

## **Length of pregnancy in inseminated Zwartbles sheep with previously synchronized oestrus cycle**

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*Abstract:* Easier breeding and shortening of the lambing period is mainly achieved with the use of insemination with the preceding oestrus cycle synchronization. With these biotechnical methods we are thus able to estimate the time span of lambing. However, another topic closely tied to this issue is the length of pregnancy, which can depend on the breed, the age of the individual animal, the number of fetuses, etc. Higher accuracy in the pregnancy length estimation might again lead to easier breeding and higher quality reproduction. In this research were observed thirty five Zwartbles ewes (from 2017 to 2019), first synchronized using tough intravaginal sponges (hormonal) Chronogest, Ovigest and CIDR (Controlled internal drug release). Afterwards, the ewes were inseminated with fresh, diluted and chilled semen taken from four ram lineages. Due to writing down the exact time of insemination and, subsequently, the exact time of lambing in each individual animal, it was possible to evaluate and compare the pregnancy lengths in individual animals. The total pregnancy rate after insemination was 50% in 2017 and 64.7% in 2018. The pregnancy length ranged between 142 and 148 days, and most of the animals lambed down between the 143<sup>rd</sup> and 146<sup>th</sup> day of pregnancy.

*Key Words:* lambing, length of pregnancy, reproduction, sheep

### **INTRODUCTION**

Similarly to cattle breeding, either natural breeding or artificial insemination is used in sheep breeding (Louda and Hegedüšová 2009). Artificial insemination without synchronization of the oestrus cycle, with the synchronization, but also the natural breeding with preceding synchronization are all biotechnical methods used mainly to lower the demand on time and effort needed for breeding, to achieve easier and more precise record keeping of the animals, to achieve balance in the flock, as well as to shorten the lambing period (Čunát et al. 2013, Sándor et al. 2011).

Methods used for synchronization can be divided into natural (light regimen control, flushing and the ram effect) and artificial (additives in the feed, intravaginal sponges, subcutaneous implants) (Čunát et al. 2013, Horák et al. 2012, Louda et al. 2002). To reach better results, these individual methods can be combined (for example intravaginal sponges + flushing, intravaginal sponges + the ram effect, the ram effect + flushing, etc.) (Čunát et al. 2013, Kuchtík 2013, Říha 1999).

The insemination itself is performed mainly using fresh, chilled semen. Significantly less frequent is insemination with use of doses frozen and stored in liquid nitrogen over a longer period of time. These doses, however, show a significant lack of fertility after defrosting, thus being used mainly within the gene reserve program. Methods of insemination can be divided by the place of the semen insertion (the further away from the sexual apparatus of the ewe, the higher probability of pregnancy, but also the higher price and difficulty of the operation) into intravaginal (into the upper part of the vaginal fornix) and intrauterine (laparoscopically, into the uterus) (Čunát et al. 2013, Sándor et al. 2011).

After performing these reproduction methods, it is crucial to know the most exact data about the pregnancy length for the estimation of lambing. Such data then allows us to shorten the preparation period for lambing to an optimal length and, thanks to the provisional numbers of future lambs acquired through the pregnancy scanning, it leads to an optimal subsequent lamb care,

an optimal utilization of space, as well as lowering (or elimination) of possible complications, which are usually caused by unpreparedness and poor time arrangement of the lambing.

The pregnancy length in ewes varies. According to Vaněk and Štolc (2002) and Kuchtík (2013) and Kuchtík (2015), it can vary between 143 and 157 days, making 147–150 days the average pregnancy length. According to Gajdošík and Polách (1984), the upper limit is one day shorter (156 days). The average length of 147 days, with a difference of a few days, is introduced by Tzanidakis et al. (2014), Gootwine (2016).

The correct selection of biotechnical methods and their execution (semen extraction, the insemination doses creation, the insemination itself, or the synchronization of oestrus) is crucial for reproduction optimising. This research focuses on evaluation of the pregnancy lengths in individual ewes, and the subsequent optimising of the lambing process.

