Recent insights into the production of colostrum, the carry over of maternal immunity and effects on these by perinatal feeding

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Introduction

Genetic selection for the increase of litter size results in the decrease of the birth weight of piglets. In a large comparison of birth weight of piglets under practical circumstances (2011-2012) a decrease in birth weight was observed with increasing litter size. Offspring of gilts had lower birth weight compared to other parities which differed to a less extend(Figure 1). Each extra born piglet resulted in the lowering of the birth weight with 35 grams (Figure 2) and still born piglets were significant lower in their birth weight compared to live born piglets (1090 vs 1357 gram respectively)[1].



Figure 1 Effect of sow parity on piglets' birth weight. Differences between bars with different superscript are significant (at least <0.05) [1].



Figure 2 Effect of the increase of littersize on the decrease of piglets' birth weight. A decrease of 35 gram of birth weight for each extra born piglets is observed [1]

Piglets with lower birth weights are more vulnerable, especially in the first days of their lives. Piglets are born with the lowest energy reserve of all farm animals. The main energy source is glycogen, stored in the liver and in the muscles. The total of energy available is barely enough to survive for the first 12 hours of life, if the temperature off the surroundings of the piglet are of sufficient warmth. This low amount of energy makes piglets very dependent on a quick and adequate colostrum intake [2, 3]. Despite the importance of colostrum for the piglets, research in the field of colostrum production of sows is scarce. The reason for this is that methods to measure the production of colostrum from a sow are very labour intensive. The two accepted methods to measure colostrum production from sows and the uptake of colostrum of piglets are the deuterium oxide method and the 24hrs weight increase of piglets [4], from which the first method is not possible under practical circumstances. However, the weighing of piglets at birth and at 24 hours of weight can give a under estimation of the colostrum uptake [5]. The optimal colostrum intake for piglets is more than 250 grams [6, 7]. Piglets with an uptake of 160 grams or lower are prone to die the first day(s) of life [8]. The colostrum production of sows is 3,3 kg average but with a large variation of 1.6-5.6 kg [9]. Besides the effect of quantity of colostrum, also the quality is important. The IgG concentration decreases rapidly after birth with a half life time of about 6 hours [10]. The increase in litter size makes the equal distribution of colostrum over the siblings even more difficult. Recent research of the Ghent University focussed on the peripartal feeding strategy [11]. Sows were divided in two groups with either a low (1.5 kg) or a high (4.5 kg) feed intake during the last week of gestation. The groups of sows with a high feed intake had a 0.5 kg higher colostrum production compared to the low feed intake sows (4.0 vs 3.5 kg resp.). The low feed intake sows had a catabolic serum profile indicative for a negative energy

balance. The sows also varied in colostrum production based on their body condition. Skinny and fat sows produced less colostrum compared to the moderate sows. The results show the importance of a homogenous body condition of the sows within a herd, which can be challenging under practical circumstances. The group housing of sows can give feed intake competition among sows resulting in a heterogenic body condition distribution. Besides its negative effect on colostrum production, this is also negative for the speed of farrowing, resulting in an increase in the percentage of still born piglets [12]. Over muscled sows and fat sows are also an important risk factor of the postpartum dysgalactia syndrome [12].

Due to the difficulty in measuring colostrum production of sows and the uptake of colostrum of the piglets, it is hard to "measure" the colostrum management leaving the percentage of pre weaning mortality as one of the most practical indicators. Besides energy, colostrum contains also a lot of important nutrients like growth factors, vitamins, enzymes and last but not least, immunoglobulins. The sow's epitheliochorial placenta type does not transfer immunoglobulins to the piglets, which are solely dependent on colostrum as a source of maternal immunity. The immunoglobulins in colostrum decrease rapidly. Six hours after birth of the first piglet the IgG concentration already decreases with 50% pointing out the importance of a quick farrowing process and a quick access to the udder after birth. The measurement of IgG in piglets serum can be a way of assessing the colostrum management. Recently a new, cheap and reliable "immunocrit" assay came available to measure IgG in piglets serum [13] which was validated by our ForFarmers laboratory [14]. Briefly, by mixing 50 µl of serum with 50µl of a 40% ammonium sulphate solution, mainly the immunoglobulins will precipitate. The resulting emulsion is, after mixing using a vortex mixer, drawn into a haematocrit capillary and centrifuged for 10 minutes with 15,000g. After centrifugation the sedimented immunoglobulins can be measured as well as the total volume in the capillary called the immunocrit ratio and is expressed as the percentage of the total capillary content. This new method makes it possible to carry out larger investigations for the IgG uptake of new born piglets at acceptable costs.

