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EFFECT OF BREED OF FALCON ON SEMEN QUALITY TRAITS

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Abstract

This study was conducted to determine the effect of falcon breed on semen quality traits. A total of 12 sexually matured males from three breeds of falcon (4 falcon males from each breed), which were Gyr, Saker and Peregrine falcons, were used in this study. Birds were reared at typical falcon houses which were provided with all standard requirements for falconry. During the reproductive season (January – April) all males were trained on semen collection procedure by using special protocol to handle these birds. Semen samples were collected from all males on a fortnightly basis. Semen traits involved in this study were ejaculate volume, sperm concentration, total number of spermatozoa, mass motility, individual motility and percentage of dead spermatozoa, abnormal spermatozoa and acrosomal abnormalities. Result revealed that there are significant differences between three breeds of falcons in regards to the semen quality characteristics involved in this experiment. Peregrine falcon recorded the highest values ($P \leq 0.05$) with respect to ejaculate volume, sperm concentration, total number of spermatozoa, mass motility and individual motility followed by the results of Saker falcon, while Gyr falcon recorded the lowest values as concerning these traits. Results also indicated that Peregrine falcon surpasses ($P \leq 0.05$) Saker and Gyr falcons concerning the percentages of live spermatozoa, normal spermatozoa and normal acrosomes. However, there were no significant differences ($P \geq 0.05$) between Saker and Gyr falcons with relation to mass motility and percentages of dead spermotoza and acrosomal abnormalities. In conclusion, results of this study clearly indicated that there were significant differences between Gyr, Saker and Peregrine falcons regarding semen quality traits involved in this study and Peregrine falcon excelled the other two breeds of falcon in relation to these semen traits.

Keywords: *Breed bird; Gyr, Saker and Peregrine falcon; Standard requirements; Bird houses; Spermatozoa and acrosomal abnormalities.*

Introduction

Many species of raptors are listed as endangered [1, 2] and captive propagation and release programs have been successful with some [3]. Raptors can pose special challenges to captive propagators: small captive populations, incompatible and infertile pairs, and small semen volumes. Artificial insemination has been used successfully with many of these species [3-5]. Wildlife managers need a reliable method for the preservation of semen to support these captive raptor propagation programs [6].

The best quality semen is donated voluntarily, either during natural copulation, or by copulation with another object (which the bird has been previously trained to use [generally a hat, knee, cushion or glove]). Natural copulation only takes 10-20 seconds but may be repeated up to twice every hour during the breeding season [7]. Birds tend to copulate more in the early

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morning and during warm or mild weather. Early season semen is often of poor quality. Larger male falcons (e.g. Peregrine, Saker and Gyr) start to produce semen in their second season but reach peak production by their fourth season [8]. The 'conditioning' of a male semen donor is very time consuming, as a genuine breeding relationship must be forged and maintained throughout the season, between bird and the same keeper [9]. A good voluntary donor is invaluable, but a breeder should never rely on one donor alone. If a male will not donate semen voluntarily, he may be 'stripped', although this does require considerable experience as well as a good working knowledge of male avian anatomy and tends to produce inferior quality semen. Considerable practice and experience is required to achieve this in an effective and a traumatic manner [10].

Captive breeding has been used effectively for the conservation of birds of prey for many years. Artificial insemination has been a useful technique in captive breeding programs since the early 1970's [11]. There is a significant trade in, and demand for, birds of prey, especially falcons Falco spp., for the sport of falconry and for captive collections. Techniques for captive breeding of peregrine and other falcons were originally developed in North America, and to a lesser extent in Europe, in the 1960s and 1970s and focused on the peregrine falcon, firstly because of its desirability for falconry and, secondly, to provide birds to re-populate, through re-introduction, some declining or extirpated peregrine populations [12].

In Iraq we have private natural preserve for breeding and proliferation of falcons, called Babylon Preserve for Breeding and Proliferation of Ostrich, Falcon and Deer. In this preserve we bred three breeds of falcons, namely Gyr, Saker and Peregrine. Therefore, the present study was conducted to determine the effect of falcon breed on certain semen quality traits of Gyr, Saker and Peregrine falcons.