## MATERIAL AND METHODS

A total number of 54 Zwartbles ewes from two to eight years of age and with the average body condition score (BCS) 3 was used for this research. These animals were observed at the Ing. Martin Hošek, Ph.D. family farm in Mohelno and the research itself took place from September 2017 to March 2019.

In the first year of research, the synchronization was performed at the end of September 2017. The subsequent insemination took place fourteen days later (9. 10. 2017). Pregnancy scanning was performed on the 46<sup>th</sup> and 89<sup>th</sup> day after insemination using OVI-SCAN (BCF technology, Scotland). The subsequent lambing took place from the end of February to the start of March 2018, in the span of six days.

At the end of September 2018, the second year of research, the ewes were again synchronized and then inseminated fourteen days later (13. 10. 2018). The pregnancy scanning took place on the 43<sup>rd</sup> and 83<sup>rd</sup> day after insemination, similarly to the scanning in 2017. The lambing took place in the first third of March in the span of six days.

The synchronization in both years was performed using an intravaginal device (term from literature e.g. Kesler 2002) CIDR (Controlled internal drug release) and intravaginal sponges Chronogest and Ovigest:

- CIDR: analogue progesterone – synthetic progestogen, 300 mg progesterone/insert, Pfizer
- Chronogest: progestogen, 20 mg cronolone/sponge, Intervet
- Ovigest: 60 mg medroxyprogesterone acetate/sponge, Laboratorios Hipra, Spain

These hormonal progesterone preparations or synthetic progestogens obstruct the oestrus cycle. The sponges (Chronogest and Ovigest) and CIDR were inserted into the vagina for 14 days using a specialized applicator. 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). In 56–60 hours after this injection the ewes were inseminated.

Two rams from the Zbyšek (ZBS 6 years) and Zbyslav (ZBY 7 years) lineages were used for the ID (insemination dose) creation in 2017, while four rams from the Zbyslav (ZBY 2 years), Zbyšek (ZBS 3 years and 7 years) and Zoubek (ZOU 2 years) lineages were used in 2018. 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 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 plastic containers and cooled down to 3 °C in a cooling box, where it stayed until the insemination itself. 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).

Both ewes and rams were fed with grass silage and hay ad libitum. Additionally, 400 g of grains and 200 g of fodder potatoes per animal was fed to the animals one month before and one and a half

month after the insemination, serving as the flushing to increase the synchronization efficiency and the pregnancy rate.

The insemination took place 56 hours from the administration of PMSG. From 12:30 to 14:00 in 2017 and from 12:40 to 15:15 in 2018. Each ewe was fixed to a fixing pad by a collar and the tail was fixed as well for better access. Before the insemination, each ewe was evaluated due to its oestrus symptoms (amount of mucus, stiffness and openness of cervix, overall activity, etc.). The insemination itself was performed with a plastic tube fixed to a plastic syringe. The semen was drawn into the syringe in a 0.4 ml amount without taking the container with semen out of the cooling box. This way the viability of the ID was not reduced and the ID was not contaminated. Before the insemination, the outer genitalia were cleaned and disinfected. The vagina and cervix were inspected using a 12 cm 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.

For statistical evaluation of the results were used Microsoft Excel.

## RESULTS AND DISCUSSION

Figure 1 and Table 1 shows the lengths of pregnancy in individual sheep inseminated 9. 10. 2017 between 12:30 and 14:00. During this phase a total of 20 ewes were inseminated. After the insemination, the pregnancy rate was 50%, that is 10 ewes (the pregnancy length was deduced from the time of lambing for accurate determination).

