquinones and Glycosides were absent across all plant extract, however, phenols, flavoniod, Taninns and steroid were present in some plant extract and absent in some (Table 1). The data obtained From the result of the glucose level test of all treatment groups, (Table 2), show that all plant extracts significantly reduce the blood sugar increase caused by polycystic ovarian syndrome when compare to the PCOS group, except for *G. max* at 100mg/ml and *L. meyenii* at 100mg/ml that had no reduction in blood sugar level.

From the result of lipid profile (Table 3), *S. indicum* 300mg/ml, S. indicum 100mg/ml and *G.glabra* 100mg/ml significantly reduced the increase in total cholesterone caused by polycystic ovarian syndrome, however *S. indicum* at 300mg/ml show statistical significant difference in the reduction of total cholesterone level, increase in high density lipoprotein and reduced low density lipoprotein.

The result of the hormonal assay (Table 4), indicate that *T. graenum* at 100mg/ml, *G. max* at 300 and 100mg/ml, *S. indicum* at 300 and 100mg/ml and *G. glabra* at 300 and 100mg/ml caused increase in follicle stimulating hormone. Whereas, *T. graenum* at 300mg/ml and *L. meyenii* at 300 and 100mg/ml reduced the high testosterone level caused by polycystic ovarian syndrome.

Clinical signs observed during the administration of letrozole

The clinical signs observed after the administration of letrozole include the following

i. Increase in their body weight across all treatment groups
ii. Lost of their body hair across all the treatment groups
iii. Increase in pubic hair across all the treatment groups

All treatment group show decrease in blood sugar except for *Glycine max* 100mg/kg and *Lepidiummeyenii* at 100mg/kg that show increase in glucose like the PCOS group however, *Trigonella* spp and *Glycyrrhiza glabra* at 100mg/kg that show good reduction in glucose level having the same superscripts like the normal control.

Sesamum indicum at both 300mg/kg and 100mg/kg, *Trigonella* at 300mg/kg and *Glycine max* at 100mg/kg show reduction in lipid profile when compare to the PCOS group however, *Sesame indicum* at 300mg/kg is the best of them all.

Table 1 Phytochemical screening result

Samples	Tannins	Saponin	Anthro quinine	Glycosides	Cardia glycoside	Alkaloid	Flavoniod	Phenolic	Steroid
Trigonella foenum graecum	+	+++	-	-	+	+++	+++	+	+
Gycine max	-	+++	-	-	+	+++	+	+	+++
Sesame indicum	-	+	-	-	+++	+++	+	-	-
Lepidium meyenii	-	+	-	-	+	+	-	-	+++
Glycyrrhiza glabra	-	++	-	-	++	+++	-	-	-

Legend
- = Negative
+ = Present in small quantity
++ = Present in moderate amount
+++ = Present in high amount

Citation: Ozioko EN, Ajibade GA, Vantsawa PA, et al. Fertility potentials of five medicinal plants in the treatment of infertility caused by polycystic ovarian syndrome in female wistar albino rats. *Int J Complement Alt Med.* 2022;15(4):213–218. DOI: 10.15406/ijcam.2022.15.00610

Table 2 The glucose analysis done on the rats after treatment with the plant extract for fifteen days

Treatment groups	Glucose
Trigonella spp 300mg/kg	5.10±0.10a
Trigonella spp 100mg/kg	5.50±0.06b
Glycine max 300mg/kg	6.10±0.10c
Glycine max 100mg/kg	7.63±0.12d
Sesamum indicum 300mg/kg	5.67±0.06b
Sesame indicum 100mg/kg	6.03±0.06c
Glycyrrhiza glabra 300mg/kg	6.03±0.15c
Glycyrrhiza glabra 100mg/kg	5.23±0.31a
Lepidium meyenii 300mg/kg	6.03±0.06c
Lepidium meyenii 100mg/kg	7.73±0.06d
PCOS	7.60±0.10d
Normal Control	5.03±0.06a
p value	< 0.0001

Values are given as mean ± standard deviation (SD). In each column, values with different superscripts have statistical significant difference ($p< 0.05$).

