

The Effect of *Cyperus esculentus* on Sperm Function Parameters in Prepubertal Mice as a Model for Human

Essraa M. A. Al Essawe*

Amaal A.M.Almashhadani *

Date of acceptance 1/3 / 2010

Abstract

The objective of this work was to study the effect of oral administration of *Cyperus esculentus* (CE) and its alcoholic extract on sperm function parameters in prepubertal mice as a model for human. The animals were divided into three groups each contains 6 animals. Group 1 was treated with 150 mg/kg body weight /day of crude CE, group 2 was treated with same dose of alcohol extract of CE and group 3 regarded as control throughout six weeks period. The results showed a significant ($p < 0.05$) increase in the mean of sperm concentration, sperm motility percent and progressive sperm motility between treated groups and control. There was no differences among groups in the mean of sperm normal morphology and sperm viability. No significant differences was recorded in the mean of body weight among groups throughout the study. The results revealed that the administration of *Cyperus esculentus* may enhance certain sperm characters in prepubertal mice without affecting body weight.

Key words: *Cyperus Esculentus*, sperm, concentration, motility, mice

Introduction

Cyperus esculentus (Chufa Sedge, Yellow Nut sedge, Tiger nut Sedge, Earth almond) is a member of the grass family Cyperaceae to which nut sedge weeds also belong. This plant was originally native to the Mediterranean region but its cultivation has now spread to many other warm countries [1]. The oldest cultivated plants in Ancient Egypt. Chufa was no doubt an important food element in ancient Egypt during dynastic times, [2]. The tubers are edible, with a slightly sweet, nutty flavour, The nuts are either consumed raw or in the dried form, mainly as a snack [3,4]. The tubers are consumed either a nut or grated. They are quite hard and are generally soaked in water before they can be eaten, thus making them much softer and giving them a better texture [4]. They were used to make cakes in ancient Egypt

and used for ice cream and beverage making. Tiger nuts have excellent nutritional qualities with a fat composition similar to olives and a rich mineral content, especially phosphorus and potassium. The oil of the tuber was found to contain 18% saturated (palmitic acid and stearic acid) and 82% unsaturated (oleic acid and linoleic acid) fatty acid [5]. The tubers contain about 25% oil, which are resistant to peroxidation, 50% digestible carbohydrates, 4% protein and 9% crude fiber [6,7]. Recently, a university study has identified a time window between eight and 12 weeks of fetal development during which reproductive problems, including low sperm count, are determined. [8] and due to nutritional qualities of *Cyperus esculentus* the objective of the present study is to find out the effect of oral administration of its on sperm function

parameters in prepubertal mice as a model for human.

Materials and methods:

Prepubertal male Balb/C mice (35 days old) were placed in an air conditioned room at 22 – 24 °C with light period of 14 hr. The mice were divided into three groups (control group, treated groups G1 and G2). Group 1 was administered with drinking water 150 mg/kg BW/day of crude *Cyperus esculentus* after grinding and soaked in warm water. Groups 2 was administered with drinking water same dose of alcohol extract of *Cyperus esculentus* for 6 weeks. While the control group was given distilled water. The dose used for human by (herbalist) was estimated to be about 100 mg- 200 mg / KgBW / day (i.e., 1 to 2 tea spoonful three times daily), according to that, the same dose was calculated for mice [9]. Body weight for each mouse was recorded at the beginning, during and end of the experimental period, using a mechanical balance (Tefesa, Germany). The animals of each group were sacrificed by cervical dislocation at the end of the experiment. The epididymis were quickly freed from the attached fat and connective tissue, the tail of epididymis was minced, using fine and sharp microsurgical scissor in to 200 small pieces in a small well of Petri-dish containing 0.5ml of normal saline, until getting a homogenized solution which contained the spermatozoa and incubated at 37°C until examination [10], then 10 µL drop of this solution was put on a warm clean slide and protected by cover slip (22x 22 mm) to be examined under a high power magnification of 40X objective [11,12], to estimate sperm concentration, sperm motility % and

progressive sperm motility %. The mean number of spermatozoa in 10 random microscopically fields were considered. Eosin nigrosin stain used for viability & abnormality estimation by mixed 10 µL drop spermatozoal solution +2 drops of 1 % eosin stain after 30 second 3 drops of 10% nigrosin added and mixed, a drop of mixture placed on a microscope slide and smear prepared within 30 second (count at least 100-200). **Statistical analysis** Statistical analysis was performed using SPSS (Statistical Package for Social Science; Version 7.5). Crude data analysis was done using paired sample t test for tables with mean and standard error of mean (S.E.M.). The differences between the values were considered statistically significant at ($P < 0.05$) [13].

