Full Length Articles

ALLEVIATIVE EFFECT OF AQUEOUS EXTRACT OF *ASPARAGUS RACEMOSUS* ON THE HAEMATO-BIOCHEMICAL PARAMETERS OF EXPERIMENTALLY INDUCED POLYCYSTIC OVARIAN SYNDROME IN WISTAR RATS

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ABSTRACT

Polycystic ovarian syndrome (PCOS) is a combination of endocrine and metabolic syndrome contributing to both the terms as a clinical feature in women. The present study was conducted to find the alleviative effect of aqueous extract of Asparagus racemosus against the induced PCOS in Wistar rats. About 60 Wistar rats (n=10) are randomly divided into 6 groups: Group 1 (control group), Group 2 (Asparagus racemosus control), Group 3 (PCOS control estradiol valerate 5 mg/rat), Group 4 (estradiol valerate + 1000 mg/kg Asparagus racemosus Prophylactic treatment from day 1 of induction of PCOS), Group 5 (estradiol valerate + 100 mg/kg Asparagus racemosus treatment, after induction of PCOS), Group 6 (Metformin 100 mg/kg). Haematological parameters such as glucose, total protein, albumin, ALT, AST, ALP, triglycerides, cholesterol, C reactive proteins, BUN and creatinine were studied in polycystic ovarian syndrome. Metformin group showed highly significant (P<0.01) reduction in haemoglobin, Packed Cell Volume (PCV) and significant (P< 0.05) reduction in Total Erythrocyte Count (TEC) compared to prophylactic, therapeutic and PCOS groups while PCOS group showed elevation in platelet count (P< 0.05). Significant increase (p < 0.05) in serum glucose, cholesterol and CRP noticed in the PCOS groups compared to prophylactic, therapeutic and metformin groups. The results indicated that Asparagus racemosus had partial alleviating effect on haematobiochemical parameters caused by polycystic ovarian syndrome in Wistar rats.

Keywords: Polycystic ovarian syndrome, haemato-biochemical parameters, *Asparagus racemosus*, Wistar rats.

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INTRODUCTION

Polycystic ovarian syndrome is one of the common hormones related disease among women (Yang *et al.,* 2021). The prevalence of PCOS was 8-13 % (Mansour *et al.*, 2016). Polycystic ovary has various symptoms of ovarian dysfunction caused by disturbed production of GnRH followed by imbalance of FSH and LH. The level of testosterone and LH is increased in PCOS women but FSH level is in normal range (Yang *et al*., 2021). Reduction of FSH and LH leads to irregular menstruation followed by imbalance which causes formation of numerous cysts in the antral follicle. Major clinical signs of PCOS include ovarian cyst, weight gain, irregular menstrual cycle and fertility problems (Strowitzki, 2021). Ovarian cyst formation leads to disturbances in ovulation followed by infertility.

Metformin and clomiphene citrate are the common drugs used for women with PCOS (Leanza *et al*., 2014). Metformin and clomiphene citrate treatment produces several side effects such as diarrhoea, nausea, vaginal/ uterine bleeding, breast tenderness, hot flashes and abdominal pain. Different drugs are in use to restore irregularity of cycle and reduce the hyperandrogenic symptoms and also to treat infertility. At present PCOS is incurable and lifelong treatment required to eliminate the symptoms. The treatment is done with oral contraceptives, progesterone therapy, clomiphene citrate, metformin and gonadotropins (Sidra *et al*., 2019).

Several medicinal plants have been used for women fertility problems. *Asparagus racemosus* is a medicinal plant with worldwide recognition. Shatavari helps in promoting normal development of ovarian follicles, regulates menstrual cycle and revitalizes the female reproductive system mainly due to its phytoestrogen (Kumar *et al*., 2008). *Asparagus racemosus* is used to treat various conditions like nervous disorders, tumours, inflammation, neuropathy and hepatopathy. It also has antiulcer, antioxidant, antidiarrhoeal, immune modulatory and anti-ageing activities. In addition, it increases longevity and improves mental health. Hence, the present study was undertaken to find out the alleviative effect of aqueous extract of *Asparagus racemosus* on haemtao-biochemical parameters of experimentally induced polycystic ovarian syndrome in Wistar rats.

