



# Ectopic Kidney and Lymph Nodes and Intra-abdominal Testicular Structure in a Freemartin Holstein Friesian Calf

Derar Refaat<sup>1,2,\*</sup> Ahmed Ali<sup>2</sup> and Mohamed Tharwat<sup>1,3</sup>

<sup>1</sup>Department of Veterinary Medicine, College of Agriculture and Veterinary Medicine, Qassim, Saudi Arabia

<sup>2</sup>Department of Theriogenology, Faculty of Veterinary Medicine, Assiut University, Egypt

<sup>3</sup>Department of Animal Medicine, Faculty of Veterinary Medicine, Zagazig University, Egypt

\*Corresponding author. [derar40@yahoo.com](mailto:derar40@yahoo.com)

## Abstract

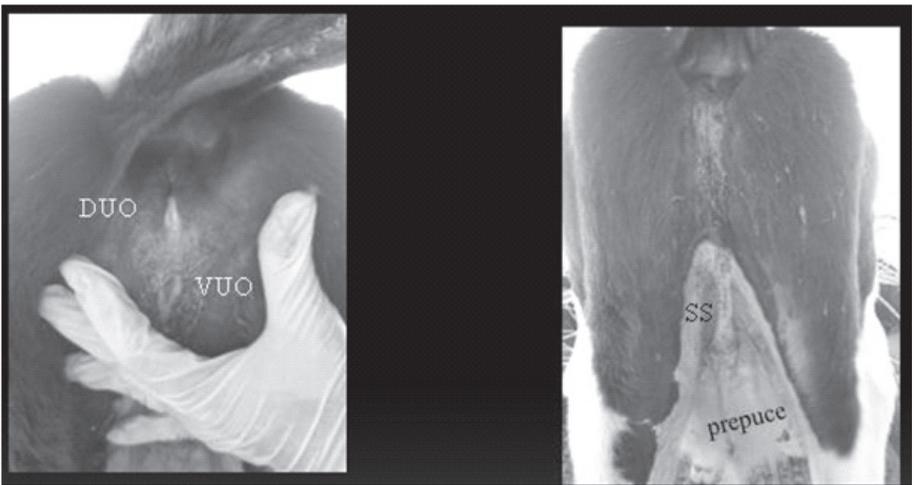
Freemartinism in a 3.5 months-old Holstein Friesian calf was investigated. External examination of the animal revealed the presence of two vulvar openings ventral to the anus. The urine is voided from the lower aperture. Transrectal palpation and ultrasonography revealed an ectopic left kidney in the caudal abdomen. At autopsy, an ectopic left kidney, right unilateral cryptorchidism, left testicular aplasia and intra-abdominal edema were the main findings observed. Histopathologically, narrowing of the seminiferous tubules and degenerative changes of the sertoli cells were recorded. This is the first report of testicular cryptorchidism and aplasia and ectopic kidney in free martin calf.

**Keywords:** intersexuality; cattle freemartin; ectopic kidney; ectopic lymph node; cryptorchidism

Freemartins arise when vascular connections occur between the placentae of developing heterosexual twin foeti; XX/XY chimerism develops and ultimately there is masculinisation of the female tubular reproductive tract to varying degrees (Gonella-Diaza *et al.*, 2012). The freemartin condition represents the most frequent form of intersexuality found in cattle, and occasionally other species (Fricke, 2001; Padula, 2005). The earliest developmental abnormalities of the

female reproductive tract resulting in freemartinism occur between 49 to 52 d post-fertilization. The condition has been studied in cattle (Ghanem *et al.*, 2005) goats (Szatkowska *et al.*, 2004), Sheep (Gonella-Diaza *et al.*, 2012) and mare (Vandeplasseche *et al.*, 1970; Juras *et al.*, 2010).

