Indian Journal of Animal Sciences **86** (10): 1150–1152, October 2016/Short communication

Age related changes in basal concentrations of FSH, LH and testosterone in indigenous and crossbred bovine males

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Received: 4 February 2016; Accepted: 7 March 2016

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Key words: Age, Crossbred, FSH, Indigenous, LH, Testosterone

Testicular development, puberty, sexual maturity and onset of spermatogenesis depend upon the interaction among follicle stimulating hormone (FSH), luteinizing hormone (LH) and testosterone (O'Donnell *et al.* 2006). Spermatozoa quality in bovine is influenced by the close interaction between Sertoli and germ cells, which is reliant on gonadotropins and testosterone (Bardin *et al.* 1994). Due to episodic secretion of GnRH, wide variations exist in the secretory pattern of FSH, LH and testosterone hormones among different breeds of bovine (Moura and Ericson 1997, Finnerty *et al.* 1998, Moura *et al.* 2011), which may be attributed to the differences in semen quality among different breeds. Further, it was found that the secretion of gonadotropin and testosterone hormone during prepubertal period determines the sperm producing capacity of bull in later stages of life (Moura and Ericson 1997) and more than 1 ng/ml of testosterone concentration in peripheral blood indicates testis growth and sperm production (Moura *et al.* 2011). Although the role of gonadotropins in sexual maturity and sperm production is well studied in exotic breeds, the basal concentration of gonadotropins and testosterone in crossbred and indigenous males has not been established. In the present study, we analyzed the age related changes in basal blood plasma concentration of FSH, LH and testosterone hormone in indigenous and crossbred males.

Peripheral blood samples from different animals at different age groups were utilized for the experiment. Three blood samples were collected in a single day at 1.5 h intervals from crossbred (Holstein Frisian × Tharparkar, 6) and indigenous (Tharparkar, 6) males at 1, 6, 12, 18 and 24 months of age at Livestock Research Centre, National Dairy Research Institute, Karnal, Haryana. Blood plasma was separated by centrifugation @ 3,000 rpm for 10 min at room temperature and stored at –20°C until analysis. Concentrations of FSH and LH in blood plasma were estimated using the FSH and LH Elisa kits as per the procedure given by the manufacturer. The minimum detectable level for FSH was 1.14 ng/ml and for LH was 0.3 ng/ml. The intra-assay variation was 7.65% for FSH and 5.82% for LH. The inter-assay variation was 11.45% for FSH and 8.14% for LH. The intra and inter-assay coefficient of variation (CV) was expressed in per cent by calculating the mean and standard deviation of replicates in one or more assay plates, respectively. Concentration of blood plasma testosterone was estimated using Bovine testosterone Elisa kit as the procedures given by the manufacturer. The minimum detectable level of testosterone was 0.02 ng/ml. The intra and inter-assay variations were 4.38 and 7.23%, respectively. Data obtained were analyzed through Sigma Plot version 11.0®. The data were expressed as mean±SE. One way analysis of variance and student t test (unpaired) were used to analyze the differences among the age groups and within the same age between breeds, respectively.

The mean FSH concentrations (ng/ml) in crossbred and indigenous males are given in Fig. 1. The concentrations of FSH increased from 1 to 18 months of age and then declined at 24 months of age in both crossbred and indigenous males. Although the reason for this phenomenon could not be explained with the available knowledge, it is possible that the age related changes in inhibin concentrations may play a role. It was reported that the concentrations of inhibin in serum and testicular tissue were high in neonatal calves and tended to decrease with age (Matsuzaki *et al*. 2001). In both the breeds, the concentration of FSH was found to be higher at 18 months of age. Significantly ($P<0.05$) higher concentration of FSH (ng/ml) was observed in crossbred males compared to indigenous males at 18 and 24 months of age. The role of FSH is well established in the process of spermatogenesis. The concentrations of FSH were reported to increase before the onset of spermatogenesis (Evans *et al.* 1996, Moura

