



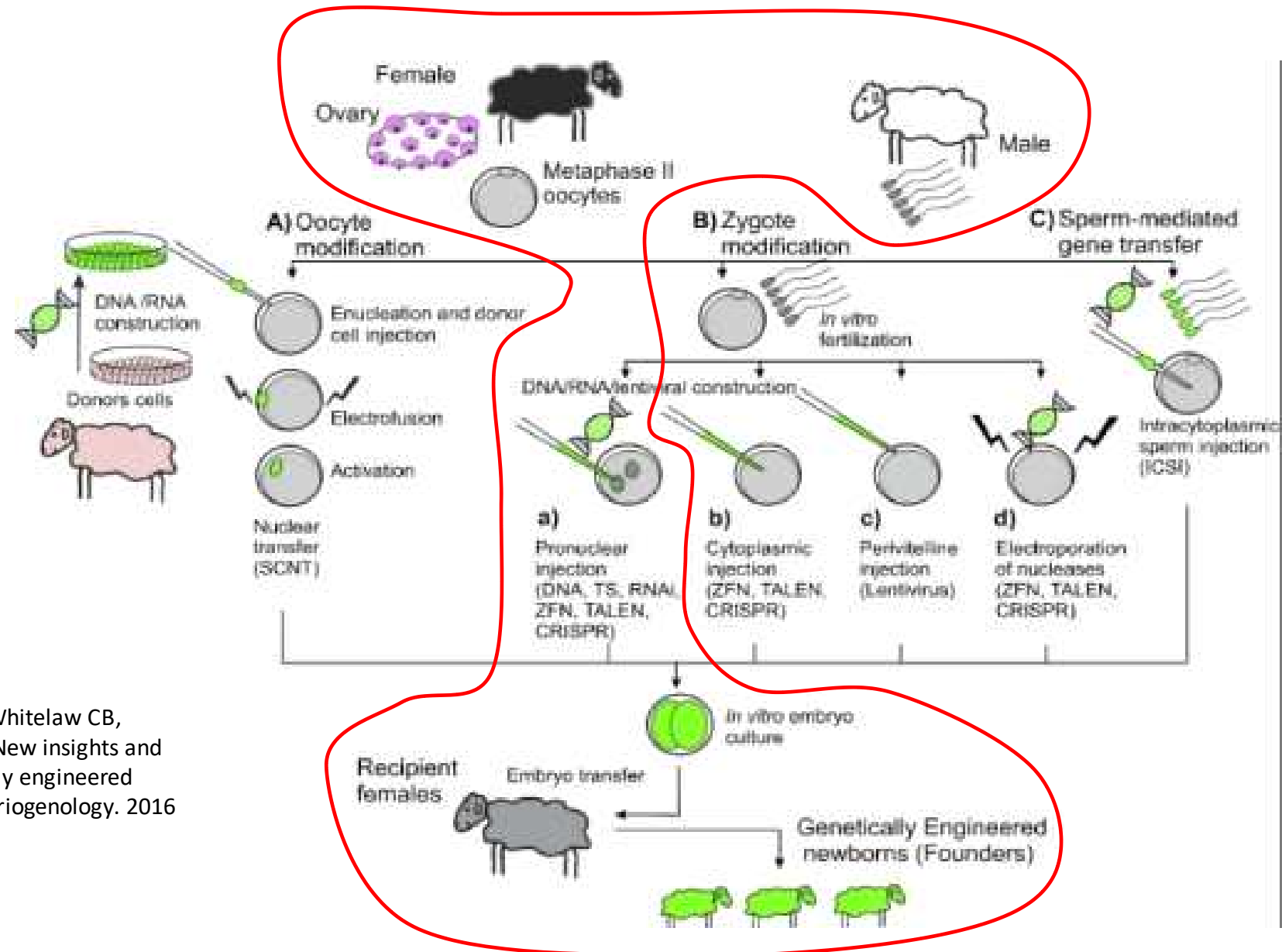
The influence of the age of young goats on the results of hormonal stimulation of folliculogenesis.

