The impact of oestrus synchronization by hormone medication and varying numbers of artificial insemination sessions, determined by sow reproductive indices

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Summary

This study was conducted on a central Albanian pig farm with a capacity of 80 sows. A uniform distribution of piglets born and the number of reared piglets sold every week and month was the target. To achieve this goal, a hormone-based synchronization scheme was used, combined with the use of one, two, or three artificial insemination sessions during the same oestrus. The main reproduction indices of sows were monitored, and the results indicated that the farrowing rate after the first insemination was 78.57%, 83.64% and 83.33% for the three groups in which one, two and three artificial insemination sessions were used during the same oestrus, obtained by inoculation with the hormone Sergon PG 400/200 IU. Average litter size (the number of live born piglets) per farrowing sow was 10.22 ± 0.33, 10.8 ± 0.2 and 10.6 ± 0.3 in the three groups, respectively. The fecundity index (FI) was 803, 905.8 and 883.3 in the three experimental groups of sows, respectively. A total of 1436 piglets from 136 farrowing sows were sold during 2018, achieving uniform sales of over 100 pigs per month with a weight over 90 kg. Therefore, it can be concluded that oestrus synchronization of sows with the Sergon PG 400/200 hormone can achieve a uniform distribution of piglets born and consequently a uniform distribution of piglets sales per week or month, regardless of the fact that this should be achieved at a larger scale. The best result as seen in the reproductive indicators was achieved by the use of artificial insemination with two sperm inoculations at a 12-hour interval during the same oestrus, synchronized by the Sergon PG 400/200 hormone.

Key words: artificial insemination; oestrus synchronization; hormone; reproduction indexes; sows

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Introduction

Pork producers in Albania are interested in increasing reproductive performance on their farms. Greater control and distribution of born piglets is implemented with the aim of better planning of monthly sales. The use of hormonal drugs for the synchronization of oestrus and consequently piglets, and the use of biotechnology for the artificial insemination of sows, remain challenges for farmers in this sector. The advantages of artificial insemination in sows are numerous. There are different techniques available, such as intra cervical insemination (Cassar et al., 2005; Caravaca et al., 2012), Post-Cervical Insemination (PCI; Rath et al., 2000; Knox et al., 2011; Fontana et al., 2014) and Deep intrauterine insemination (DIU), or intra uterine horns techniques (Watson et al., 2001; Dimitrow et al., 2007).

Different factors influence the fertility of liquid boar semen, including storage time, conservation, season and the constitutions of dilutions (Koh et al., 1976; Sulo, 1985; Alexopoulos et al., 1996; Roca et al., 2006). Different techniques use different volumes of liquid inseminate, and in turn different numbers of spermatozoa per dose (Wiggins et al., 1951; Krueger et al., 1999; Martinez et al., 2001).

The target of these techniques is the number of doses collected per boar during the year. The number of boars on the farm improve the quality of semen and quality of boars (Table 1).

According to the literature, the number of spermatozoa per dose for artificial insemination is very important. In cervical procedures of the artificial insemination (CAI) in sows, billions of spermatozoa are used (2.5-4 x 10^9 spermatozoa per insemination dose) in varying volumes of liquid per dose (70-100 mL). Approximately 90% of the spermatozoa inseminated cannot be recovered from the uterus 2 hours after artificial insemination (AI) (Pursel et al., 1978; Viring, 1980). According to Mburu et al. (1996), only 1 x 10^5 spermatozoa reach the uterotubal junction and about 1 x 10^3 reach the sperm reservoir in the caudal 1-2 cm of the isthmus (Hunter, 1981).

Table 1. Different techniques of artificial insemination in sows, their volume and number of spermatozoa

|                              | Traditional artificial insemination Intra Cervical (CIA) | Post Cervical Insemination (PCI) | Intra Uterine horn insemination or Deep intrauterine insemination (DIU) |
|------------------------------|----------------------------------------------------------|---------------------------------|-------------------------------------------------------------------------|
| Number of Spermatozoa       | (3x10^9/80-100 mL)                                       | (1.5x10^9/40-50 mL)             | (0.6-1x10^9/20-30 mL)                                                   |
| Volume of liquid             | 100 mL                                                   | 50 mL                           | 20 mL                                                                   |
| Frequency of Ejaculation per boar/week | 2 times/week                                           | 2 times/week                    | 2 times/week                                                            |
| Doses per ejaculation        | 20                                                       | 40                              | 100                                                                     |
| Doses per year               | 2,000                                                    | 4,000                           | 10,000                                                                  |
| Proportion of boars          | 10                                                       | 5                               | 2                                                                       |

Table adapted from the literature
where the cells can be stored without reducing their fertilizing ability (Suarez et al., 1991) until just before ovulation (Hunter, 1984). Sufficient spermatozoa to ensure subsequent fertilization are established in the isthmus reservoir within one hour of mating (Hunter, 1981, 1984). Some of the principal reasons for loss of spermatozoa after insemination are backflow (Steverink et al., 1998; Matthijs et al., 2003), phagocytosis by polymorphonuclear neutrophils (PMN), which influx the porcine uterus after insemination (Rozeboom et al., 1999; Matthijs et al., 2003), and sperm losses in the uterine horns caused by the adhesion of sperm to the ciliary epithelial cells of the uterus (Levis et al., 2001).