There are a number of clinical and biological methods that have been introduced or developed for the diagnosis of freemartinism. A typical clinical examination for the diagnosis of freemartinism is the determination of the vaginal length as well as the examination of the presence or absence of the uterus, cervix and long and coarse vulval hair (Sohn *et al.*, 2007). This method is simple and easy, but in doubtful cases, additional laboratory examinations are necessary to confirm the judgment based on the clinical/physical indications. The present report investigated the findings associated with freemartinism in a Holstein Friesian calf using different diagnostic tools.

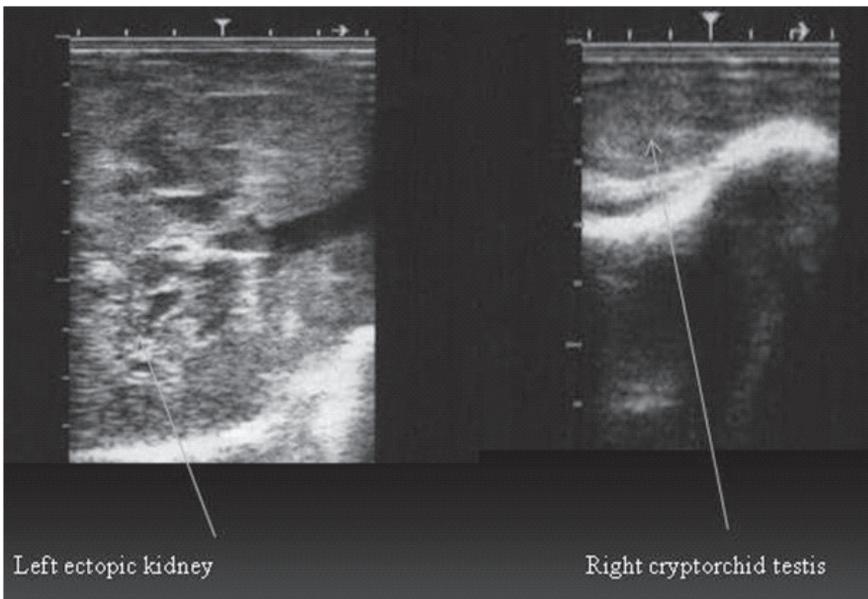


**Figure 1. Morphological findings in a 3.5 months-aged free martin calf with intersexual external organs: DUO, dorsal urogenital opening; VUO, ventral urogenital opening; SS, scrotal sac and prepuce.**

A 3.5-month-old Holstein Friesian calf born co-twin to a normal male calf was presented for diagnosis and treatment of general anorexia, abdominal discomfort and abnormal voidage of urine via an opening below the anal opening. External examination of the animal revealed that the animal is dull, feverish and anorexic with abdominal distension since 15 days. Two vulvar openings were present ventral to the anus (Fig. 1). The upper one is a blind 1.5 cm length and the second

is below the first one by 7 cm and 2.5 cm length. The urine is voided from the lower aperture which enfolded the external urethral orifice. The distance between the anal opening and the upper and lower urogenital apertures was 3 and 11.5 cm, respectively. Few scalds and crusts were seen around the lower aperture due to the voidage of urine from this opening. Cranial to the lower aperture and run alongside the caudal abdomen, a narrow penile tissue which lacked sigmoid flexure and ended with a preputial orifice could be palpated. A skin fold could be seen without any structures inside (Fig. 1).