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Nowadays, the solutions to the contemporary problems of animal breeding appear to be inconceivable without the use of biotechnological methods. One of the most effective tools in modern biotechnology is the CRISPR / Cas9 method, that allows to introduce targeted modifications into the genome of an organism. This tool allows to replace the lengthy process of natural selection, thus new breeds of animals receive targeted mutations that could arise naturally.

Current tools for genetically engineered sheep and goats *



*Menchaca A, Anegón I, Whitelaw CB, Baldassarre H, Crispo M. New insights and current tools for genetically engineered (GE) sheep and goats. *Theriogenology*. 2016 Jul 1;86(1):160-9.

- Among livestock, goats are perhaps the most convenient object for genome modification. They are prolific, have a relatively short gestation period, early to mature and have a wide range of economically demanded products such as milk, meat, wool, fluff etc.



The aim of this study was to define the minimum possible age for optimal hormonal stimulation of female goats.

- Materials and methods:
- Group 1: 3,5 old month goats.
- Group 2: 4,5 old month goats.
- Group 3: 5-6 old month goats.
- Group 4: 6-7 old month goats.

Stimulation of growth follicle was carried out with the injection of pharmacy "Sergon" (follicle-stimulating hormone, FSH) intramuscularly, once. Animals of groups 3 and 4, used to obtain zygotes, were injected intravenously with human chorionic gonadotropin (hCG) after 71 hours in order to induce ovulation of mature follicles. Later, after another 10 hours they were inseminated naturally.

Table 1. Results of sOmulaOon of folliculogenesis in young goats.

Data	Group 1	Group 2	Group 3	Group 4
Number of goats	3	3	3	3
Age, month	3.5	4.5	5-6	6-7
Weight, kg.	15-18	20-25	21-27	25-33
Dose of FSH unit/goat	300	500	1500	1500
Dose of FSH unit/kilo	15-20	15-20	50-60	50-60
Demonstrated the oestrus, unit	0	3	3	3
Number of antral follicles > 3 mm, number	42	66	91	54
Number of antral follicles > 6 mm, number	8	17	82	42

Oocyte-cumulus complexes and mature goat oocytes

Animals from groups 1 and 2 were used to obtain oocytes. For this, the ovarian follicles were opened and the resulting oocyte-cumulus complexes (OOC) were put on maturation in vitro in JVM medium at 38 ° C with 5% CO₂ in the gas phase. The results were evaluated after 24 hours. Cumulus cells were removed in a hyaluronidase solution. Oocytes with a released polar body were considered mature.

