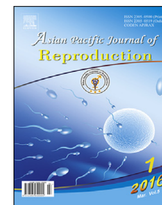
Contents lists available at ScienceDirect

Asian Pacific Journal of Reproduction

journal homepage: www.apjr.net



Original research http://dx.doi.org/10.1016/j.apjr.2015.12.009

Phytochemical screening and evaluation of anti-fertility activity of *Dactyloctenium aegyptium* in male albino rats

B. Sreedhar Naik[1*], Nim Bahadur Dangi[1], Hari Prasad Sapkota[1], Nabin Wagle[1], S. Nagarjuna[1], R. Sankaranand[2], B. Anantha kumari[3]

[1]*Division of Pharmacology, Raghavendra Institute of Pharmaceutical Education and Research, (RIPER), Krishnam Reddy Palli Cross, Chiyyedu, Anantapuramu 515721, Andhra Pradesh, India*

[2]*Lusaka Apex Medical University, Kasama Road, Lusaka, Zambia*

[3]*SK University, Anantapur, India*

ARTICLE INFO

Article history:
Received 10 Jun 2015
Received in revised form 17 Oct 2015
Accepted 25 Oct 2015
Available online 17 Dec 2015

Keywords:
Antifertility
Dactyloctenium aegyptium
Testosterone
Sperm count
Sperm motility
Anti implantation

ABSTRACT

Objective: To find out the effect of ethanol extract of *Dactyloctenium aegyptium* (*D. aegyptium*) on the fertility of male albino rats.
Methods: Experimental animals were divided in four groups of six each. Group first received normal saline and served as control. The second, third and fourth groups of animals were administered with ethanolic extract of *D. aegyptium* extract at a dose of 200, 400, and 600 mg/kg body weight, respectively for a period of 30 days.
Results: A non significant increase in the bodyweight and a significant decrease in weight of testes, accessory sex organs, and reduction in sperm count, increase motility and abnormality were observed. Some serum biochemical parameters showed significant variations and were as the serum hormonal levels are significantly decreased.
Conclusions: The decreased levels of sperm count, weight of reproductive organs, serum hormonal levels and number of implantations in female rats reveals the antifertility activity of *D. aegyptium* and it was with dose dependent manner.

1. Introduction

Population control is an issue of global and national public health concern. Birth control is an essential part of our life. A variety of synthetic contraceptive agents are available in the market only for women's and their use is associated with severe side effects, the progress and possibilities on male are still slow and limited [1]. The World Health Organization suggested that practice of usage of traditional medicine for the control of fertility, instead of synthetic drugs, as cost effective management for Birth control [2]. *Dactyloctenium aegyptium* (*D. aegyptium*) is a common coarse grass belonging to Poaceae family [3], found commonly throughout India, Nepal, Pakistan, Afghanistan, Israel, Lebanon, Turkey, Sri Lanka, Malaysia, Myanmar, Philippines, China, Japan, Singapore, Thailand, Vietnam, New Guinea, Algeria, Morocco, Egypt, Sudan, Tunisia and Libya. It is reported to have astringent, bitter tonic, anti-anthelmintic and used to treat gastrointestinal, biliary, urinary ailments, polyurea, fevers, smallpox, heart burn, immunodeficiency, urinary lithiasis, spasm of maternity, renal infections, gastric ulcers, wounds healing and the seeds are used by tribesmen to prepare liquor as well as famine food with unpleasant taste [4] and it is also used against cough [5]. And, it also found that plant also reported to have antimicrobial activity [5], diuretic, antipyretic, and anti-proliferative activity [6]. The main objective of the present study is to evaluate antifertility activity of ethanolic extract of *D. aegyptium*.

2. Materials and methods

2.1. Procurement and identification of plant materials

Plant of *D. aegyptium*, family Poaceae, was collected from Madanapalle, chittoor district of A.P. The plant was identified and authenticated by Dr. J. Raveendra Reddy, M.Pharm Ph.D., Raghavendra Institute of Pharmaceutical Education and

*Corresponding author: Sreedhar Naik B., Division of Pharmacology, Raghavendra Institute of Pharmaceutical Education and Research, (RIPER) Krishnam Reddy Palli Cross, Chiyyedu, Anantapuramu 515721, Andhra Pradesh, India.
Tel: +91 9676942197, +91 8554 255646.
E-mail: bsreedhar07@gmail.com
Peer review under responsibility of Hainan Medical College.

