

EMBRYO TRANSFER FROM SEROPOSITIVE GOATS FOR CAPRINE ARTHRITIS-ENCEPHALITIS VIRUS (CAEV) WITH BIRTH OF SERONEGATIVE KID

(Transferência de embriões de cabras soropositivas para artrite-encefalite caprina a vírus (CAEV) com nascimento de cria soronegativa)

Vicente José de Figueirêdo FREITAS^{1*}, Tânia Vasconcelos CAVALCANTE¹, Hévila Oliveira SALLES² & Maria Fátima da Silva TEIXEIRA¹

¹ Universidade Estadual do Ceará – Faculdade de Veterinária, ² Centro Nacional de Pesquisa em Caprinos (EMBRAPA)

ABSTRACT

The aim of this study was to determine whether recipient goats would seroconvert following transfer of embryos collected from donor goats seropositive for CAEV and if kids produced would be clinically normal and seronegative for CAEV. Four does (Saanen and Alpine), seropositive for CAEV, were used as donors, receiving superovulation treatment consisting of progestagen, cloprostenol and FSH. During estrus the donors were mated with seropositive bucks. Seven days after estrus, embryo recovery was performed by surgery. The embryos were then frozen according to the protocol of the International Embryo Transfer Society (IETS). Twelve seronegative does were used as recipients and received an estrus synchronization treatment consisting of progestagen, cloprostenol and eCG. Embryo transfer was performed seven days after synchronized estrus using the semilaparoscopy technique. Pregnancy was verified in one of twelve recipients, that remained seronegative until six months after giving birth and the kid until six months of age. The preliminary success in producing CAEV negative kids and failure of seroconversion in previously seronegative recipients in this study suggests that embryo transfer technology may offer an alternative in the prevention of CAEV transmission in goat herds as well for the utilization of genetic patrimony of seropositive goats.

KEY WORDS: goat, CAEV, embryo transfer.

RESUMO

O objetivo deste estudo foi verificar se cabras receptoras sofreriam soroconversão após a transferência de embriões colhidos de cabras soropositivas e se as crias nascidas seriam clinicamente normais e soronegativas para CAEV. Foram utilizadas quatro cabras (Saanen e Alpina), todas soropositivas para CAEV e que receberam um tratamento superovulatório que consistiu no uso de progestágeno, cloprostenol e FSH. Durante o estro, as doadoras foram cobertas por bodes soropositivos. Sete dias após o estro, a colheita de embriões foi realizada utilizando a técnica cirúrgica. Os embriões colhidos foram congelados de acordo com o protocolo da Sociedade Internacional de Transferência de Embriões (IETS). Doze cabras soronegativas foram utilizadas como receptoras e receberam um tratamento de sincronização do estro consistindo de progestágeno, cloprostenol e eCG. A transferência de embriões foi realizada sete dias após o estro sincronizado usando a técnica de semi-laparoscopia. A gestação foi confirmada em uma das doze receptoras, a qual permaneceu soronegativa até seis

*Author for correspondence:
Av. Paranjana, 1700, 60740-000 Fortaleza-CE,
e-mail: vjff@uece.br

meses após o parto e sua cria até os seis meses de idade. O sucesso preliminar na produção de crias soronegativas para CAEV e a não soroconversão em receptoras soronegativas sugere que a tecnologia de transferência de embriões pode oferecer uma alternativa para a profilaxia da CAEV em rebanhos caprinos, bem como para o aproveitamento do patrimônio genético de animais soropositivos.

PALAVRAS-CHAVE: caprino, CAEV, transferência de embriões.

INTRODUCTION

The economic loss resultant from the slaughter of dairy goats seropositive for CAEV (Caprine Arthritis-Encephalitis Virus) is responsible for a great prejudice in the milk goat industry in Brazil.

The CAEV virus is a member of the Retroviridae family and Lentivirinae subfamily of slow viruses. This group of cell-associated viruses is characterized by the presence of an RNA-dependent DNA polymerase, and the CAEV is functional only when the viral RNA is transformed to viral DNA and is integrated into the host cell DNA (FROENKEL-COURAT & KIMBUL, 1982). The virus is morphologically and biochemically closely related to the agents causing ovine progressive pneumonia (OPPV) and maedi-visna of sheep (CLEMENTS et al., 1980; DAHLBERG et al., 1981).

The CAEV has been reported to be most efficiently transmitted to kids through colostrum and milk. In one study, 2 of 32 cesarean-derived kids that did not suckle seropositive does became seropositive, indicating the possibility of intrauterine infection (ADAMS et al., 1893).

The use of embryo transfer for virus disease control has been discussed in several species. Recipient cows failed to develop serum antibodies or viremia following transfer of embryos from bluetongue viremic seropositive donors (BOWEN et al., 1983). Recipients of embryos from bovine leukemia virus seropositive donors remained seronegative as did the calves produced by this procedure (EAGLESOME et al., 1982). Embryos collected from goats seropositive for bluetongue failed to cause seroconversion when transferred to seronegative recipients (CHEMINEAU et al., 1986).

There has been limited work carried out regarding the use of embryo transfer for control of caprine viral diseases. The objectives of the present study were to determine whether recipient goats utilized in standard embryo transfer procedures would seroconvert following transfer of embryos collected from donor goats clinically affected with CAEV and whether kids produced by this procedure would be clinically normal and seronegative for CAEV.