Table 3 Lipid profile analysis done on the rat after treatment with the plant extract for fifteen days

Treatment group	Cholesterone	Triglycerides	HDL	LDL
Trigonella spp 300mg/kg	1.57±0.06d	0.49±0.01d	0.87±0.06e	0.29±0.01a
Trigonella spp 100mg/kg	1.87±0.06e	0.32±0.01b	0.59±0.01b	0.39±0.01a
Glycine max 300mg/kg	5.97±0.06f	0.56±0.01fg	0.59±0.01b	0.39±0.01a
Glycine max 100mg/kg	1.50±0.01d	0.54±0.01ef	0.99±0.02f	0.39±0.01a
Sesame spp 300mg/kg	1.19±0.01bc	0.24±0.01fg	0.88±0.01b	0.39±0.49a
Sesame spp 100mg/kg	0.98±0.02b	0.31±0.01b	0.59±0.03e	0.40±0.02a
Glycyrrhiza 300mg/kg	1.38±0.03cd	0.56±0.01fg	0.69±0.01c	0.49±0.01a
Glycyrrhiza 100mg/kg	0.73±0.47a	0.58±0.01gh	0.79±0.01d	0.39±0.02a
Lepidium spp 300mg/kg	1.49±0.02d	0.51±0.00de	0.79±0.02d	0.40±0.01a
Lepidium spp 100mg/kg	1.39±0.01a	0.48±0.07d	0.69±0.12c	0.29±0.01a
PCOS	1.63±0.05de	0.62±0.01h	0.45±0.05a	0.61±0.01a
Normal control	1.45±0.05bc	0.39±0.02c	1.17±0.06g	0.23±0.06a
P Values	<0.0001	<0.0001	<0.0001	0.266

Values are given as mean ± standard deviation (SD). In each column, values with different superscripts have statistical significant difference ($p< 0.05$).

Table 4 Hormonal assay analysis done on the rats after fifteen days of treament with plant extract

Treatment groups	Progesterone	Estradiol	Testosterone	FSH	LH
Trigonella 300mg/kg	6.71±0.18c	48.4±0.07a	5.33±0.29a	49.6±0.03a	58.8±0.05c
Trigonella 100mg/kg	10.7±0.31d	206.8±12.8d	24.2±0.44h	160.3±1.0d	58.5±0.65c
Glycine 300mg/kg	10.2±0.38d	76.2±0.47b	26.1±0.29i	101.1±0.31b	58.5±0.65c
Glycine 100mg/kg	12.2±0.56e	90.7±0.62c	20.7±0.79g	101.8±0.51b	79.1±0.63f
Sesame 300mg/kg	12.7±0.44e	303.9±3.74e	21.2±0.99g	125.0±37.5bc	84.2±0.5g
Sesame 100mg/kg	12.8±0.62e	62.4±1.55b	14.7±0.63e	162.4±3.10d	97.7±1.2h
Glycyrrhiza 300mg/kg	12.7±0.69e	101.7±1.83c	12.0±0.57d	149.9±2.0c	72.8±1.9e
Glycyrrhiza 100mg/kg	20.4±0.89g	402.5±11.3f	16.2±0.32f	94.8±46.8b	66.6±1.7d
Lepidium 300mg/kg	4.14±0.74a	94.8±1.16c	7.10±0.42b	42.1±2.84a	96.9±1.8h
Lepidium 100mg/kg	4.69±0.61a	91.9±1.52c	6.48±0.46b	43.9±1.05a	48.9±0.1b
PCOS	16.9±0.10f	825.9±22.6g	34.6±0.56j	36.6±0.09a	98.6±0.6h
Normal control	5.65±0.02b	35.4±0.02a	8.83±0.03c	36.9±0.67a	45.8±0.3a
P Values	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Values are given as mean ± standard deviation (SD). In each column, values with different superscripts have statistical significant difference ($p< 0.05$).

