Evaluation of pregnancy rate and length of pregnancy after artificial insemination in Zwartbles sheep

Vojtech Pesan, Martin Hosek, Radek Filipcik
Department of Animal Breeding
Mendel University in Brno
Zemedelska 1, 613 00 Brno
CZECH REPUBLIC
vojtech.pesan@mendelu.cz

Abstract: The oestrus cycle synchronization and artificial insemination are some of the most frequently used biotechnical methods of reproduction. They are used mainly to achieve easier breeding and shortening of the lambing period. An exact determination of the lambing period (according to the length of the pregnancy) is essential for the breeding process optimization, namely it allows a precise timing of the lambing period. The experiment monitors forty eight Zwartbles ewes, whose oestrus cycles were first synchronized with the use of hormonal intravaginal sponges Ovigest. The subsequent intracervical insemination was done using fresh, diluted and chilled semen from rams of the Zbyšek and Zachari lineages extracted into an artificial vagina. The pregnancy rates after insemination and after the subsequent breeding of the barren ewes in a harem were determined according to the pregnancy detection and the subsequent lambing times. The overall pregnancy length in the artificially inseminated animals was determined through marking the exact date and time of insemination and the lambing period of each animal. The pregnancy rate after insemination was 63.6% for ram Zbyšek and 23.1% for ram Zachari. The overall pregnancy rate after insemination was 41.7%, and after the subsequent natural breeding of the barren ewes in harems the overall pregnancy rate was 93.8%. The pregnancy periods generally ranged between 141 and 147 days and the lambing itself took place between the 144th and 146th day in 70% of the cases.

Key Words: insemination, pregnancy rate, reproduction, sheep, synchronization of Oestrus, Zwartbles

INTRODUCTION

Similarly to other ruminants, sheep breeding is usually done through natural breeding or artificial insemination (Louda and Hegedušová 2009). Both the natural breeding with prior oestrus cycle synchronization and artificial insemination (with or without prior oestrus cycle synchronization) are amongst the most often used biotechnical methods used in sheep breeding. These methods can assure decrease of the work load and time restraints of the breeding process, they can assure a better record keeping and health monitoring of the animals, as well as shortening of the lambing period, which subsequently leads to a better balance in the flock (Čunát et al. 2013, Sándor et al. 2011).

The methods used for oestrus cycle synchronization can be divided into the artificial (additives in the feed, intravaginal sponges, subcutaneous implants), and natural ones (light regimen control, flushing, the ram effect); (Čunát et al. 2013, Horák et al. 2012, Louda and Ježková 2002). For better efficiency, these individual methods can be combined (for example the ram effect + flushing, the ram effect + intravaginal sponges, flushing + intravaginal sponges); (Čunát et al. 2013, Říha 1999).

The artificial insemination is performed using fresh, chilled, diluted semen. Previously extracted doses stored in liquid nitrogen for a longer period of time are used only rarely, since this method of conservation leads to a rapid decrease of fertility of the insemination dose after its defrosting (Louda et al. 2001, Ntemka et al. 2018). In the Czech Republic, frozen insemination doses are currently created only for certain gene reserve programs for the Valachian sheep and Šumavská sheep breeds. Rarely, they are imported from abroad on demand of the breeder or created from top-level rams of domestic origin (Čunát et al. 2013; Louda and Hegedušová 2009).

Insemination methods are further divided according to the place of the semen insertion - generally, the deeper into the sexual apparatus of the ewe it is inserted, the higher probability of pregnancy, but also the higher price and difficulty of the method. These methods are thus divided into intravaginal, intracervical, and intrauterine ones (Čunát et al. 2013, Sándor et al. 2011).
The insemination doses are created from sperm extracted at the day of insemination. Semen is usually extracted into an artificial vagina after a jump on an ewe in heat. To eliminate the time restraint and to eliminate the need of an ewe being present, an electroejaculator can be used. The extracted semen is then macroscopically and microscopically evaluated. For the dilution, commercial thinners, as well as milk-based thinners, can be used (Čunát et al. 2013). The milk used for dilution was pasteurized (min. 95 °C for 10 minutes) to inactivate lactein, an antibacterial agent that would act as toxic towards the semen in its active state (Salamon and Maxwell 2000).

Subsequently, it is crucial to know the exact data concerning the pregnancy length. This data allows for an estimation of lambing times, and thanks to the estimated number of expected lambs, the care for the new-borns can be optimized and complications, which usually arise due to the bad lambing times management, can be eliminated.

