Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 – 6131
Vol. 23(4): 227 - 237 (2019)
www.ejabf.journals.ekb.eg

Effect of α-Chlorohydrin water-bait on the fertility of captive males of the Egyptian fruit-bat (*Rousettus aegyptiacus*) and the proper time for controlling its free-ranging populations in Egypt

Ayat Taha* and Sohail Soliman
Zoology Department, Faculty of Science, Ain Shams University, Abbassia, Cairo, Egypt
* Corresponding author: ayattaha@yahoo.com

ARTICLE INFO
Article History:
Received: Sept. 22, 2019
Accepted: Oct. 14, 2019
Online: Oct. 16, 2019

Keywords:
Riverine habitats
Water-bait
Egyptian fruit-bat
Rousettus aegyptiacus
α-chlorohydrin
antifertility
control

ABSTRACT

The Egyptian fruit-bat (*Rousettus aegyptiacus*) is a serious vertebrate pest that lives in riverine and agricultural habitats of the Nile Delta and Valley of Egypt. The present work evaluates the acceptability and effect of a water bait of α-chlorohydrin (ACH), which is a well-known species-specific male chemosterilant, on the fertility of males of this bat species. A dose of 85.7mg/kg consumed by captive males for four consecutive days has damaged their testicular tissues, caused a significant decrease in sperm count and motility and a significant increase in sperm abnormalities. Preliminary results of the study of the reproductive cycle of this bat species in one of the districts of Greater Cairo Area showed that males had reached peak sexual activity during autumn mouths. For best control results, it is suggested that control campaigns be implemented during these months. It is recommended that ACH water-baits be set in roosting caves and near fresh water courses where bats used to drink.

INTRODUCTION

The Egyptian fruit-bat (*Rousettus aegyptiacus*) is the only magachiropteran bat species known from Egypt (Qumsiyeh, 1985). It is known from the Western Mediterranean Coast and from agricultural areas in and near the Nile Delta and Valley south to Aswan (Gaisler *et al*., 1972). Fruit bats consume average daily amounts of food that equal to 50–150% of their body mass thus causing large economic losses to cultivated crops and fruit trees (Qumsiyeh, 1996). These bats are, therefore, considered to be serious agricultural vertebrate pests (Hadjisterkotis, 2006).

Traditional methods used for the control of this bat species have largely depended on the use of poisonous fumes (Makin and Mendelssohn, 1987). Poisonous bait formulations were also suggested by Eissa (2007). The use of poisons, however, not only targets fruit-bats but it may also destroy the entire cave ecosystem in which bats live (Hadjisterkotis, 2006).

The use of antifertility compounds, as an alternative approach to the control of vertebrate pests, started several decades ago when α-chlorohydrin (ACH) was presented as a rat chemosterilant (Ericsson, 1970). ACH, also known as (3-chloro-1,2-

propanediol), (3-monochloropropane-1,2-diol), and (3-MCPD), is a well-known contaminant found in many food products (El Ramy *et al*., 2007; Hamlet *et al*., 2011; Zhang *et al*., 2012; Jędrkiewicz *et al*., 2016) and is often present in tap water (Velisek *et al*.,1978). The chemical has proved to be species-specific; producing sterility in males of some mammalian species such as rats (Tsunoda and Chang, 1976; Toth *et al*., 1989; Slott *et al.*, 1990), hamsters (Lubicz-Nawrocki and Chang, 1974 a, b) and rat-tailed bats (Dixit and Lohiya, 1976), not in others. Although ACH shows rapid antifertility effects at low doses and prolonged or permanent sterility at high doses (Jones, 1983; El-Gohary *et al*., 2014), it does not affect libido or appetite of treated males (Jones, 1983; Yamada *et al.*, 1995). For these reasons and some others, ACH was considered to be an ideal male antifertility control agent for a wide range of vertebrate pests (Jones, 1983).

ACH was first tested with male Egyptian fruit-bats by Mahmoud *et al*. (2018). They applied the chemical in the form of fruit-baits and at concentrations lower than that applied in the present study. The results of their study were positive and promosing. In the present piece of work, ACH is used in the form of water baits. This is based on the observations of several authors that fruit-eating bats drink water. They observed that roosts of fruit bats, other than the Egyptian fruit-bat, were located near riverine habitats and other water bodies (Thomas and Fenton, 1978, Monadjem and Reside, 2008). They also described their drinking behavior (Harvey *et al*., 1999). Water baits are of course much cheaper than fruit baits and are thus more suitable for use in mass control programs of Egyptian fruit-bats.

MATERIALS AND METHODS

Chemicals

ACH was purchased from Sigma Chemical Co. (St. Louis, MO), and diethyl ether was supplied by SD Fine Chem Limited (Mumbai, India). Doses of ACH solutions were prepared in distilled water just before use. All other chemicals were of analytical grade and were obtained from El-Nasr Pharmaceutical Chemicals Co. (Cairo, Egypt) unless otherwise specified.