Study1: Effect of birth order and sow feed on the intake of colostrum [15]

With the objective to increase the birth weight of piglets an experiment was conducted with increased amounts of amino acids in the sow feed during the last weeks of gestation and this was compared with a control diet. Piglets were weighed directly after birth. Piglets were weighed again 24 hours after birth of the first piglet to estimated the colostrum intake of the piglets [4]. A colostrum sample was taken after birth of the first piglet for the measurement of colostrum IgG concentration with radial immune diffusion (Triple J Farms). From in total 70 litters, three piglets per litter were bled between 24 hours and 4 days of age by jugular venepuncture: the first live born, the middle live born and the last live born piglet. Serum was measured for IgG concentration using the immunocrit assay as described above.

However birth weights were not increased by the high amino acid diet, an effect on the speed of farrowing and the subsequent quality of colostrum uptake was observed. Sows fed a higher amount of amino acids during the last weeks of gestation farrowed faster resulting in a higher serum IgG concentration of the piglets compared to the offspring of sows fed a control diet. Colostrum IgG concentration between the two treatment groups did not differ. Also effects were seen related to the birth order of piglets. First born piglets had higher IgG concentrations compared to last born piglets. Piglets who died during the suckling period were lighter compared to the piglets which survived (1.048 kg vs 1.306 kg) and also the colostrum intake (CI) differed in total, and as expressed as the weight differences in the first 24 hours of life (166 gram CI and 4,9% weight <u>decrease</u> for the lost piglets and a 281 CI and a 5,1% weight <u>increase</u> for the surviving piglets) (Figure 3).



Figure 3 Differences in colostrum intake (CI) during the first 24 hours of life between piglets which died before weaning (mortality, 166 gram CI; 4.9% body weight <u>decrease</u>) and piglets which survived until weaning (survived, 281 gram CI; 5.1% bodyweight <u>increase</u>) (p<0.001).

Study 2: Analysing sow colostrum for IgG using different methods [16]

During the analysis of the colostrum samples originated from study 1 awkward results were seen when the colostrum was measured for IgG using an Elisa (Bethyl). To measure IgG with an elisa, huge dilutions had to be made in the range of 1:1,000,000 resulting in dilution errors to be magnified. In bovine, radial immunodiffusion is considered as the golden standard to measure IgG while the use of refractometer or specific gravity meters give acceptable results estimating the quality of colostrum [17]. However, for swine no comparison of methods to measure colostrum IgG is available. To evaluate different methods of measuring colostrum for the IgG content, the colostrum samples of study 1 were reanalysed with different methods. The colostrum samples were stored at -32°C and were thawed in a warm water bath. To homogenize fat, colostrum samples were warmed up to 36° C for 30 minutes and cooled down to 20°C before analysis. Colostrum was analysed with a refractometer (VWR, 0-50% Brix), radial immunodiffusion (Triple J Farms Swine IgG) and with an Elisa (Bethyl Swine IgG). The simple refractometer technique was relative good correlated with IgG concentration in the colostrum with a R² of 0.63(Figure 4). The correlation with total protein was even higher with a R² of 0.90 (data not shown). The Elisa method was poor correlated with a R² of 0.27(Figure 5). The mean IgG concentration of colostrum at time of birth of the first piglet was 60.2 mg/ml and had a large variation between sows from a minimum of 32.3 mg/ml to a maximum of 70.1 mg/ml (Table 1).

In conclusion, the refractometer proved to be a robust method to measure IgG under field conditions. For research proposals however, RID is an accurate and relative simple method to measure IgG. When considering the use of Elisa tests, validation has to be done in advance to evaluate test performance and working procedures in the laboratory.