The pregnancy lengths vary. Vaněk and Štloc (2002), Kuchtík (2015) state that the pregnancy can last between 143 and 157 days, while the average pregnancy length is somewhere between 147 and 150 days according to them. Gajdošík and Polách (1984) state that the upper limit is one day shorter (156 days). The average of 147 days of pregnancy with a deviation within a few days is stated in the works of Tzanidakis et al. (2014) and Gootwine (2016).

The correct choice and application of biotechnical methods is crucial for optimization of reproduction outcomes. The aim of this work is to evaluate the pregnancy lengths of individual sheep.

MATERIAL AND METHODS

This experiment focused on 48 Zwartbles breed sheep from private farm belonging to dr. HOSEK in the MOHELNO, CZECH REPUBLIC. All of the sheep were of age ranging between 2 and 8 years, with an average body condition score (BCS) of 3. The experiment itself took place between September 2019 and April 2020. The synchronization of the oestrus cycle using intravaginal sponges, insemination doses extraction and dilution, the insemination itself and the pregnancy scanning all took place in September 2019. In April 2020, the experiment ended with the lambing.

The rams, as well as the ewes, were fed grass silage and hay ad libitum. Additionally, 400 g of grains and 200 g of fodder potatoes per animal were fed to the animals one month before and one and a half month after the insemination, serving as a flushing.

For the oestrus cycle synchronization performed in 2019, only intravaginal sponges Ovigest (60 mg medroxyprogesterone acetate/sponge, Laboratorios Hipra, Spain) were used. These hormonal progesterone preparations or synthetic progestogens obstruct the oestrus cycle. Using a specialized applicator, these sponges were inserted inside the ewes’ vaginas for fourteen days. After removal of the sponges, each ewe was injected with a lyophilized serum gonadotropin PMSG (0.2 ml/ewe = Sergon 200 IU, Bioveta Ivanovice, CZ), which works similarly to the hormone-stimulating follicles (FSH) and the luteinizing hormone (LH). 56–60 hours after this injection, the ewes were inseminated.

Two rams, one from the Zbyšek (ZBS 0001 – 7 years) and one from the Zachari (ZAC0001 – 8 months) lineages were used for the ID (insemination dose) creation. An ewe was fixed to a fixing pad and the semen was extracted into an artificial vagina (Minitübe, Germany) in one jump (two jumps in case of a lower amount of extracted semen) on the day of insemination.

The semen was then macroscopically and microscopically evaluated and the degree of dilution was determined due to the observed sperm concentration and motility. For the dilution, pasteurized cow milk with 3.5% fat content was used in a 1:1–2 (milk:semen) ratio. The diluted semen was then placed in small labelled plastic containers and cooled down to 3 °C in a cooling box.

The insemination itself was performed 56 to 60 hours after removal of the intravaginal sponges and PMSG application. The ewes were divided into two groups and the insemination took place on October 11, 2019, from 13:00 to 14:30 and on October 13, 2019, from 12:20 to 14:40.

Each ewe was fixed to a fixing pad by a collar and, for better access, the tail was fixed as well. Each ewe was then evaluated due to its oestrus symptoms (the colour and amount of mucus, blood perfusion, stiffness and openness of cervix, overall activity, etc.).

The insemination itself was performed with a plastic tube fixed to a plastic syringe. The diluted semen was drawn into the syringe in a 0.4 ml amount. During the time of insemination, the ID was not
taken out from the cooling box to assure its viability, to reduce temperature changes and to eliminate possible contamination.

Before the insemination, the outer genitalia were cleaned and disinfected. The vagina and cervix were inspected using a 12cm long sheep vaginal speculum and a LED torch. Excess mucus was removed where needed. The ID was then inserted through the speculum 1 to 2 cm deep into the cervix.

Several days after the insemination, the ewes were put into groups (harems) with the rams whose semen was used for the insemination. This was done to assure an increase in the pregnancy rate in the ewes in which the artificial insemination was not successful.

The pregnancy scanning took place on the 43rd and 83rd day after insemination using OVI-SCAN (BCF technology, Scotland) and, along with the data on pregnant/barren ewes, the number of foetuses in the uterus was noted.

Due to marking the exact day and time of insemination and lambing in individual ewes, the overall pregnancy durations were subsequently calculated.

STATISTICA 12.0 software and MS Excel 2016 were used for statistical assessment of the data.

