

CYTOGENETIC AND CLINICAL STUDY OF A GOAT FREEMARTIN CASE

STUDIU CITOGENETIC ȘI CLINIC AL UNUI CAZ DE FREEMARTINISM LA CAPRINE

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ABSTRACT | REZUMAT

The research was oriented towards clinical and cytogenetics analysis of a intersexuality case in goat, turned out in the end to be a freemartin.

The presence of sex chromatin in 38% of the oral mucosa cells, in 4% of neutrophils, of cromosomal mosaicism in bone marrow cells, 65% - specific to female sex (54, XX) and 35% - specific to male sex (54, XY) represents a proof of freemartin status. The lack of a male congener at birth, leads us to the conclusion of a state generated by oocyte double fertilization.

In circumstances in which the incidence of clinically, cytogenetic or molecular diagnosed freemartin cases is reduced in goats, the present study may represent a contribution to a more accurate characterization of intersexuality in domestic goat.

Keyword: sterility, freemartin, goat

Cercetarea s-a orientat spre o analiză clinică și citogenetică a unui caz de intersexualitate la capră, dovedit la final ca fiind un freemartinism.

Prezența cromatinei sexuale în 38% din celulele raclatului de mucoasă bucală, în 4% din neutrofile, a mozaicismului cromozomal în celulele măduvei osoase, 65% - specific sexului femel (54,XX) și 35%-specific sexului mascul (54,XY) reprezintă dovada stării de freemartinism. Lipsa unui congenen de sex mascul la naștere conduce spre concluzia unei stări generate de fecundația dublă a ovocitei.

În conjunctura în care incidența cazurilor diagnosticate clinic, citogenetic sau molecular, de freemartinism la caprine este redusă, studiul de față poate reprezenta un aport pentru o mai precisă caracterizare a stărilor de intersexualitate la capra domestică.

Cuvinte cheie: sterilitate, freemartinism, capră

According to data from literature, freemartin is a complex state of intersexuality, sometimes encountered in natural monotocic species, in case of double heterosexual pregnancy (1).

The cause of the occurrence of this intersexuality has been a challenge for researchers from related fields: biology, cytogenetic, reproduction etc.

The results of their investigations generated several hypotheses. Among these, the influence of the androgens made possible by anastomosis of chorionic vessels of heterosexual fetuses – Lillie's hypothesis (1916), was widely accepted, although it cannot explain all freemartins cases.

In the past, two groups of researchers, Gartler (1962) and Josso (1965), claimed a hypothesis in which the double fertilization of the oocyte could cause an intersexual product. Involvement polispermia and atypical divisions that could generate the appearance of intersexuality, also freemartin, it was rarely been incriminated, although this hypothesis, as of Lillie, relies

more on cytological grounds and not on embryogenetic experiments. In goats, the incidence of freemartin is generally low, less than 1% (6), which led us to investigate clinical and cytogenetic the possible causes for the appearance of the investigated case. The results of the investigations have led to the conclusion that double oocyte's fertilization represents a cause of freemartin occurrence, at least in this species.

MATERIALS AND METHODS

The investigations have targeted a 2 years and 6 months years old crossbreed, from a single birth, of a 7 years old mother, whose products from previous years have not shown reproductive abnormality incompatible with reproduction. Animal presented external female features, which is why it was kept on the farm to be reproduced. The absence of reproductive behavior, namely the absence of heats, at the age of 1 year, had not alerted the farmer, but their absence in the following breeding season, determined the owner to use the services of specialists from the Faculty of Veterinary Medicine of Timișoara.

Initially, the animal has been subjected to a more

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rigorous clinical examination of the genital region.

Cytogenetic examinations as smear of the oral mucosa and from the peripheral blood were done.

These examinations focused on the presence and frequency of sex chromatin (Barr corpuscles, Smith formations), in 100 somatic cells, respectively polymorphonuclear cells. After slaughtering, the components of the genital tract have been examined clinically and cytogenetic smears were made from bone marrow using Whang and Tjio technique for examination of chromosomes in metaphase. For this purpose 20 metaphases were analyzed.

RESULTS AND DISCUSSION

Clinical results.

Analyzing the sex components from phenotypic and somatic point of view there were found a clitoris highly developed (Fig. 1), prominent in a rudimentary vulva continued by a vestibule and vagina.



Fig. 1. Clinical aspect of goat vulva: hypertrophic clitoris

Clinical examination after slaughtering highlighted the absence of cervix, replaced by a cylindrical structure, 4 cm long, flanked by two glandular formations probably vestiges of the seminal vesicles.