Experimental animals

Twenty adult male fruit-bats with scrotal testes were collected during October 2018 from Giza Governorate, Egypt. The body weight of collected males ranged between 127.5g and 138.5g. The bats were housed individually in special wooden cages (30 x 30 × 20 cm) with metal-mesh sides in the animal house, Faculty of Science, Ain Shams University, Cairo. The top of each cage was open and surrounded by a collapsible thick cloth that permitted an easy access to bats without permitting them to escape. Bats were maintained at a constant room temperature of 25°C. Caged bats were provided with water and fresh fruits *ad libitum*. Bats were observed for one week before providing the chemical bait to give them enough time to acclimate to laboratory conditions and to be sure that they were healthy. Animal handling was in accordance with the ethical standards of Ain Shams University Research Ethics Committee, WHO guidelines for animal care, and the Helsinki Declaration of 1975 as revised in 1983.

Experimental design

Average individual daily consumption of fresh water was first estimated to help determine the concentration of ACH in water baits. The twenty Egyptian fruit-bats were randomly divided into two groups; ten individuals each, a treated group and a control group. Bats of the control group were allowed free access to fresh water and

fresh fruits of the season. The concentration of ACH in water baits was calculated on the basis of two parameters; the average individual daily consumption of fresh water and the daily amounts of ACH intented to be given to bats (100 mg/kg/day for four consecutive days). To calculate the actual daily amount of ACH consumed by each individual bat, a known amount of ACH water-bait was provided to each bat in a 4x10 cm shallow dish. The remaining amount of water bait was measured after 24h to determine the actual daily amount of water bait, and consequently the exact daily amount of ACH, consumed by individual bats. ACH water-baits were presented to bats for four consecutive days. The acceptability of the ACH water-bait formulation was calculated using the equation of Mason *et al.* (1989):

$$\text{Acceptability (\%)} = \frac{\text{Average daily consumption of ACH water-bait (ml)}}{\text{Average daily consumption of (ACH water-bait + fresh water) (ml)}} \times 100$$

Sample collection

Twenty-four hours after the last dosing, all bats were sacrificed by an overdose of diethyl ether and weighed. At necropsy, the reproductive organs; i.e., testes, epididymides, seminal vesicles and prostate glands, were excised, cleaned of adhering fat and weighed in grams. Organ index (relative weight) was calculated according to the formula: [(organ weight/body weight) ×100].

The right testis was routinely processed for histological examination and stained with hematoxylin and eosin (H&E). The right cauda epididymis was placed in 2 ml modified Tyrode's medium (MT6) (125 mM NaCl, 2.7 mM KCl, 0.5 mM $MgCl_2$. 6H2 O, 0.36 mM NaH_2PO_4. $2H_2O$, 5.56 mM glucose, 25 mM $NaHCO_4$, 1.80 mM $CaCl_2$, 100 units penicillin and 4 mg/ml BSA) at 35°C for later determination of sperm characteristics (Saad and Mahmoud, 2014).

Epididymal sperm count and sperm motility

The right cauda epididymis was minced with small scissors in a 35 mm Petri dish containing 2 ml MT6 medium, and left for 15 min at 35°C for sperm release. An aliquot of the diluted sperm was infused into a Neubauer-type hemocytometer for microscopic examination. Sperm were counted in the four corner-squares as well as in the central square. The data were expressed as the number of sperm/ml (Saad and Mahmoud, 2014).

For evaluation of differential sperm motility, an aliquot of sperm in MT6 medium was layered onto a clean, grease-free microscope slide and was examined using a light microscope at 400 x. Percentage motility was estimated at three different fields in each slide. The mean of the three estimations was calculated and used as the final motility score (Saad and Mahmoud, 2014).

Evaluation of sperm abnormalities

Sperm morphology was determined using the same samples used before for the evaluation of sperm motility. Sperm were smeared onto microscope slides, air dried, then fixed with methanol. After fixation, samples were stained with 1% (w/v) aqueous eosin Y for 1h, washed with distilled water, dehydrated, cleared, then mounted in neutral resin under a coverslip. Two hundred spermatozoa from each sample were evaluated at 1,000 x using a light microscope with oil immersion objective lens (Saad and Mahmoud, 2014). A sperm was considered to be abnormal if it was double-headed, tailless, headless, and with bent, highly folded, or twisted tail (WHO, 2000). Sperm abnormalities were recorded as percentages of total number of spermatozoa counted.

Histopathological examination

Microscopic sections of 5 µm of the right testis were prepared according to the method used by Saad and Mahmoud (2014). The right testis was fixed in Bouin's solution, dehydrated through an ascending series of ethanol, cleared in terpineol and embedded in paraffin wax. Sections were cut at 5 µm, deparaffinized in xylol, rehydrated through a descending series of ethanol, and stained with H & E. For each testis section, 30 tubules were evaluated for histopathological changes. Thickness of testis capsule and tubule diameter were measured in µm at 400 x.