MATERIALS AND METHODS

Animal study

Sixty female 4 – 6 weeks old Wistar rats were obtained from the Laboratory Animal Medicine Unit of Tamil Nadu Veterinary and Animal Sciences University, Madhavaram with the approval (Approval Lr. No. 508/ DFBS/IAEC/2022) from the Institutional Animal Ethics Committee (IAEC). Aqueous extract of *Asparagus racemosus* was purchased from the pharmaceutical company Chemiloids and estradiol valerate was procured from TCI, India (CAS RN: 979-32-8). Control group was given *ad libitum* water and feed. *Asparagus racemosus* extract (1000 mg/kg/day) was administered for 28 days as

oral gavage for group 2. Single intramuscular administration of estradiol valerate (5 mg/ rat) was given for the rats in group 3. In group 4 plant extract was administered orally from the day 1 of administration of estradiol valerate as prophylactic treatment for 28 days. In group 5 plant extract was administrated after the induction of polycystic ovarian syndrome (18^{th} day) and continued up to 28^{th} day. Metformin (100 mg/kg/day) was administered orally in group 6 for 28 days. Animals were sacrificed on the 29^{th} day of trial. Blood samples were collected by retrobulbar plexus method in EDTA and plain vacutainers for haematological and serum biochemical studies respectively.

Haematology

The haematological parameters such as PCV, Haemoglobin (Hb), TEC, Total Leukocyte Count (TLC), Differential Leukocyte Count (DLC) and Platelet count (PLTs) were estimated in the EDTA samples by using auto analyser BC vet 2800 (Mindray).

Serum biochemistry

The blood collected in plain vacutainer with clot activator was allowed to settle for 30 minutes and centrifuged at 2500 rpm for five minutes. The separated serum was collected and subjected to serum biochemical evaluation in A15 BioSystem auto biochemical analyzer **(**BioSystem**).** The parameters such as glucose, total protein, albumin, ALT, AST, ALP, triglycerides, cholesterol, C reactive proteins, BUN and creatinine were estimated.

Statistical analysis

The data generated from the different parameters of the experimental study were subjected to one way analysis of variance (ANOVA) test using statistical package for the social sciences (SPSS) software version 20 of windows.

RESULTS AND DISCUSSION

During the study period, it was observed that there is no appreciable clinical signs and mortality**.** The results of haematological parameters (Table 1) of haemoglobin and PCV showed a highly significant reduction ($P< 0.01$) in the PCOS group compared to the metformin group. Results indicated that PCOS has decreased the haematocrit values as noticed by Simmonds *et al.* (2016). PCV and haemoglobin values reduced in metformin group which indicates that metformin suppresses heme production (Li *et al*., 2019).

A significant ($P<0.05$) elevation in total erythrocyte count in PCOS group compared to treatment and metformin groups was noticed. A significant ($P<0.01$) increase in platelet values in the PCOS group compared to treatment and metformin groups was also observed.

MCV, MCH, MCHC, lymphocytes and monocyte showed no significant difference between the control and treatment group whereas a significant ($P<0.05$) reduction of neutrophil was observed in PCOS group compared to metformin group. White blood

Table 1. Mean (± SE) haematological parameters in 28 days study in estradiol valerate induced PCOS in Wistar rat model (n=10)

	Control	*Asparagus racemosus* control	PCOS Control	Prophylactic group	Treatment Group	Metformin group	F	Sig
Haemoglobin (g/dl)	12.8^{b}± 0.247	12.37^{b}± 0.267	12.45^{b}± 0.262	12.322^{b}± 0.184	12.27^{b}± 0.212	11.32^{a}± 0.312	4.182	**
TEC (x 10^6/µl)	7.68^{b}± 0.157	7.56^{b}± 0.14	7.53^{b}± 0.108	7.514^{b}±0.117	7.32^{b}± 0.163	7.03^{a}± 0.162	2.634	*
PCV (%)	40.7^{c}± 0.732	39.4^{bc}± 0.96	39.12^{bc}± 0.66	39.13^{bc}±.604	37.92^{a}± 0.9	36.26^{a}± 0.8	3.728	**
WBC (x 10^3/µl)	11177.8± 1511.2	11800± 1400.2	10766± 1204.9	11088.89± 898.5	10300± 1280.19	8211.11± 1059.2	1.014	NS
Platelet (x 10^3/µl)	1018.778^{ab}± 48.37	1086.556^{b}± 60.55	1136.333^{b}± 575.00	1159.778^{b}± 98.1	1002.333^{a}± 116.426	817.889^{a}± 72.03	2.449	*
Lymphocyte (%)	74.22± 2.7	79.88±1.58	78.33± 1.64	78.44±1.52	76.77±1.92	73.22±2.7	1.548	NS
Monocyte (%)	4.4± 0.376	3.6± 0.471	4.11± 0.388	3.66±0.33	3.66±0.33	4.22±0.32	0.832	NS
Neutrophil (%)	21.22^{ab} ±2.64	15.33^{a}± 0.707	17.44^{ab} ± 1.46	17.88^{ab} ±1.3	19.55^{ab}±1.7	22.55^{b}±2.5	1.945	*
MCV (fl)	53.14± 0.758	52.13± 0.46	51.96± 0.58	52.08 ±0.214	51.78 ± 0.3417	51.59± 0.347	1.256	NS
MCH (pg)	16.77± 0.139	16.36± 0.087	16.54± 0.309	16.40±0.106	16.80± 0.267	16.11± 0.301	1.379	NS
MCHC (g/dl)	31.58± 0.327	31.45± 0.264	31.83± 0.399	31.49±0.119	32.43±0.46	31.22± 0.18	1.33	NS

