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Comparative study on physical characteristics of semen of PB2 x Indigenous and Dahlem red chicken

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Abstract
A total of 100 ejaculates had been collected from PB2 x Indigenous and Dahlem Red chicken being maintained at AICRP on Poultry Breeding, Assam Agricultural University, Khanapara, Guwahati-781022 to study the different physical characteristics of semen. The normal colour of semen was creamy white to milky white in both groups. The overall mean ejaculate volume, initial sperm motility, live sperm and sperm concentration was 0.27 ± 0.03 ml, 77.60 ± 1.36 percent, 80.80 ± 1.00 percent and 3487.00 ± 498.49 million per ml in PB2 x Indigenous chicken and 0.29 ± 0.01 ml, 77.10 ± 1.60 percent, 81.20 ± 0.97 percent and 2295.00 ± 120.51 million per ml in Dahlem Red chicken, respectively. The mean ejaculate volume, initial sperm motility and live sperm did not differ significantly but there is a significance difference in sperm concentration between both the groups.

Keywords: Chicken, poultry semen, sperm, physical character

Introduction
The assessment of semen quality characteristics of poultry birds gives an excellent indicator of their reproductive potential and has been reported to be a major determinant of fertility and subsequent hatchability of eggs. For breeding programme selection of males is different between avian and mammalian species. In mammals, the testes are located outside the body, while in poultry, the testes are located within the body. Thus breeding soundness evaluation in poultry does not include examination of the scrotum but must instead rely on macroscopic and microscopic examination of the spermatozoa to determine fertility of the cock (Abdul *et al*).[2] For a cock to be successful, it must be physically able to copulate and transfer semen to the female. Furthermore, fertilization in poultry depends on age, sperm collection, timing between mating, oviposition and sperm quality (Brilard and McDaniel) [4]. The standard semen analysis involved evaluating a number of parameters such as motility, sperm concentration, live or dead count and morphology of spermatozoa etc. Several reports on semen characteristics of the domestic fowls have indicated that breed and strain significantly affects semen quality and quantity (Peters *et al.*) [11], (Tuncer *et al*) [13]. Although, the results of several studies on semen characteristics of the domestic fowls have been published, little or none has been reported on the PB2 x Indigenous and Dahlem red chicken. This study therefore focuses on the comparative evaluation of semen quality between these two groups.

Materials and methods
A total of 100 ejaculates collected from 10 nos. of PB2 x Indigenous and 10 nos. of Dahlem Red chicken aged 10 months maintained at the poultry shed of AICRP on Poultry Breeding, Assam Agricultural University, Khanapara, Guwahati-22 were used to study the different physical characteristics of semen. The PB2 is a synthetic broiler parent and indigenous is a native bird of Assam, India. The PB-2 x Indigenous is a crossbred bird where male PB-2 is selected to cross with female Indigenous bird. The birds (Pb-2 x indigenous and Dahlem red) were reared under cage system and maintained under uniform feeding and managemental practices through out the period of study. All cocks were kept in individual cages and fed with balanced poultry ration and water was provided ad libitum. The semen samples were collected from each bird once in a week because the time range required for semen to pass from the testes to the distal region of the ductus deferens varies from 1-4 days (Munro) [10]. The time of semen collection was between 9.00am to 10.00 am.

The semen samples were collected by the abdominal manual massage method as per Burrows and Quinn [5]. First, the cloacal area was cleaned. The back and tail feathers and the abdominal region were then stroked gently and repeatedly, which resulted in the erection of the phallus. Semen was ejaculated after slight pressure was applied to the inverted cloaca. The semen was carefully collected in a test tube and brought to the laboratory and placed in a water bath maintained at 37º C prior to evaluation.

The colour of semen was recorded based on visual observation in the collecting tube. The total volume of semen was measured in the collecting tube itself. For evaluation of sperm motility, a small drop of semen was placed on a clean grease free slide maintained at 37˚C by placing it over a biotherm. A cover slip was placed over the drop to prevent overflow and allowed a uniform film to form and prevented quick drying of the semen. The slide was examined at 400X magnification under the microscope. Sperm motility was recorded from 0-100 based on percentage of progressively motile spermatozoa. The concentration of the spermatozoa was estimated using a haemocytometer and was recorded in million per ml. The technique involved mixing semen with appropriate diluents at a dilution rate of 1:200 with an eosin solution. The diluting fluid was prepared as Sodium chloride 1.00g, Eosin Y 0.05g, Formalin 1.00 ml and distilled water 100 ml. For estimation of live sperm a drop of semen was mixed with two drops of Nigrosin_Eosin stain on a clean microslide. After incubation of semen stain mixture for one minute, a thin smear was prepared on a clean microslide. Two hundred spermatozoa were counted in each smear under 1000X magnification to determine the percentage of live spermatozoa.