Statistical analysis

Numerical data are reported as means ± S.E. Data were analyzed using one-way ANOVA followed by Tukey's post hoc multiple comparisons test for comparative analysis among groups. Values for $p \leq 0.05$ were considered statistically significant.

RESULTS

Average daily consumption of ACH

Calculations for estimating the average actual daily consumption of ACH by individual male bats are shown in Tables 1 and 2. It is observed from these tables that the average daily consumption of ACH water-bait is significantly lower than that of fresh water and its acceptability is 46.2%. The average actual daily consumption of ACH is 11.4 mg/individual (equivalent to 85.7 mg/kg body weight).

Table 1: Calculations for the concentration of ACH in water-baits to be applied to male Egyptian fruit-bats (*n* =10).

Average body weight of treated males (g)	133 ± 1.5 (1)
Average observed daily consumption of fresh water (ml/individual)	75.8 ± 5.9 (2)
Average actual daily consumption of fresh water (ml/individual) (2)X0.90*	68.2 (3)
ACH intented to be consumed by bats (mg/kg)	100 (4)
ACH intented to be consumed by individual bats (mg/individual) (4)X(1) / 1000	13.3 (5)
Concentration of ACH in water bait (mg/ml) (5)/ (3)	0.195 (6)

*The average actual daily consumption of fresh water is supposed to be 90% of the average observed daily consumption.

Table 2: Average actual daily amount of ACH consumed by individual male Egyptian fruit-bats (*n* =10) and acceptability of ACH water-baits.

Average observed consumption of ACH water-bait (ml/individual)	65 ± 8.20* (7)
Average actual daily consumption of ACH water-bait (ml/individual) (7)X0.90**	58.5 (8)
Average actual daily consumption of ACH (mg/individual) (8)X(6)	11.40 (9)
Acceptability (%) (8) X (100) / (3)+(8)	46.2

*Significantly different compared to fresh water group ($P < 0.05$).

**The average actual daily consumption of ACH water-baits is supposed to be 90% of the average observed daily consumption.

Morphology and histology of the testis of the control group

Testes of normal bats of the control group were ovoid in shape with normal size and color. Microscopic examination of testis sections indicated that it had the normal histological structure known for mammalian testes (Fig.1-a and -b).

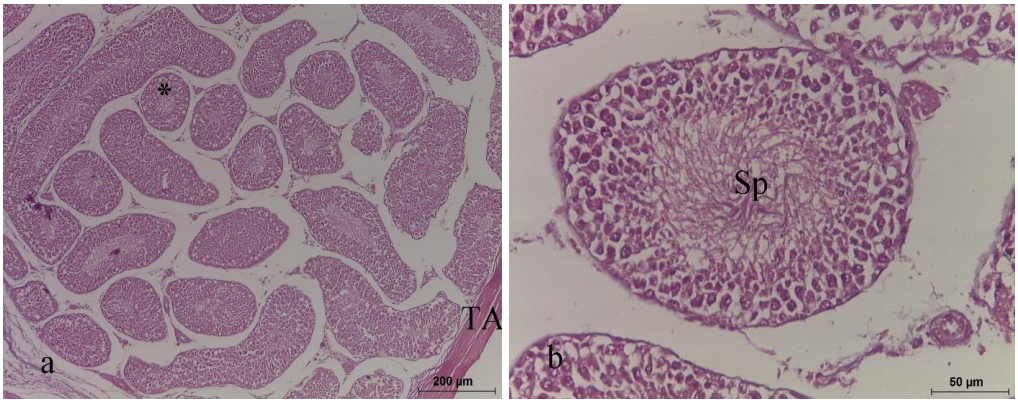

Fig. 1: Photomicrographs showing sections of the testis of control male Egyptian fruit-bats. a, shows the oval seminiferous tubules surrounded by thin tunica albuginea (TA), lined with stratified germinal epithelium (asterisk); b, shows normal seminiferous tubules containing numerous sperms (Sp).

Effect of ACH on treated bats

No clinical signs appeared during the course of treatment. Treated fruit-bats appeared active, showed normal behavior and consumed normal daily amounts of food and water. The relative weights of reproductive organs of treated males did not show significant changes compared to those of the control group (Table 3).

Table 3: Effect of ACH on the relative weights of reproductive organs of male Egyptian fruit-bats (*n* =10). Mean is followed by S.E. and range (in parentheses).

Groups	Testis	Epididymis	Vesicula seminalis	Prostate
Control	0.966 ± 0.083	0.23 ± 0.02	0.49 ± 0.017	0.081 ± 0.020
	(0.633-1.19)	(0.19-0.34)	(0.083-1.19)	(0.023-0.148)
Treated	0.903 ± 0.11	0.25 ± 0.02	0.398 ± 0.055	0.081 ± 0.006
	(0.43-1.19)	(0.15-0.33)	(0.26-0.65)	(0.072-0.116)

Histopathological effects on the testis

Testes of treated males showed thickened tunica albuginea, tubular diminution and spermatogenic arrest. Sertoli cells, spermatogonia and spermatocytes suffered from severe cytoplasmic vacuolization and nuclear pyknosis. Multinucleated stromal giant cells were also observed in the testes of treated males (Fig. 2-a and -b).

