Behavioral, hormonal, and testicular histological study to evaluate the effect of season on mating in local breed free housing tom cats in Al-Diwaniya city

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Abstract

The current study was conducted in the center of Al-Diwaniya city, Al-Qadisiya province in Iraq during a period of one year from (15/1/2012 to 15/1/2013) divided according to the sexual activity of males into three periods: the 1st period extended from mid of January to February, the 2nd period from February to November, and the 3rd period from November to mid of January. During these entire periods forty seven male local breed cats, (ten for each of the 1st and 3rd periods, while the other twenty seven were included in the 2nd period). All males were monitoring during the night and early morning to evaluate the behaviors, and then hunted to evaluate hormonal and histological changes in the male genital system. The results of behavioral study during the 1st period revealed that the male showed caterwaul sound, urination and following of the females with foreplay but without mating; in the 2nd period the same notes were observed with a huge desire to mating and obligating females for mating; the behaviors in the 3rd period were as the same of the 1st period. The ratios of studied periods during year were (4.16%, 75% and 20.84%) respectively. The results of the hormonal assay revealed that the testosterone levels were (0.42, 1.31 and 0.35 ng/ml) for the three periods of study respectively, while the LH concentrations were (1.31, 4.86 and 1.23 ng/ml) respectively. The histological study of testis showed that the seminiferous tubules characterized by very active vacuolated Sertoli cells filled with secretions with presence of all developmental stages of spermatogenesis, while the 1st and 3rd period characterized by thickening of connective tissue and peritubular tissue as well as not all developmental stages of spermatogenesis.

Key words: local male cats, behavioral, hormonal and histological study, effect of season.

دراسة سلوكية وهرمونية ونسجيه للخصى لتقييم تأثير الموسم على تزاوج ذكور القطط المحلية حرة المعيشة في مدينة الديوانية

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الخلاصة

أجريت الدراسة الحالية في مركز مدينة الديوانية على مدار سنة كاملة للفترة من 2012/1/15 إلى 2013/1/15 . قسمت فترات الدراسة حسب النشاط الجنسي للذكور إلى ثلاث فترات: الفترة الأولى والتي امتدت من منتصف كانون الثاني وحتى شباط ، الفترة الثانية من شباط إلى تشرين الثاني والفترة الثالثة من تشرين الأول إلى منتصف كانون الثاني. اشتملت الدراسة على سبعة وأربعين ذكرا من القطط (السلالة المحلية)، بواقع عشرة ذكور لكل من الفترتين الأولى والثالثة وسبعة وعشرون للفترة الثانية. تم مراقبة الذكور لرصد نشاطها خلال فترة الليل وفي الصباح الباكر لتقييم التغيرات السلوكية ومن ثم اصطيدت لدراسة التغيرات الهرمونية والنسجية في الجهاز التناسلي الذكري. أظهرت نتائج رصد سلوك الذكور خلال الفترة الأولى: أصوات المواء ، و التبول ومن ثم مداعبة الإناث ولكن من دون التزاوج ، بينما لوحظ نفس

السلوك في الفترة الثانية على الذكور ولكن مع رغبة جامحة للجماع وإرغام الإناث على التزاوج وكان السلوك خلال الفترة الثالثة مشابها للسلوك خلال الفترة الأولى. النسبة المئوية لفترات الدراسة خلال السنة كانت (4.16%،75%و20.84%) على التوالي. و على مستوى الفحص الهرموني كان مستوى هرمون التستوستيرون 0.42 ng/ml ، 1.31 ng/ml و 0.35 ng/ml في الفترات الثلاثة على التوالي ، في حين كانت تراكيز الهرمون اللوتيني 1.31ng/ml ، 4.86ng/ml و 1.23 ng/ml على التوالي. كما أظهرت الدراسة النسيجية للخصى خلال الفترة الثانية من الدراسة أن النبيبات المنوية تميزت بنشاط واضح من خلال تميز خلايا سرتولي بكبر حجمها وامتلائها بالإفرازات مع وجود جميع مراحل نمو الحيوانات المنوية ، في حين تميزت الدراسة الشكلية لنسيج الخصية خلال الفترة الأولى والثالثة بسمك النبيبات المنوية حيث لوحظ سمك النسيج الخلالي الضام وكذلك عدم اكتمال في مراحل تطور الحيوانات المنوية.

