

Effect of Date Palm Pollen Grains (*Phoenix Dactylifera*) on Testes Function and Fertility in Rats

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Summary

This study presents the effects of Date Palm Pollen grains (DPP) (*Phoenix dactylifera*) on testes function, fertility and pregnancy index in male adult rats. Forty adult male albino rats were divided randomly into four equal groups. The first group was not received DPP and considered as control group and the other groups (T1, T2 and T3) were received daily gavages of aqueous suspension (DPP) containing 30, 60 and 120 mg/kg B.W for 56 days. At the end of experiment blood were collected to determine serum testosterone level. Sexual reaction was determined and reproductive indices (fertility, pregnancy) index were examined in normal fertile 32 female rats mated with treated male rats. Sperm concentration, mortality and morphology of treated groups were, also determined. The results showed that treatment with DPP for 56 days led to significant ($P < 0.05$) increase in serum testosterone concentration, testicular weights, mortality and sperm concentration in (T2 and T3) groups compared with control and T1 groups, but sperm morphology, concentration was significantly decrease in the T2 + T3 groups compared with control and T1 groups. Also, treatment with DPP cause significant ($P \leq 0.05$) increase in fertility and pregnancy index for all treated groups, as compared with control group, furthermore, sexual reaction was significantly ($P \leq 0.05$) decrease in the 3 treated groups compared with control group. conclusion, DPP suspension plays a role in improvement sperm quality and enhances fertility in adult rats.

Key word :- Date Palm Pollen Grains (*Phoenix Dactylifera*) , Testes Function , Fertility in Rats
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تأثير حبوب لقاح التمر الحلو في وظيفة الخصية وخصوبة الجرذان البالغة

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الخلاصة

تناول هذا البحث دراسة تأثير المعلق المائي لحبوب اللقاح لنخيل التمر الحلو على وظيفة الخصى ودليل الخصوبة والحمل في الجرذان البالغة. استعمل 40 ذكر جرذ بالغ بعمر 6-8 اسابيع قسمت عشوائياً الى اربعة مجاميع متساوية المجموعة الاولى لم تعامل بالمعلق المائي لحبوب اللقاح واعتبرت مجموعة سيطرة، اما المجموع الثلاث الاخرى فقد تم تجريعها بـ 30 ، 60 ، 120 ملغم/كغم من وزن الجسم على التوالي من المعلق المائي لحبوب اللقاح ولمدة 56 يوما على التوالي. من قلب الحيوان مباشرة تم جمع نماذج الدم لغرض قياس تركيز هورمون الشحمون الخصوي في مصل الدم. وتم فحص وقت الاثارة الجنسية ودليلي الخصوبة والحمل بالنسبة للاناث السليمة بعد مزاجتها مع الذكور المعاملة بحبوب اللقاح وبعد التضحية بالحيوانات تم قياس تركيز النطاف والنسبة المئوية للنطاف اللاسوية من بربخ الخصى اليسرى. اوضحت النتائج ان المعاملة أدت الى زيادة معنوية في تركيز هورمون الشحمون الخصوي ووزن الخصى في مجموعتي الجرعة مقارنة بمجموعة السيطرة و كما حصلت زيادة معنوية في النسبة المئوية لحركة النطاف ومعدل تركيزها في مجموعتي المعاملة مقارنة بمجموعة السيطرة، اما النسبة المئوية للنطاف اللاسوية ومعدل الاثارة الجنسية فقد انخفض في جميع مجاميع المعاملة مقارنة بمجموعة السيطرة، من جانب اخر فقد حصلت زيادة واضحة في معدل دليل الخصوبة والحمل في المجاميع الثلاثة المعاملة مقارنة بمجموعة السيطرة. نستنتج من هذه الدراسة بأن المعلق المائي للقاح نخيل التمر الحلو أدى إلى تحسين نوعية النطاف وخصوبة ذكور الجرذان البالغة وزيادة معدلات الحمل وعدد المواليد.