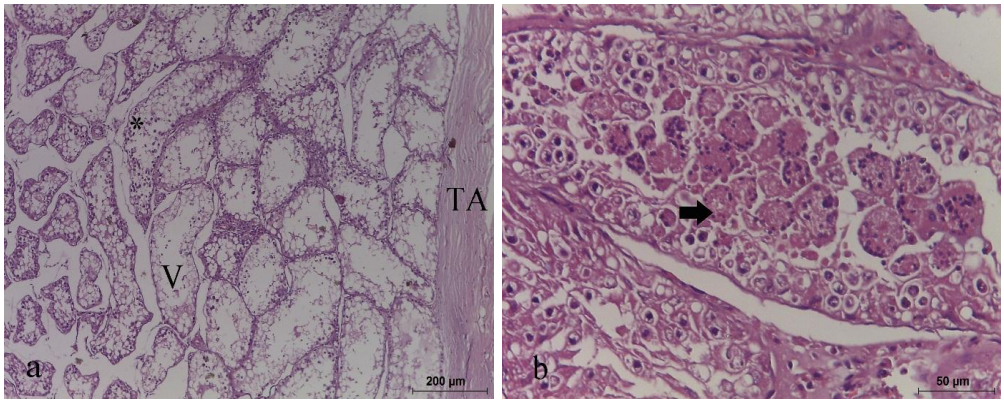

Fig. 2: Photomicrographs showing sections of the testis of treated male Egyptian fruit-bats. a, shows thickened tunica albuginea (TA), tubular vacuolization (V), and spermatogenic arrest with absence of sperm (asterisk); b, shows the presence of multinucleated stromal giant cells (arrow).

Effect on testicular morphometrics

Histological examination of the testis showed a significant decrease in the diameter of seminiferous tubules and epithelial height, and significant increase in the thickness of tunica albuginea (Table 4 and Fig. 2-a).

Table 4: Effect of ACH on testicular morphometrics of male Egyptian fruit-bats (n =10). Mean is followed by S.E. and range (in parentheses).

Group	Tubule diameter (µm)	Tunica albuginea thickness(µm)	Epithelial height (µm)
Control	148.14 ± 2.96 (133.33-177.77)	67.77 ± 1.62 (62.22-71.1)	66.66± 2.9 (53.33-80)
Treated	130.95 ± 4.5* (88.88-151.11)	137.75± 2.9* (124.4-151.11)	39.99± 2.3* (26.66-44.44)

* Significant difference ($P < 0.05$).

Effects on sperm parameters

Sperm count and motility were significantly reduced, while the number of abnormal sperm has significantly increased (Table 4 and Figs. 3 & 4). Various abnormalities of sperm morphology were observed including double heads, tailless heads, headless tails, bent tails, highly folded tails, and twisted tails (Table 4 and Fig. 5).

Table 5: Effect of ACH on sperm parameters of male Egyptian fruit-bats (n=10). Mean is followed by S.E. and range (in parentheses).

Groups	Sperm count ($x10^6$/ml)	Sperm abnormality %	Progressive motile %	Non-progressive motile %	Immotile %
Control	160.5± 6.25 (146-188)	12.5± 0.9 (10-15)	39.3 ± 4.3 (30-60)	48 ± 5.5 (23-63)	12.3 ± 1.5 (7-17)
Treated	45.5 ± 0.3* (35-56)	35.5 ±5.05* (23-55)	____	____	100 ± 0* (100-100)

* Significant differences ($P < 0.05$).

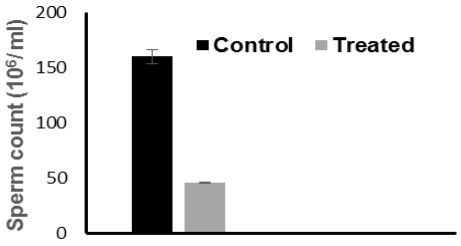

Fig. 3. A histogram showing the effect of ACH on sperm count in male Egyptian fruit-bats.

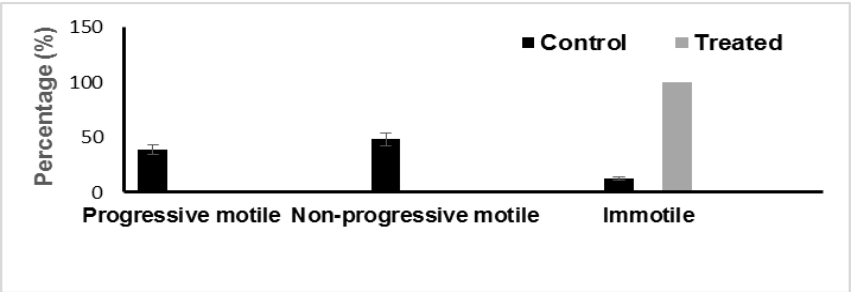

Fig. 4: A histogram showing the effect of ACH on sperm motility in male Egyptian fruit-bats.