Mean bearing similar superscripts do not differ significantly within a row -One-way ANOVA-Duncan's test; NS – Non significant; ** ($P<0.01$),* ($p<0.05$)

Table 2. Mean (± SE) serum biochemistry parameters in 28 days study in estradiol valerate induced PCOS in Wistar rat model (n=10)

	Control group	*A. racemosus* group	PCOS control Group	Prophylactic group	Treatment Group	Metformin group	F	S
Glucose (mmol/L)	161 ^{ab} ±24.43	134.66 ^{ab} ±48.8	172.66^{b}±32.16	169^{a}±17.34	171.33 ^{ab} ±13.9	159 ^{ab} ± 5.13	2.044	*
Total protein (g/dL^{-1})	6.76± 0.26	6.63±0.24	6.366±0.16	6.20±0.1	6.73±0.484	6.36± 0.06	0.800	NS
Albumin (g/dL^{-1})	3.76± 0.133	4.93±0.08	4.9±0.11	3.80±0.05	4.06±0.08	3.96± 0.0.08	1.265	NS
ALT (U L^{-1})	67.66± 4.9	57.66±6.8	68.66±9.3	66.66±7.2	54.33±5.2	77.66±20	0.625	NS
AST (U L^{-1})	196^{cd}±5.5	181^{bc}±11.06	144.33^{a}±6.3	153 ^{ab} ±3.51	177^{bc}± 6.08	219.33^{d}± 18.33	7.708	**
ALP (U L^{-1})	125.33 ^{bc}±19.35	180^{b}±37.3	57.33^{a}±8.2	104 ^{ab} ±17.05	50^{a}±2	91.6 ^{ab} ±14.5	5.855	**
Cholesterol (mmol/L)	56^{a}±3.2	73.66 ^{ab} ±3.75	77.33 ^{b} ±3.84	63.33 ^{ab} ±12.1	77^{b} ±3.6	63.33 ^{ab} ±1.2	2.292	*
Triglyceride (mmol/L)	165.33± 11.09	194.667±43.8	237.6±41.68	185.66±17.3	195.6±82.1	173±7.54	0.692	NS
BUN (mg/dL^{-1})	16.64± 0.94	15.53±0.86	18.01±0.63	17.29±1.53	18.23±0.99	78.37±0.53	1.278	NS
Creatinine (mg/dL^{-1})	0.49± 0.08	0.32±.017	0.41 ±0.04	0.4 ±0.023	0.51±0.043	0.42±0.014	2.643	NS
CRP (mg/dL)	6.96 ^{ab} ± 0.83	7.65 ^{ab} ±0.84	8.8 ^{b} ±0.37	7.04 ^{ab} ±1.14	6.72 ^{ab} ±0.79	5.62 ^{a}±0.74	1.658	*

Means bearing similar superscripts do not differ significantly within a row -One-way ANOVA-Duncan's test; NS –Non significant; S-Significant *(P<0.05), ** (P<0.01)

cell count decreased in the PCOS group as reported by Barath *et al.* (2022) could be the cause of reduction of neutrophil.

Significant (P<0.05) increase in the serum glucose value in PCOS group was noticed in comparison to the control and prophylactic group which might be due to metabolic disorder of glycolysis/ gluconeogenesis (Huang *et al*., 2020). There is highly significant reduction (P< 0.01) in serum liver enzymes such as AST and ALP in the PCOS induced group compared to control group, which could be due to altered enzyme metabolism. Total protein, albumin, ALT, BUN and creatinine levels showed no significant difference between the control and treatment group. Lipid profile values of cholesterol showed significant elevation (P< 0.05) in the PCOS group compared to the prophylactic groups. The values were found to be decreased in the prophylactic groups as reported by Haslan *et al*. (2021) in which significant changes in the lipid profile values of letrozole induced PCOS rats was observed. Serum triglyceride values showed no significant difference between the control and treatment groups.

Elevation of CRP value was observed in the PCOS group compared to control and treatment groups and the levels were reversed in the treatment groups of *Asparagus racemosus* which indicated that *A. racemosus* possess reproductive function restorative and antioxidant properties (Pandey *et al*., 2018).

It is concluded that *Asparagus racemosus* might have partial alleviative effect on the altered haemato-biochemical parameters induced by polycystic ovarian syndrome in Wistar rats.

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