Materials and Methods

This study was conducted at Babylon Preserve for Breeding and Proliferation of Ostrich, Falcon and Deer, Babylon Governorate, Iraq from January to April, 2014-2015 (breeding seasons). A total of 12 sexually matured males from three breeds of falcon (4 falcon males from each breed), which were Gyr, Saker and Peregrine were used in this study. Birds were reared at typical falcon houses which were provided with all standard requirements for falconry (Fig. 1).

Fig. 1. Falcon pens and the requirements used in the falconry.

All males were fed on live pigeons which were introduced to males directly after been slaughtered and cleaned from feather especially the regions of chest and abdomen of pigeons as the falcon prefer to feed on live birds or fresh and recent slaughtered bird (Fig. 2). However, mixtures of vitamins and fish oil were added on the pigeon carcass to enhance the reproductive performance of male falcon. Males where supplied with photoperiod schedule of 16 L: 8 D.

Fig. 2. Providing the slaughtered pigeon as a main food meal for falcon

During the reproductive season (January – April) all males were trained on semen collection procedure by using special handling protocol for these birds which include extreme caution when dealing with these birds as they are raptor birds and have a very strong beak which can remove fresh meat very easily from the body and they have very strong claws used to grab prey and assist in cutting prey. However, they are very nervous and fierce birds and therefore special ways to deal with them must be found. The procedure of collecting semen from falcon males could be summarized as follows: a person that has experience in dealing with these birds grips the falcon and then puts the head cap on the top of the male head to calm him as this not allow him to see what is happening around him (Fig. 3). After the male gets settled, repeated back massage is performed to ensure the male is calm on the one hand and on the other hand to start the stimulation (Fig. 4).

After that the male has been grabbed from behind, using the towel to control the bird and to avoid injuring the person that does the semen collection procedure by the beak or strong and sharp claws of the falcon (Fig. 5a), the falcon is placed in the lap of the person holding him with his back down in order to be brought under control (Fig. 5b). After that, the edge of the towel is put on his head to calm him down and to avoid any injuries to the person handling it (Fig. 5c). After the falcon settles down, his legs are caught between the left hand fingers of the handler (Fig. 5d). This process allows full control on the falcon's legs and ensures that he can't use his sharp claws to hurt the technician and in the same time enables the technician to use his fingers that caught the falcon's legs to start the massage around the falcon's vent and start the stimulation process and semen collection (Fig. 5e).

Fig. 3. Holding the falcon male and put the bonnet on his head.

Fig. 4. Massage of falcon back to calm him down and stimulate.

Semen collection process begins through the simultaneous massage of the higher part of the vent using the fingers of the left hand that caught the falcon's legs and behind the vent, using the right hand fingers (Fig. 5f). This process continues until the male papillae emergence from the vent, which indicates a good response from the male (Fig. 5g and h) and then a capillary tube was placed on the edge of papillae as the semen flows inside the capillary tube by capillary property (Fig. 5i). Several capillary tubes would fill (Fig. 6a, b). However, an appropriate pipette can be used to suck the semen flowing from the male vent (Fig. 6c). After the semen collection procedure was completed, the semen samples were evaluated by using standard methods. Semen quality traits included in this study were mass and individual motility of spermatozoa, spermatozoa concentration and percentage of dead spermatozoa, abnormal spermatozoa and acrosomal abnormalities (Fig. 7 and 8). These semen quality traits were evaluated according to standard procedures reported by Weaver and Gee [3, 4]. Semen samples evaluation was done on a monthly basis from January to April, 2014-2015 (breeding seasons).

Fig. 5. The Falcon manipulation: a - holding the falcon male from his behind by using the towel; b - place the falcon with his back to the bottom in the lap of the person carrying out semen collection; c - covering the head of falcon male with a towel to avoid his sharp peak; d - holding the legs of falcon with left hand of a technician to avoid his sharp claws; e - starting the stimulation of male falcon by massaging around the vent of male with the left hand fingers; f - simultaneous massage of higher part of the vent with constant massage behind the vent of male falcon g and h - The emergence of papillae from the vent of male in response to massage process; i - semen flowing from male papillae

Fig. 6. Semen collection by: a and b - using capillary tubes; c - using micropipette

