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Seasonal effects on androgens metabolism in Dromedary Male Camel (*Camelus dromedarius*)

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Abstract

Dromedary male camel displays unique sexual behavior and secretion of poll gland during breeding season. This study aimed to determine the effect of season on metabolism of androgens and the role played by the poll gland there on. Blood and tissues from testes, poll gland and liver were collected from camels during breeding and non breeding seasons to estimate steroid concentrations testosterone (T), 5α- dihydrotestosterone (DHT) and estradiol (E2) and enzyme activity. Appreciable the activity of enzymes were found in testes and poll gland of rutting camels. Activity of α-reductase was equivalent in testes of rutting and non- rutting camels. However, it was significantly higher in poll gland of rutting than in non rutting camels. The activity of the enzyme in rutting camels was significantly higher in poll gland than in testes. Aromatase activity was significantly greater in testes and poll gland of breeding males as compared to non- breeding ones. The liver of both breeding and non-breeding camels has low level of enzyme activity. The peripheral serum concentrations of testosterone 5α- dihydrotestosterone (DHT) and estradiol-17β (E2) were significantly higher in breeding compared to non- breeding camels. The Results obtained indicate that breeding season leads to activation of poll gland with increasing the two pathways; α-reductase and aromatase activities and excessive DHT and E2 production in the camel.

Keywords: androgens, aromatase, camel, season, α-reductase

Introduction

The breeding season of dromedary camels in Arabic Peninsula is the coldest season of the year, from November to February (Tingari et al., 1984). The rutting camel becomes unpredictable and aggressive towards other male camels and humans. Three main neurotransmitters of the neuroendocrine system regulating the aggression behavior in the brain while testosterone stimulates the subcortical areas of the brain to produce aggression, cortisol and serotonin play antagonistic roles in inhibiting testosterone effects. (Menelaos and Batrinos, 2012).

The typical aggressive behavior of a rutting camel includes grinding of teeth accompanying extrusion and inflation of soft palate (dulaa), gargling sounds and profuse salivation. Secretion of occipital gland (poll gland) which is blackish in colour, thick in consistency and has strong unpleasant odor. Typical scent markings in which poll gland secretion is rubbed on to shrubs and sand. Hind legs are wide

spread that the camel looks bigger and more threatening. Urine is dispersed and sprinkled across the body by movements of the tail, which creates strong odour Flehmen is displayed after having picked up the female urinary smell.

Seasonal breeders like camels (Musa, 2004) have dormant phases of reproductive cycle, which could suggest a potential difference in testosterone metabolism, reflected in different hormone levels during the cycle. The androgen concentrations in blood of camels were increased during rutting season far higher than in bovine bulls and were correlated with radical changes in behavior of the animals (Yagil and Etzoin, 1979). The high concentration of testosterone correlates practically with antisocial behavior (Jack and Dennis, 2007). In previous studies, during rutting season of camels, androgenic effect and hyperactivity of poll glands were interrelated (Yagil and Etzoin, 1980; Tingari and Rahma, 1984 and Musa, 2004). In the brain, such stimulating behavior of testosterone is interlinked with two transformation pathways (i) aromatase enzyme metabolize testosterone into estradiol-17 β (E2) (Lephart, 1996) and (ii) reductase enzyme 5 α –converts T to dihydrotestosterone (DHT) (Martini 1982). This study was carried out to investigate the effect of breeding season on metabolism of androgen in rutting camels and the role played by the poll gland there on.

Materials and Methods

Animals

Twelve mature healthy male camels (*Camelus dromedarius*) were used, aging 7-8 years and weighing 500- 600 kg. Animals were kept under nomadic conditions in an open yard in a private farm sponsored by University of Dammam during the breeding season or non- breeding season. Each one was fed daily on 2kg of mixture of barley and wheat bran, and Hay and water were provided *adlibitum.*

Experimental design

Camels were allocated to 2 experiments, each containing 6 animals as Follows:

Experiment 1: Rutting camels were slaughtered at mid breeding season to obtain blood and tissues from testes, liver and poll glands situated at the occipital region between the two ears. Tissues were placed in liquid nitrogen. Jugular blood was obtained, centrifuged at 1500g, and serum was stored at -30° C until analysis.