2305-0500/Copyright © 2015 Hainan Medical College. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Research, Ananthapuramu and the voucher specimen **(02/15)** was preserved in the department of Pharmacology, RIPER, for further reference.

2.2. Preparation of extract

The whole plants of *D. aegyptium* were dried in shade. Powdered using a grinder and sieved. Weighed quantity (1 000 g) of fresh, finely grounded powder was mixed with ethanol (95%) and subjected to maceration with intermittent shaking for seven days. The extract then obtained was collected by filtration using muslin cloth. Thus obtained filtrate was subjected to solvent evaporation to obtain solid extract which was weighed and stored in an air tight container.

2.3. Preliminary phytochemical screening

Freshly prepared ethanolic extract was subjected to quantitative analytical tests for the detection of various chemical constituents like carbohydrates, proteins, amino acids, saponins, flavonoids, tannins, terpenoids and alkaloids [6].

2.4. Animals

Male Albino Wistar Rats weighing 200–250 g were used for the present study. The animals were collected from Central Animal Facilities, Indian Institute of Science, Bangalore. The animals were maintained under controlled conditions of temperature (22 ± 2 °C), humidity [(50 ± 5)%] and 12 h light–dark cycles. All the animals were acclimatized for seven days before the study. The animals were housed in sanitized polypropylene cages containing sterile paddy husk as bedding. They had free access to standard pellets as basal diet and water *ad libitum*. All the studies conducted were approved by the Institutional Animal Ethical Committee (IAEC) of Raghavendra Institute of Pharmaceutical Education and Research (RIPER), Ananthapuramu. Approval No. 878/ac/05/CPCSEA/003/2015 dated 06/01/2015, according to prescribed guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India.

2.5. Acute toxicity study of the extract

The test performed according to OECD guidelines 423. Swiss albino mice were administered with ethanolic extract up to 2 000 mg/kg. Animals were observed for gross behaviour changes as well as for motility for 14 days [7].

2.6. Treatment protocol

The animals were equally divided into four groups (6/group). Group I animal were given normal saline for 30 days and they served as control. Group II, III, and IV animals received ethanolic extract at dose levels of 200, 400, 600 mg/kg body weight daily per oral for 30 days.

2.7. Determination of body and reproductive organ weight

The initial and final body weight of animals was recorded. Blood samples were collected by retro orbital puncture, then the testes, epididymis, vas deferens, seminal vesicle and ventral prostate were dissected out, freed from adherent tissue and weighed.

2.8. Sperm concentration, motility and abnormality

The caput and cauda regions of epididymis was chopped separately in two petridishes and 1 mL of normal saline at temperature of 36 °C was added to the semen to enhance sperm survival *in-vitro* during the period of the study. The semen mixture was then sucked into a red blood pipette to the 0.5 mark, diluted with warm normal saline, sucked up to the 101 mark. The normal saline at the stem of the pipette was discarded and the content was mixed thoroughly. A drop of the semen mixture was placed on the neubauer counting chamber which spread under the cover slip by capillary action. The chamber was mounted on the stage of microscope, viewed under the magnification of ×40 and counted and expressed in million per mL [8]. A drop of the sperm saline mixture was taken in a separate glass slide. One slide was covered with a cover slip and examined under the microscope. Sperm motility at the caudal epididymis was then assessed by calculating the motility spermatozoa per unit area. A smear was made on another slide and total morphological abnormalities were observed [9].

2.9. Serum biochemical profile

Serum protein, albumin, globulin, urea, creatinine and liver marker enzymes like SGPT, SGOT, and ALP was estimated by using standard procedure [10–13].

2.10. Estimation oxidative parameters in blood serum

Blood were collected from retro orbital method and serum was separated by using micro-centrifuge.

2.10.1. Catalase (CAT)

Two milliliter of serum diluted with 1 mL of H_2O_2 and take the absorbance at 240 nm for 3 min with 30 s intervals. (Add H_2O_2 just before taking O.D) [Aebi-1984].

2.10.2. Reduced glutathione (GSH)

The serum was precipitated with 20% trichloro acetic acid (TCA) and centrifuged. 0.25 mL supernatant was taken for GSH estimation using freshly prepared DTNB solution (2 mL) and volume up to 3 mL with phosphate buffer (pH 8). The intensity of the yellow colour formed was read at 412 nm against blank for each sample without reagent was run. The GSH content was calculated by using $\varepsilon = 13.6 \times 10^3\ M^{-1}\ cm^{-1}$ expressed as nmol/g serum [14].