Figure 1 Length of pregnancy in individual sheep (2017–2018)

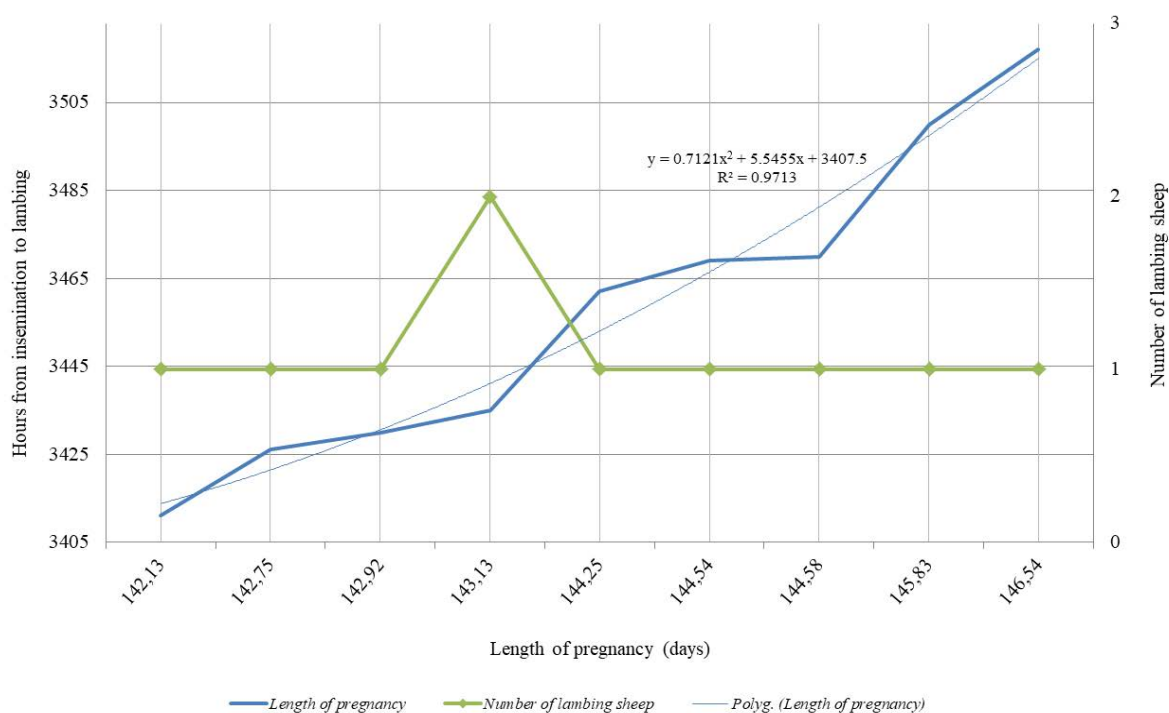


Table 1 Length of pregnancy in individual sheep (2017–2018)

Length of pregnancy (hours)										Average	Sx	Vx
<b>3411</b>	3426	3430	3435	3435	3462	3469	3470	3500	<b>3517</b>	3455.50	32.463	0.9395
Length of pregnancy (days)												
<b>142.13</b>	142.75	142.92	143.13	143.13	144.25	144.54	144.58	145.83	<b>146.54</b>	143.98	1.353	

Legend: Sx – Standard deviation, Vx – Coefficient of variation, Minimum and Maximum in bold

The first lambing took place 142 days and 3 hours after insemination, the last lambing took place 148 days and 13 hours after insemination. Thus, the whole lambing took place within 106 hours (4 days and 10 hours). As visible in Figure 1, the main lambing period took place from the end of day 142 to the middle of day 144 of pregnancy.

Figure 2 Length of pregnancy in individual sheep (2018–2019)