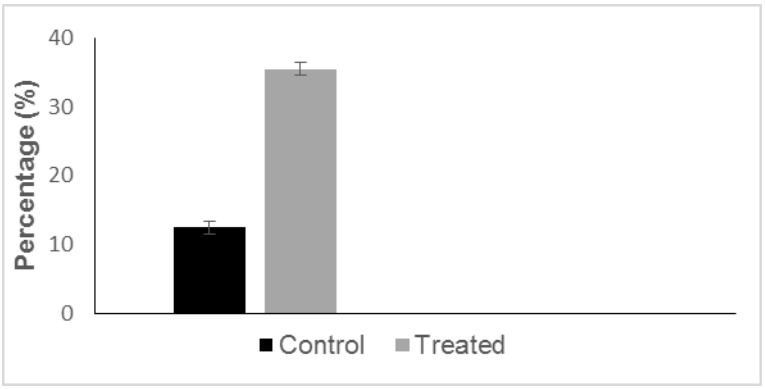

Fig. 5: A histogram showing the effect of ACH on sperm abnormalities in male Egyptian fruit-bats.

Sexual activity cycle of males

Preliminary results of the study of the sexual activity cycle of male fruit-bats in one of the districts of Greater Cairo Area indicate that they reach peak of sexual activity in autumn months. Males are completely sexually quiescent in summer months.

DISCUSSION

Egyptian fruit-bats are serious vertebrate pests that live in agricultural areas and riverine habitats. Captive male bats readily accepted ACH in the form of a water-bait and consumed an average daily dose of 85.7mg/kg for four consecutive days. This dose is much higher than that (59.2 mg/kg for four consecutive days) consumed by males of the same bat species in the form of fruit-baits (Mahmoud *et al*., 2018). The increase in the actually consumed amount of ACH was accompanied by an increase in its antifertility effects. These effects are consistent with those reported for other vertebrate pests (Kwack *et al*., 2004; Cho *et al*., 2008; Madhu *et al*., 2011; El-Gohary *et al*., 2014).

The testes of ACH-treated bats exhibited shrinkage in tubular diameter (88.4% of the control group) which is greater than that observed in the previous study (72.8% of the control group). Shrinkage of tubules is usually associated with the loss of germ cells (Manjit and Parshad, 1993; Creasy, 2005; Gupta *et al.*, 2006; Saad and Mahmoud, 2014). This is due to the disturbance of the Sertoli-germ cell junctions (Mesbah *et al.*, 2008). The average thickness of the tunica albuginea in treated male bats (203.2% of the control group) was less than that observed in the previous study (271.6% of the control group) (Mahmoud *et al*., 2018). The increased thickness of the tunica albuginea could be explained on the basis of the decreased volume of the testicular parenchyma as suggested by Arenas *et al.* (1997).

In the present work, the effect of ACH dose on sperm parameters is greater than that of the dose used before by Mahmoud *et al*. (2018). ACH-treated males exhibited significantly decreased sperm count (28.3% of the control group) and significantly increased sperm deformities (284% of the abnormal sperm of the control group) compared to the previous study (31.7%, and 186.2%, respectively). It is noticed that both doses of the present and previous studies have caused complete arrest of sperm motility. Alterations in sperm parameters are directly associated with the histopathological changes in testicular tissue that cause reproductive dysfunction (de Souza *et al*., 2010). Sperm parameters are, therefore, important signs of infertility of treated males (Shen and Lee, 1994; Orisakwe *et al.*, 2004). It was suggested that ACH

has blocked the metabolic pathways of epididymal sperm (Jelks and Miller 2001; Nakajima *et al.,* 2007; Nascimento *et al.,* 2008; Frayne *et al*., 2009). It inhibited androgen-dependent enzymes such as adenosine triphosphatase and acetylcholine esterase in the epididymis (Jelks *et al.,* 2001; Zhang *et al.,* 2012) and increased alkylation of cysteine residues in sperm, which inhibits sperm motility (Kalla and Bansal, 1977; Kalla and Singh, 1981).

Morphology of sperm is a vital parameter that reflects their normality and maturity and is directly correlated with male fertility (Memon *et al.,* 1986). Abnormalities of the head and mid-piece of sperm have been identified as primary defects of spermatogenesis (Schumacher and Moll, 2011), and are good indicators of testicular degeneration (Bloom, 1950). Similar effects have been reported for other animals such as rats (Madhu *et al.,* 2011).

Free-ranging males showed peak sexual activity in autumn months. ACH water-baits would, therefore, be highly effective in producing sterility in these males if they are applied during months of highest sexual activity.