Other studies have investigated the use of one, two or three artificial insemination sessions during the same oestrus. The number of insemination sessions can affect the cost of insemination (Alexopoulos et al., 1996; Caravaca et al., 2012; Driancourt et al., 2013; Ulguim et al., 2016). Depending on the situation, the most effective technique for the best fecundity index should be selected at the individual farm level, and this depends on farrowing rate and the number of piglets born alive per litter. For this reason, different hormones have been used in the past to stimulate oestrus synchronization (Cox et al., 1983; Dial and BeVier, 1986; Britt et al., 1989). Based on the above techniques, a new group of hormones is used in protocols for oestrus synchronization (De Rensis and Kirkwood, 2003; Baer and Bilkei, 2004; Degenstein et al., 2008; Manjarin et al., 2010; Martinat-Botte et al., 2010; de Jong et al., 2013; Ulguim et al., 2016).

The purpose of this study was to assess the differences in reproductive parameters in sows following artificial insemination with one, two or three sperm inoculations during the same oestrus, synchronized using the Sergon PG 400/200 hormonal drugs produced by the Bioveta Cz company (lyophilized powder of two hormonal substances: human chorionic gonadotropin (hCG) and equine chorionic gonadotropin (eCG)). Sergon PG 400/200 was applied in accordance with the manufacturer’s recommendations (Bioveta, 2018).

The study was conducted on a pig farm in central Albania with a capacity of 80 sows during 2018. The aim of the study was to evaluate the fecundity index (FI) which depends on the farrowing rate and the number of piglets born alive per litter, by insemination with one, two, or three artificial insemination sessions during the same oestrus, synchronized by the Sergon PG 400/200 hormone (Bioveta, Czech Republic; Bioveta, 2018).

**Material and Methods**

The study was conducted on a pig farm in central Albania with a capacity of 80 sows during 2018. The aim of the study was to evaluate the fecundity index (FI) which depends on the farrowing rate and the number of piglets born alive per litter, by insemination with one, two, or three artificial insemination sessions during the same oestrus, synchronized by the Sergon PG 400/200 hormone (Bioveta, Czech Republic; Bioveta, 2018).

**Hormonal treatment**

The second day after piglets were born, sows were injected with 2 mL I/M Sergon PG 400/200 IU. Oestrus was detected 2-3 days later. Despite the appearance of oestrus, sows were not fertilized at this time. At 21 days after oestrus, artificial insemination was performed. Sows that did not respond after the first treatment were subjected to a second treatment over a 25-day period with the same hormone (Sergon PG 400/200 IU, 2 mL I/M). Sows that did not display oestrus after the second treatment were excluded from the study (Bioveta, 2018).

Serum gonadotropin acts similarly to follicle stimulating hormone (FSH) and the luteinizing hormone (LH) and induces the growth of ovarian follicles. Chorionic gonadotropin acts similar to
LH and supports ovulation and corpus luteum growth. The combination of these hormones induces the fertile oestrus cycle in sows (Bioveta, 2018).

**Sperm collection**

The same boar was used for the insemination of the three groups of sows (large white). Sperm was diluted with BTS (Beltsville Thawing Solution, Minitüb, Germany).

The boar was used to obtain semen once a week using the gloved-hand technique and semen was filtered to remove the gel. The average number of spermatozoa/AI dose was controlled by visual estimation of density according to colour, and measured with a colorimeter. Semen quality assessment, including viability, motility, progressive motility and morphology of spermatozoa, was estimated using a bipolar microscope. After calculation and dilution, doses were stored in 100 mL bottles containing 3 billion ($3 \times 10^9$) spermatozoa per dose. Doses were stored at 16°C and used within 24 h, according to Levis et al. (2001). CAI was performed with disposable spiral tip catheters (Minitüb, Germany).

**Sperm inoculation**

Sergon PG 400/200 IU was inoculated into 166 large white sows after farrowing for the synchronization of oestrus. Oestrus detection was performed twice daily by experienced workers and by allowing sows nose-to-nose contact with mature boars and applying backpressure. The occurrence of oestrus was defined by the standing reflex in front of a teaser boar and reddening and swelling of the vulva, according Levis et al. (2001).