Experiment 2: Non-rutting camels during non- breeding season were slaughtered to obtain blood and tissues from testes, liver and poll glands as in experiment 1. Pieces of liver, testes and poll glands were homogenized in 250 mM sucrose/ 50mM potassium phosphate buffer, and stored at- 60°C ready for the assay.

Analytical procedures

Enzyme activity:

Aromatase and 5α-reductase were measured by modified methods of Lephart et al. (1989) and Wade (1997),

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Hormonal measurements

Testosterone, E2 and 5α-DHT were estimated by methods previously described and validated for the camel serum (Homeida et al., 1988). The intra-assay co-efficient of variation for testosterone, E2 and DHT were 9.2%, 8.4% and 7.8%, the sensitivity was 5.1, 4.1 and 6.2 pg per tube, respectively. Efficiency of radioactive hormone recovery was 78% for testosterone, 83.3% for E2 and 76% for DHT and values were corrected for extraction losses.

Statistical method

The data subjected to two tailed *t* test to analyze the seasonal differences.

Results

Activity of α- reductase was equivalent in testes of breeding and non-breeding camels. However, it was significantly ($P<0.001$) higher in poll gland of breeding than in non-breeding camels. The activity of the enzyme in breeding camels was significantly ($P<0.05$) higher in poll gland than in testes.

Aromatase activity was significantly ($P<0.05$) greeter in testes and poll gland of breeding, than in non- breeding males.

The liver of breeding and non-breeding camels has low level of enzyme activity. Testes, poll gland and serum concentrations of T, DHT and E2 are shown in Fig 3. A, B, C respectively Significantly ($P<0.001$) higher levels of T, DHT and E2 were shown in breeding compared to non- breeding ones.

pmol/min.mg protein

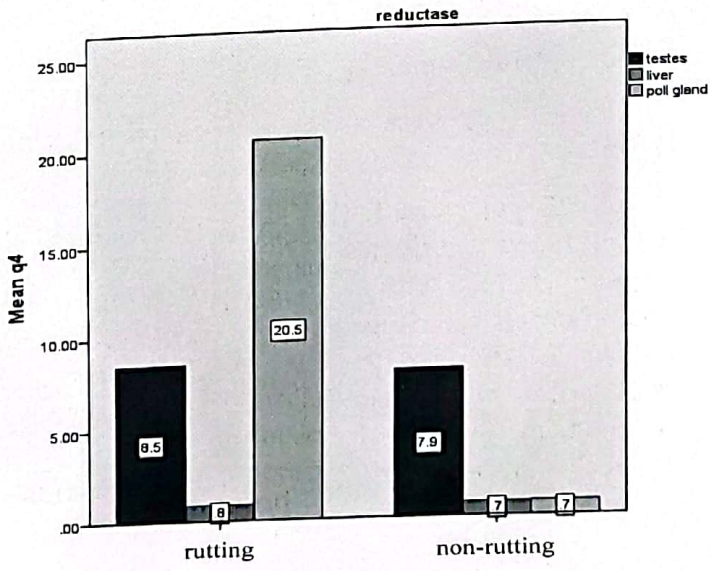

Fig.1: Mean activity of aromatase (rate of estradiol17β production, pmol/min.mg protein) in testes, liver and poll gland of rutting and non-rutting camels (N=6).