The structure is continued with a uterine body, 3 cm long and with rudimentary uterine horns (Fig. 2).

Clinically, oviducts, mesosalpinx and infundibulum have not been found and in the top of each uterine horn was positioned a testicle, attached to the uterine horn by a developed pampiniform plexus.

Macroscopic examination of testicular parenchyma revealed an amorphous structure, without clear evidence of Haller network, different from a normal testicle.

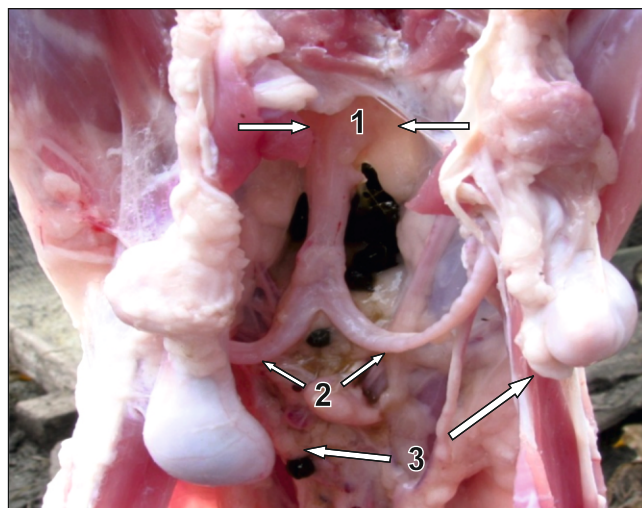


Fig. 2. Rudimentary appearance of the specific female genital tract, along with testicle: 1- vestiges of the seminal vesicles; 2 - uterine horns; 3 - testes

In goats compared to other ruminants, there are fewer reports of freemartin cases.

Changes of the external genital segments are similar to those from cattle and sheep, and consist in an overly developed clitoris and vestibular hypoplasia (9). Their existence is the result of influence of intense activity of existing gonad. The same influences are also upon internal genital tract (5, 8) and there are many similarities between the species.

According to many opinions (1, 2, 8), deficiency in secreting antimüllerian hormone and androgen protein leads to rudiments of the specific female genital tract, as a result of a spontaneous embryogenesis and to non-forming of Wolff embryonic structures, fact noted by the lack of specific male genital tract in freemartins.

For cytogenetic analysis, the biological samples have been processed and examined in the Laboratory of Cytogenetics and molecular genetics from Complex "Horia Cernescu" of Research Laboratories, within the Faculty of Veterinary Medicine of Timișoara.

Sex chromatin has been identified in 38% of the oral mucosa cells, which indicates the existence of genetical female sex (XX). This value is similar with those reported by Szatkowska et al. (7), values obtained from the analysis of three cases of freemartin in goats.

Examination of the peripheral blood smears, for the presence of Smith and Davidson units in leukocytes, confirmed the existence of genetically female sex, although the proportion of cells bearing the genetic marker female was only 4% (the reference value in the literature being 10%).

Cytogenetic analyses of metaphases from bone

marrow cells have revealed chromosomal mosaicism. In 13 metaphases (65%) were identified 6 metacentric and 48 acrocentric chromosomes (54, XX), and the rest, 7 cells (35%) had 6 metacentric and 47 acrocentric chromosomes and a small metacentric chromosome (54, XY). The presence of anatomical structures belonging to female specific genital tract, with male gonads correlated with chromosomal mosaicism of both sexes and the absence of a male congener, leads us to believe that the product analyzed is a freemartin result of a double fertilization, followed by further unpredictable divisions.

Freemartin as a result of polyspermia, has been described also by Cîrlan (2), after multiple anatomical, histological and cytogenetic investigations of a cattle freemartin case. According to the author polyspermia was possible perhaps due to oocyte immaturity.

Since the goats have a lower incidence of diagnosed cases of freemartin compared to other ruminants, it is important to conduct extensive studies on intersexuality cases in domestic goats.

CONCLUSIONS

1. Internal and external examination of genital tract revealed existence of an intersexual state, incompatible with reproduction.

2. The presence of sex chromatin (in 38%) and of drumstick (in 4%) proves the female genetic sex of the animal.

3. Cytogenetic analyses of metaphases from bone marrow cells have revealed chromosomal mosaicism: in 65% out of metaphase, there were chromosomes

specific to female karyotype (54, XX) and in 35% - chromosomes specific to male karyotype (54, XY).

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