الكلمات المفتاحية: ذكور القطط المحلية ، دراسة سلوكية وهرمونية ونسيجية ، تأثير الموسم.

Introduction

Domestic cats are seasonally polyestrous and the most important factor in its reproduction is increasing daylight length (1, 2, and 3). Under natural temperate photoperiod, domestic cats have estrous cycles throughout the year showing peak activity in the months with increasing photoperiod (4). Free-living non-pedigree and feral cats are seasonally polyestrous, some have regular estrous, but others may show non-regular pattern (3). Some studies consider that the tom cats are not seasonal breeder (5) and the quantity or quality of sperm of male cats are not affect by increase or decrease in photoperiod (6), but may has an effect on testicular weight (5), while others suggested that there is a moderate effect for photoperiod on sperm quantity and variation in hormone production in the tom (7) with moderate variation in serum testosterone concentration (8). The morphology of testes cells of male cats may be related to seasonal change with seasonal sperm production (9). Fertility of toms extend on all year seasons, but at the female fertility season, the behaviors of toms totally changed such as become more aggressive and wandering for long distances especially at night (10). The aim of this study was to evaluate the behavioral, hormonal and histological changes in the male genital system of male cats (Free-living non-pedigree) in the center of Al-Diwaniya city, Al-Qadisiya province.

Materials and methods

Forty seven local breed free housing male cats hunted from the streets of the center of Al-Diwaniya city were included in this study. The study was hold during a period of one year (from 15/1/2012 to 15/1/2013) and divided according to the sexual activity of the male cats into three periods including: 1st period which extended from mid-January to February, the 2nd period from February to November and the 3rd period from November to mid-January; Ten male cats were used in the 1st period and ten in the 3rd period, while the other twenty seven were included in the 2nd . The cats were monitoring during the night and early morning by car touring in the streets of the city and the sexual behavior was filmed by using digital camera (Sony Cyber-Shot DSC-H70 16.1 MP Digital Still Camera with 10x Wide-Angle Optical Zoom G Lens and 3.0-inch LCD- Black) and then the male cats were hunted by a special trap made locally for that purpose and after capture general anaesthesia by i.m injection with a combination of Ketamine hydrochloride (15mg/Kg BW; Ketamine, Trittav, Germany), Xylazine (1.1mg/Kg BW; Xyla, Interchemie, Holland) and Atropine sulphate (0.05mg/Kg BW; Atropine, Al-Sharq company, Syria) (11), after that, 3 ml of blood sample from the jugular vein was drew. The blood samples were centrifuged for 20 minutes to obtain serum for hormonal assay which include testosterone and luteinizing hormone (LH) by using Tosoh AIA-360 apparatus (Tosoh company, Japan). The castration also done immediately after animal anesthetized. The tissue autopsies were fixed in 10% formalin till doing the routine histological process. The postoperative care including i.m. injection of Amoxicillin trihydrate (7mg/Kg BW; Duramox 20LA, Medmac, Amman-Jordan) for three days, then released from capture (12). **Statistical analysis:** Data were viewed

as mean ± standard error (M±SE), and analyzed using computerized SPSS system. Statistical difference was considered significant at $p<0.05$. (13).

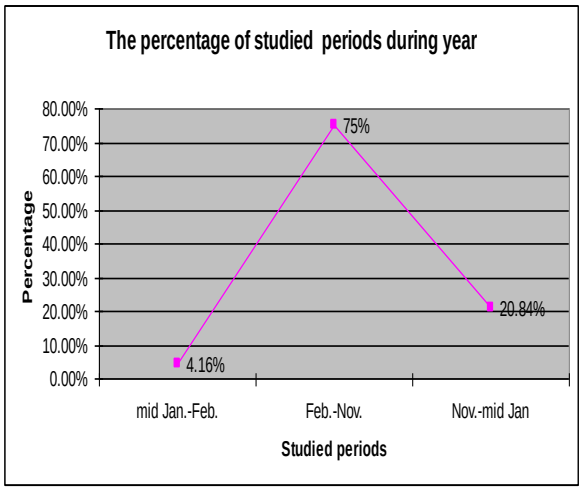
Results

1-Behaviors of Toms:

The results of the 2nd period showed behaviors include caterwaul sound, urination and following of the females with foreplay and mating were observed obviously. The same behaviors were observed in the 1st and 3rd periods except the matting which didn't happened due to refuse of the female (table 1). The percentage of studied periods during year (mid-Jan. to Feb., Feb. to Nov. and Nov. to mid-Jan) were 4.16%, 75% & 20.84%) respectively (fig. 1).

