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Research Article

Effect of Acacia Nilotica on FSH, LH and Prolactin Levels of Female Albino Rats

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Abstract

Introduction: Acacia Nilotica is one of famous herb use in Sudan and other many countries all over the world as a medicinal herb in the treatment of wide diversities of medical and health problems. Most of hazards of uses of medicinal herbs not detected, discovered, or even known from the experiences of users of the communities. This study aimed to investigate the effect of using different extract concentrations of Acacia Nilotica on FSH, LH and Prolactin levels in female Albino rats.

Material and Methods: This was experimental study in which aqueous extract of Acacia Nilotica administrated to three groups of albino rats for 60 days in doses (25, 50 and 100 mg/ml) the fourth group was control. To determine the levels of FSH, LH and Prolactin special rats ELISA kits used according to the manufacturer's instructions. Statistical analysis performed by using SPSS (IBM version 25), descriptive statistics and ANOVA were employed.

Results: FSH mean values were (8.48 ± 1.15 mlU/ml) for control group, (6.56 ± 1.07, 5.58 ± 0.7 and 4.54 ± 0.46 mlU/ml) for treated groups (B, C and D) respectively. LH hormone mean values were 14.11 ± 1.26 mlU/ml for control group, (10.13 ± 0.52, 7.58 ± 1.07 and 4.82 ± 0.2 mlU/ml) for groups B, C and D respectively. Prolactin hormone mean values were (9.3 ± 0.93 ng/ml) for control group, (7.11 ± 0.59, 4.84 ± 0.46 and 2.83 ± 0.47 ng/ml) for the other groups B, C and D respectively. Group D showed the lowest values of mean, minimum and maximum for all tested hormones. FSH, LH and prolactin levels showed significant decrease with increase of Acacia concentration ($P < 0.000$).

Conclusion: Aqueous extract of Acacia Nilotica in three different concentrations used in this study. The significant decrease in levels of FSH, LH and prolactin hormones was according to dose concentration, it might indicate that Acacia may affect the pituitary gland and alter the secretion of these hormones, even further investigations needed.

Keywords: Acacia Nilotica; FSH; LH; Prolactin; Female Reproductive System

Introduction

Medicinal herbs use in traditional medicine is the sum total of knowledge, skills and practices based on the theories, beliefs and experiences indigenous to different cultures that are used to maintain health, as well as to prevent, diagnose, improve or treat physical and mental illnesses. Many people believe that because herbal medicines natural or traditional they are safe (or carry no risk for harm). Increased patient awareness about safe usage is important, as well as more training, collaboration and communication among providers of traditional and other medicines [1]. Studies reveal that uses of the traditional medicine drugs may lead to hazards and side effects that cause changes in the body systems [2,3]. However, 70 to 80% of people Worldwide rely chiefly on traditional, largely herbal, medicines to meet their primary healthcare needs. The global demand for herbal medicine is not only large but growing [4].

Acacia Nilotica is easily accessible source of natural antioxidants, which can be used as supplement to aid the therapy of free radical mediated diseases [5,6]. The pods of Acacia Nilotica contain gallocatechin 5-0-gallate, methyl gallate, gallic acid, catechin 5-0-gallate,1-0-galloyl-β-D-glucose,1-6-di-0-galloyl-β-D-glucose and digallic acid, therefore can be promoted for usage in pharmaceutical and nutraceutical products in near future [7]. In addition, pods of Acacia Nilotica contain sufficient mineral constituents Cu and Zn is low in concentration. Ca followed by K showed highest concentration, and concentration of P about (12.3 × 103 mg\g). The ethyl acetate extract of Acacia Nilotica showed significant activity against K. aerogenes and P. mirabilis [8]. Furthermore, Acacia Nilotica leaf improves insulin resistance and hyperglycemia associated acute hepatic injury and nephrotoxicity by improving systemic antioxidant status in diabetic mice [9]. Studies have confirmed anti-diabetic activities [10]. Glucose lowering effect may be due to presence of phenols flavonoids, alkaloids, tannin, phylobatanins and saponins [11]. The aqueous stem bark extracts of Acacia Nilotica at high dose of 1 g/kg body weights, which is far from the therapeutic dose tends to cause toxicological effects [12]. Moreover, both methanolic and aqueous extracts of different parts of the plant showed anti-hypertensive activities [13,14].