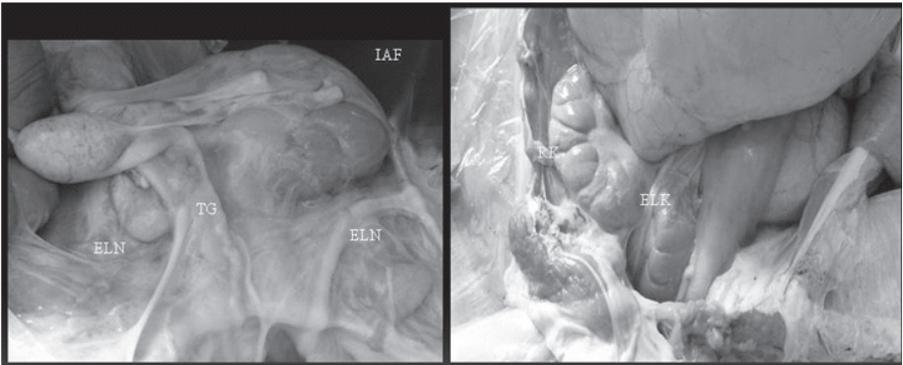
Transrectal palpation and ultrasonography revealed an ectopic left kidney in the caudal abdomen, fully-filled intact urinary bladder and right testicle-like structure and accumulation of intra-pelvic and intra-abdominal fluid (Fig 2).



**Figure 2. ultrasonographic findings in a 3.5 months-aged free martin calf with ectopic left kidney and right cryptorchid testis**

The calf was euthanized and the abdominal topography was evaluated. The abdominal organs were topographically, morphologically and morphometrically normal. An ample amount of intrapelvic and intrabdominal jelly-like fluid was found (Fig. 3). The presence of right testicle-like structure just caudal to the right kidney was recorded (Fig. 3). In addition, right and left ovoid structures caudal to

the cryptorchid testis were monitored. A cord-like body was present on the surface of both structures. This body was continued with a vascular and sinuous structure that thought to be the spermatic cord. The testicle-like structure was joined to the penis (penile urethra) across a series of ducts. Cord-like structures adjacent to the male tubular tract was also identified replacing the female tubular genitalia extended from the right and left sides at the level of the cryptorchid right testis till the blind vestibule.

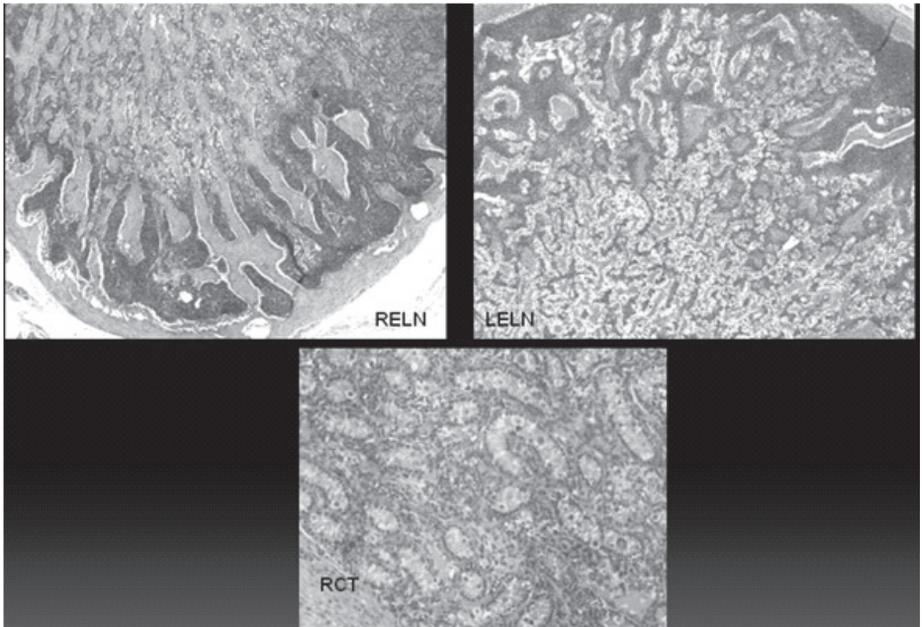


**Figure 3. Necropsy findings in a 3.5 months-aged free martin calf with mixed internal sex organs: IAF (intra-abdominal fluid; CT (cryptorchid testis); ELN (ectopic lymph node); TG (tubular genitalia) EK (ectopic kidney).**

Samples from the suspected male and female gonads were fixed in 10% formalin, and histological sections were stained with hematoxylin and eosin. The right and left ovoid structures were ectopic proliferative lymph nodes. The testicle-like structure found inside the abdominal cavity showed Degeneration of the hypoplastic seminiferous tubules and sertoli cell dysgenesis (Fig 4). The parenchyma consisted of atrophic seminiferous tubules coated with a poor germinal epithelium.