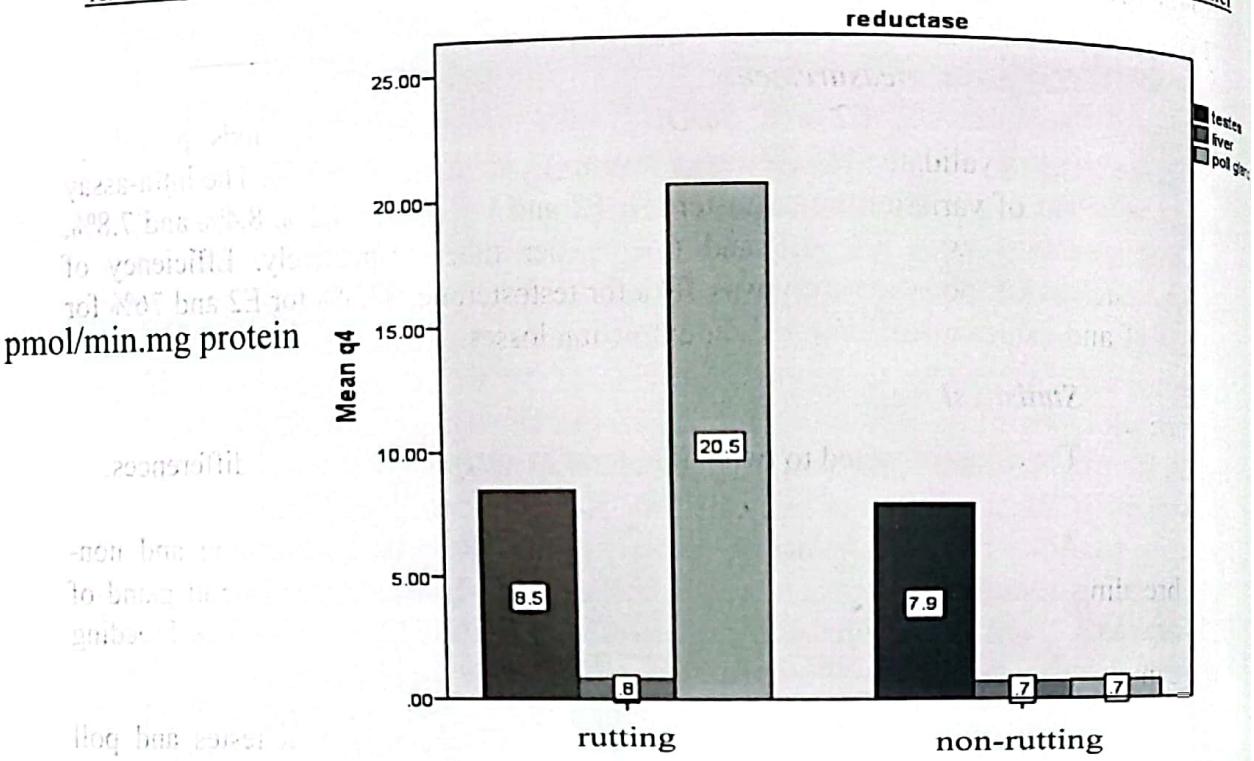

Fig.2: Mean activity of 5α-reductase (rate of 5α-dehydrotestosterone production, pmol/min.mg protein) in testes, liver and poll gland of rutting and non-rutting camels (N=6).