After thorough cleaning and drying the sow’s vulvar labia area, the insemination dose was introduced slowly in the sow’s uterus. The catheter remained in the cervix 2–4 min after insemination to reduce backflow, according to Levis et al. (2001).

Of the 166 sows, 56, 62 and 48 were artificially inseminated with one, two, or three inoculations of sperm, respectively. The age of the sows in the three groups did not differ significantly ($P > 0.1$) with respect to the number of gestations. Other breeding conditions, such as nutrition, prophylactic programmes, etc., were the same for all three groups of sows.

In the first group in which only one dose of semen was inoculated, insemination was performed 12 hours after the discovery of oestrus when the sow stayed with the boar. For the second and third doses, the first insemination was carried out 12 hour after the discovery of oestrus, and the second or third insemination was performed at 12-hour intervals after the first insemination.

**Results and discussion**

The study was conducted to assess the fertility parameters and the total number of piglets born alive per sow, after the insemination of 166 sows with one, two or three insemination sessions during the same oestrus following hormone synchronization.

**Table 2. Number of sows for each group and the hormonal medication used**

| Groups  | No. of sows | Number of inseminations during the same oestrus | Hormone medication used for oestrus synchronization |
|---------|-------------|-----------------------------------------------|---------------------------------------------------|
| Group A | 56          | 1                                             | Sergon PG 400/200 IU                              |
| Group B | 62          | 2                                             | Sergon PG 400/200 IU                              |
| Group C | 48          | 3                                             | Sergon PG 400/200 IU                              |
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**Table 3. Fertility after the first insemination session for three groups of sows**

| Groups | No. of sows | Number of inseminations during the same oestrus | Farrowing on the first oestrus | Hormone medication used for oestrus synchronization |
|--------|-------------|-----------------------------------------------|--------------------------------|-----------------------------------------------------|
| Group A | 56          | 1                                             | 44 sows (78.57%)             | Sergon PG 400/200 IU                                 |
| Group B | 62          | 2                                             | 52 sows (83.87%)             | Sergon PG 400/200 IU                                 |
| Group C | 48          | 3                                             | 40 sows (83.33%)             | Sergon PG 400/200 IU                                 |

**Table 4. Number of piglets born live for each group and for each litter**

| Groups | No. of sows | Number of inseminations during the same oestrus | Live born litter size |
|--------|-------------|-----------------------------------------------|----------------------|
| Group A | 56          | 1                                             | 10.22±0.3 per sow    |
| Group B | 62          | 2                                             | 10.8±0.2 per sow     |
| Group C | 48          | 3                                             | 10.6±0.3 per sow     |

**Table 5. Summary table for fertility and number of piglets born live per sow in three groups**

| Groups | No. of sows | Number of inseminations during the same oestrus | Farrowing on the first oestrus | Live born litter size | Hormone medication used for oestrus synchronization |
|--------|-------------|-----------------------------------------------|--------------------------------|----------------------|-----------------------------------------------------|
| Group A | 56          | 1                                             | 44 sows (78.57%)             | 450 piglets born (10.22±0.3/litter)                  | Sergon PG 400/200 IU                                 |
| Group B | 62          | 2                                             | 52 sows (83.87%)             | 562 piglets born (10.8±0.2/litter)                  | Sergon PG 400/200 IU                                 |
| Group C | 48          | 3                                             | 40 sows (83.33%)             | 424 piglets born (10.6±0.3/litter)                  | Sergon PG 400/200 IU                                 |

The table above shows the percentage of fertility in three groups of artificial insemination of sows receiving one, two or three semen inoculations during the same oestrus, following synchronisation using the hormone Sergon PG 400/200 IU.

The percentage of sow fertility after artificial insemination during the first oestrus was 78.57%, 83.64 and 83.33% for the three groups, respectively, receiving one, two or three semen inoculation during the same oestrus, and the use of the same hormone.

The number of piglets born live per litter was 10.22 ± 0.33, 10.8 ± 0.2 and 10.6 ± 0.3 piglets for the three groups of sows, respectively. In the second and third group of sows, there was no difference in the fertility percentage, or in the average number of piglets born live per litter. These results are consistent with other reports (Britt et al., 1989; Bates et al., 1991).

However, in the first group, which received artificial insemination only once during the same oestrus, a difference of
about 5% was observed in the farrowing rate (78.57%), with a difference of approximately 0.5 piglets on average per litter, less than in the second and third groups of sows (Tables 5 and 6).