The present report described a case of intersexuality in a free martin Holstein Friesian calf with a first time record of ectopic kidney, lymph nodes. Similar interspecies differences have been observed regarding the abnormal development of the genital apparatus of unlike-sexed twins of mammals, being practically always a masculinisation of the genetical female towards a chiefly male with rudimentary female genital apparatus called in cattle (Ghanem *et al.*, 2005), sheep (Gonella-Diaza *et al.*, 2012), goats (Szatkowska *et al.*, 2004) and horse (Vandeplassche *et al.*, 1970; Juras *et al.*, 2010). An ectopic lymph node (found in the scrotal sac), two intra-abdominal testes and vestiges of the female reproductive system were found

in a 1.5 year-aged ewe (Gonella-Diaza *et al.*, 2012). However, the freemartins that are described in all previous reports were not associated with congenital abnormalities in the renal or lymph systems. The phenotypic morphological findings for freemartins can vary, depending on the degree of masculinisation (Gonella-Diaza *et al.*, 2012).



**Figure 4. Histopathological findings in a 3.5 months-aged free martin calf showing (A) RELN (Right ectopic lymph node and (B) LELN (left ectopic lymph node) proliferative lymph nodes X10 obj. and (C) RCT (right cryptorchid testis) X40 obj.**

Degeneration of the hypoplastic seminiferous epithelium occurs because of dysgenetic changes. Focal orchitis was associated with tubular ectasia and sertoli cell dysgenesis, disruption of the blood-testis barrier and gave rise to an autoimmune process (Nistal *et al.*, 2002).

To the best of the authors' knowledge, this is the first time a case has been reported where kidney and lymph nodes have been found ectopic.

## References

Fricke PM. 2001. Review: twinning in dairy cattle. The professional animal scientist, **17**: 61–67.

- Ghanem ME, Yoshida C, Nishibori M, Nakao T, Yamashiro H. 2005. A case of freemartin with atresia recti and ani in Japanese Black calf. *Anim. Reprod. Sci.*, **85**(3-4): 193–199.
- Gonella-Diaza AM, Duarte LZ, Dominguez S, Salazar PA. 2012 Abnormal position of lymph nodes in a freemartin sheep. *Veterinary Medicine: Research and Reports*, **3**: 1-6.
- Juras R, Raudsepp T, Das PJ, Conant E, Cothran EG. 2010. XX/XY Blood Lymphocyte Chimerism in Heterosexual Dizygotic Twins from an American Bashkir Curly Horse. Case Report. *J equine V Sci.* **30**(10): 575–580.
- Nistal M, Riestra ML, Paniagua R. 2002. Discussion of pathologic mechanism of tubular atrophy. *Arch. Pathol. Lab. Med.*, 126: 64-9.
- Padula AM. 2005. The freemartin syndrome: an update. *Anim. Reprod. Sci.*, **87**(1–2): 93–109
- Sohn SH, Cho EJ, Son WJ, Lee CY. 2007. Diagnosis of bovine freemartinism by fluorescence in situ hybridization on interphase nuclei using a bovine Y chromosome-specific DNA probe. *Theriogenology*, **68**: 1003–1011.
- Szatkwska I, Zych S, Uda J, Dybus A, Aszczyk PB, Sysa B, Dnbrowsk T. 2004. Freemartinism: Three Cases in Goats. *Acta Vet. Brno.*, **73**: 375–378.
- Vandeplassche M, Podliachouk L, Beaud R. 1970. Some Aspects of Twin-gestation in the Mare. *Can. J comp. med.*, **34**: 218-226.