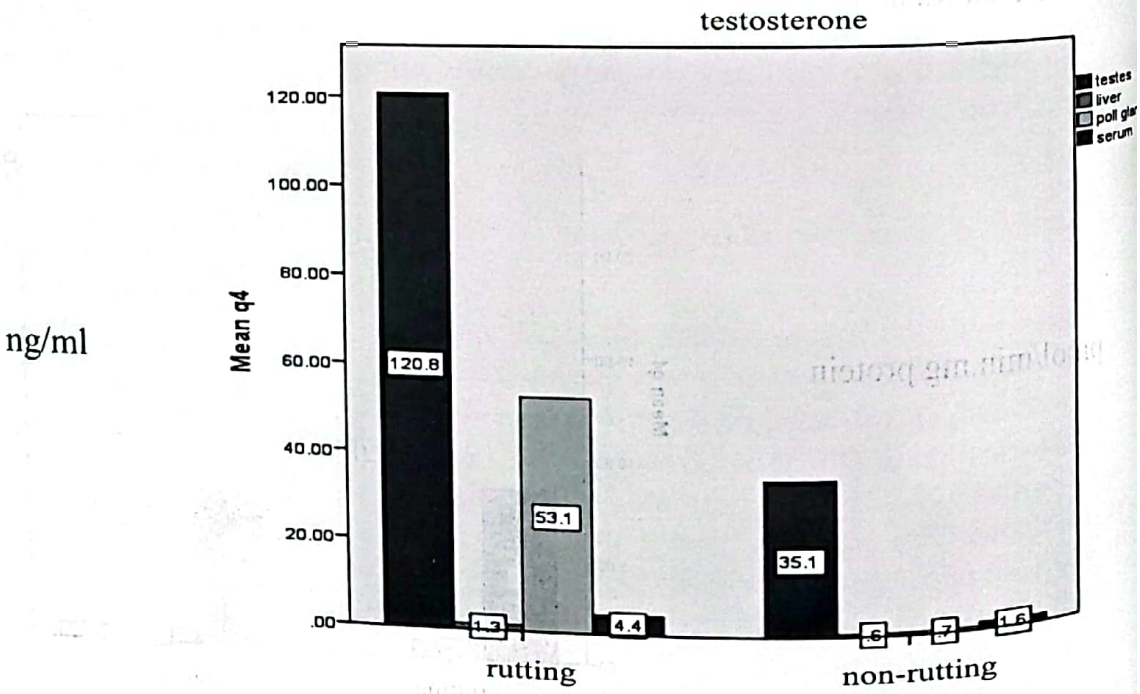

Fig 1 .A

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pg/ml

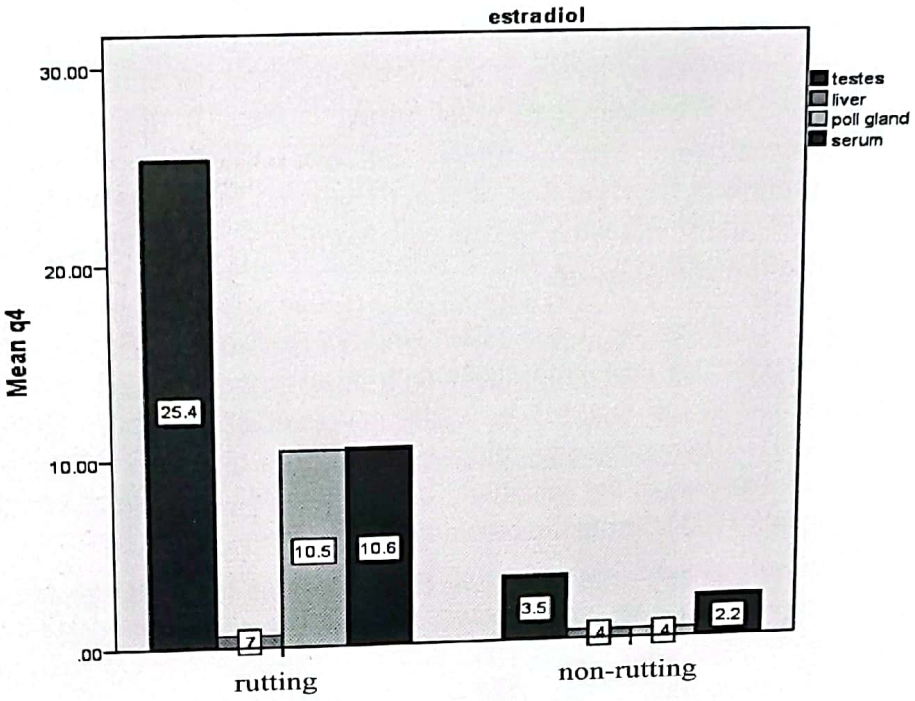

Fig 3 .B

ng/ml

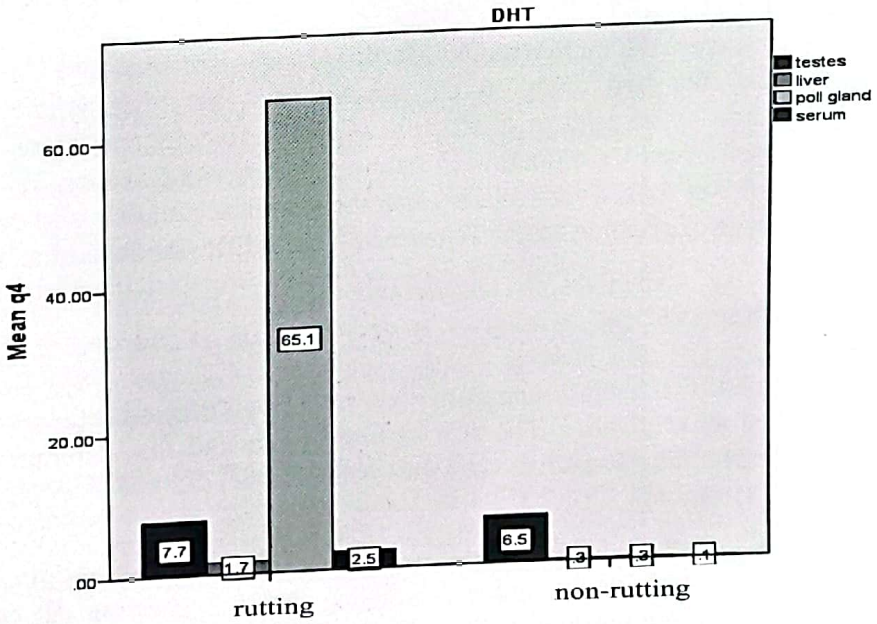

Fig 3 .C

Fig.3. A, B, C: Mean concentrations of testosterone (ng/ml), estradiol17β (pg/ml) and 5α-dihydrotestosterone (DHT) (ng/ml) in testes, liver, poll gland and serum of camels in rutting and non-rutting.

Discussion

In the present study, and during the breeding season, the rutting camel was characterized by having higher levels of testicular and peripheral serum testosterone and DHT compared to non- rutting camels. In line with previous findings, the concentration of testosterone in testicular tissue (Berndston et al., 1983; Johnson and Thompson, 1987) and in plasma (Yagil and Etzion, 1980; Nasr and El- Azab, 1990; El- Harairy and Attia, 2010) was found to increase during breeding season than in non- breeding season.

However, these authors did not measure DHT in their camels. The increased secretion of these androgens may be as a result of effects of one or more factors including the increase in leydig cell number per testes (Johnson and Thompson, 1987). The increase in volume of interstitial tissue (El- Harairy and Attia, 2010) and the increase in the sensitivity of leydig cells to enhanced secretion of LH (Agarwal et al., 1991) during the breeding season.

The results also show that the metabolism of androgen was increased during breeding season. Testosterone is rapidly metabolized to estrogens (aromatizing pathway) or to 5α- DHT (5α- reductase pathway) (Collotti et al., 1980; Wilson and Brouchovsky, 1968). Higher activity of 5α- reductase was demonstrated in poll glands and consequently higher concentration of DHT was produced in rutting camels. The fact that 5α- DHT is more potent than testosterone (Godfrey et al., 1992)

Massa and Maritin (1974); Homeida and Cooke (1984) demonstrate that the poll gland in the present case has high activity of α- reductase and contributed a lot to production of DHT. Histological examination showed that the gland may be endocrine in nature (Tingari and Rahama, 1984). However, from the present data, it seems likely that the glands accumulate and extensively metabolize T to DHT. Further research is needed to confirm the endocrine nature of the glands.