CONCLUSION

ACH proved to be effective in producing sterility in captive male Egyptian fruit-bats when individual bats consumed a dose of 85.7 mg/kg for four consecutive days in the form of water baits. It is recommended that water baits be set inside roosting caves and beside fresh water courses where bats used to drink. ACH water-baits are cheap and more suitable for use as part of integrated pest management programs. For best control results, it is suggested that control campaigns be implemented during months of highest sexual activity in males.

ACKNOWLEDGMENT

The authors would like to thank Prof. Yomna Mahmoud, professor of histology and histochemistry for her interest in the work and for valuable comments on the manuscript.

REFERENCES

Arenas, M. I.; Bethencourt, F. R.; De Miguel, M. P.; Fraile, B.; Romo, E. and Paniagua, R. (1997). Immunocytochemical and quantitative study of actin, desmin and vimentin in the peritubular cells of the testes from elderly men. J. Reprod. Fertil., 110: 183–193.

Bloom, E. (1950). Fertility in male animals. J. Fertil. Steril., 1: 223-224.

Cho, W. S. ; Han, B. S. ; Lee, H. ; Kim, C. ; Nam, K. T. ; Park, K. ; Choi, M. ; Kim, S. J. ; Kim, S. H.; Jeong, J. and Jang, D. D. (2008). Subchronic toxicity study of 3-monochloropropane-1, 2-diol administered by drinking water to B6C3F1 mice. Food Chem.Toxicol., 46: 1666–1673.

Creasy, D. M. (2005). The male reproductive system. In: Turton J, Hooson J, Eds. Target Organ Pathology: a Basic Text. Taylor and Francis, London, Bristol. pp. 334–365.

de Souza, P. F. ; Diamante, M. A. and Dolder, H. (2010). Testis response to low doses of cadmium in Wistar rats. Int. J. Exp. Pathol., 91:125–31.

Dixit, V. P. and Lohiya, N. K. (1976). Mechanism of action of α-chlorohydrin on the testes and caput epididymis of rat, gerbil (*Meriones hurrianae*), bat and mouse. Acta Anat., 95 (1): 50–7.

Eissa, Y. A. (2007). Ecological, biological and toxicological studies on fruit bat. Ph.D. Thesis, Fac. Agric., Al-Azhar Univ.

El-Gohary, F.; Soliman, S. and Sobeiha, M. K. (2014). Use of the male chemosterilant: α-chlorohydrin in the control of house rats, *Rattus rattus*. Egypt. J. Agric. Res., 92: 947–987.

El Ramy, R.; Elhkim, M. O.; Lezmi, S. and Poul, J. M. (2007). Evaluation of the genotoxic potential of 3-monochloropropane-1, 2-diol (3-MCPD) and its metabolites, glycidol and β-chlorolacetic acid, using the single cell gel/comet assay. Food Chem. Toxicol. 45: 41–48.

Ericsson, R. J. (1970). Male antifertility compounds: U-5897 as a rat chemosterilant. J. Reprod. Fertil., 22 (2): 213-22.

Frayne, J.; Taylor, A.; Cameron, G. and Hadfield, A. T. (2009). Structure of insoluble rat sperm glyceraldehyde-3-phosphate dehydrogenase (GAPDH) via heterotetramer formation with Escherichia coli GAPDH reveals target for contraceptive design. J. Biol. Chem., 284: 22703–22712.

Gaisler, J.; Madkour, G. and Pelikan, J. (1972). On the bats (Chiroptera) of Egypt. Acta Sc. Nat.Brno., 6 (8):1-40.

Gupta, R. S.; Kachhawa, J. B. S. and Chaudhary, R. (2006). Antispermatogenic anti-androgenic activities of Albizia lebbeck (L.) Benth bark extract in male albino rats. Phytomedicine. 13: 277–283.

Hadjisterkotis, E. (2006). The destruction and conservation of the Egyptian fruit bat *Rousettus aegyptiacus* in Cyprus: a historic review. Eur. J.Wildl. Res., 52: 282–287.

Hamlet, C. G.; Asuncion, L.; Velíšek, J.; Doležal, M.; Zelinková, Z. and Crews, C. (2011). Formation and occurrence of esters of 3-chloropropane-1, 2-diol (3-CPD) in foods: what we know and what we assume. Eur. J. Lipid Sci. Technol. 113: 279–303.

Harvey, M. J.; Altenbach, J. S. and Best, T. L. (1999). Bats of the United States. Arkansas Game and Fish Commission.

Jędrkiewicz, R.; Kupska, M.; Głowacz, A.; Gromadzka, J. and Namieśnik, J. (2016). 3-MCPD: a worldwide problem of food chemistry. Crit. Rev. Food Sci. Nutr. 56: 2268–2277.

Jelks, K. B. and Miller, M. G. (2001). α-Chlorohydrin inhibits glyceraldehyde- 3-phosphate dehydrogenase in multiple organs as well as in sperm. Tox. Sci., 62: 115–123.

