THE EFFECT OF LAMBS DIET SUPPLEMENTATION WITH COW COLOSTRUM AND COW COLOSTRUM PROTEIN CONCENTRATE ON SERUM IgG LEVEL AND BODY GAINS

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Abstract. The study on availability of cow colostrum (C) and cow colostrum protein concentrate (CC) derived from membrane filtration was conducted on Olkuska breed lambs. The lambs were assigned to 4 groups receiving IgG addition in cow colostrum or colostrum concentrate: group I – control, group II – 7.49g IgG in 150 ml C, group III – 7.09 IgG in 125 ml CC, and group IV – 14.18 IgG in 250 ml CC. The lambs were fed directly after the birth. IgG concentration was significantly higher on the 2nd and 12th days of lambs life in the control group compared to group IV supplemented with an increased concentrate dose. The results point that an addition of cow colostrum concentrate obtained as a result of membrane filtration may strengthen lambs immune system, which may be of a special significance especially in case of multiple pregnancies.

Keywords: immunoglobulins, cow colostrum, microfiltration, lambs

Introduction. Low immunoglobulins level in the blood of newborns may be caused by their low concentration in the colostrum, mammary gland disorders, too low amounts of colostrum produced by the mother, especially in the case of multiple lambing, as well as the newborn’s digestive system deficiency. The incidence of the digestive and respiratory diseases in neonates which were not provided with proper passive immune status is subject to a significant increase (Godden et al., 2008; Klobasa and Werhahn, 1989; Nowak et al., 2012; Scott et al., 1979; Szewczyk et al., 1979; Szewczuk et al., 2011). Supplementation of the newborns with immunoglobulin containing preparations does not always provide the expected results, or the cost of globulin obtaining does not offset the gains from such supplementation (Quigley, 2002; Quigley et al., 2002; Witkowski et al., 2005). Quigley (2002) points out that despite many commercially available supplements or colostrum products substitutes, the definition of such products is not clear and well defined. As a result, the products which do not contain adequate amounts of immunoglobulins and other nutrients ensuring proper nutritional and immune status of sucklings can be found on the market.

It has been observed that maternal colostrum often does not provide adequate immunoglobulin levels for neonatal livestock due to their low content (Nowak et al., 2012; Quigley, 2002; Rauprich et al., 2000). In such cases, an alternative may be to provide preserved colostrum or cow colostrum preparation (Klobasa et al., 1992; Klobasa et al., 1994; Machado-Neto et al., 2011; Moretti et al., 2010a) which can improve the immune status of the newborn and ensure its protection in the first period of life. Dairy cows produce 12–15 kg of colostrum during the first two milkings of which 5–6 kg is enough to fulfil the immunological demands of their calves. Colostrum remaining from the first and the second day after calving may be used for support of the immunological system of other mammal sucklings, since it is fully utilized by them. This constitutes large, untapped reserve of natural immunoglobulins (Moretti et al., 2010b). The conditions of cows’ maintenance environment that bovine colostrum is characterized by considerable concentration of the aforementioned proteins, what increases the safety of its application to other animal species, since it can infect the newborns fed with it. An attempt of effective colostrum pasteurization using the temperature causes its jellification, loss of feeding usefulness and considerable biological value lowering just at a temperature of 70°C (Godden et al., 2003; Meylan et al., 1996; Śmietana et al. 2007; Varman and Sutherland, 1994). An application of colostrum membrane filtration is a chance of elimination of bacteria, mould and fungi, what increases the safety of its application to other animals.

The aim of this study was to analyze the possibility of bacterial flora reduction in cows colostrum as a result of thermal treatment, application of focused microwave field, pulsed electromagnetic field and membrane microfiltration, as well as to assess the effects of lambs supplementation after birth with cows colostrum and colostrum protein concentrate, obtained in the process of cow colostrum filtration, on serum IgG level and their gains rate.
Material and methods

Colostrum was collected on the first day after calving from 20 cows of Polish Holstein-Friesian breed, black-and-white variety, which were in 2–4 lactation, and it was frozen at a temperature of -20 °C. Cow colostrum without external symptoms of color and consistence changes was qualified for freezing.

The effectiveness of thermal treatment application, focused microwave field and pulsed electromagnetic field using SU-1 reactor, as well as microfiltration on ceramic membranes, was analyzed in order to eliminate bacterial microflora. Among the methods applied, only microfiltration appeared to be effective, and therefore it was used in the process of preparation for lambs manufacturing.

After thawing, colostrum was mixed and examined for chemical composition and part of it was frozen in portions for the lambs. The other part was subjected to membrane filtration process. The filtration on ceramic membranes was conducted on the whole and fat-free colostrum. To avoid the formation of fat filter on the membranes and inhibition of other colostrum filtration components, the process was performed after removal of colostrum fat in centrifugation process. The concentrate and colostrum were portioned for the lambs in the planned doses, and then frozen.