Introduction

Pollen grains carry the male genetic material, suspension of *phoenix dactylifera* date palm. Pollen (DPP) is an herbal mixture that is widely used as a folk remedy for treatment of sexual incapacity and weakness in human and animals (1 and 2). Pollen grains of date palm were also used to promote and enhance fertility in women in ancient Egypt (3). It has been suggested that pollen grains contain a non-crystalline estrogenic substance which involved in the regulating the renewal of spermatogonial stem cells (4), and improve male infertility (2 and 5). Reports have also pointed that isolation of microelements from DPP has estrone, sterols and other agents that may influence male fertility (6, 7 and 8). The literature does show little reports on its effects in sperm parameters or fertility index. Therefore, the present study was designed to determine the effect of DPP on the testis, few sperm parameters and fertility index of adult male rats.

Materials and Methods

The DPP was collected from date palms in the Najaf area south of Baghdad during April and May 2008 . The pollen grain were dried at 50 C° before utilized. Forty sexually mature 6-8 weeks old male albino rats were used in the present study. The animals were housed in stainless steel cages in a room temperature 22-25 C°, with 12 hours dark/ light cycle and had access to food and water *adlibitum*. After the adaptation period, the rats were randomly divided into four equal groups .

Control Group: Animals of this group were received orally 1ml daily distilled water by gavage's needle.

Treated groups T1,T2 and T3

Animal of these groups were received 1 ml of suspension of DPP in distilled water orally contain 30,60 and 120 mg /kg B.W. respectively daily for 56 days.

At the end of experiment the animals, were anesthetized by 1/m injection of ketamine 90 mg/kg B.W., xylozine 40 mg/kg B.W. Blood samples were obtained via cardiac puncture, centrifuged, and then serum samples were stored in freezer at -18 C° for measurement of testosterone hormone by using immunoassay technique using testosterone kits (BIOMERIEUX- Paris / FRANCE). The analysis, were archived in radioactive isotope clin-laboratory at Al-Kindy street- Baghdad.

Sperm Collection : After the animals were sacrificed , the tail of the left epididymus were exposed and embedded in one ml of normal saline at 37C° in a glass watch, before the tail was sliced into at least 200 sections by /special microsurgical scissor. Sperm motility were performed according to (9), sperm morphology (10), sperm viability (11) and sperm concentration (12) .

Latency of Copulation: After the end of treatment 32 virgin female rats (8 females per each group) were examined to detect estrus cycles, before mixed with treated males in one cage and the time span from the moment of introducing male into female cage, until the first trail of copulation were measured. This time was considered as latency of copulation (13). While the reproductive indices were studied according to (14), which includes fertility and pregnancy index.

Statistical Analysis: Results are expressed as mean $\pm$ SE. statistical analysis of data was performed on the basis of Chi square (X^2), and one- way analysis of variance. Group differences were determined using least significant difference (LSD) test at ($P < 0.05$) (15).

Results and Discussion

The effect of different doses (30, 60 and 120mg/kg B.W.) of DPP on testicular weight, sperm parameters were demonstrated in table (1). There is a significant increase ($P<0.05$) in testes weight in the rats treated with DPP concentration of 60 and 120 mg/kg B.W. compared with 30 mg/kg B.W. and control group, this effect might be due to increase of testosterone concentration in the two treated groups (T2, T3) which has an important role in improvement the weight of reproductive organs (16), and stimulation of anabolic metabolism by stimulation of body tissues tantelize glaeer (17). Also this effect may be due to the presence of gonadotropin like substances or/and steroidal component (5 and 8). Also table (1) showed that DPP concentration of 60 and 120 mg/kg B.W. led to significant increase in testosterone level in rats serum, this effect may be due to properties of DPP which contain flavonoid component (10 and 11) which have positive effect on testes and seminal vesicle weight and activity in the rats due to fluid resorption effect of estradiol which improved testosterone and fertility.