2.10.3. Superoxide dismutase (SOD)

0.5 mL of serum was diluted with 0.5 mL distilled water, to this add all chilled reagents, 0.25 mL ethanol, 0.5 mL of chloroform and shaken for 1 min and centrifuged at 2 000 rpm for 20 min supernatant enzymatic activity was determined to it 0.05 mL of carbonate buffer (0.05 M pH 10.2) and 0.5 mL EDTA (0.49 M) was added. The reaction was initiated by addition of 0.4 mL epinephrine and the change in optical density/mm was measured at 480 nm. SOD was expressed as U/mg protein [15].

Table 1
Effect of extract of *D. aegyptium* on body weight and reproductive organs weight.

Groups	Body weight (g)		Testis (g)	Epididymis (mg)		VD (mg)	SV (mg)	Prostate (mg)
	Before	After		Caput	Cauda			
Group I (Normal)	220.000 ± 6.325	252.000 ± 15.940	3.288 ± 0.415	768.10 ± 18.13	762.50 ± 12.50	240.00 ± 10.00	1050.00 ± 10.00	240.00 ± 10.00
Group II (200 mg/kg)	230.000 ± 10.000	270.000 ± 13.040	1.955 ± 0.3244^{a}	440.10 ± 39.90^{b}	467.00 ± 33.00^{b}	166.50 ± 13.50^{a}	941.5 ± 8.50^{b}	166.50 ± 13.50^{ns}
Group III (400 mg/kg)	250.000 ± 10.370	292.000 ± 4.899	1.604 ± 0.1596^{b}	325.10 ± 24.87^{b}	336.60 ± 13.40^{c}	105.00 ± 5.00^{b}	772.50 ± 7.50^{c}	105.00 ± 5.00^{ns}
Group IV (600 mg/kg)	250.000 ± 4.472	274.000 ± 10.300	1.061 ± 0.066^{c}	226.60 ± 26.63^{c}	310.40 ± 10.40^{c}	94.00 ± 4.00^{b}	726.00 ± 4.00^{c}	94.00 ± 4.00^{a}

All values are expressed as mean ± SEM, one-way ANOVA followed by Tukey's multiple comparison test. $^{a, b, c \& ns}$ indicates $P < 0.05$, $P < 0.01$, $P < 0.001$ and non-significant, respectively when compared to normal control.

2.11. Fertility of male albino rats

Fertility was estimated in adult male rats treated with whole plant ethanolic extract of *D. aegyptium* and in the control male counterparts. Each male rat was placed in an individual cage with two virgin untreated females rats of the same strain. They were left together for 10 days during which two oestrone cycles had elapsed in female rats. And after 10 days the exposed male rats are removed, pregnant females were killed by cervical dislocation under light ether anaesthesia and the number of pregnant rats, implantation sites, and the number of foetuses was recorded [16].

2.12. Sex hormones and pituitary gonadotropins in male albino rats

Blood samples were collected by retro orbital puncture. Blood was centrifuged at 3 000 rpm to separate the serum for the measurement of testosterone, Luteinizing (LH), oestrogen and follicle stimulating hormone (FSH). The quantitative determination of hormones was done by using Enzyme Immuno Assay method (EIA).

2.13. Histopathology of testis

After the experiment rats were sacrificed by cervical dislocation. Their testis were removed and weighed individually. For the evaluation of histopathological changes in testis, it was separated and immediately fixed with 10% formalin. Thereafter, the specimens were embedded in paraffin, sectioned at 5 µm and stained with haematoxylin and eosin.

2.14. Statistical analysis

The analysis of data was done by using one way analysis of variance (ANOVA), followed by Tukey's multiple comparison test by using Graphpad prism 5.0 software (Graphpad, San Diego, CA).

3. Results

3.1. Extraction

Extraction was carried out by maceration technique by using ethanol and percentage yield of extract was found to be 11.6%.

3.2. Preliminary phytochemical screening

Ethanolic extract revealed the presence of carbohydrates, proteins, amino acids, saponins, flavonoids, tannins, terpenoids and alkaloids.

3.3. Acute toxicity studies

For ethanolic extracts, no toxicity was found up to 2 000 mg/kg. Hence extract was selected as lower dose, intermediate dose and higher dose, i.e., 200 mg/kg, 400 mg/kg & 600 mg/kg-p.o, of body weight, respectively.

3.4. Effect of ethanolic extract of D. aegyptium on body weight and reproductive organs weight

The final body weight of rats of all groups showed no significant increase in body weight when compared with initial

Table 2

Effect of extract of *D. aegyptium* on sperm concentration, motility and abnormality in the epididymis of adult male albino rats.