MATERIALS AND METHODS

Animals

Four adult Saanen (n=2) and Alpine (n=2) does were utilized as embryo donors. All four donors were seropositive for CAEV and had clinical evidence of arthritis in one or more joints. These does were mated to two bucks (1 Saanen and 1 Alpine) that were also seropositive and also showed clinical evidence of arthritis. Twelve adult does of undefined breed that were clinically normal and serologically negative for CAEV were used as embryo recipients.

Superovulation and estrus synchronization treatment

Sponges impregnated with 45 mg of fluorogestone acetate (Chrono-gest, Intervet, France) were placed intravaginally in donors and recipients and left for 11 days. On day 9 of progestagen treatment all does received (intramuscularly) 50 µg of cloprostenol (Estrumate, Pitman-Moore, France). The superovulation in donors was induced using twice daily intramuscular injections of FSH (Université de Liege, Belgium) beginning on day 9 and continuing for 3 days. A total dose of 16 mg of FSH was administered using a decreasing dosage regimen.

Recovery and freezing of embryos

Following a 24 h fast donor does were submitted to a laparotomy. The number of corpora lutea were counted in each ovary. Each uterine horn was flushed separately with 40 mL of P.B.S. and the recovered embryos were observed using stereomicroscopy at 12x magnification. Embryos were frozen according to the protocol used by BARIL et al (1993) and according to patterns of the IETS (1992).

Embryo transfer

Estrus-synchronized recipients were tranquilized with Xylazina (Rompum, Bayers, Brazil) and prepared for aseptic surgery for embryo transfer. The embryo transfer was performed 7 days after synchronized estrus, using the semilaparoscopy technique (MCMILLAN & HALL, 1994). The embryos were then deposited in the lumen of the uterine horn near the uterotubal junction ipsilateral to the corpora lutea.

Serological testing

The agar gel immunodiffusion test described by GOUVEIA & MAGALHÃES (1994) was used to evaluate the serum for antibodies to CAEV. At the beginning of the study, serum was collected from each donor, recipient and bucks twice at 30 day intervals. A serum sample was collected from each recipient doe every month until 6 months after kidding. The kid was sampled before and 24 h after ingestion of pasteurized colostrum, then every

month until 6 months of age.

Statistical analysis

Data were submitted to descriptive statistics (mean, standard deviation and range) in order to provide a better presentation of results concerning ovulation rate and recovered structures.

RESULTS

All donors showed estrus 16 h after sponge removal. The mean (\pm s.d.) ovulation rate was 12.5 ± 0.6 . These results are in accord with estrus induction and superovulation obtained with seronegative donors (BARIL et al., 1993).

From a total of 50 corpora lutea counted at laparotomy, 33 structures (range 4 to 10) were collected (Tab.1). The results show that the sanitary condition of donor is irrelevant to both embryo production and embryo development during the few days after fertilization.

One of twelve recipients became pregnant resulting in one live kid. All seronegative recipients remained seronegative throughout the study. Additionally, the kid remained seronegative until 6 months of age.

DISCUSSION

This study has shown that the transfer of embryos from superovulated seropositive does mated with seropositive bucks did not produce seroconversion in previously seronegative

Table 1. Results of embryo recovery from goats seropositive for CAEV.

Donor	Breed	Corpora lutea (n)	Recovered structures (n)	Structure development			
				Mo	EB	ExB	NF
3	Alpine	13	10	1	1	4	4
26	Alpine	13	4	-	3	1	-
37	Saanen	12	9	1	1	7	-
40	Saanen	12	10	1	-	-	9

Mo: morula EB: early blastocist ExB: expanded blastocist NF: nonfertilized

recipient does. Additionally, the one live kid produced remained seronegative beyond weaning. The poor pregnancy rate could not be attributed to CAEV.

In this study, viremia in the donor does was not ascertained, although animals with antibodies are assumed to be actively infected with the virus (NARAYAN et al., 1984). Viral persistence in infected goats may be due in part to infection in the macrophage-monocyte-cell system, and further investigation is needed to ensure that these virus-containing cells do not pose a potential source of infection in embryo transfer techniques. Additionally, further studies are indicated to determine if later stage embryos that have hatched from the zona pellucida are capable of becoming infected with or of transmitting CAEV, as has been shown with bovine herpesvirus-1 and bovine diarrhea virus in cattle (ARCHBALD et al., 1979; BOWEN et al., 1985).

Current recommendations for preventing CAEV infection in kids are to remove all kids from the doe immediately at birth, being careful to prevent contact of the newborn kid with secretions from the doe and providing colostrum from known seronegative does or colostrum that has been heated to 56°C for 1 h. Additional suggestions are to isolate kids and separate them from infected goats with serological testing at 6 month intervals coupled with removal of all seropositive animals from the herd (ADAMS et al., 1983).

In conclusion, the preliminary success in producing a CAEV negative kid and failure of seroconversion in previously seronegative recipients in this study suggest that embryo transfer technology may offer an alternative for the prevention of CAEV transmission in goat herds. The results indicate that embryo transfer techniques of genetic patrimony of seropositive goats for CAEV.

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