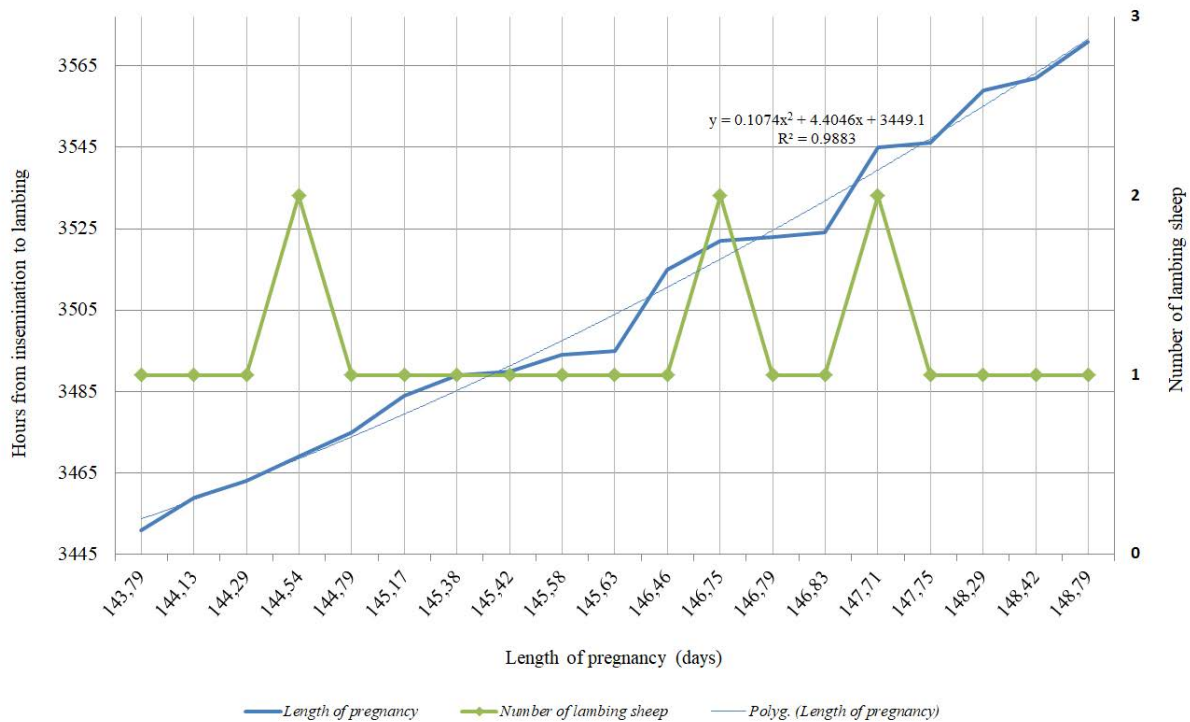


Table 2 Length of pregnancy in individual sheep (2018–2019)

Length of pregnancy (hours)											Averagea	Sx	Vx
<b>3451</b>	3459	3463	3469	3469	3475	3484	3489	3490	3494	3495	3507.82	35.689	1.0174
3515	3522	3522	3523	3524	3545	3545	3546	3559	3562	<b>3571</b>			
Length of pregnancy (days)											146.16	1.487	
<b>143.79</b>	144.13	144.29	144.54	144.54	144.79	145.17	145.38	145.42	145.58	145.63			
146.46	146.75	146.75	146.79	146.83	147.71	147.71	147.75	148.29	148.42	<b>148.79</b>			

Legend: Sx – Standard deviation, Vx - Coefficient of variation, Minimum and Maximum in bold

Figure 2 and Table 2 shows the lengths of pregnancy in individual sheep inseminated on 13.10.2018 between 12:40 and 15:15. In this phase a total of 34 ewes was inseminated. After the insemination the pregnancy rate reached 64.7%, that is 22 ewes (the pregnancy length was deduced from the time of lambing for accurate determination), which is an above-average outcome for a pregnancy rate after insemination. The average of pregnancy after insemination is 60% (Kuchtík et al. 2007).

The first lambing took place 143 days and 19 hours after the insemination, while the last one took place 148 days and 19 hours after the insemination. The whole lambing thus took place within 120 hours (5 days). Figure 2 shows that the main lambing period was split into two parts. The first peak occurred between the beginning of day 144 and the middle of day 145. After a 20-hour break, the second peak occurred in the middle of day 146 and ended in the middle of day 148 after the insemination.