Antifertility activity of Acacia Nilotica has been reported, aqueous extract of dried pods at the dose of 200 mg/kg when administrated to male Wistar rats showed significantly reduction in sperm motility, progressive motility and sperm concentration. In addition, testosterone level decreased in treated rats. Consequently, histopathology of reproductive organs showed that dose of Acacia Nilotica disrupted seminiferous tubule architecture and spermatogenesis process. However, the plant severely affects male animal fertility parameters even it not yet clear whether its antifertility effects are reversible [15]. Moreover, the aqueous extract of Acacia Nilotica has demonstrated contraceptive properties by interrupting sperm transportation through the reduction in progressive motility; the contraceptive properties have been demonstrated *in vitro* by inducing premature spontaneous acrosome reaction, which could affect sperm-egg interaction. *In vivo* studies would confirm the contraceptive properties of Acacia Nilotica [16].

Acacia Nilotica is one of famous herb use in Sudan and many countries all over the world as a medicinal herb in the treatment of wide diversities of medical and health problems. Most of the hazards of uses of medicinal herbs not detected, discovered, or even known from the experiences of users of the communities. Therefore, this research highlights uses of medicinal herbs particularly the safeness of uses Acacia Nilotica regarding female reproduction.

Materials and Methods

In this experimental study about 48 Adult female wistar rats obtained from the Animal Research Center of King Khalid University, Abha, Saudi Arabia. All animals kept under a photoperiod of 12 h light: 12 h darkness with automatically regulated temperature (22-23 Co) and free access to water and food allowed. This study carried out in accordance with the local regulations set, by which follow the regulations of laboratory animal care and use published by the US National Institutes of Health (NIH publication No. 85-23, revised 1996).

For our experiments, Acacia Nilotica fresh pods dried in shade and crushed using a blender, and then, always freshly dissolved overnight in distilled water for 12 hours. Different concentrations of the extract were prepared based on the experimental procedure. Accordingly, animals divided into four groups (n = 12/group)

- **Group A:** received distilled water only set as control group.
- **Group B:** received the aqueous extract of Acacia Nilotica at a final concentration of 25 mg/ml,
- **Group C:** received the aqueous extract of Acacia Nilotica at a final concentration of 50 mg/ml and
- **Group D:** received the aqueous extract of Acacia Nilotica at a final concentration of 100mg/ml.

Treatment continued for 60 days on daily basis and all treatments were daily and freshly prepared. Special rats ELISA kits were used to measure serum levels of Prolactin (Cat. No. ab113351, Cambridge, UK). Follicular stimulating hormone (FSH) (Cat. No. ab108641, Cambridge, UK). Luteinizing hormone (Cat. No. CSB-E12654r, Cusabio Biotech Co., Ltd., China).

A statistical analysis for all obtained parameters performed by using SPSS (IBM version 25). Data presented as means with their standard Deviation (mean ± SD). Normality and homogeneity of the data confirmed before ANOVA and the differences of significance among the groups assessed by one-way ANOVA followed by LSD test for multiple comparison.

Results and Discussion

Descriptive statistic including mean, minimums and maximums for each separate group of Acacia Nilotica treated animals (A control, B 25mg/ml, C 50mg/ml, D 100mg/ml). Group D showed the lowest values of mean, minimum and maximum for all tested hormones table 1 and figure 1. In addition, the mean decreases as the dose of Acacia Nilotica increases.