Results :

The effect of orally administration of crude *Cyperus esculentus* and alcohol extract of it in a dose of (150 mg/kg BW/day) on certain sperm function parameters of prepubertal male mice was shown in table 1. There was a significant $P < 0.05$ increase in the mean of sperm concentration, sperm motility percent and progressive sperm motility between treated groups and control. There was no differences among groups in the mean of sperm normal morphology and sperm viability. No significant differences $P > 0.05$ was recorded in the mean of body weight among groups through out the study table 2.

Table 1: The effect of (150 mg/kg BW/day)orally administration of crude plant and alcoholic extract of(*Cyperus esculentus*) on Sperm Function parameters of pre pubertal male mice.

Groups	Sperm Function parameters				
	Sperm concentration ($\times 10^6 \times \text{ml}$)	Sperm motility (%)	Progressive sperm motility (%)	Normal sperm morphology (%)	sperm viability (%)
Control group	12.6 \pm 2.18	17.00 \pm 4.63	13.00 \pm 2.0	85.26 \pm 2.65	84.88 \pm 2.46
Treated group(1)	33.75 \pm 11. 61 *	63.75 \pm 8.003 *	57.00 \pm 10.40 *	87.17 \pm 1.61	88.40 \pm 1.57
Treated group(2)	28.00 \pm 3.39 *	63.00 \pm 6.819 *	54.0 \pm 6.965 *	82.93 \pm 3.31	89.55 \pm 2.23

* $P < 0.05$ a significant differences from the control group Number of animals per each group = 6

Table 2: Changes in the body weight associated with daily administration of (150 mg/kg BW/day) crude plant and alcoholic extract of(*Cyperus esculentus*) to prepubertal male mice.

Groups	Body weight (g) mean \pm SE			
	weight before treatment	weight after 2week treatment	weight after 4week treatment	weight after 6week treatment
Control group	16.411 \pm 2.00	22.155 \pm 2.292	24.706 \pm 1.882	26.57 \pm 1.38
Treated group(1)	15. 94 \pm 1.102	19.701 \pm 1.18	25.137 \pm 0.694	26.387 \pm 0.868
Treated group(2)	16.711 \pm 0.446	22.43 \pm 1.453	25.103 \pm 1.535	25.308 \pm 1.462

Number of animals per each group = 6

Discussion :

In this investigation ,it has been found that oral administration of crude plant and alcoholic extract of *Cyperus esculentus* significantly enhanced certain sperm function parameters such as sperm concentration ,sperm motility percent and progressive sperm motility . *Cyperus esculentus* locally known as Hhabb el aziz which used to treated male infertility and increased sperm count [14]. On the other hand it affects semen volume as well as testes weight, vassdeferens, testis length, right testis circumference, epididymal length, and weight of reproductive tract [15] although the percentage of motile

spermatozoa and their progressiveness usually give a good indication on sperm quality and are important in predicting mammalian fertility[16]. The seeds of *Cyperus esculentus* established as a very nutritious[17,18]. Rich mineral content, especially phosphorus and potassium, oil resistant to peroxidation and fatty acids (palmitic acid , stearic acid, oleic acid and linoleic acid) [5,6,7].In addition to its anti-inflammatory properties upon inflammation, and immunostimulatory effects [19]may play an important role in spermatogenesis and enhancement of sperm motility parameters with out affecting sperm morphology and

viability. Even though high nutritional quality of *Cyperus esculentus* the result of this investigation recorded no significant differences in the mean of body weight this may be due to low protein content[5,18]. In conclusion the results revealed that the administration of *Cyperus esculentus* in this dose enhanced certain sperm characters in prepubertal mice without affecting body weight, so it may be recommended to use small amounts from this sedges as food supplementation in prepubertal stage to improve reproductive system maturity.

References:

1. Mokady, S.H. and Dolev. A. 1970. Nutritional evaluation of tubers of *Cyperus esculentus* L. J Sci Fd Agric.(21):211-14.
2. Zohary, D. and Hopf. M. 2000. Domestication of plants in the Old World, ed.No. 3 Oxford : pp 198.
3. Okladnikov, I.; Vorke,I.L.; Trubachev,I.; Vlasova,N.and Kalacheva, G. 1977. Inclusion of chufa in the human diet as a source of polyunsaturated fatty acids. Voprosy Pitaniia (3):45-48.
4. Simpson, D.A and Inglis, C.A. 2001. Cyperaceae of economic, ethnobotanical and horticultural importance. Kew Bulletin (56):257-360.
5. Eteshola,E. and Oraedy,A.C.I. 1996. Fatty acid compositions of tiger nut tubers(*Cyperus esculentus* L.) Baobab seeds, and their mixture. JAOCS (73) :255-257.
6. Shilenko, M. ;Kalacheva, G.; Lisovskii, G. and Trubachev, I. 1979. Chufa (*Cyperus esculentus*) as a source of vegetable fat in seald life-support system. Kosm Biol Aviakosm Med .(13):70-74.
7. Emmanuel, O.A. and Edward, E. 1984. Nutritive value of a mixture of tigernut tubers (*Cyperus esculentus* L.) and baobab seeds (*Adansonia digitata* L.). J Sci Food Agric.(35).
8. Richard,S. 2008. Male infertility may be decided in womb .The Medical Research Council(MRC) Human Reproductive Sciences Symposium. www.ed.ac.uk./news.
9. Charton, E. 1999.Reduction and replacement of animal tests in the European Pharmacopoeia: recent developments for monographs on biological substances. Pharmaeuropa.(11):197-199
10. Al-Dujaily, S.S, 1996._In vitro sperms activation and Intra-Bursal insemination in_mice. Ph.D.Thesis College of Veterinary Medicine, Baghdad University. Pp62-64.
11. Hinting, A. 1989. Method of semen analysis. In assessment of Human Sperm Fertilization ability. Ph.D. thesis, University of Michiganstares.
12. Al-Taii, H. A. 1994. Sperm Activation and Intrauterine Insemination: The Effect of Serum Concentrations and Culture Media on Sperm Activation Potential *In vitro*. M.Sc. thesis, University of Baghdad.
13. Sorlie, D.E. 1995.Medical Biostatistics and Epidemiology: examination and board review. First edition .Appleton and Lang, Norwalk, Connecticut.Pp47-88.
14. Bedevian ,A. K. 1994. Illustrated polyglottic dictionary of plant. pp 222.
15. Oguike,M.A.;Aboaja,C.U.and Herbert,U. 2008. Influence of tiger nut (*Cyperus esculentus* L.) on the semen characteristics and testicular parameters of rabbits. Bulletin of Animal Health and Production in Africa. Vol 56, No 1.
16. Makler, A. 1988. Modern methods in semen analysis evaluation. In: Progress in Infertility. Behrman, S.J.; Kistner, R.W. and Patton, G.W. (eds.). Little, Brown and

17. Company, Boston, Toronto. pp: 633-66.
18. Monago, C.C. and Uwakwe, A.A. 2009. Proximate Composition and In vitro Anti-sickling Property of Nigerian *Cyperus esculentus* (Tiger Nut Sedge). Trees for Life Journal 4(2): 19
19. Glew, R.H.; Glew, R.S.; Chuang, L.; Huang, Y.S.; Millson, M.; Constance, D. and Vanderjagt, D.J. 2006. Amino acid minerals and fatty acid content of pumpkin seeds (*Cucurbita* spp) and *Cyperus esculentus* nuts in the Republic of Niger. Plant Foods for Human Nutrition. (61): 51-56.
www.chfadevalencia.org.
20. Salem, M.L.; Zommara, M. and Imaizumi, K. 2005. Dietary supplementation with *Cyperus esculentus* L (tiger nut) tubers attenuated atherosclerotic lesion in apolipoprotein E knockout mouse associated with inhibition of inflammatory cell responses. American Journal of Immunology 1 (1): 60-67.

تأثير استخدام عشبه لوز الأرض في المعايير الوظيفية لنطف ذكور الفئران غير البالغة كموديل للإنسان

أمال عبد الواحد محمد المشهداني *

أسراء محسن عبد الهادي العيساوي *

*معهد أبحاث الاجنه وعلاج العقم \جا معه النهرين

الخلاصة:

هدفت الدراسة الحالية إلى معرفة تأثير إعطاء عشبه لوز الأرض (CE) ومستخلصها الكحولي فمويًا على معايير النطف لذكور الفئران قبل البلوغ. قسمت حيوانات التجربة (18) إلى ثلاثة مجاميع كل مجموعة تضم ستة حيوانات. المجموعة الأولى أعطيت 150 ملغم/كغم/يوم من العشبه الخام والمجموعة الثانية عوملت بنفس الجرعة من المستخلص الكحولي للعشبه واعتبرت المجموعة الثالثة مجموعته سيطرة خلال فترة مقدارها ستة أسابيع. أظهرت النتائج زيادة معنوية ($p < 0.05$) في معدلات عدد النطف ونسبه المتحركة منها وحركتها التقدمية بين المجموعة الأولى والثانية مقارنة مع مجموعته السيطرة. ولم تشاهد فروقا معنوية بالنسبة لمعدلات النطف السوية ومقياس عيشويه النطف بين المجاميع. كما أظهرت النتائج عدم وجود فروقا معنوية في معدلات أوزان الحيوانات خلال فترة الدراسة. نستنتج من الدراسة الحالية إن التجريع الفموي للعشبه عزز معايير معينه للنطف في ذكور الفئران قبل البلوغ دون أن يؤثر على معدلات أوزانها.