The sexual reaction time table (1) is significantly ($P<0.05$) improved in all treated groups than that of control group, this decrement in time may be due to the effect of DPP active ingredient on centers associated with libido and/or due to the a significant increase in testosterone serum concentration. Present result agreed with suggested reported by (20,21 and 23) there is a strong relationship of testosterone with male libido. As well as The testosterone effects neurobehavior function such as sexual arousal, aggression, emotional tone and cognition. Sperm concentration, motility and viability were improved due to oral administration of various DPP concentration, a significant differences were observed in animals groups (T2, T3) compared with T1 and control groups (table 2). The results of table 2 agreed with suggestion of (15 and 16). that improvement of sperm parameters may be due to estradiol and flavonoid component of DPP which have positive effects on sperm quality as well as (18) reported that flavonoid as antioxidant and its role as scavengers has the main important effects on the sperm parameters, also the presence of phytoestrogen as a steroidal component. The suspension of DPP reduced the sperm DNA denaturation and improve its DNA quality the sperm chromatin stability could be improve sperm quality (19). The result of fertility and pregnancy index (table 3 and 4) showed that DPP suspension of non treated females rats mating with treated male rats reflect significant ($P\leq 0.05$) increase. The improvement of sperm quality (mortality, morphology and testosterone hormone) is most important in determining by fertilization rate (23) and for evaluating male fertility (24).

Table (1): Effect of date palm pollen (DPP) on testes weight, Testosterone level and sexual reaction time. $\pm$ S.E, n=10 rats /group.

Groups	Control group	T1 group	T2 group	T3 group
Parameters				
Testis weight (gm)	1.44 $\pm$ 0.03 A	1.46 $\pm$ 0.11 A	1.80 $\pm$ 0.10 B	1.92 $\pm$ 0.07 B
Testosterone conc. (mg/ml)	0.80 $\pm$ 0.04 A	0.86 $\pm$ 0.02 A	1.92 $\pm$ 0.02 B	1.88 $\pm$ 0.03 B
Sexual reaction time (min)	2.16 $\pm$ 1.02 A	1.88 $\pm$ 0.01 B	1.32 $\pm$ 0.07 B	1.16 $\pm$ 0.14 B

Capital letter denote significant differences between groups ($P<0.05$).

Table (2): Effect of date palm pollen (DPP) on sperm parameters. $\pm$ S.E,n = 10 rats /group

Groups Parameters	Control group	T1 group	T2 group	T3 group
Sperm conc. ($\times 10^4$)	6310.2 $\pm$ 17.2 A	6580 $\pm$ 15.2 A	721.2 $\pm$ 2.01 B	811 $\pm$ 16.2 C
Sperm motility (%)	66.2 $\pm$ 5.2 A	70.40 $\pm$ 4.2 A	81.06 $\pm$ 5.1 B	96.18 $\pm$ 4.6 B
Sperm morphology (%)	20.16 $\pm$ 2.3 A	20.22 $\pm$ 1.15 A	16 $\pm$ 1.32 B	11.14 $\pm$ 2.18 C

Capital letter denote significant differences between groups ($P < 0.05$)

Table (3): Effect of date palm pollen (DPP) on fertility index (%) in non treated female rats.(n=8/group).

Groups Parameters	Control group	T1 group	T2 group	T3 group
No. of females mated successfully	8	8	8	8
No. of pregnant animals	5	6	8	8
Non- pregnant animals	3	2	0	0
Fertility index	62.5 %	75 %	100 %	100 %

Capital letter denote significant differences between groups ($P < 0.05$)

Table (4): Effect of date palm pollen (DPP) on pregnancy index (%) in non treated female rats. n = 10 rats /group

Groups Parameters	Control group	T1 group	T2 group	T3 group
No. of pregnant rats	5	6	8	8
No. of pregnant rats gives life baibes	4	4	8	8
pregnancy Index	80 %	66 %	100 %	100 %

Capital letter denote significant differences between groups ($P < 0.05$).

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