Group	Sperm concentration (counts × 10^6 mil)		Sperm motility (FMI) @ (Cauda)	Sperm abnormality (%)	
	Caput	Cauda		Head	Tail
Group I (Normal)	273.000 ± 1.732	390.300 ± 6.064	114.700 ± 2.906	6.333 ± 1.453	7.333 ± 1.453
Group II (200 mg/kg)	225.000 ± 2.887[c]	361.700 ± 7.265[a]	77.000 ± 1.732[b]	9.333 ± 2.028[ns]	9.333 ± 0.882[ns]
Group III (400 mg/kg)	205.000 ± 2.887[c]	347.000 ± 1.528[b]	50.330 ± 5.487[c]	16.330 ± 2.028[a]	13.000 ± 0.577[a]
Group IV(600 mg/kg)	190.000 ± 5.774[c]	338.300 ± 4.410[c]	40.000 ± 5.774[c]	20.670 ± 0.881[b]	15.000 ± 1.000[b]

All values are expressed as mean ± SEM, one-way ANOVA followed by Tukey's multiple comparison test. [a, b, c & ns] indicates $P < 0.05$, $P < 0.01$, $P < 0.001$ and non-significant, respectively when compared to normal control.

Table 3

Effect of extract of *D. aegyptium* on serum biochemical parameters.

Parameter	Group I	Group II	Group III	Group IV
Protein (g/dL)	8.267 ± 0.393	6.433 ± 0.233[ns]	6.033 ± 0.636[a]	5.667 ± 0.353[a]
Albumin (g/dL)	3.433 ± 0.260	3.667 ± 0.167[ns]	3.533 ± 0.273[ns]	5.437 ± 0.173[b]
Globulin (g/dL)	2.400 ± 0.231	3.400 ± 0.265[a]	3.600 ± 0.173[a]	3.933 ± 0.0882[b]
Creatinine (mg/dL)	0.900 ± 0.0577	0.533 ± 0.145[ns]	0.667 ± 0.0882[ns]	0.600 ± 0.173[ns]
SGOT (U/L)	250.300 ± 5.783	265.000 ± 5.774[ns]	283.300 ± 8.819[a]	304.300 ± 5.364[b]
SGPT (U/L)	57.000 ± 1.732	52.000 ± 1.732[ns]	51.330 ± 0.666 [ns]	57.000 ± 0.577[ns]
ALP (U/L)	260.300 ± 1.000	258.000 ± 0.666 [ns]	265.000 ± 2.887[ns]	251.700 ± 3.756[ns]

All values are expressed as mean ± SEM, one-way ANOVA followed by Tukey's multiple comparison test. [a, b & ns] indicates $P < 0.05$, $P < 0.01$ and non-significant, respectively when compared to normal control.

body weights. A significant decrease in weight of testis, epididymis, i.e. caput and caudal, vas deferens, seminal vesicle and prostate were noted in all treatment groups when compared with Group I animals and are shown in Table 1.

3.5. Effect of ethanolic extract of D. aegyptium on sperm concentration, motility and abnormality

A significant reduction of total sperm count and increase in motility, abnormality of sperm in caput and caudal was observed in all treatment groups when compared to Group I are shown in Table 2.

3.6. Effect of ethanolic extract of D. aegyptium on serum biochemical parameters

The serum biochemical parameters like protein, albumin, globulin, creatinine and liver marker enzymes like SGOT, SGPT, ALP of control and treated rats were represented in Table 3.

3.7. Effect of extract of D. aegyptium on oxidative parameters

The results in Table 4 showed a significant decrease in SOD, catalase, GSH when compared to Group I animals represented in Table 4.

3.8. Histopathology of testis

Group-I (Normal control): Section of testis of normal rat showing spermatogenic activity in the seminiferous tubules, normal population of the leydig cell (H & E ×400) (Figure 1).

Group-II (200 mg/kg): Section studies shows that slightly reduction in the diameter of seminiferous tubules, with reduced layering, with less spermatozoa, showing mild hyper-cellularity of leydig cells (H & E ×400).

Group-III (400 mg/kg): Section studies shows that significantly reduction in the diameter of seminiferous tubules, immature and less no. of spermatozoa, showing severe hyper-cellularity of leydig cells and presence of large multinucleated cells (H & E ×400).