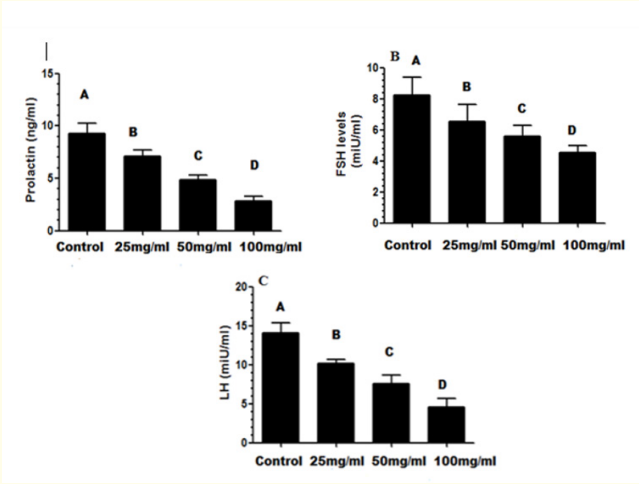

Figure 1: Levels of Prolactin, Follicular stimulating hormone (FSH) and Luteinizing hormone (LH) for all groups of rats. Values expressed as Mean ± SD for 12 rats/group. Values considered significantly different at $P < 0.05$.

Hormones	Groups	N	Minimum	Maximum	Mean	Std. Deviation
FSH	A	12	7.12	10.06	8.4800	1.14514
	B	12	4.24	7.94	6.5583	1.06952
	C	12	4.59	6.76	5.5742	0.69696
	D	12	3.88	5.18	4.5400	0.45941
LH	A	12	12.83	16.51	14.1050	1.25806
	B	12	9.44	11.25	10.1275	0.51967
	C	12	6.36	10.12	7.5775	1.06623
	D	12	3.80	6.36	4.8233	0.91827
Prolactin	A	12	8.46	11.85	9.3017	0.92783
	B	12	6.08	8.23	7.1142	0.58773
	C	12	4.08	5.85	4.8350	0.46348
	D	12	2.23	3.92	2.8267	0.47429

Table 1: Levels of FSH, LH (mlU/ml) and Prolactin (ng/ml).

There is significant and dose concentration dependent regarding levels of FSH, LH, and Prolactin as showed in table 2.

The maximum decreases in serum levels of FSH seen with the extract at 100mg/ml. In Multiple Comparisons, table 3 there is a significant different in level of FSH in Group A comparing to other groups ($P < 0.000$). All groups show significant differences to each other supporting that mean difference increase according to dose concentration.

Hormones		Sum of Squares	df	Mean Square	F	Sig.
FSH	Between Groups	101.316	3	33.772	42.858	0.000
	Within Groups	34.672	44	.788		
	Total	135.988	47			
LH	Between Groups	560.401	3	186.800	194.947	0.000
	Within Groups	42.161	44	.958		
	Total	602.562	47			
Prolactin	Between Groups	282.818	3	94.273	229.087	0.000
	Within Groups	18.107	44	.412		
	Total	300.924	47			

Table 2: ANOVA test for FSH, LH and Prolactin.

* The mean difference is significant at the 0.05 level.

Groups of the study	Groups of the study	Mean Difference	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
A	B	1.92167*	0.3624	0.000	1.1913	2.6520
	C	2.90583*	0.3624	0.000	2.1755	3.6362
	D	3.94000*	0.3624	0.000	3.2096	4.6704
B	A	-1.92167*	0.3624	0.000	-2.6520	-1.1913
	C	.98417*	0.3624	0.009	.2538	1.7145
	D	2.01833*	0.3624	0.000	1.2880	2.7487
C	A	-2.90583*	0.3624	0.000	-3.6362	-2.1755
	B	-.98417*	0.3624	0.009	-1.7145	-.2538
	D	1.03417*	0.3624	0.007	.3038	1.7645
D	A	-3.94000*	0.3624	0.000	-4.6704	-3.2096
	B	-2.01833*	0.3624	0.000	-2.7487	-1.2880
	C	-1.03417*	.3624	0.007	-1.7645	-.3038

Table 3: Multiples Comparisons between groups regarding FSH.

* The mean difference is significant at the 0.05 leve0l.

The maximum decreases in serum levels of LH seen with the extract at 100mg/ml. In Multiple Comparisons, table 4 there is a significant different in level of LH in Group A comparing to other groups ($P < 0.000$). All groups show significant differences to each other supporting that mean difference increase according to dose concentration.

The maximum decreases in serum levels of Prolactin seen with the extract at 100mg/ml. In Multiple Comparisons, table 5 there is a significant different in level of LH in Group A comparing to other groups ($P < 0.000$). All groups show significant differences to each other supporting that mean difference increase according to dose concentration.