Jelks, K. B.; Berger, T.; Horner, C. and Miller, M. G. (2001). α-Chlorohydrin induced changes in sperm fertilizing ability in the rat: association with diminished sperm ATP levels and motility. Reprod.Toxicol., 15: 11–20.

Jones, A. R. (1983). Anti-fertility actions of α-chlorohydrin in the male. Aust. J. Biol. Sci., 36: 333–350.

Kalla, N. R. and Bansal, M. P. (1977). In vivo and in vitro alkylation of testicular cysteine by α-chlorohydrin administration. Indian J. Exp. Biol., 15: 232–233.

Kalla, N. R. and Singh, B. (1981). Synergistic effect of alpha chlorohydrin on the influence of copper ions on human spermatozoa. Int. J. Fertil., 26: 65–67.

Kwack, S. J. ; Kim, S. S. ; Choi, Y. W. ; Rhee, G. S.; Lee, R. D. ; Seok, J. H.; Chae, S. Y.; Won, Y. H.; Lim, W. J.; Choi, K. S.; Park, K. L. and Lee, B. M. (2004). Mechanism of antifertility in male rats treated with 3-monochloro-1, 2-propanediol (3-MCPD). J. Toxicol. Env. Heal. A., 67: 2001–2011.

Lubicz-Nawrocki, C. M. and Chang, M. C. (1974a). Effect of a-chlorohydrin on the fertilizing ability of hamster epididymal spermatozoa. J. Reprod. Fertil., 38:65-71.

Lubicz-Nawrocki, C. M. and Chang, M. C. (l974b). The onset and duration of infertility in hamsters treated with α-chlorohydrin. J. Reprod. Fertil., 39: 291-295.

Madhu, N. R.; Sarkar, B.; Biswas, S. J.; Behera, B. K. and Patra, A. (2011). Evaluating the antifertility potential of á-chlorohydrin on testis and spermatozoa in the adult male wild Indian house rat (*Rattus rattus*). J. Environ.Pathol. Tox. and Oncol., 30: 93–102.

Mahmoud, Y. I.; Taha, A. and Soliman, S. (2018). 3-Monochloropropane-1, 2-diol (alpha-chlorohydrin) disrupts spermatogenesis and causes spermatotoxicity in males of the Egyptian fruit-bat (*Rousettus aegyptiacus*), Biotech- Histochem. 93:4,293-300, DOI: 10.1080/10520295.2018.1437471.

Makin, D. and Mendelssohn, H. (1987). Israel wipes out its bats. Ecologist., 16: 1-2.

Manjit, S. and Parshad, V. R. (1993). Field evaluation of alpha chlorohydrin against the Indian mole rat: studies on toxic and antifertility effects. Ann. App. Biol., 122: 153–160.

Mason, J. R.; Avery, L., and Otts, D. L. (1989). Standard Protocol for Evaluation of Repellent Effectiveness in Birds. Bird Section Res. No. 20, B, DWRC, pp. 1- 20.

Memon, M. A.; Bretzlaff, R. N. and Ott, R. S. (1986). Comparison of semen techniques in goats. Theriogenology., 26: 823-827.

Mesbah, S. F.; Shokri, S.; Karbalay-Doust, S. and Mirkhani, H. (2008). Effects of nandrolone decanoate on ultrastructure of testis in male adult rats. Iran. J. Med. Sci., 33: 94–100.

Monadjem, A. and Reside, A. (2008). The influence of riparian vegetation on the distribution and abundance of bats in an African savannah. Acta Chiropterol., 10 :339–348.

Nakajima, H.; Amano, W.; Fujita, A.; Fukuhara, A.; Azuma, Y. T.; Hata, F.; Inui, T. and Takeuchi, T. (2007). The active site cysteine of the proapoptotic protein glyceraldehyde-3- phosphate dehydrogenase is essential in oxidative stress-induced aggregation and cell death. J. Biol. Chem., 282: 26562–26574.

Nascimento, J. M.; Shi, L. Z.; Tam, J.; Chandsawangbhuwana, C.; Durrant, B.; Botvinick, E. L. and Berns, M. W. (2008). Comparison of glycolysis and oxidative phosphorylation as energy sources for mammalian sperm motility, using the combination of fluorescence imaging, laser tweezers, and real-time automated tracking and trapping. J. Cell. Physiol., 217: 745−751.

Orisakwe, O. E.; Husaini, D. C. and Afonne, O. J. (2004). Testicular effects of subchronic administration of Hibiscus sabdariffa calyx aqueous extract in rats. Reprod. Toxicol., 18: 295–298.

Qumsiyeh, M. B. (1985). The Bats of Egypt. 1st ed., Special Publication 23, Texas Tech University Press, Lubbock, TX. p. 1–102.

Qumsiyeh, M. B. (1996). Mammals of the Holy Land. Texas Tech University Press, Lubbock, TX. p. 389.