Figure 4 Comparison of Radial Immuno Diffusion (RID; IgG mg/ml) and refractometry (Brix%) of in total 70 colostrum samples. Linear regression resulted in a significant correlation with a R² of 0.63



Figure 5 Comparison of Radial Immuno Diffusion (IgG mg/ml) and Elisa (IgG mg/ml) of in total 70 colostrum samples. Linear regression resulted in a significant correlation, however with a poor R² of 0.27.

Colostrum composition	Potassium g/kg	Sodium g/kg	Crude Protein g/kg	Fat g/kg	Lactose g/kg	IgG (RID) mg/ml
Mean	1.34	0.76	173.8	58.7	24.3	60,2
Standard Deviation	0.117	0.118	24.1	16.2	3.97	8,56
Minimum	1.11	0.53	130.7	26.8	15.29	32.3
Maximum	1.58	1.23	225	102.9	33.85	77.1

Table 1 Colostrum composition of in total 70 sows. A colostrum sample was taken from the anterior teats directly after birth of the first

Study 3: Field observations of 9,000 new born piglets and their IgG concentration [18]

Based on the findings of study 1 and 2 a monitoring program was started in the autumn of 2013 called the Colostrum Score[®]. Farmers were given the opportunity to send in blood samples, taken by their own vet, to the ForFarmers laboratory. A protocol was supplied to the vet. Briefly, six litters were selected (cross fostered litters to be excluded). From each litter, six piglets were bled between the age of 24 hours and 5 days. Piglets were weighed at the time of bleeding and farmers were given the opportunity to weigh the piglets again after 7 days and at weaning. The farmers received a detailed report of their results and growth compared to the total of the database. In total from over 9,000 piglets, serum was analysed for IgG. The mean IgG was 37.6 mg/ml. For each litter the coefficient of variation (CV) was calculated. Mean CV was 26.9% and the best 20% of the Farmers had a CV <20%. Of in total 2271 piglets the growth was followed until weaning. The first 7 days after blood sampling the growth was 190 gram/day. Mean growth until weaning was 227 gram/day. The 20% low birth weight piglets (LWP; <1.1 kg) had an OR of 5.0 for having very low concentrations of IgG (<10 mg/ml) and a OR of 2.4 for having low concentrations of IgG (<20 mg/ml) compared to the 20% heavy birth weight piglets (HWP; >1.82 kg). LWP and HWP were different in their mean IgG concentration, but differences weren't large (35.5 and 36,5 mg/ml respectively; p<0.05). Offspring of gilts had lower IgG concentrations compared to multiparous sows (34.3 vs 38.0 mg/ml respectively; p<0.001). There was, only for gilts and 4th parity sows, a very weak relationship between the number of live born piglets and the IgG concentration of these piglets. There was no relationship with IgG concentrations of the piglets and the outside temperature. Piglets with low IgG concentrations (<20 mg/ml) had a 20 grams lower growth per day during the suckling period. Preliminary results of a recent trial showed lower IgG concentrations in piglets that died before weaning compared to the other piglets (21.5 vs 37.5 mg/ml respectively; p<0.001). In this recent trial the total offspring of 70 sows were analysed for serum IgG. The immunocrit method proved to be a practical way in assessing colostrum management on sow farms.



Figure 6 The 20% Light weight piglets (LWP;<1.1 kg; n = 1728) had a significant OR for having very low (<10 mg/ml; OR 5.0) and low IgG concentrations (<20 mg/ml; OR 2.4) compared to heavy weight piglets (HWP;>1.82 kg; n = 1728) p<0.0001



Figure 7 IgG concentration in the first days of life and the growth until weaning. Piglets having an IgG of <20 mg/ml grow approximately 20 gram less compared to the other piglets.