Group-IV (600 mg/kg): Section studies shows that significantly lesions of seminiferous tubules, significantly reduction in the diameter of seminiferous tubules, immature and less no. of spermatozoa, showing severe hyper-cellularity of leydig cells and presence of large multinucleated cells (H & E ×40).

3.9. Effect of ethanolic extract of D. aegyptium on sex hormones and pituitary gonadotropins in male albino rats

The administration of ethanolic extract of *D. aegyptium* showed significant decrease in serum testosterone levels and

Table 4

Effect of extract of *D. aegyptium* on oxidative parameters.

S.No.	Group	SOD (units)	CAT (μ moles of H_2O_2 decomposed/min/mg protein)	GSH (nmol GSH/mg of protein)
01	Group I (Normal)	33.000 ± 1.528	9.333 ± 0.882	13.330 ± 0.882
02	Group II (200 mg/kg)	24.670 ± 0.8819[a]	6.000 ± 0.577[ns]	9.667 ± 0.882[a]
03	Group III (400 mg/kg)	20.670 ± 1.764[b]	5.333 ± 0.882[a]	7.333 ± 0.882[b]
04	Group IV(600 mg/kg)	19.330 ± 1.333[c]	4.000 ± 0.577[b]	6.333 ± 0.333[b]

All values are expressed as mean ± S.E.M ($n = 6$). One-way ANOVA followed by Tukey's multiple comparison test. [a, b, c & ns] indicates $P < 0.05$, $P < 0.01$, $P < 0.001$ and non-significant, respectively when compared to normal control.

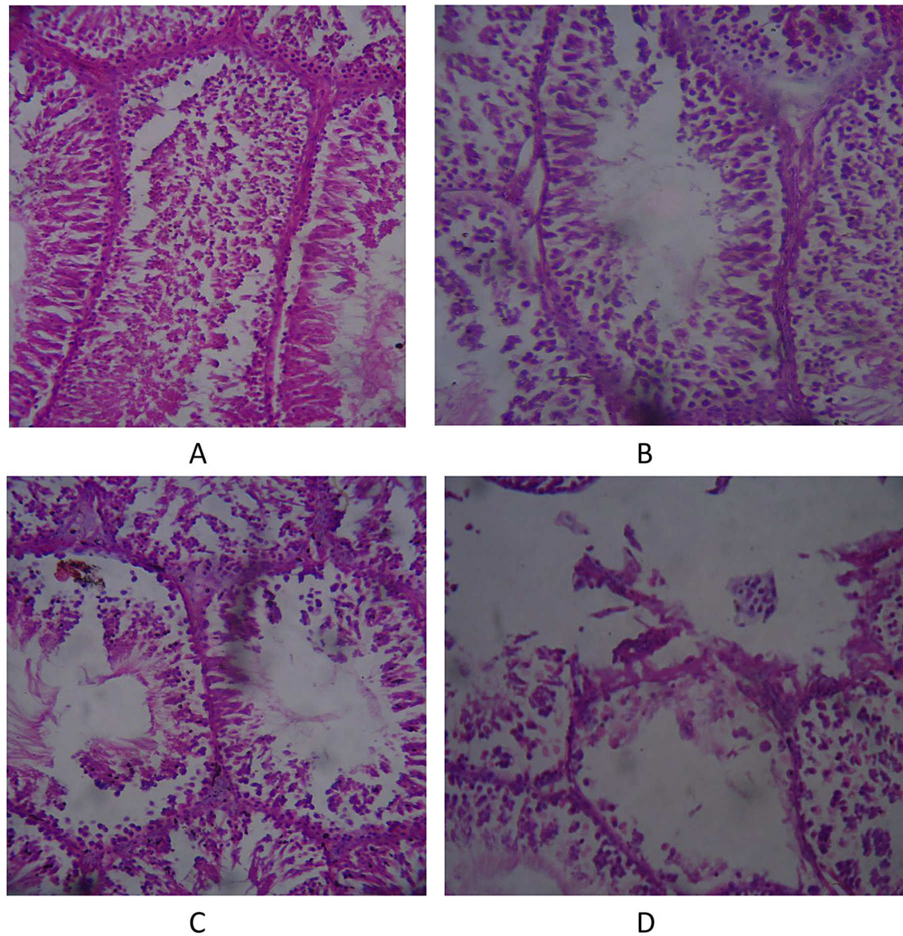


Figure 1. Histopathology of testis where A, B, C & D indicates normal control, 200 mg/kg, 400 mg/kg and 600 mg kg, respectively (H & E ×400).