Table 5 Pregnancy and delivery across all treatment groups

Treatment groups	Rat 1 Latency	Rat 1 Litter size	Rat 2 Latency	Rat 2 Litter size
Normal control	27 Days	5 pups	31 Days	4 pups
Trigonella 300mg/kg	33 Days	4 pups	32 Days	2 pups
Sesame spp 300mg/kg	41 Days	3 pups	45 Days	3 pups
Sesame spp 100mg/kg	73 days	3 pups	75 Days	3 pups

Citation: Ozioko EN, Ajibade GA, Vantsawa PA, et al. Fertility potentials of five medicinal plants in the treatment of infertility caused by polycystic ovarian syndrome in female wistar albino rats. *Int J Complement Alt Med.* 2022;15(4):213–218. DOI: 10.15406/ijcam.2022.15.00610

Table 6 Pregnancy and delivery across all treatment groups second delivery

Treatment groups	Rat 1 Latency	Rat 1 Litter size	Rat 2 Latency	Rat 2 Litter size
Normal control	38 Days	5 pups	38 Days	6 pups
Trigonella 300mg/kg	32 Days	5 pups	35 Days	4 pups
Sesame indicum 300mg/kg	35 Days	3 pups	37 Days	3 pups
Sesame spp 100mg/kg	45 Days	3 pups	50 Days	3 pups

Table 7 Misscarriages that occur across all treatment groups

Treatment group	Day of miscarriage after mating
Trigonella spp 100mg/kg	20
Glycine spp 300mg/kg	25
Glycine spp 100mg/kg	23
Sesame spp100mg/kg	20
Glycyrrhiza spp300/mg	25
Lepidium spp 300mg/ml	25
Lepidium spp 100mg/kg	26

Table 8 Ranking of the plant extract according to their impact on the parameters of PCOS

Plant extract and their ranks	Decrease glucose	Decrease lipid	Decrease number of cyst	Increased FSH	Decreased testosterone	Life birth
Trigonella 300mg/kg 5/6	+	+	+		+	+
Sesamum 300mg/kg 4/6		+	+	+	+	+
Sesamum 100mg/kg 4/6		+	+	+	+	+
Glycyrrhiza 100mg/kg 3/6	+	+		+		
Trigonella 100mg/kg 3/6	+		+	+		
Glycine 300mg/kg 2/6			+	+		
Glycine 100mg/kg 2/6			+	+		
Lepidium 300smg/kg 2/6			+		+	
Lepidium 100mg/kg 2/6			+		+	
Glycyrrhiza 300mg/kg 1/6				+		

Discussion

The phytochemical results obtained for *Glycine max* show abundant presence of Saponins, Alkaliods, Steriods and moderate presence of Cardiac glycosides, Flavoniods and Phenols, this report is in agreement with the results obtained by Salem et al 2018, they made reference to the fact that it is these phytochemicals present in this plant that is responsible for its estrogenic properties. Also, the result obtained from *T. graecum* show abundant presence of Saponins, Alkaliod, and Flavonoid with moderate presence of Tannins, Cardiac glycosides, phenols and steroids,is in agreement with the result obtained by.[7] Alkaliod, saponins and Cardiaic glycosides were found to be present in all five plants extracts. Flavoniod on the other hand was found to be present in only T. *graecum*, *G. max* and *S indicum*, but with very high amount in *T. graecum*. Flavoniod has been reported by many researchers to possess numerous health benefits such as anticancer, antioxidant and above all estrogenic properties.[8] The outcome of this research also buttress the fact that flavoniod possesses estrogenic properties as experimental animals treated with *T. graecum* had more delivery and a very short latency period when compare to the other groups that had prolong latency period and several miscarriages. Increase in glucose level is one of the parameters of polycystic ovarian syndrome; this is because of the increase in androgen level which then leads to insulin resistance and increase in blood sugar. From the result of the glucose level test, *T. graecum* at 300mg/kg reduced the blood sugar level from 7.60mmol/L in the PCOS group to 5.50mmol/L in *T. graecum* to the level of that of the normal control, this report is in agreement with the report of Genet et al.[9] Next after *T. graecum* is *G. glabra* at 100mg/kg followed by *T. greacum* at 100mg/kg. All other plant extract were able to reduce the blood sugar level when compared to the PCOS group, except for *G. max* at 100mg/kg and *L. meyenii* at 100mg/kg.