Results and Discussion

The normal colour of fresh semen of PB2 x Indigenous and Dahlem Red chicken was milky white. This was in agreement with the findings of Peters *et al.* [11]. The color of semen is generally an indicator of the density of the ejaculate. The semen of the domestic fowl varies from a dense opaque suspension to a watery fluid secreted by various reproductive glands. It ranges from a relative high sperm density or degrees of clear to milky white, with declining sperm numbers (Peters *et al*.) [11]. The color of semen may depend on the species of bird used, but generally semen should be creamy which indicates a high sperm concentration (Cole and Cupps). [6]

The overall mean ejaculate volume of semen in PB2 x Indigenous chicken was 0.27 ± 0.03 ml and in Dahlem Red chicken was 0.29 ± 0.01 ml which was comparable with that reported by Haunshi *et al.* (2010) [9] in Vanaraja bird. However, the value was lower than that values reported by Peters *et al*. (2008) [11] in Giriraja, White leg horn, Frizzle feathered, Nera black, naked neck. On the other hand the overall mean ejaculate volume was within the range as reported by Renganathan [12]. The cockerel produces between 0.1 ml and 1.5 ml per ejaculation, with 0.6 ml being the average ejaculate volume recorded (Cole and Cupps) [6]. Different cockerels of the same species often produce different volumes of semen at different times (Anderson) [3]. The average volume ejaculated using the abdominal massage technique is approximately 0.25ml (Gordon) [7]. The variation in total ejaculate volume in the present study from that reported earlier studies might be due to managemental practices, difference in breed, strain, season, environmental factor, frequency and technique of semen collection etc. The mean ejaculate volume did not differ significantly between the group of PB2 x and Dahlem Red chicken. The overall mean sperm motility in PB2 x Indigenous chicken was recorded as 77.60 ± 1.36 percent and in Dahlem Red chicken was 77.10 ± 1.60 percent. The present value was in close proximity to the value reported by Tuncer *et al.* [13] in New Hampshire chicken. However, the value was higher than that reported by earlier workers in Miri, Vanaraja, Mizo local and Grampriya cocks (Haunshi *et al*) [9]. There is no significance difference for mean sperm motility in between the PB2 x Indigenous and Dahlem Red cocks.

The overall mean live sperm was found to be 80.80 ± 1.00 percent in PB2 X Indigenous bird and 81.20 ± 0.97 percent in Dahlem Red birds which was close proximity with that reported in El-Salam pedigreed cock by Abd El Ghany *et al* [1]. The present value of mean live sperm percent was higher than that the values reported by Haunchi *et al* [9] in local Indigenous, Vanaraja, Mizo local and Grampriya cocks.

The mean percentage of live sperm did not differ significantly between the group of birds of PB2 x Indigenous and Dahlem Red.

The overall mean concentration of spermatozoa was recorded as 3487.00 ± 498.49 million per ml in PB2 x Indigenous cock and 2295.00 ± 120.51 million per ml in Dahlem Red chicken which was higher than that reported by Haunshi *et al* [9] in Vanaraja, local indigenous, Mizo local and Grampriya bird. The present value in case of PB2 x Indigenous was in close proximity with the values reported by Peters *et al* (2008) [11] in white leg horn and Fizzle feathered bird.

The overall mean concentration of spermatozoa differed significantly ($P \leq 0.05$) between the group of birds PB2 x Indigenous and Dahlem Red chicken. The significance difference in semen concentration between PB2 x Indigenous and Dahlem Red bird could be due to bred and strain. Gordon [7] stated that semen collected from domestic cockerel contains an average sperm concentration of 5000 $\times 10^6$ sperm/ml. On the other hand Hafez and Hafez [8] stated that semen collected from domestic cockerel contains an average sperm concentration of 3000-7000 $\times 10^6$ sperm/ml.

Summary

From the different physical characteristics of semen studied between PB2 x Indigenous and Dahlem Red chicken, it has been found that the overall mean ejaculate volume, initial sperm motility, live sperm did not differ significantly among them but the sperm concentration was differ significantly between PB2 x Indigenous and Dahlem Red chicken respectively.

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