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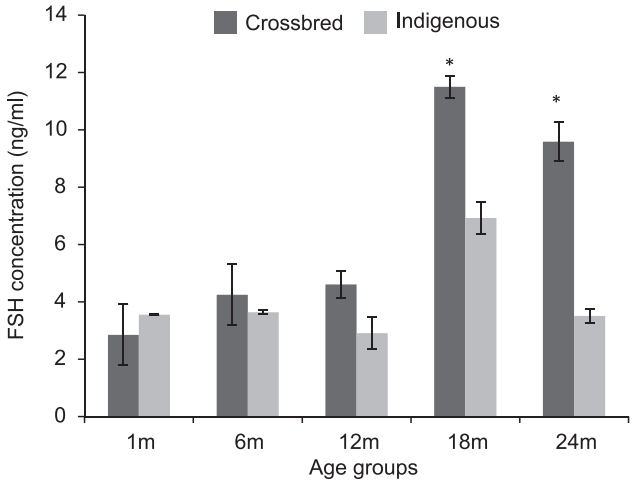

Fig. 1. Age related changes in FSH concentration in crossbred and indigenous males. *indicate significant difference between breed (P<0.05).

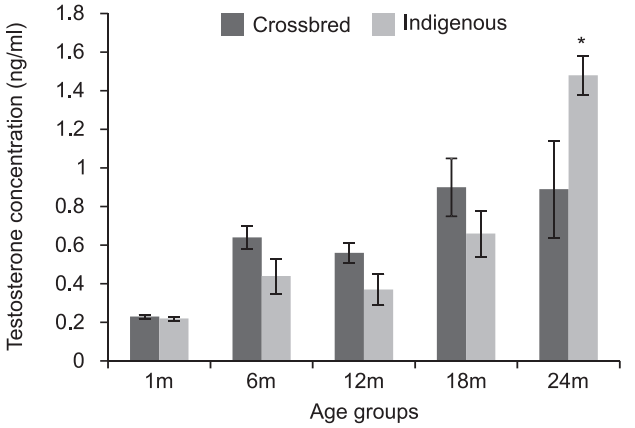

Fig. 2. Age related changes in LH concentration in crossbred and indigenous males. *indicate significant difference between breed (P<0.05).

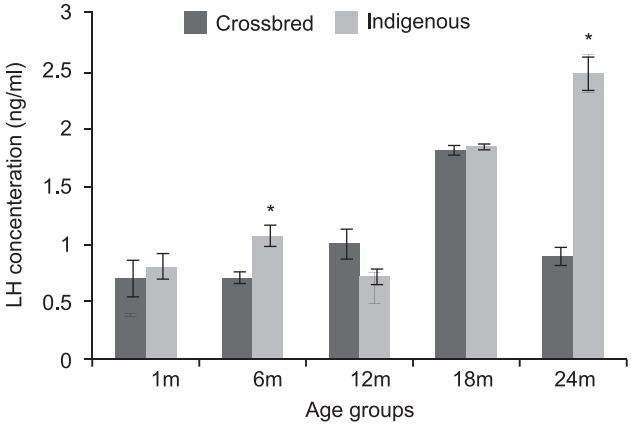

Fig. 3. Age related changes in testosterone concentration in crossbred and indigenous males**.** *indicate significant difference between breed (P<0.05).

and Erickson 1997, Moura *et al.* 2011) and the findings of the present study also confirm these observation i.e., highest FSH concentrations were observed at 18 months of age in both crossbred and indigenous males. FSH concentration observed in the study are in accordance with Finnerty *et al.* (1998), who reported that FSH concentrations were 1.03 ng/ml at 7 and 8.8 ng/ml at 11 months of age, respectively, in HF bulls. A wide variation was reported in the age related changes in FSH concentration in bulls. Evans *et al.* (1993) reported a peak FSH concentration in the bull at 20 weeks of age but D'Occhio *et al.* (1990) did not observe any change in mean serum FSH concentrations in bulls from 1 to 10 months of age. Mac-Donald *et al.* (1991) reported that the concentration of FSH increased from 0.32 to 0.43 ng/ml between 4 and 10 weeks of age, but declined from 0.43 to 0.33 ng/ml between 10 and 12 weeks of age. The reason reported for increased FSH concentration after 12 weeks was decreased concentration of inhibin at that time.