The composition of colostrum and colostrum concentrate was analyzed using Infrared Milk Analyzer 150 device of Bentley Company. The concentration of G class immunoglobulins in colostrum and colostrum concentrate as well as in lambs’ serum was determined using immunoenzymatic ELISA test with an application of the set of kits for determining bovine IgG. Each set of the test contained IgG standard allowing determining IgG concentration of the set of kits for determining bovine IgG. Each set of the test contained IgG standard allowing determining IgG concentration of the standard curve. Similarity of bovine and ovine immunoglobulins makes this test application possible, which was demonstrated in the study by Moretti et al. (2010b). Microbiological examinations on the presence of E. coli and coliform were performed in cow colostrum after thawing and in the solution obtained after filtration, using Coli ID culturing media of Biomerieux company, the results were counted using aCOLyte reader of Symbiosis company.

The research material consisted of 33 lambs of Olkuska breed sheep born from January to April 2012. After the birth, the lambs were randomly assigned to particular groups. After additional administration, the lambs stayed with mothers. There were four research groups:

- Group I – control (8 lambs);
- Group II – fed with 150 ml of bovine colostrum – 7.49 g of IgG (8 lambs);
- Group III – fed with 125 ml colostrum filtrate – 7.09 g of IgG (9 lambs);
- Group IV – fed with 250 ml colostrum filtrate – 14.18 g of IgG (8 lambs).

Sheep with the lambs stayed in the new sheepfold building and were housed on a deep litter. After the birth, the lambs with mothers stayed in a separate pen for 3 days, and then were included in the group of mothers with lambs. Starting with the 2nd week of life, the lambs had an access to the complete mixture C and hay in a separated pen. Within a period of 1–3 hours after birth, the lambs from the experimental groups (II – IV) were given bovine colostrum or colostrum protein concentrate using the bottle with nipples. The lambs from group IV were given the colostrum in two portions, and the lambs from the control group were fed directly with maternal colostrum. The lambs stayed with their mothers throughout the whole experiment. On the 2, 12, 22 and 32 days of life, blood samples were taken from the right jugular vein in order to determine the level of serum IgG. On these days, and additionally on the 56th day of life, at the end of the experimental period, the body weight of lambs was measured in order to determine the growth rate of lambs in the examined groups. The animals were subjected to a constant zootechnical and veterinary observation.

The results obtained were subjected to statistical analysis using one-factor analysis of variance based on statistical model (group effect) for each of the four dates of blood collection for serum IgG determination. GLM procedures of the statistical package Statistica 8.0 were applied. The significance of the differences between the groups was determined using Duncan’s test.

Results and discussion

The cow colostrum used in the study contained 21.28% IgG immunoglobulin in the dry matter (Table 1), which corresponds to the average quality of the bovine colostrum (Nowak et al., 2012). In turn, the content of the IgG in the colostrum concentrate, obtained by microfiltration, was almost 2.3-fold higher in a dry matter compared to initial colostrum.

<table>
<thead>
<tr>
<th>Specification</th>
<th>Dry matter</th>
<th>Total protein</th>
<th>IgG</th>
<th>Casein</th>
<th>Lactose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cow colostrum</td>
<td>23.49</td>
<td>55.30</td>
<td>21.28</td>
<td>21.78</td>
<td>9.53</td>
</tr>
<tr>
<td>Cow colostrum concentrate obtained in membrane filtration</td>
<td>11.78</td>
<td>81.25</td>
<td>48.16</td>
<td>2.49</td>
<td>12.13</td>
</tr>
</tbody>
</table>

Fresh cow colostrum contained on average $2.8 \times 10^7$ E. coli and $6.9 \times 10^7$ coliform in 1 ml, which constitutes a considerable contamination and may be a health threat when fed to the newborns. Those bacteria were not
recorded in cow colostrum concentrate obtained by microfiltration. Thermal treatment appeared to be useless in bacterial flora reduction in colostrum due to colostrum jellification in higher temperatures. Similar situation was found in the cases of focused microwave field and pulsed electromagnetic field since they reduced bacterial flora only of about 50% in total.

The concentration of IgG in serum of lambs supplemented with cow colostrum and cow colostrum filtrate obtained as a result of membrane filtration is presented in Table 2. The highest IgG concentration was noted in group IV on days 2 and 12 after calving compared to the control group. The differences between these groups on the 2nd and 12th days after birth were 5.43 g/l and 3.81 g/l, respectively, what accounts for 25.4% of the growth and is statistically significant.

