TECHNICAL ASPECTS OF ACCEPTING THE EMBRYOS OF SHEEP-DONORS USING MEDIAN LAPAROTOMY AND LAPAROSCOPY

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Abstract: Nevertheless of the possibilities which the methods of embryotransfer (ET) offer in sheepbreeding, they have limited application in nowadays selection programs. More often this is due to the low level of superovulation response (Whyman et al., 1980), lowered vitality of the fertilized eggs (Graham, 1986), and comparatively low percentage of fertilization (Robinson et al., 1989). Disadvantages of the method of accepting embryos with median laparotomy are other essential problem, which is difficult to be completed and is connected with risks of traumatisation of internal genital organs and their adhesion. This leads to impossibility of second use as donors (Vlachov et al., 1991).

In this study our aim is to research comparatively the possibilities of the two methods for accepting embryos, especially with median laparotomy and laparoscopy.

The results proved the advantages of the method for accepting embryos with laparoscopy in three aspects: washing of embryos, risks of surgery manipulation, easy to be accessed and completed.

The laparoscopy is high effective. The risk of surgery intervention is minimal. The necessity of very expensive technique is a disadvantage of the method.

Key words: sheep, laparoscopy, embryotransfer, embryos.

Introduction and literature review

The contemporary stage of the development of sheep breeding is characterized with search for new methods of approach for forming of bred composition and breeding of sheep. The increased interest in the international market of production of high quality lamb meat requires the breeding of specialized breeds of sheep for meat. Regarding to this recently in Bulgaria there is large interest of the producers to the import of male and female animals of the following French breed - ile de France, Mouton Charolais and others. In veterinary aspect this is connected with risk of the spreading of some chronic viruses and bacterial infections, such as Pseudotuberculosis, Vibriosis, Brucellosis, Paratuberculosis and some others. The risk of spreading of the disease scrapies is also essential. The low adaptive abilities of sheep are essential precondition for their act. As it refers to the mentioned diseases they are genetically determined. This imposed spending of many resources for putting under quarantine animals and taking some extra loses of its adaptation. To surmount the mentioned difficulties, methods of embryotransfer of valuable animals are applied. Besides achieving high genetic progress with this method the danger of carrying of infection diseases can be limited, and at the same time the risks during the adaptive period of animals are decreased.

Independently of the possibilities, which the methods of ET in sheep breeding suggest us, they have limited application in the nowadays selection programs.

Mostly often this is due to the low degree of superovulation response (Whyman et al., 1980), reduced vitality eggs (Graham, 1986), and comparatively low percentage of fertility (Robinson et al., 1989).

In considerable degree, the effectiveness of ET is dependent on the method of accepting embryos. In the wide practice, the used for this aim median laparotomy is difficult for technical implementation and very often it is connected with risks of traumatisation of inner genital organs and their subsequent adhesion. This leads to impossibility of secondary use as donor (Vlachov et al., 1991).

As these mentioned problems to be solved and the traumatic effect to be limited, there are applied several methods of accepting embryos from laparoscopy.

The aim of this study is to research comparatively the possibilities of the two methods, and at the same time to define their positive and negative features.

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Materials and methods

Routine technical approaches are applied for accepting embryos with median laparotomy and laparoscopy of sheep-donors in 5\textsuperscript{th} – 6\textsuperscript{th} day after their insemination. The technical fulfillment of both methods is attended by application of 12-hours hunger and water diet, fixing of the animal upon operation table, mechanical and surgery manipulation of the operation field. The inhalation narcosis of animals is completed with Halotan.

1. Accepting embryos, with median laparotomy.

![Image 1]

The operation field is placed in the backward part of the abdomen. The operation access is to 2-3 cm in front of the Linea alba. The abdominal cavity is opened by section which length is 6-7 cm. The horns uteri and the ovaries are taken out from abdominal cavity. An overview of the ovarian surfaces is done if there are available corpus luteum or of other organoids (nonovulated follicles, ovarian cysts and others). The number and the morphology of the available corpus luteum is observed. In this way superovulation response is reported. If there is no corpus luteum on the two ovariums, then no washing of the horns uteri is made. Such situation is called lack of superovulation response. In this case the abdominal cavity is closed.

Positive superovulation response is observed in these animals who possesses well-formed as morphology corpus luteum. Washing of uterus cavity is done one by one for each uterus horn. Flexible sond is input in infundibulum at depth 3-4 cm. The washing liquid is applied in the direction from horn uterus to infundibulum. In this way, the available embryos located on the uterus horn are pushed from oviduct to infundibulum. One leading operator and two assistant-operators are doing the mentioned manipulations for approximately 30-40 minutes.

2. Accepting embryos, with laparoscopy.

![Image 2]
The operation field is placed in the backward part of the abdomen. The operation access is at 3-4 cm bilaterally of Linea alba. Three trocars are place in the abdominal cavity. The endoscope is entered through the lumen of the first trocar. Assistant instruments are entered in the other two trocars. In the beginning only the endoscope is place.