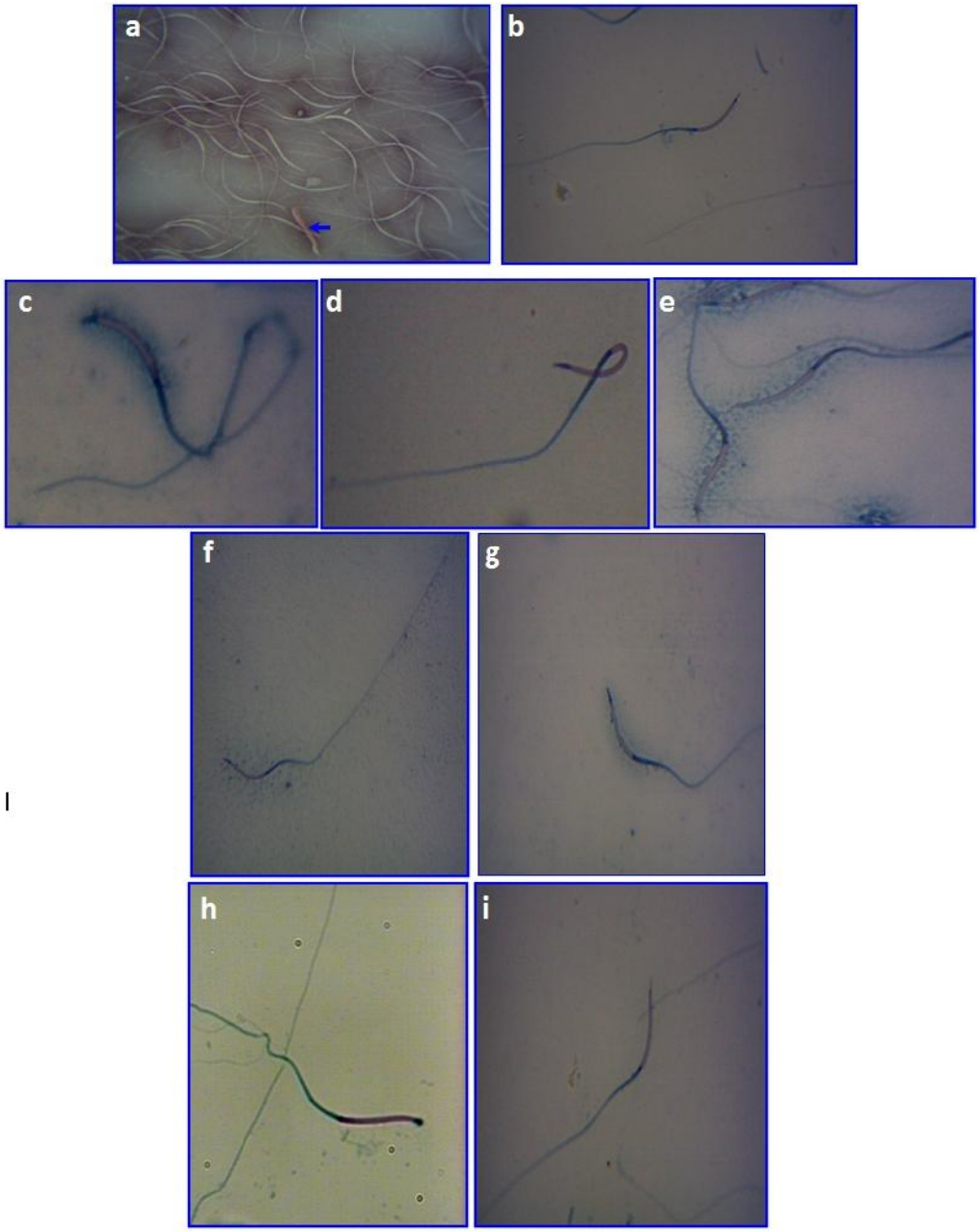

Fig. 7. Semen morphology on microscope: a - live sperms stained with white stain while dead sperms stained with red stain (blue arrow); b - sperm with normal morphology; c, d and e - sperms with different deformities; f and g - sperms with normal acrosomes; h and i - sperms with abnormal acrosomes

Statistical comparisons based on 8 semen samples for each male were done on a monthly basis from January to April, 2014-2015 (breeding seasons) in regards to ejaculate volume, sperm concentration, mass motility, individual motility, dead sperm, abnormal sperm and abnormal acrosome. The data was assessed by analysis of variance using the General Linear

Model method [15]. Test of significance for the difference between means of different oil treatments was done by Duncan's multiple range test [16].