RESULTS AND DISCUSSION

Figure 1 shows the pregnancy rates after insemination/breeding with the individual ram lineages. The pregnancy rate after insemination was 63.6% for ram Zbyšek (ZBS 0001) – 14 pregnant sheep out of the 22 inseminated – which was an above-average outcome compared to the 60% presented by Kuchtík et al. (2007) as an average for the Czech Republic. On the other hand, with ram Zachari (ZAC 0001) the pregnancy rate after artificial insemination was only 23.1% – 6 pregnant ewes out of the 26 inseminated), which is a very below-average outcome caused by the worsened quality of the IDs. The overall pregnancy rate after artificial insemination was thus 41.7%, which is a below-average outcome.

Figure 1 Comparison of the pregnancy rates after insemination and the overall pregnancy rates in individual rams

After the insemination itself and after letting the barren ewes naturally breed in the next oestrus cycle, the ewes were put into harems with the rams whose semen was used for the insemination, to assure an increase in the pregnancy rate. The subsequent overall pregnancy rate was 95.5% with ram Zbyšek and 92.3% with ram Zachari. The overall pregnancy rate was thus 93.8% (45 pregnant ewes out of 48 used in the experiment). This can be considered a relatively good outcome, since Louda and Hegedušová (2009) state the average pregnancy rate in natural breeding to be 95%.

Vallejo et al. (2019) states in his study that the pregnancy rates at sheep after oestrus synchronization done with hormonal preparations and after subsequent intracervical insemination were
between 45% and 65%. He also states that after the follow-up ten days of natural breeding of the barren sheep in a harem the overall pregnancy rate reached more than 80%.

Within the individual rams, a highly statistically provable difference was noted in the individual pregnancy rates. On the contrary, within the data on overall pregnancy rates, there was no notable statistical deviation.

Figure 2 Length of pregnancy in sheep lambing in 2020

Figure 2 show the lengths of pregnancy in individual sheep inseminated on October 11, 2019 between 13:00 and 14:30 and October 13, 2019 between 12:20 and 14:40. During these phases a total of 48 ewes were inseminated. After the insemination, the pregnancy rate was 41.7%, that is 20 ewes (in which an exact pregnancy duration could be determined after lambing, which is shown in Figure 2), which can be considered a below-average outcome compared to the average pregnancy rates after artificial insemination (Kuchtík et al. 2007, Louda and Hegedüšová 2009). This average rate after insemination is usually 60% (Kuchtík et al. 2007).

The first lambing took place 141 days and 13.5 hours after the insemination. The last lambing took place 147 days and 19.5 hours after the insemination. The lambing thus took place within a 150 hour span (6 days and 6 hours). In Figure 2 and Table 1 we can observe that the most frequent lambing times came in the middle of day 144 and lasted until the middle of day 146 of pregnancy.

Table 1 Frequency of lambing according to pregnancy length

<table>
<thead>
<tr>
<th>Days range after insemination</th>
<th>141.0–141.9</th>
<th>142.0–142.9</th>
<th>143.0–143.9</th>
<th>144.0–144.9</th>
<th>145.0–145.9</th>
<th>146.0–146.9</th>
<th>147.0–147.9</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of lambing sheep (%)</td>
<td>1</td>
<td>2</td>
<td>4</td>
<td>7</td>
<td>3</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>(%</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>35</td>
<td>15</td>
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</table>

The pregnancy length in ewes that were lambing in 2019/2020 (as seen in Figure 2 and Table 1) varied between 141 and 147 days. However, very little ewes go into lambing during the two threshold days and these cases are considered an exception. In comparison with the average pregnancy lengths in sheep mentioned in the introduction (143–157 days), the average pregnancy length observed in this research was approximately 145 days, which is two days less than stated by e.g. Ingoldby and Jackson (2016).
The pregnancy length can be influenced by a number of factors, such as the breed, the feed composition, microclimatic and macroclimatic conditions, sex or the number of foetuses. For this reason, to gain more exact data, it is necessary to replicate this research and carry it out again on a larger number of animals and, subsequently, to evaluate the influence of these factors on the pregnancy length itself.

CONCLUSION

Within this research, the pregnancy length in Zwartbles breed sheep was determined to last between 141 and 147 days, while majority of the animals went into lambing between days 144 and 146.

These numbers are lower than the average pregnancy lengths usually stated for sheep in general, without taking the breed into consideration.

The pregnancy rate after insemination was 63.6% for ram Zbyšek and 23.1% for ram Zachari. The total pregnancy rate after insemination was 41.7% and the overall pregnancy rate after insemination and natural breeding in a harem was 93.8%.

The results of this research will be used for its replication and broadening, mainly for an even more exact determination of the lambing times and better time management for the lambing times preparations.

REFERENCES