Table 5
Effect of extract of *D. aegyptium* on sex hormones and pituitary gonadotropins in male albino rats.

Group	Parameters			
	Testosterone (ng/dL)	LH (mIU/mL)	Estrogen (pg/mL)	FSH (mIU/mL)
Group I (Normal)	3.367 ± 0.353	3.500 ± 0.289	26.000 ± 1.155	0.250 ± 0.0231
Group II (200 mg/kg)	2.067 ± 0.260[a]	2.333 ± 0.441[ns]	29.000 ± 0.577[ns]	0.200 ± 0.0231[ns]
Group III (400 mg/kg)	1.433 ± 0.233[b]	2.900 ± 0.0577[ns]	33.000 ± 1.155[b]	0.123 ± 0.0186[a]
Group IV (600 mg/kg)	1.067 ± 0.0882[c]	2.200 ± 0.153[a]	33.330 ± 0.882[b]	0.127 ± 0.0176[a]

All values are expressed as mean ± SEM, one-way ANOVA followed by Tukey's multiple comparison test. [a, b, c & ns] indicates $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively when compared to normal control.

increase in serum estrogen levels when compared to Group I. The pituitary gonadotropins levels in serum i.e. LH and FSH showed slight decrease in their levels compared to Group I animals, as represented in Table 5.

3.10. Effect of extract of *D. aegyptium* on fertility of male albino rats

The administration of ethanolic extract of *D. aegyptium* showed dose dependent decrease in number of pregnant females and number of foetuses represented in Table 6 and Figure 2.

4. Discussion

For a developing country, the lack of basic education and access to health care services, limits male involvement in family planning. Scientists are trying to develop male contraceptive from plants which will be more acceptable for economic and safety reasons. Our present studies showed that *D. aegyptium* has antifertility effect on male reproductive system of rats and caused morphological alteration of spermatozoa which was reflected through significant reduction in fertility rate. The result of this investigation demonstrated that the ethanolic extract of *D. aegyptium* showed no significant increase in body weight of male albino rats which are administered orally for 30 days. And, also found that there is a significant decrease in weight of testis and other accessory sex organs along with the reduction in sperm count of epididymis, increase motility and abnormality of sperm. The reduction of the weight of testis and other accessory organs might be due to low levels of androgen, which was not enough to maintain the weight of gonads and other accessories [16]. It is known that the accessory sex organs via, epididymis

Table 6
Effect of extract of *D. aegyptium* on the fertility of male albino rats.

Group	No. of males	No. of females	No. of pregnant females	No. of implantation
Group I (Normal)	6	12	11/12 (91.66%)	11.670 ± 1.202
Group II (200 mg/kg)	6	12	8/12 (66.66%)	7.000 ± 0.577[a]
Group II (400 mg/kg)	6	12	5/12 (41.66%)	5.000 ± 1.155[b]
Group IV(600 mg/kg)	6	12	2/12 (16.66%)	3.000 ± 0.577[c]

All values are expressed as mean ± SEM, one-way ANOVA followed by Tukey's multiple comparison test. [a, b, c & ns] indicates $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively when compared to normal control.

and vas deference are androgen dependent target organs and manifest differential sensibility to androgens for maintenance of the structure and function. It is also known that any change in circulatory androgens would affect the internal environment of epididymis and thereby lead to alteration in sperm motility and metabolism [17].

The development of normal and matured sperm is the key to optimum male fertility. The production of sperm cell (spermatozoa) and testosterone in testis are mainly regulated by FSH and LH which are released from anterior pituitary [18]. FSH stimulates spermatogenesis in the steroli cell, while the LH stimulates the production of testosterone in the leydig cells of testis [19]. The result of the present study suggested that ethanolic extract of *D. aegyptium* for 30 days may affect the normal function of the steroli and leydig cells. Sexual cells can occur during the reproductive phase, mitotic division of spermatogonia or during maturation of spermatozoa, there by affecting the no. of quantity of sperm cell production in the testis. The ethanolic extract of the treated group II, III and IV (200, 400 and 600 mg/kg body weight) produced a significant reduction in sperm count and viable sperm. This may be as a result of the ability of extract at given doses, to either interfere with spermatogenetic process in the seminiferous tubules, epididymal function or activities of testosterone on hypothalamic release factor and anterior pituitary secretion of gonadotropin which may result in alteration of spermatogenesis [20,21].