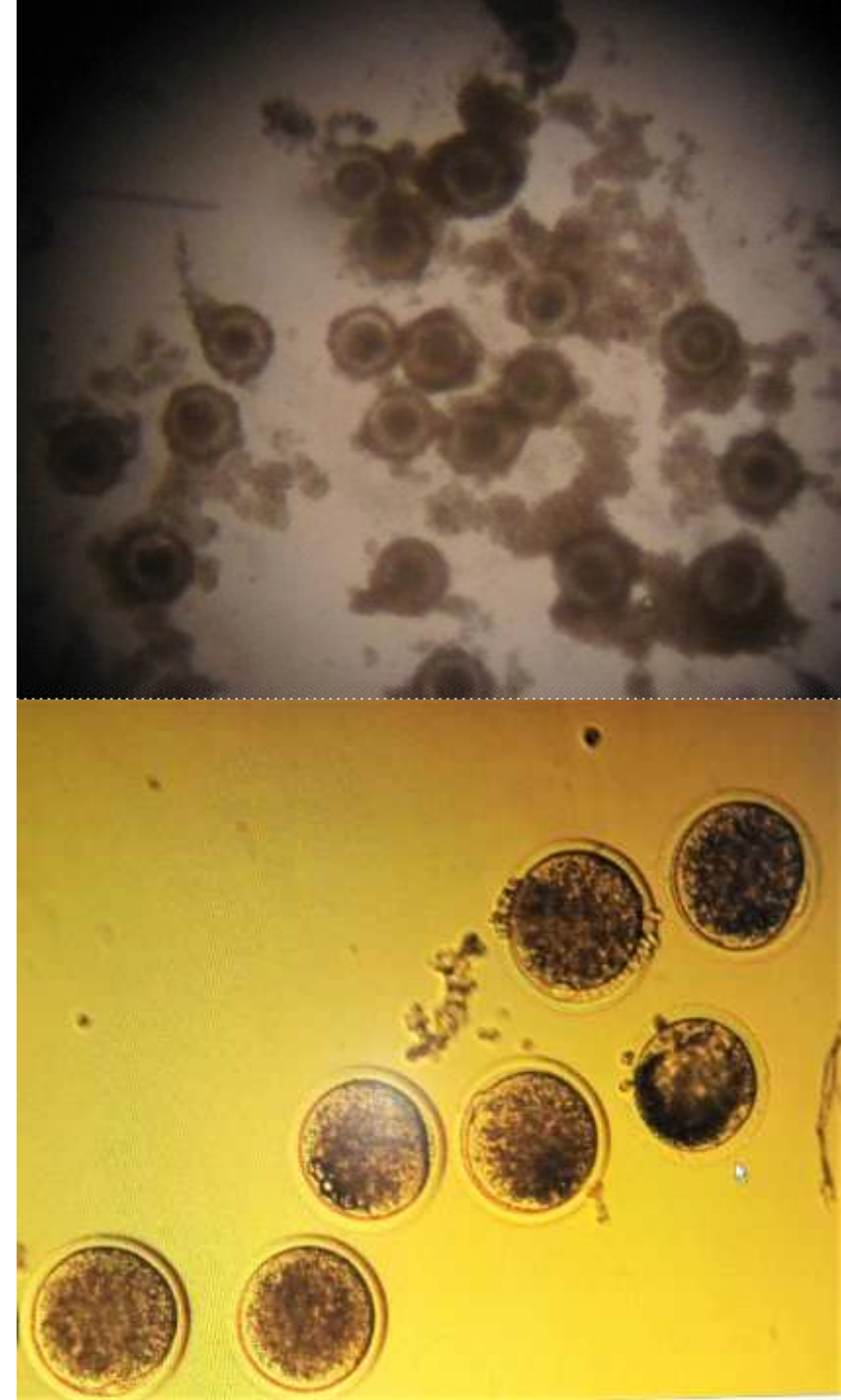


Table 2. Obtaining and in-vitro maturation of oocytes of goats that have not reached sexual maturity.

Data	Group 1	Group 2
Number of goats	3	3
OOC, number	178	92
Mature oocytes, number	15	83
Maturation, %	8	90



Goat ovaries after
superovulation (from the
left, number of ovulations
reached 29)

and without hormonal
stimulation (from the
right).