Increase in cholesterone level is another diagnostic feature of polycystic ovarian syndrome, and it has been attributed by many researchers to obesity, hormonal imbalance and insulin resistance in people with polycystic ovarian syndrome.[10,11] From the results obtained from Lipid profile test, *S. indicum* at 300 and 100mg/kg and *G.glabra* at 100mg/kg showed significant reduction of cholesterone level and increase in High Density Lipoprotein when compare to the PCOS group. *S.indicum* at 300mg/kg is the novel treatment group because it did not just reduced the high cholesterone level, it also increased the HDL and reduced the LDL and Triglyceride, when compared to the PCOS group and these attributes is what makes it the best when it comes to control of Lipid level.

The result of the hormonal assay explains the reason behind every other result obtained in the cause of this research, this is because hormones are the major determinant factors of glucose level, cholesterone level, rate of conception and delivery and miscarriages in patients with polycystic ovarian syndrome. From the results obtained from the hormonal assay, the PCOS group (the group administered letrozole for 21 days without any treatment), when there is increase in LH, there will be an automatic decrease in FSH and an increase in testosterone and Estradiol this fact is in agreement with the report of Rojas et al. 2014, Ricard Lucida (2019) and (Center for Reproduction 2019),[12] this however was not found in *T. graecum* at 300mg /ml as there was a reduced LH, a decreased Testosterone and an increased FSH, this is the reason behind the increased pregnancy and delivery in members of this group and also from the report of the toxicity test carried out by Ozioko et al.,[13] *T. graenum* appear to be the best plant

extract as it was able to reversal the effect of letrozole on the liver and the kidney and show no toxic effect on the liver and kidney. *S. indicum* at 100mg/ml, have moderate LH level but has an extremely high above normal FSH and a reduced Testosterone level, although had a prolong latency period of about 38 days, at least they were able to conceive and have delivery unlike the other groups that conceived and had miscarriages. *Glycine* spp at both 300 and 100mg/ml and *Glycyrrhiza glabra* at both 300 and 100mg/kg were able to conceived because of the high level of FSH and Estradiol, however the high level of testosterone prevent the body from producing the necessary level of progesterone that will sustain the pregnancy to term, this is the actual cause of miscarriages among all the members of these groups and it is for this same reason that when women with PCOS conceive they are placed on progesterone supplement until three month when the placenta is matured enough to start production of progesterone. Members of the group in *L. meyenii* at 300mg/kg and 100mg/kg had miscarriages because of low progesterone level. In modern medical practice, Obstetrician/gynecologist places their patience on several medications that help in addressing each one of these parameters, the reason behind this mode of treatment was made clear by the outcome of this research as each of the plant extract controlled one or two of this parameters however, *Trigonella feonum graenum* plant extract was exceptional as it was able to effectively control five out of the six parameters of PCOS.

Conclution

T. graecum and *G. glabra* show good control over insulin resistance, *S. indicum* and *G. glabra* effectively controlled high cholesterone level, *L.meyenii* also showed effective control of increased Androgen level and hirsitism, ability to increase follicle stimulating hormone was show in *S. indium, T. graecum*, *Glycine max*, and *G.aglabra*, whereas the ability to ovulate, go through a successful gestation period, and deliver live babies was seen in *S indicum* and *T. greacum*, this could be attributed to the high flavoniod (phytoestrogens) seen in the two plant extracts. The best of all the plants extracts is the extract of *T. graenum* because of the continuous fertility recorded in this group and also the none toxic effect on the vital body organs.

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Conflicts of interest

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