Table (1): The notes of behavioral study during the periods of study (mid Jan. 2012-Feb.; Feb.-Nov. and Nov.-mid Jan. 2013).

parameters Periods	caterwaul sound	Fighting	urination	Follow &foreplay of ♀	Mating	Noting of dams and offspring
1st P.	+	+	+	+	-	-
2nd P.	+	+	+	+	+	+
3rd P.	+	+	+	+	-	-



(Fig. 1): The percentage of studied periods during year (mid Jan. 2012-Feb.; Feb.-Nov. & Nov.-mid Jan. 2013).

2-Hormonal study:

The results of the hormonal assay showed that the mean levels of testosterone were (0.42±0.21 ng/ml, 1.31±0.33 ng/ml and 0.35±0.22 ng/ml) in the three periods respectively, while the mean concentrations of LH were 1.31±0.47, 4.86±1.03 and 1.23±0.43 ng/ml for the three periods respectively (table 2). Our results revealed a significant effect in 2nd period as comparative with 1st and 3rd periods at ($P<0.05$).

Table (2): The Concentration of (Testosterone and LH) ng/ml in the serum of experimental animals, during the periods of study (mid Jan. 2012-Feb.; Feb.-Nov. & Nov.-mid Jan. 2013).

Hormones / Periods	Testosterone (mean ± SD) ng/ml	LH (mean ± SD) ng/ml
1st P.	0.42±0.21 a	1.31±0.47 a
2nd P.	1.31±0.33 b	4.86±1.03 b
3rd P.	0.35±0.22 a	1.23±0.43 a

Different letters denote a significance difference ($P<0.05$).

3-Histological study:

The results of testes histology showed that the seminiferous tubules characterized by presence of all developmental stages of spermatogenesis that include: spermatogoneum, primary and secondary spermatocytes and spermatids (fig. 2), with very active vacuolated Sertoli cell filled with secretions, as well as very active Leydig cells in the peritubular tissue (fig. 3), while the 1st and 3rd period characterized by thickening of connective tissue and peritubular tissue as well as not all developmental stages of spermatogenesis were observed the spermatogoneum, primary and secondary spermatocytes (fig. 4,5), but not spermatids.

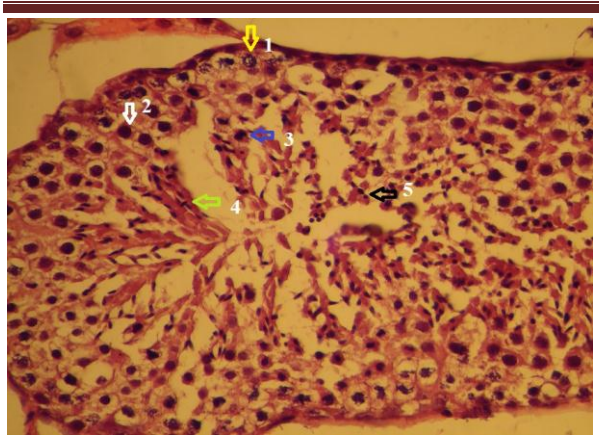


Fig. (2): (Testis of tom) section of seminiferous tubules showed very active vacuolated Sertoli cell filled with secretion (1) with presence of all developmental stages of spermatogenesis that include: spermatogoneum (2), primary and secondary spermatocytes (3&4) and spermatids (5). H&E, 400X.

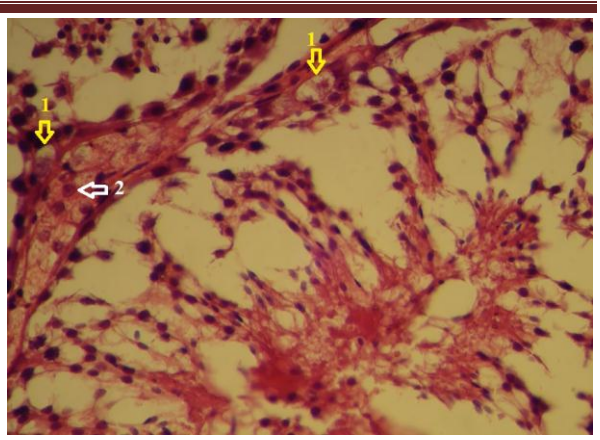


Fig. (3): (Testis of tom) section of seminiferous tubules showed very active vacuolated Sertoli cell filled with secretion (1), as well as very active Leydig cell in the peritubular tissue (2). H&E, 400X.