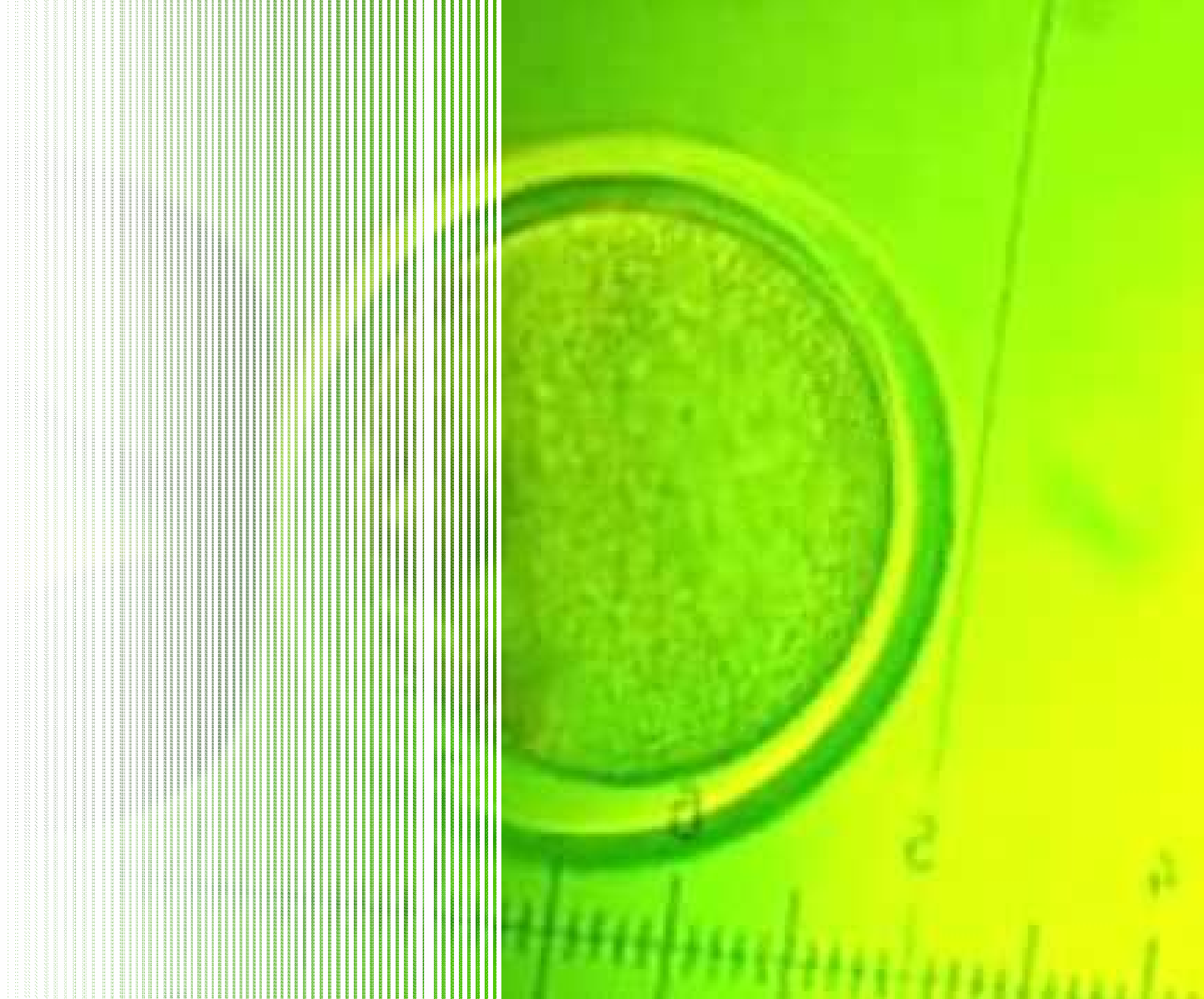


Table 3. Results of superovulation of goats that have reached puberty.

Data	Group 3	Group 4
Number of goats	3	3
Ovulations observed, number	57	19
Large non-ovulated follicles , number	25	23
Washed zygotes, number	39	14

Goat zygote at the pronucleus stage

- a) er centrifugation.



- **Conclusions:**

- Despite the high content of oocyte -cumulus complexes in the ovaries of immature goats, the efficiency of their maturation under in vitro conditions is very low, and their use as donors of germ cells is resulted to be impractical.
- Young goats at the beginning of puberty (4.5-7 months) are good donors of germ cells for microinjection procedures.
- The highest superovulation obtained as a result of hormonal treatment is goats at the age of 5-6 months, which makes it possible to use animals of this age as donors of zygotes in order to obtain genetically edited goats with new economically useful characteristics.

Thank you for your attention!