Table 2. Mean IgG concentration in serum of lambs supplemented with cow colostrum and cow colostrum concentrate compared to the control group

<table>
<thead>
<tr>
<th>Group</th>
<th>IgG content, [g/l]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day 2</td>
</tr>
<tr>
<td>I</td>
<td>21.39 a</td>
</tr>
<tr>
<td>II</td>
<td>24.33</td>
</tr>
<tr>
<td>III</td>
<td>24.12</td>
</tr>
<tr>
<td>IV</td>
<td>26.82 b</td>
</tr>
</tbody>
</table>

In other experimental groups, there was also a distinct increase in immunoglobulin concentration; in group II, on day 2, 2.94 g/l, i.e. 13.7%, in group III, 2.73 g/l, i.e. 12.7%, and 13.8% and 12.8%, respectively on day 12; however the differences were not statistically significant. On days 22 and 32 of life, the differences between the groups were on a similar level in favour of the experimental groups, but were still not statistically significant. Moretti et al. (2010a) also reported higher levels of immunoglobulins in the blood serum of lambs supplemented with cow colostrum. Machado-Neto et al. (2011) reported no negative impact of cow colostrum on digestive system development and intestinal absorption in lambs. Bovine colostrum immunoglobulins may additionally bond pathogens causing diarrhea and other diseases. Similar activity is also demonstrated in case of IgY immunoglobulins obtained from egg yolk, especially in calves diseases prevention. Lack of significance of the differences between the control group and groups II and III may also result from lower immunoglobulins supply in the supplements compared to group IV which received their double amount.

The results obtained indicate the possibility of lambs passive immunity improvement as a result of administration of cow colostrum or cow colostrum filtrate in the first hours of their life which was also demonstrated by other authors (Klobasa et al., 1994; Machado-Neto et al., 2011; Moretti et al., 2010a; Tsiligianni et al., 2012) who noted an improvement in health status in lambs fed with cow colostrum. The results obtained in the case of lambs from single births feeding point that this form of supplementation may be important in case of multiple pregnancies, when lambs are to a lower degree supplied with IgG by the mother. They can also be also used in case of mothers udder diseases, low quality or absence of colostrum in ewes, as well as in order to increase lambs immune status via single administration of cow colostrum or colostrum concentrate obtained as a result of filtration process.

Table 3. Effect of lambs’ diet supplementation with cow colostrum and cow colostrum preparation on lambs body weight gains

<table>
<thead>
<tr>
<th>Group</th>
<th>Body weight of lambs after birth, kg</th>
<th>Body weight after birth expressed as 100%*</th>
<th>Body weight in subsequent periods [%]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day 12</td>
<td>Day 22</td>
<td>Day 32</td>
</tr>
<tr>
<td>I</td>
<td>4.74</td>
<td>100</td>
<td>168.60</td>
</tr>
<tr>
<td>II</td>
<td>4.63</td>
<td>100</td>
<td>125.45</td>
</tr>
<tr>
<td>III</td>
<td>4.86</td>
<td>100</td>
<td>177.14</td>
</tr>
<tr>
<td>IV</td>
<td>4.35</td>
<td>100</td>
<td>144.66</td>
</tr>
</tbody>
</table>

* weight at birth was assumed as 100%
The results of an application of cow colostrum and colostrum filtrate addition on lambs’ body weight gains are presented in Table 3. It was observed that random selection of the calves to particular groups did not assure their similar body weight in the groups after birth which would have affected their further gains. The highest difference in body weight after birth was noted between groups III and IV (0.51 kg). On day 12, their body weight was lower by 16.5%, and on day 56 decreased up to 9.4% compared to the control group. Analysis of the gains up to day 56 of rearing did not reveal any significant differences between the groups. The highest body weight on the 12 day of life was observed in the lambs from groups I and III, while the lowest in the groups supplemented with cow colostrum and increased addition of colostrum concentrate. At the age of 56 days, body weight of the lambs in groups I and III was significantly lower compared to the control group, and may be applied in order to enhance immunological status of lambs from multiple and other births. No diseases occurrence was noted in all examined groups of the lambs. Similar results were obtained by other authors (Klobasa et al., 1992, Klobasa and Werhahn, 1989).

Conclusion
It was found that supplementation of lambs after single birth with cow colostrum and cow colostrum filtrate with an increased IgG concentration in dry matter, obtained as a result of filtration, allowed an increase in immunoglobulin level in their blood serum, however a statistically significant relationship was only noted in case of higher immunoglobulins addition (14.18 g of IgG). This points an increase in their passive immunity level, and may be applied in order to enhance immunological status of lambs from multiple and other births. No significant effect of feeding with cow colostrum and its protein concentrate on growth rate of lambs was noticed. It was established that colostrum membrane microfiltration results in complete elimination of E. coli and Clostridium bacterial flora which makes that the additive in this form may be safe in feeding other animal species. No satisfactorily results in bacterial flora reduction were obtained in the case of fresh colostrum thermal treatment, as well as using focused microwave field or pulsed electromagnetic field.

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