The mean concentration of LH in crossbred and indigenous males is given in Fig. 2. The concentration of LH was found to be highest at the age of 18 and 24 months in crossbred males and indigenous males, respectively. The LH concentration, irrespective of age, ranged from 0.71 to 1.82 ng/ml in crossbred males and from 0.72 to 2.49 ng/ml in indigenous males. A significant (P<0.05) difference was observed between crossbred and indigenous males in the concentration (ng/ml) of LH at 6 and 24 months of age. The basal concentrations of LH observed in the present study are in consensus with Moura and Ericsion (1997), who reported that LH concentration ranged from 0.47± 0.03 to 0.66 ± 0.06 ng/ml in bull calves between 2 to 4 months of age, which increased to 1.09 ± 0.05 ng/ml at 9 months of age. Finnerty *et al.* (1998) reported that the mean plasma LH concentrations in HF bull calves at 4–5 months, 7 and 11 months of age were 1.8, 1 and 1.8 ng/ml, respectively. Bagu *et al.* (2004) observed the mean LH concentration at the age of 4 and 8 weeks of age in bull calves was 0.50± 0.1 and 0.70± 0.10 ng/ml, respectively. Wide variations in LH concentration was reported by Dixit *et al.* (1998) in buffalo bulls (0.92 to 9.91 ng/ml).

The mean testosterone concentrations (ng/ml) in crossbred and indigenous male are given in Fig. 3. The concentrations of testosterone ranged from 0.23 to 0.90 ng/ml in crossbred males, with no significant difference among different age groups. In indigenous males, testosterone concentrations ranged from 0.22 to 1.48 ng/ml and significantly (P<0.05) higher testosterone concentrations were observed at 24 months (1.48±0.32) of age. The concentrations of testosterone did not differ significantly between crossbred and indigenous males in any age group studied except at 24 months of age. The concentrations of testosterone observed in the present study are in consensus with the value observed by Finnerty *et al.* (1998) in HF bull calves at 4–5 month (0.8 ng/ml), 7 months (1 ng/ml) and 11 months (0.8 ng/ml) of age. The mean concentration of testosterone in Holstein bull calves, crossbred young bulls and crossbred adult bulls were reported to be 0.49±0.66 ng/ml, 1.96 to 4.08 ng/ml, 0.44 to 5.60 ng/ml and 0.65±0.11 to 0.69±0.12 ng/ml, respectively (Santra *et al.* 1999, Kumar *et al.* 2006, Gholami *et al.* 2010). In our previous studies,

we observed that the concentration of testosterone at 6 and 15 months of age in crossbred bulls was 2.28 ±0.09 and 1.42 ± 0.22 ng/ml, respectively (Rajak *et al.* 2014). Wide range of testosterone concentrations (0.89 to 14.59 ng/ml) was reported in Cholestiani bulls (Mahmood *et al.* 2011).

SUMMARY

Collectively, the findings of the present study revealed that the concentrations of both FSH and LH were highest at 18 months of age in crossbred males, while in indigenous males, the FSH and LH concentrations were highest at 18 and 24 month of age, respectively. Higher concentrations of testosterone were observed at 18 months of age in crossbred males and at 24 months of age in indigenous males. These findings may explain the endocrinological support for comparatively late sexual maturity in indigenous males.

ACKNOWLEDGEMENT

The authors are thankful to the Director and Vice Chancellor, National Dairy Research Institute, Karnal for providing all research facilities for the successful completion of this study. The work was funded by the National Agricultural Science Fund (NFBSFARA/BS-3009) of ICAR. The first author is a recipient of Rajiv Gandhi National Fellowship (UGC-RGNF) from University Grant Commission, New Delhi.

REFERENCES

Bagu E T, Madgwick S and Dugawathi R. 2004. Effects of treatment with LH or FSH from 4 to 8 weeks of age on the attainment of puberty in bull calves. *Theriogenology* **62**(5): 861–73.

Bardin C W, Cheng C Y, Mustow N A and Gunsalus G L. 1994. The Sertoli cell. *Physiology of Reproduction.* 2 edn, pp. 1291–333. (Eds) Knobil E and Neil J D. New York, Raven Press.