Figure 3 shows the year-on-year difference in pregnancy lengths in ewes who became pregnant after insemination in both years 2017 and 2018. The pregnancy length varies significantly in the individual ewes. However, the year-on-year comparison shows that in half of the ewes, the year-on-year pregnancy length difference was quite low, 10 hours at highest. The other half of ewes however showed a striking year-on-year difference, up to 5 days.

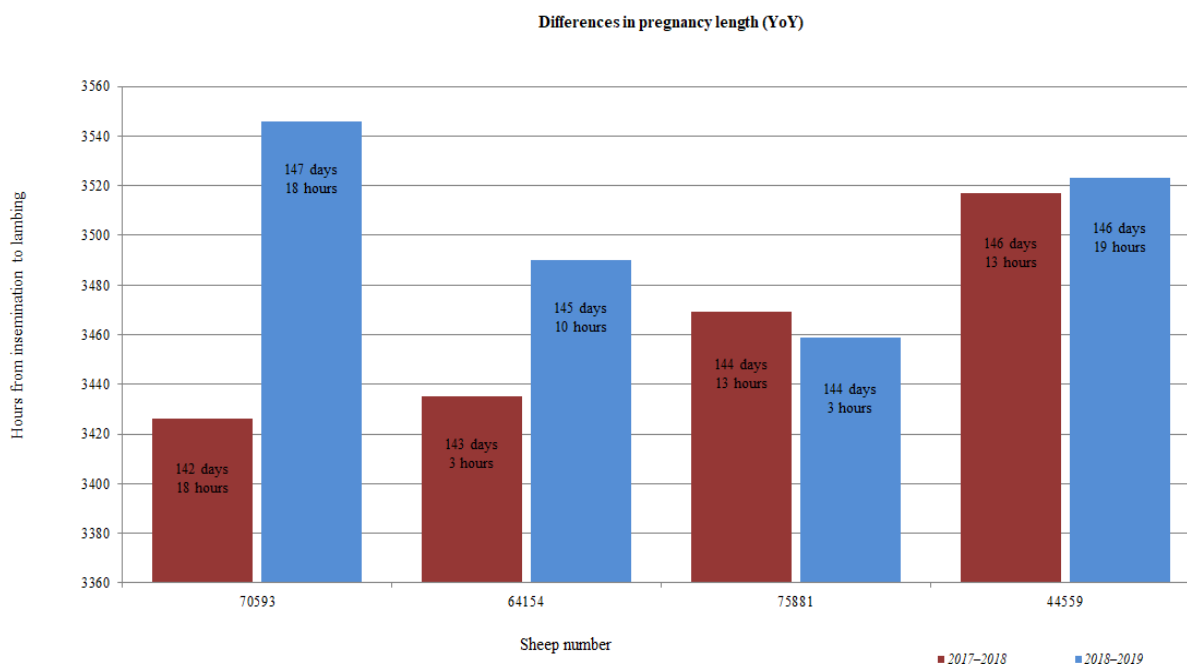
The pregnancy length in ewes that were lambing in 2018/2019 (as seen in Figures 1 and 2) varied between 142 and 148 days. However, very little ewes go into lambing during the two threshold days, and the lambing is mainly an exception. In comparison with the average pregnancy lengths in sheep mentioned 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 difference in pregnancy lengths in 2018/2019 might be caused by differences in temperature, alternatively by a different numbers and genders of the foetuses in individual ewes.

Since the pregnancy length can be affected by a number of factors, from the feed composition to the microclimate, the amount of foetuses or the breed of sheep, it is necessary this research is replicated on a bigger number of animals with the subsequent evaluation of the impact of these factors on the pregnancy length itself.

Figure 3 Year-on-year differences in the length of pregnancy in sheep



## CONCLUSION

This research found that the pregnancy length in Zwartbles sheep varied between 142 and 148 days and the lambing took place between the 143<sup>rd</sup> and 146<sup>th</sup> day of pregnancy in majority of the ewes. These values are lower than the average pregnancy lengths usually stated for sheep without a breed specification.

Data acquired in this research will be used for its replication and extension, mainly to determine the lambing period more precisely and to achieve a better time arrangement during lambing.

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