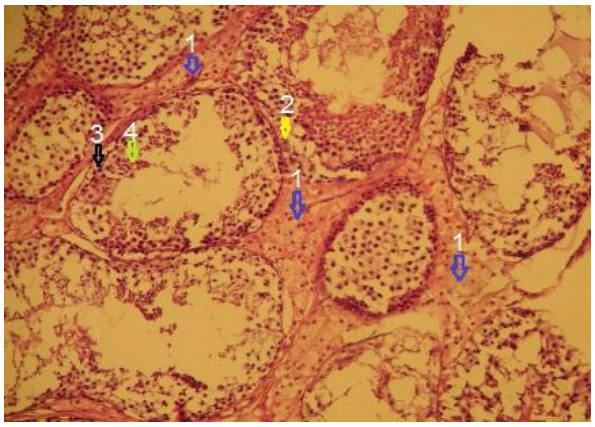


Fig. (4): (Testis of tom) section of seminiferous tubules showed thickening of connective tissue and peritubular tissue (1) with observation of Sertoli cell (2), spermatogoneum (3), and primary spermatocytes (4). H&E, 100X.

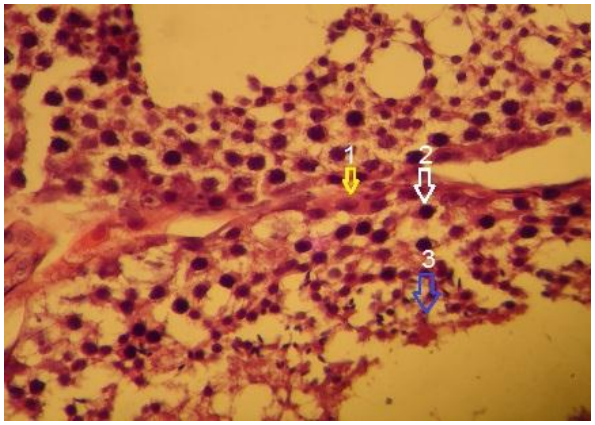


Fig. (5): (Testis of tom) section of seminiferous tubules showed Sertoli cell (1), spermatogoneum (2), and primary & secondary spermatocytes (3). H&E, 400X.

Discussion

1-Behaviors of Toms:

In the table (1), the results reveal caterwaul sound, urination and following of the females with foreplay and mating in 2nd period , while 1st and 3rd periods have same behaviors except the mating, these results agree with results are obtained by (3) who found that the cat affected by season, and with (10) who found that fertility of toms extend of all year seasons, but the females refuse mating until the beginning of breeding season (estrus phase). The periods during this study (mid-Jan. to Feb., Feb. to Nov. and Nov. to mid-Jan.) as show in fig. (1) are recode (4.16%, 75% & 20.84%) respectively, the percentages of our results are in accordance with (8) who found that natural photoperiod affect in sperm quality, as well as, compatible with (2) who mentioned

increasing daylight length is affect in breeding in domestic cats.

2-Hormonal study:

Hormonal levels show significant effect in 2nd period for each (Testosterone and LH) comparative with 1st and 3rd periods (table 2), this result agree with (14) who indicate that plasma LH and testosterone concentrations were increased during breeding season in male cats, as well as (15) who notice that serum LH concentrations are higher during breeding season in male Pallas' cats, but we differ with (8) who mentioned that natural photoperiod moderately varied in serum testosterone concentrations. Our interpretation of the 2nd period is show a significant effect when compared with the other periods in that, the increased levels or concentrations are related with the increased sex desire and mating.

3-Histological study:

Testes histology in 2nd period shows the presence of all stages of spermatogenesis (spermatogoneum, primary and secondary spermatocytes and spermatids) in the seminiferous tubules, and developing Sertoli cells with filling secretion and Leydig cells are very active in the peritubular tissue, while in 1st and 3rd periods the histological changes shows seminiferous tubules with thickening of connective tissue and peritubular tissue as well as not all developmental stages of spermatogenesis are observed (spermatogoneum, primary and secondary spermatocytes), our results are in accordance with (9) that showed seasonal changes in testis cell morphology and sperm production in tom, and with (8) who found that natural photoperiod affect in sperm quality.

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