Saad, R. A. and Mahmoud, Y. I. (2014). Ursodeoxycholic acid alleviates cholestasis-induced histophysiological alterations in the male reproductive system of bile duct-ligated rats. Reprod. Toxicol., 50: 87–97.

Schumacher, J. and Moll, H. D. (2011). Collection of semen. In: A Manual of Equine Diagnostic Procedures (Eds.: Schumacher, J. and Moll, H.D.). Teton NewMedia, Jackson, WY, USA (www.tetonnm.com/). Internet Publisher: International Veterinary Information Service, Ithaca NY (www.ivis.org). Document No. A5420.1011.

Shen, R. S. and Lee, J. P. (1994). Selected testicular enzymes as biochemical markers for procarbazine induced testicular toxicity. Arch. Toxicol., 55: 233–238.

Slott, V. L.; Suarez, J. D.; Simmons, J. E. and Perreault, S. D. (1990). Acute inhalation exposure to epichlorohydrin transiently decreases rat sperm velocity. Fundam. Appl. Toxicol., 15:597-606.

Thomas, D. W. and Fenton, M. B. (1978). Notes on the dry season roosting and foraging behaviour of *Epomophorous gambianus* and *Roussettus aegyptiacus* (Chiroptera: Pteropodidae). J. Zool., 186:403–406.

Toth, G. P.; Stober, J. A.; Read, E. J.; Zenick, H. and Smith, M. K. (1989).The automated analysis of rat sperm motility following subchronic epichlorohydrin administration: methodological and statistical considerations. J Androl., 10:401-415.

Tsunoda, Y. and Chang, M. C. (1976).Fertilizing ability in vivo and in vitro of spermatozoa of rats and mice treated with α-chlorohydrin. J. Reprod. Fertil., 46:401-406.

Velisek, J.; Davidek, J.; Hajslova, J.; Kubelka, V.; Janiceck, G. and Mankova, B. (1978). Chlorohydrins in protein hydrolysates. Z. Lebensm. Unters. Forsch., 167:241–244.

World Health Organization (WHO) (2000). Laboratory manual: for the examination of human semen and sperm-cervical mucus interaction. 4thEd. Cambridge: Cambridge University Press.

Yamada, T.; Inoue, T.; Sato, A.; Yamagishi, K. and Sato, M. (1995). Effects of short-term administration of αchlorohydrin on reproductive toxicity parameters in male Sprague-Dawley rats. J. Toxicol. Sci., 20:195–206.

Zhang, H.; Yu, H.; Wang, X.; Zheng, W. ; Yang, B.; Pi, J.; He, G. and Qu, W. (2012). (S)-α-chlorohydrin inhibits protein tyrosine phosphorylation through blocking cyclic AMP-protein kinase a pathway in spermatozoa. PLoS One., (8): e43004.

ARABIC SUMMARY

تأثير الطعم المائى لمادة ألفا-كلوروهيدرين على خصوبة ذكور خفاش الفاكهة المصرى (*Rousettus aegyptiacus*) المأسورة والوقت الأمثل لمكافحة عشائره الطليقة فى مصر

آيات طه* و سهيل سليمان

قسم علم الحيوان - كلية العلوم - جامعة عين شمس - العباسية -القاهرة - مصر

*ayattaha@yahoo.com

يعتبر خفاش الفاكهة المصرى (*Rousettus aegyptiacus*) من الآفات الفقارية الخطيرة التى تعيش فى البيئات النهرية والزراعية على امتداد دلتا ووادى النيل فى مصر . ويهتم البحث الحالى بتقييم أستساغة وتأثير الطعم المائى لمادة ألفا-كلوروهيدرين، وهى مادة معروفة بتأثيرها المانع للخصوبة فى ذكور بعض أنواع الحيوانات، على خصوبة ذكور هذا النوع من الخفافيش .ولقد تبين أن ابتلاع افراد الخفافيش المأسورة لكمية من هذه المادة تقدر بـ 85,7 مجم/كجم من وزن الجسم لمدة أربعة أيام متوالية قد تسبب فى إلحاق الضرر بأنسجة الخصية، وانخفاض معنوى فى عدد الحيوانات المنوية وحركتها، وزيادة معنوية فى نسبة المشوه منها.

ولقد أظهرت النتائج الأولية لدراسة الدورة التزاوجية لهذا النوع من الخفافيش بإحدى مناطق القاهرة الكبرى أن الذكور تصل لقمة نشاطها الجنسى أثناء شهور الخريف. وللحصول على أفضل النتائج فى مكافحة هذه الخفاش يقترح أن تتم حملات المكافحة أثناء هذه الشهور. ويوصى بوضع الطعوم المائية لمادة ألفا-كلوروهيدرين داخل الكهوف التى تعيش بها هذه الخفافيش أو على مقربة من الجداول المائية التى اعتادت أن تشرب منها.