Dixit V D, Singh B, Singh P, Georgie G C and Dixit V P. 1998. Circadian and pulsatile variations in plasma levels of inhibin, FSH, LH and testosterone in adult Murrah buffalo bulls. *Theriogenology* **50**(2): 283–92.

D'Occhio M J, Neish A and Broadhurst L. 1990. Differences in gonadotropin secretion post-partum between Zebu and European breed cattle. *Animal Reproduction Science* **22**(4): 311–17.

Evans A C O, Pierson R A, Garcia A, McDougall L M, Hrudka F and Rawlings N C. 1996. Changes in circulating hormone concentrations, testes histology and testes ultrasonography during sexual maturation in beef bulls. *Theriogenology* **46**(2): 345–57.

Evans A C O, Currie W D and Rawlings N C. 1993. Opioidergic regulation of gonadotrophin secretion in the early prepubertal bull calf. *Journal of Reproduction and Fertility* **99**: 45–51.

Finnerty M, Enrighrt W J and Roche J F. 1998. Testosterone, LH and FSH episodic secretory patterns in GnRH-immunized bulls. *Journal of Reproduction and Fertility* **114**: 85–94.

Gholami H, Towhidi A, Shahneh Z and Dirandeh E. 2010. The relationship of plasma leptin concentration and puberty in Holstein bull calves (*Bos taurus*). *Journal of Animal Physiology and Animal Nutrition* **94**: 797–802.

Kumar N, Verma R P, Singh L P, Varshney V P and Dass R S. 2006. Effect of different levels and sources of zinc supplementation on quantitative and qualitative semen attributes and serum testosterone level in crossbred cattle (*Bos indicus* × *Bos taurus*) bulls. *Reproduction Nutrition and Development* **46**: 663–75.

MacDonald R D, Deaver D R and Schanbacher B D. 1991. Prepubertal changes in plasma FSH and inhibin in Holstein bull calves: responses to castration and (or) estradiol. *Journal of Animal Science* **69**: 276–82.

Mahmood S A, Ijaz A, Ahmad N, Rehman H U, Zaneb H U and Farooq U. 2011. Studies on the libido and serum testosterone concentration of Cholistani AI bulls under stress free and stressful seasons. *Journal of Animal Plant Science* **23**(6):1491–95.

Matsuzaki S, Uenoyama Y, Okuda K, Watanabe G, Kitamura N, Taya K, Cruzana M B and Yamada J. 2001. Prepubertal changes in immunoreactive inhibin concentration in blood serum and testicular tissue in Holstein bull calves. *Journal of Veterinary and Medical Science* **63**: 1303–07.

Moura A A and Erickson B H. 1997. Age related changes in peripheral hormone concentration and their relationship with testis size and number of Sertoli and germ cell in yearling beef bulls. *Journal of Reproduction and Fertility* **111**: 183–90.

Moura A A, Souza C E A and Erickson B H. 2011. Early prepubertal testis criteria, seminiferous epithelium and hormone concentrations as related to testicular development in beef bulls. *Animal Reproduction Science.* Article in press. doi:10.1016/j.animalreprosci. 2011.01.016.

O'Donnell L, Meachem S J, Stanton P G and McLachlan R I. 2006. Endocrine regulation of spermatogenesis. *Physiology of Reproduction.* 3 edn, pp. 1017–69. (Eds) Neill J D, Knobil and Neill. Academic Press, St Louis.

Rajak S K, Kumaresan A, Gaurav M K, Layek S S, Mohanty T K, Aslam M K M, Tripathi U K, Prasad S and De S. 2014. Testicular cell indices and peripheral blood testosterone concentrations in relation to age and semen quality in crossbred (Holstein Friesian × Indigenous) bulls. *Asian Australasian Journal of Animal Science* **27**(11): 1554–61.

Santra A, Agrwal N, Kamara D N and Pathak N N. 1999. Effect of concentrate supplement on blood biochemical and testosterone levels in crossbred (*Bos indicus* × *Bos taurus*) calves. *Asian Australasian Journal Animal Science* **12**(6): 881–88.