Superovulation response is reported according to the number and quality of corpus luteum. It is accepted that if there are no corpus luteum then the animal has negative superovulation response. Then more manipulations are not completed. If corpus luteums are available it is accepted that the animal has positive superovulation response and the manipulations are completed.

The second trocar entered the abdominal cavity. With assistant instrument (foceps), the uteri horn is fixed in the field of bifurcation. Then the third trocar is applied. Using it, with sharp needle the wall of the fixed horn is perforated. The needle is taken out of the third trocar and to its place three-way catheter (IMV, Cassou, France) is entered. It is placed in uteri horn, which is perforated. Some air entered through first way of the catheter, which fixes it at the wall of the uteri horn. In this way the cavity of the horn top is closed. A flexible sond is placed in this cavity through the second way of the catheter. Washing liquid is applied through the third way of the catheter. In this way, the available embryos and the medium enter in the sond.

The laparoscopy is completed with a laparoscope of Karl-Schtorz, Endoskope, Germany, and the photo documentation with camera (Ricoh-Company, Japan).

Results and discussion

Some anatomic characteristics of the inner organs of sheep don’t allow application in uterus cavity of catheters and some other devices for accepting embryos. This is the reason, for this aims the researchers to use different surgery approaches. Their effectiveness can be defined in three aspects: accepting of embryos, complement of the washing, risks of surgery intervention.

Regarding to the accepting of embryos, essential differences between the two methods are not noticed in the studies up to now. Accepting the embryos with median laparotomy and laparoscopy, correspondently in 7 and 5 sheep-donors, is made in one of our trials. Using the first of the mentioned methods, 21 embryos are accepted from 41 corpus luteums were observed (accepting 51.2 % ±7.80). And using the second method 24 embryos are accepted of 34 corpus luteums (accepting 61.8 % ±8.33).

The observed differences are not proved mathematically (Vlachov et al., 1991). Independently of the existing differences regarding to the accepting of embryos we have reason to maintain that the two methods are not essentially different as it refers to this indicator.

In order to optimize the risks of surgery intervention when the methods of median laparotomy and laparoscopy are used, it is necessary to mention, that there is one essential disadvantage of the first method. Using this method, it is not possible the degree of superovulation response of sheep-donors to be evaluated in advance. This leads to extra surgery intervention, regarding to the surgery opening of the abdominal cavity of negative reaction donors.

Regarding to the technical fulfillment of the median laparotomy and laparoscopy it can be said that it leads to mechanical trauma of uterus, oviducts and mezovarium. There is also some danger of surgery infection. In this way, there are conditions of risks for decreasing of the reproductive status of sheep and their secondary use as donors. Sometimes, post operation complications are observed. In this case the period of recovery of the animal prolongs approximately 7 days. The mentioned risks and complications are not observed after the use of laparoscopy (Scudamore et al., 1990; Vlachov et al., 1991).

Conclusions

1. Accepting embryos with laparoscopy is highly accessed and effective method, due to which the risks of operation and postoperation complications are limited.
2. The application of both methods requires the team principle to be used and also the operators to be highly qualified.
3. The laparoscopy requires expensive technique and equipment, which makes the method difficult.
TEHNIČKI ASPEKTI PRIHVATANJA EMBRIONA OD OVACA DONATORA
KORIŠĆENJEM MEDIJALNE LAPARATOMIJE I LAPAROSKOPIJE

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Rezime

Uprkos mogućnostima koje pruža metoda embrion troška (ET) u ovčarstvu ona ima ograničenu primenu u postojećim programima selekcije i odgajivanja. Razlog tome je često nizak nivo superovulatorne reakcije (Whyman et al., 1980), lošija vitalnost oplodenih jajnih čelijskih (Graham, 1986) i komparativno nizak procenat oplodnje (Robinson et al., 1989). Nedostaci metode prihvatanja embriona medijalnom laparotomijom su takođe veoma važan problem koji se teško može rešiti s obzirom da je povezan sa rizicima povrede unutrašnjih genitalnih organa i njihovom adhezijom. To dovodi do nemogućnosti korišćenja donatora po drugi put (Vlachov et al., 1991).

Cilj ovog ispitivanja je bio uporedno ispitivanje mogućnosti dve metode za prihvatanje embriona, posebno sa medijalnom laparotomijom i laparoskopijom.

Rezultati su dokazali prednosti metode za prihvatanje embriona sa laparoskopijom u tri aspekta: ispitivanje embriona, rizik od hirurške manipulacije i veća lakoća pristupa.

Laparoskopija je veoma efikasna. Rizik od hirurške intervencije je minimalan. Neophodnost veoma skupe tehnike je nedostatak ove metode.

Ključne reči: ovca, laparoskopija, embrion trošak, embrioni.

References