Since male reproductive toxicology and male contraceptive are two side of the same coin, the negative consequence of *D. aegyptium* on the sperm may be taken advantage for further study. By the treatment employed in the study, no toxic effect was produced in the liver and kidney and reduction levels of SOD, catalase, reduced glutathione might be due to the excess production of anions in response to the extract. The present study revealed a decrease in the sperm level of testosterone this is due to the decrease serum levels of LH/ICSH observed in this investigation. Leydig cells secret testosterone by the stimulatory effect of LH [21,22]. In males reduction of testosterone level may impair spermatogenesis and cause male infertility. This study further observed significant increase in the estrogen level. This

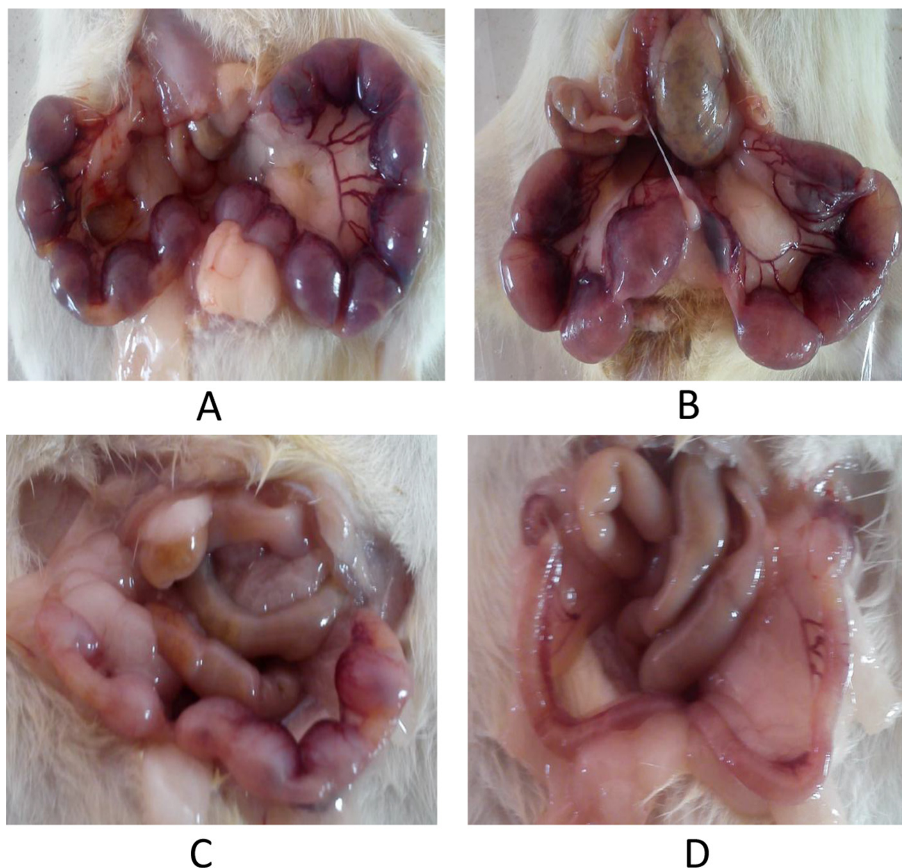


Figure 2. Showing no. of implantation where A, B, C & D indicates normal control, 200 mg/kg, 400 mg/kg and 600 mg/kg, respectively.

increase might be due to the conversion of testosterone to estrogen [23,24].

In addition, the number of implantations and the number of viable foetuses were also decreased, this reflect and may be due to the decreased in sperm motility and sperm density in the study. The preliminary phytochemical screening of the extract showed the presence of carbohydrates, proteins, amino acids, saponins, flavonoids, tannins, terpenoids and alkaloids. Literature shows that presence of saponins, flavonoids, tannins, alkaloids possess antifertility activity [25].

Histopathologically, the ethanolic extract of *D. aegyptium* caused a drastic and dose dependent decrease in the number of spermatozoa, reduction in diameter of seminiferous tubules, and severe hyper-cellularity of leydig cells.

The decreased levels of sperm count, weight of reproductive organs, serum hormonal levels and no. of implantations in female rats reveals the antifertility activity of *D. aegyptium* when compared with normal group and it was in dose dependent manner. However further research is needed to isolate and identifying the pure compound which is responsible for antifertility activity.

Conflict of interest statement

We declare that we have no conflict of interest.

References

[1] Joshi SC, Sharma A, Chaturvedi M. Antifertility potential of some medicinal plants in males: an overview. *Int J Pharm Pharm Sci* 2011; **3**: 204-217.