According to some authors, the percentage of fertility in artificial insemination may (Roca et al., 2006) or may not be influenced (Sulo, 1985) by the number of sperm inoculations within the same oestrus. This can also be influenced by other factors such as number of spermatozoa per sperm dosage (Alexopuolos et al., 1996), breeder, environmental factors (Koh et al., 1976), and so on. Another important factor that can influence artificial insemination is also the time of oestrus appearance and correct timing of sperm inoculation (Alm et al., 2006).

**Distribution of born piglets and number of rearing pigs on the market each month**

From the tables above, we can see that in the year 2018, 136 fertilized sows delivered a total of 1436 piglets (Table 5, groups a, b, c). Over 1200 rearing pigs (i.e. over 100 rearing pigs per month) were sold during 2018, with a weight of over 90 kg per pig at age near 5 months. Therefore, over 100 rearing pigs per month were obtained (thereby fulfilling the main request from the farm).

**Conclusions and Recommendations**

The results of this study and findings from the literature allow us to conclude that the percentage of sow fertility and total number of piglets born on average per sow using artificial insemination with one, two or three inoculations during the same oestrus following synchronization using hormonal drugs (Sergon PG 400/200 IU) have only inconspicuous differences. The best results were obtained using insemination with 2 or 3 sperm inoculations than with a single inoculation during the same oestrus synchronized by hormonal drugs.

Specifically, the results of sow fertility were 78.57%, 83.64 and 83.33% for the three groups, respectively, in which one, two and three inoculations were used during the same oestrus induced by the hormonal drugs like Sergon PG 400/200 IU.
The total number of piglets born live was 10.22 ± 0.33, 10.8 ± 0.2 and 10.6 ± 0.3 per litter for the three groups, respectively. The second and third group of sows showed no differences in fertility or in the number of piglets born live per litter.

In the first group, receiving artificial insemination only once during oestrus, fertility was about 5% percent lower (78.57%) with a margin of approximately 0.5 piglets per litter less than in the second and third groups.

Based on the results of this study, we recommend pig farmers should use artificial insemination with two sperm inoculations during the same oestrus, following hormonal synchronization using Sergon PG 400/200 IU.

Using only a single inoculation can give lower results. The use of three separate inoculations, however, is not economical and had no effect on increasing the reproductive index in sows.

Thus, the greatest interest in practical use in terms of fertility percentage in sows, the total number of piglets born live per litter and the economic benefit was achieved by administering two sperm inoculations in a 12-hour interval in the same oestrus, following hormonal synchronization with Sergon PG 400/200 IU.

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Utjecaj sinkronizacije estrusa hormonalnim lijekovima i različitim brojem umjetnih osjemenjivanja u reproduktivnim indeksima krmača

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Ova studija provedena je na farmi svinja s kapacitetom od 80 krmača, koja se nalazi u središnjoj Albaniji. Cilj je bila ujednačena raspodjela oprasenih i broja prasadi za uzgoj prodavanih svaki tjedan i mjesec. Za postizanje tog cilja, rabljena je shema sinkronizacije na bazi hormona u kombinaciji s uporabom jednog, dva i tri umjetna osjemenjivanja tijekom istog estrusa. Iz praćenja glavnih reproduktivnih indeksa krmača nakon pripremljene sheme rezultati su pokazali da je stopa prasenja nakon prve oplodnje bila 78,57 %, 83,64 % i 83,33 % za tri skupine u kojima su rabljene jedno, dva, odnosno tri umjetna osjemenjivanja tijekom istog estrusa postignutog injekcijama hormona Sergon PG 400/200 IJ. Veličina okota (broj živorođene prasadi) bila je 10,22 ± 0,33, 10,8 ± 0,2, odnosno 10,6 ± 0,3 prosječno rođene prasadi po oprasenim krmačama u tri skupine. Indeks plodnosti (FI) bio je 803, 905,8, odnosno 883,3 u tri eksperimentalne skupine krmača. Prodano je 1436 prasadi od 136 oprasenih krmača tijekom 2018., postigavši ujednačenu raspodjelu prodaje od više od 100 prasadi u mjesec, težine veće od 90 kg. Iz naših rezultata možemo zaključiti (bez obzira na činjenicu da je iste potrebno analizirati na većem broju grla) da se sinkronizacijom estrusa u svinja pomoću hormona Sergon PG 400/200 može postići ujednačena raspodjela oprasenih prasadi i time ujednačena raspodjela prodaje prasadi tjedno i mjesečno. Najveći uspjeh u reproduktivnim indikatorima pokazala je uporaba umjetnog osjemenjivanja s dvije inokulacije sperme u razdoblju od 12 sati tijekom istog estrusa sinkroniziranog s hormonom Sergon PG 400/200.

Ključne riječi: umjetno osjemenjivanje, sinkronizacija estrusa, hormon, reproduktivni indeksi, krmače