The data also suggest that there is a possibility that the poll gland is steroid-dependent and is involved in accumulation of androgen rather than their synthesis (Tingari and Rahma, 1984; Tingari and George, 1984) Dependency of the poll glands on testosterone from the testis is well demonstrated by the effect of castration on these glands. The size of poll glands and the secretion decrease rapidly after castration Secretions of the poll glands disappear completely 10 days post-castration.

In contrast, 5α- reductase activity was equivalent in testes homogenate from males in breeding and non-breeding season suggesting this certain enzyme activity is required for the function of testes during the whole year round.

In this study a significantly higher levels of E2 in testes than in general blood compartment has occurred favoring a testicular source of estrogen in the camel as in other species (Carreau, 2001). A measurable of aromatase activity was demonstrated in camel testes as in case of stallion (Sipahutar et al., 2003), further suggest a role of estrogen in camel line testicular function. Estradiol is now considered as a survival factor for germ cells in human testis (Pentakainen et al.,

2000). In monkeys, a blockage of spermatic maturation has occurred following treatment with aromatize inhibitors (Shetty et al., 1997). In irradiated rats and mice recovery of spermatogenesis has occurred following treatment with estradiol (Shetty et al., 2002) and even more by phytoestrogens (Adeoya- Osiguwa et al., 2003). Aromatase is demonstrated in Leydig cell of most mammalian species (Carreau et al., 2001). Moreover, aromatase in sertoli cells may change with the season as in black bear (Tsubota et al., 1997) an animal considered to be a true seasonal breeder. Therefore, during breeding season the presence of aromatase and E2 in the testes of rutting camel may be related to the initiation of testicular recrudescence and delimitation of spermatogenesis. The metabolic converting process of steroid; E2 derived from T in several species is an critical step in male control (Whalen et al., 1985, Meisel and Sachs, 1994) as well as the female (Homeida, and Cooke, 1984) sexual behaviors.

Conclusion

Looking at the pattern of changes of the hormones in camels during breeding season, it is likely that, in this animal both T metabolites, E2 and DHT, are required to activate the full complement of muscling sexual and aggressive behaviors.

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تأثير الموسم على أيض هرمونات الذكورة في الجمل العربي

بدرية الصويغ ، ابتسام السحيمي، آمال أكبر، عزيزة الخالدي ، عبدالقادر حميدة

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تعرض ذكور الجمل العربي نموذجاً فريداً للسلوك الجنسي ولإفراز غده البل خلال موسم التكاثر. هدفت الدراسه الي معرفة تاثير الموسم علي استقلاب الأندروجين والدور الذي تلعبه غده البل هناك. تم جمع عينات الدم والأنسجه من الخصيتين ،غده البل والكبد خلال فصل التكاثر واللاتكاثر. أظهرت النتائج انه يوجد نشاط ملموس ومعنوي لإنزيمي الأروماتير و٥- الفا رداكتيز في الخصيتين وغده البل في الجمل المتخدد. كان نشاط الفار- داكتيز في الخصيتين متعادل في الجمال المتخدده والغير المتخدده إلا أنه كان مرتفع بشكل معنوي في غده البل في الجمال المتخدده عنه في اللامتخدده، كان أعلي معنويا في غده البل عنه في الخصيتين. ارتفع نشاط انزيم الاروماتير بشكل معنوي واضح في الخصيتين وغده البل في موسم التكاثر عنه في موسم اللاتكاثر. كما لوحظ إنخفاض نشاطه في الكبد في كلا الموسمين. كانت تركيزات البلازما من هورمونات التسيتوستيرون والاستراديول اعلي معنوياً في فصل التكاثر عنه في فصل اللاتكاثر. أشارت النتائج التي تم الحصول عليها أن موسم التكاثر يؤدي الي تنشيط غده البل مع زياده نشاط إنزيمي الأروماتير والرداكتيز وزياده مفرطه في انتاج هورموني التسيتوستيرون والاستراديول في الجمال.