Groups of the study	Groups of the study	Mean Difference	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
A	B	3.97750*	0.39963	0.000	3.1721	4.7829
	C	6.52750*	0.39963	0.000	5.7221	7.3329
	D	9.28167*	0.39963	0.000	8.4763	10.0871
B	A	-3.97750*	0.39963	0.000	-4.7829	-3.1721
	C	2.55000*	0.39963	0.000	1.7446	3.3554
	D	5.30417*	0.39963	0.000	4.4988	6.1096
C	A	-6.52750*	0.39963	0.000	-7.3329	-5.7221
	B	-2.55000*	0.39963	0.000	-3.3554	-1.7446
	D	2.75417*	0.39963	0.000	1.9488	3.5596
D	A	-9.28167*	0.39963	0.000	-10.0871	-8.4763
	B	-5.30417*	0.39963	0.000	-6.1096	-4.4988
	C	-2.75417*	.399630	.0000	-3.5596	-1.9488

Table 4: Multiples Comparisons between groups regarding LH.

* The mean difference is significant at the 0.05 level.

Groups of the study	Groups of the study	Mean Difference	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
A	B	2.18750*	0.26189	0.000	1.6597	2.7153
	C	4.46667*	0.26189	0.000	3.9389	4.9945
	D	6.47500*	0.26189	0.000	5.9472	7.0028
B	A	-2.18750*	0.26189	0.000	-2.7153	-1.6597
	C	2.27917*	0.26189	0.000	1.7514	2.8070
	D	4.28750*	0.26189	0.000	3.7597	4.8153
C	A	-4.46667*	0.26189	0.000	-4.9945	-3.9389
	B	-2.27917*	0.26189	0.000	-2.8070	-1.7514
	D	2.00833*	0.26189	0.000	1.4805	2.5361
D	A	-6.47500*	0.26189	0.000	-7.0028	-5.9472
	B	-4.28750*	0.26189	0.000	-4.8153	-3.7597
	C	-2.00833*	0.26189	0.000	-2.5361	-1.4805

Table 5: Multiples Comparisons between groups regarding Prolactin.

* The mean difference is significant at the 0.05 level.

Discussion

Pituitary gland hormones that regulate normal female reproductive system are affected, follicle stimulating hormone (FSH) analysis in this study shows gradual decreases with the increase concentrations of Acacia extracts (25mg/ml B, 50mg/ml C and 100mg/ml D groups as compared with control group (A). In addition, luteinizing hormone (LH) level decreased with increase of the concentration of the doses of Acacia extract. Prolactin hormone lev-

el had steady decrease from 25mg/ml group B to 50mg/ml group C then 100mg/ml group D as compared with control group A.

The results of the study reported that doses of 25, 50 and 100 mg/ml of Acacia Nilotica affect FSH, LH and Prolactin levels of female rats. Although data regarding the effects of Acacia Nilotica on female reproductive system were lacking, experimental studies showed that alcohol even in moderate consumption disrupts female puberty by affecting hormonal levels [17] as we reported regarding Acacia treated animals. However, extract of Acacia Nilotica in high doses may prevent pregnancy or interfere with the development of the conceptus.

Acacia Nilotica affect male Wistar rats' fertility by lowering sperm motility, progressive motility and sperm concentration in addition, testosterone levels [15]. Furthermore, it has contraceptive properties in males, demonstrated *in vitro* by inducing premature spontaneous acrosome reaction, which could affect sperm-egg interaction [16]. Moreover, Acacia Nilotica disrupted seminiferous tubule architecture and spermatogenesis process in doses of 200 mg/kg [15]. Hereby it seemed to be high doses and concentrations affect both males and females' fertility depending on experimental studies.

Conclusion

Aqueous extract of Acacia Nilotica in three different concentrations used in this study. The significant decrease in levels of FSH, LH and prolactin hormones was according to dose concentration, it might indicate that Acacia may affect the pituitary gland and alter the secretion of these hormones, even further investigations needed. However, inappropriate use of Acacia Nilotica may cause infertility issues and may interfere with hormonal stimulation in assisted reproduction cycles. Consequently, it may affect female puberty in case of uncontrolled used pre-puberty.

Conflict of Interest

No conflict of interest exists.

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