[2] Umadevi M, Kumar PS, Bhowmik D, Duraivel S. Medicinal plants with potential antifertility activity. *J Med Plants Stud* 2013; **1**(1): 26-33.

[3] Khare CP. *Indian medicinal plants: an illustrated dictionary*. Berlin: Springer Science & Business Media; 2007.

[4] Janbaz KH, Saqib F. Pharmacological evaluation of *Dactyloctenium aegyptium*: an indigenous plant used to manage gastrointestinal ailments. *Bangladesh J Pharmacol* 2015; **10**(2): 295-302.

[5] Abdallah Emad M, El-Ghazali Gamal E. Screening for antimicrobial activity of some plants from Saudi folk medicine. *Glob J Res Med Plants Indig Med* 2013; **2**(4): 189-197.

[6] Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. Phytochemical screening and extraction: a review. *Int Pharm Sci* 2011; **1**(1): 98-106.

[7] Nagarjuna S, Gopala Krishna Murthy TE, Srinivasa Rao A. Antidiabetic activity of different solvent extracts of *Dactyloctenium aegyptium* in streptozotocin induced diabetic rats. *Res J Pharm Biol Chem Sci* 2015; **6**(3): 485-493.

[8] Singh RP, Dhanalakshmi S, Rao AR. Chemomodulatory action of *Aloe vera* on the profiles of enzymes associated with carcinogen metabolism and antioxidant status regulation in mice. *Phytomedicine* 2000; **7**(3): 209-219.

[9] Linder RE, Strader LF, Slott VL, Suarez JD. Endpoints of spermatotoxicity in the rat after short duration exposures to fourteen reproductive toxicants. *Reprod Toxicol* 1992; **6**(6): 491-505.

[10] Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein measurement with the Folin phenol reagent. *J Biol Chem* 1951; **193**(1): 265-275.

[11] James S, Bilbiss L, Muhammad B. The effects of *Catharanthus roseus* (L) G. Don 1838 aqueous leaf extract on some liver enzymes, serum proteins and vital organs. *Sci World J* 2007; **2**(1); http://dx.doi.org/10.4314/swj.v2i1.51700.

[12] Reitman S, Frankel S. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am J Clin Pathol* 1957; **28**(1): 56.

[13] King EJ, Armstrong AR. A convenient method for determining serum and bile phosphatase activity. *Can Med Assoc J* 1934; **31**(4): 376.

[14] Ellman GL. Tissue sulfhydryl groups. *Arch Biochem Biophys* 1959; **82**(1): 70-77.

[15] Beauchamp C, Fridovich I. Superoxide dismutase: improved assays and an assay applicable to acrylamide gels. *Anal Biochem* 1971; **44**(1): 276-287.

[16] Sharma N, Jocob D. Antifertility investigation and toxicological screening of the petroleum ether extract of the leaves of Mentha arvensis L. in male albino mice. *J Ethnopharmacol* 2001; **75**(1): 5-12.

[17] Khan P, Awasthy K. Cytogenetic toxicity of neem. *Food Chem Toxicol* 2003; **41**(10): 1325-1328.

[18] Steinberger E. Hormonal control of mammalian spermatogenesis. *Physiol Rev* 1971; **51**(1): 1-22.

[19] Kerr J, De Kretser D. Cyclic variations in Sertoli cell lipid content throughout the spermatogenic cycle in the rat. *J Reprod Fertil* 1975; **43**(1): 1-8.

[20] Bowman W, Rand M. The reproductive system and drugs affecting the reproductive systems. *Textb Pharmacol* 1985; **20**: 1-8.

[21] Udoh FV, Udoh PB. Hepatotoxicity of the methanol extract of Carica papaya (PawPaw) seeds in Wistar rats. *Pharm Biol* 2005; **43**(4): 349-352.

[22] Udoh F, Udoh P, Umoh E. Activity of alkaloid extract of *Carica papaya*. Seeds on reproductive functions in male Wistar rats. *Pharm Biol* 2005; **43**(6): 563-567.

[23] Carr B. Neuroendocrinology of reproduction. In: *Textbook reproductive medicine*. Norwalk: Appleton and Lange; 1993, p. 157.

[24] Chinoy N, Padman P. Antifertility investigations on the benzene extract of Carica papaya seeds in male albino rats. *J Med Aromat Plant Sci* 1996; **18**(3): 489-494.

[25] Okon UA, Etim BN. Citrus aurantifolia impairs fertility facilitators and indices in male albino wistar rats. *Infertility* 2014; **19**: 20.