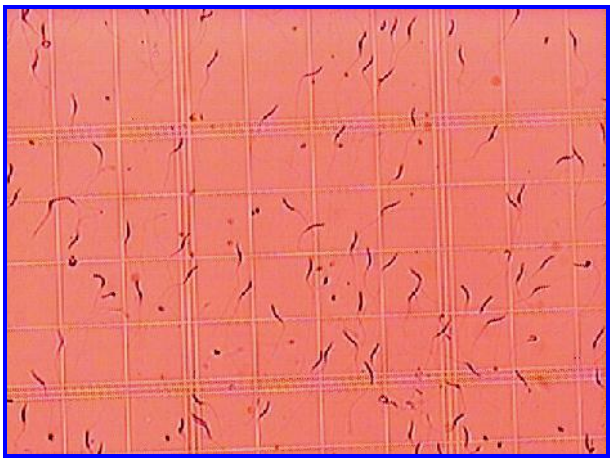

Fig. 8. Determination of spermatozoa concentration by using Haemocytometer.

Results and Discussion

Results from Table 1 revealed that there were significant differences ($P \leq 0.05$) between the three breeds of falcon, in regards to ejaculate volume, sperm concentration, total number of spermatozoa and mass motility (Table 1). Peregrine falcon recorded the highest values ($P \leq 0.05$) of ejaculate volume, sperm concentration, total number of spermatozoa and mass motility followed by the results of Saker falcon, while Gyr falcon recorded the lowest values ($P \leq 0.05$) of ejaculate volume, sperm concentration, total number of spermatozoa and mass motility (Table 1).

Table 1. Effect of breed of falcon on semen quality traits (Mean ± SE).

Traits	Breeds		
	Gyr falcon	Saker falcon	Peregrine falcon
Ejaculate volume (µl)	40.2 ± 1.8 ^{c}	55.6 ± 2.8 ^{b}	67.1 ± 3.7 ^{a}
Sperm concentration (10^6/ ml)	41.3 ± 1.5 ^{b}	44.5 ± 2.7 ^{b}	55.3 ± 3.6 ^{a}
Total number of spermatozoa (10^6)	1660.2 ± 35.9 ^{c}	2474.2 ± 58.9 ^{b}	3710.6 ± 66.8 ^{a}
Mass motility (%)	66.9 ± 2.8 ^{b}	70.8 ± 3.9 ^{b}	79. 9 ± 4.6 ^{a}

^{a,b,c}Means within a row with different superscript are different ($P \leq 0.05$).

Each value represents a mean of semen samples evaluation that was done on a monthly basis from January to April, 2014-2015 (breeding seasons).

As shown in Table 2, there were significant differences ($P \leq 0.05$) between the three falcon breeds, with respect to individual motility and percentages of dead spermatozoa, abnormal spermatozoa and abnormal acrosomes. Peregrine falcon surpasses the other two breeds of falcon (Saker and Gyr) concerning mass motility, while Gyr falcon exhibited the lowest means in relation to this trait (Table 2). However, Peregrine falcon showed the lowest values ($P \leq 0.05$) in relation with the dead spermatozoa abnormal, spermatozoa and abnormal acrosomes percentages, whereas Gyr falcon recorded the highest means ($P \leq 0.05$), followed by the results of Saker falcon (Table 2). Each value represents a mean of Semen samples evaluation that was done on a monthly basis from January to April, 2014-2015 (breeding seasons).

Table 2. Effect of breed of falcon on certain semen quality traits (Mean ± SE).

Traits	Breeds		
	Gyr falcon	Saker falcon	Peregrine falcon
Individual motility (%)	68.8 ± 3.5 [c]	74.2 ± 1.9 [b]	83.9 ± 5.6 [a]
Dead spermatozoa (%)	30.9 ± 1.1 [a]	28.5 ± 1.3 [a]	21.9 ± 2.0 [b]
Abnormal spermatozoa (%)	31.8 ± 2.0 [a]	25.1 ± 1.7 [b]	20.0 ± 1.2 [c]
Abnormal acrosomes (%)	15.3 ± 0.9 [a]	13.2 ± 1.8 [a]	8.9 ± 0.8 [b]

[a,b and c] Means within a row with different superscript are different (P≤ 0.05).

The data in relation to the effect of falcon breed on the semen quality traits are so limited or absent. The reproductive anatomy and function of raptors have attracted little attention to date. Basic information, such as the presence and location of sperm-storage tubules and the duration of the fertile period, is still unknown for most species. This paucity of knowledge not only acts as a limiting factor for improved reproductive success in captive breeding programs, but also renders it more difficult to understand the reproductive ecology of wild raptors [17]. Spermatogenesis in male birds of prey depends on follicle-stimulating hormone (FSH), testosterone, the activity of Sertoli cells and their interaction with the spermatogonial stem cells. Seasonal testicular growth usually takes up to 45 days in the majority of raptor species, a period longer than ovarian growth in the female. FSH and LH, as well as testosterone, are essential for spermatogenesis [18, 19]. The process of spermiogenesis, and the duration of the transport through the excurrent ducts are unknown, but it is clear that fluid is absorbed to concentrate sperm and to become seminal plasma. Seminal plasma differs from blood plasma in regards to electrolyte and protein composition [20, 21]. The importance of this process is not well understood, but is likely related to sperm mobility more than fertilizing ability, since testicular sperm are able to penetrate the inner periviteline membrane in vitro [22]. Semen production period varies among species and individuals, but usually last for nearly three months. Bird described an average period of 74 days for captive American Kestrels, with a maximum of 103 days [23].

Longer periods were found for Peregrine falcons (95 days)[24] and eagles (up to 110 days)[20]. Semen production in American Kestrels held in Montreal, Canada begins at about 12 hours and 45 minutes of daylight, and declines considerably at about 15 hours and 45 minutes [24]. Ejaculate characteristics vary greatly among species, individuals and collection method [11, 25, 26], male reproductive condition, nutrition [27], certain pollutants [28] and climate [23]. Concentrations ranging from 31,000 to 40,000 spermatozoa per mm^3 and volumes between 3 and 14.6µL have been reported for the American Kestrel [23, 24, 29). Expectedly, the ejaculate volume increases with species size. Semen volume in Peregrine Falcons can be as high as 95µL [30], with cell concentrations ranging from 26,000 to 81,000 sperm per µL. Sperm production varies seasonally; sperm concentration increases early during the breeding season, peaks in mid-season, and declines after. This pattern varies longitudinally. Numbers of spermatogonia, spermatids and abnormal spermatozoa are more likely to be present in the both early and late season ejaculations when testosterone levels are lower than normal. This is related to the need to ensure maximum sperm quality at the time of maximal frequency of copulation prior to egg laying [31]. Urine contamination of semen and subsequent sperm damage is frequent during collection using forced-massage techniques [23]. Fox provides a useful description, including an illustration of the various contaminants in raptor semen [7]. The use of modified diluents may help reduce deleterious effects [31]. *Escherichia coli* is the most prevalent bacteria contaminating raptor semen. Samples need to be evaluated with caution before artificial insemination to avoid the risk of ascendant salpingitis [32)]. Further research is needed to unravel some of the major questions including the spatial requirements and factors involved in the reproduction control of endangered raptors. Finally, an improved knowledge of the reproductive physiology of raptors will help us better understand the impact that the

chemicals released into their environment have on their reproduction and, ultimately, their survival.

Conclusions

It was concluded from this study that the falcon breed has a significant effect ($P \leq 0.05$) on semen quality traits included in this study, which were ejaculate volume, sperm concentration, total number of spermatozoa, mass motility, individual motility and percentages of dead spermatozoa, abnormal spermatozoa and abnormal acrosomes. Peregrine falcon recorded the best results as regards semen quality traits included in this study, while Gyr falcon recorded the worst results concerning semen quality traits included in present study. Therefore further studies with respect to semenial plasma traits and fertilizing ability of spermatozoa will be useful in this field.

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