Discussion

To increase the productivity of sow farms the number of live born piglets is an important parameter. The genetic selection of the number of live born piglets can in this way contribute towards a higher productivity. However, selection on the increase of litter size is also confounded by the decrease of the birth weight of piglets. The birth weight of piglets is an important parameter for survivability. So the increase in litter size also implicates an increase in mortality if management practises are not improved. Farmers who make a shift towards sow lines producing large litters, without changing their management practices, can be disappointed in the end result. Also the increase of sow farms in West Europe, from the more traditional family farm of 400 sows towards larger farms working with personnel, brings other needs to the working floor. With personnel available during the normal working hours, leaving often the farm unattended at night time, the induction of parturition can improve the care given to neonatal piglets as they will be born when personnel is available. However, a reliable insemination date is needed and too early induction of farrowing will result in even more light birth weight piglets.

These changes in sow productivity and the increase of herd sizes resulting in working with personnel pleads for a professional management of farms. Whereas at the traditional family farm everybody is aware of the work needed to be done, in larger farms this should be managed by clear protocols and procedures. To improve productivity, there is a need for good and reliable tools to measure management practices. A good assurance of colostrum uptake by piglets is crucial in reducing pre weaning mortality. Measuring colostrum uptake by piglets by weighing the piglets twice, directly after birth and 24 hours later, are very time consuming and labour is scarce at farms. The formula to estimate the colostrum uptake of piglets [4] is underestimating the colostrum uptake of piglets [5]. This was also clear in our dataset where a part of the piglets had a negative colostrum uptake. It also can be doubted what the extra information like time of first udder contact, adds to a reliable prediction of colostrum intake. Our data showed that the relative increase in bodyweight the first 24 hours of life is a good alternative, taking also the differences of birth weight into account. There was a great difference in weight development between the piglets which died and which survived until weaning. The new immunocrit method is suitable to measure colostrum management off sow farms and well correlated with pre weaning mortality. The data presented in this paper demonstrates clear benchmarks of the IgG concentrations of piglets under practical circumstances. Also the success of the Colostrum Score® indicates the need for a simple but effective measuring tool for the colostrum management. The risk group for poor colostrum uptake are clearly light weight birth piglets (<1100 gram) and offspring of gilts. This data can help in convincing farmers to improve the colostrum management and to evaluate management practices and protocol compliance of personnel of sow farms.

Besides the focus on the management practices in the farrowing house, also the shift towards group housing of sows gives new challenges. When sows are group housed but can't be individually fed (when electronic feeding stations are lacking) problems can arise with the distribution of the body condition. Skinny as well as fat or hyper muscular sows, produce less colostrum compared to sows with a moderate body condition [11]. Sows with a body building syndrome are also prone for dysgalactia after farrowing [12] and for an increased percentage of still birth [19]. The prevention of a negative energy balance around farrowing is important for an optimal colostrum production as is pointed out by a study of the Ghent University [11]. Although in this study a significant increase in ketones were found at sows with a low feed intake, these were so low that these can't be measured using standard laboratory methods. Other studies stated that sows, in contradiction to for instance bovine, hardly develop ketosis [20] which results to the conclusion that blood analysis using ketone bodies is not the right method of choice. However, the scoring of the body condition by an experienced person is relative easy

to do and does give useful information to improve sow productivity. Also the weighing of sows at the entry of the farrowing house, and after weaning, can give a lot of data about the status of the distribution of the body condition within a sow herd and the nutritional status during gestation and lactation. Besides the body condition also the composition of the feed is important. Obstipation/constipation around farrowing is to be avoided since this is impairing the homeorhesis from gestation towards lactation [12]. Obstipation can be prevented by the application of enough fibres in the feed during gestation and lactation. A transitional diet during the last week of gestation and the first days of lactation can be used to meet the nutritional needs of the sow during the physiological radically changes within this time frame. However, the implementation of a specific transitional diet is depending on technical facilities for automatic feeding of an extra diet or depending on the availability of labour for manual feeding.

Nutritional studies with the scope of improving colostrum production are performed by only a few research groups. The main reason for this is the tremendous amount of labour which is needed to estimate the colostrum uptake of piglets. However, when the total colostrum production of the sow is to be measured, the weighing of each individual piglet still remains the golden standard. When measuring IgG as a quality parameter of colostrum, RID is the most reliable way and to our opinion the golden standard. The different RID plates showed consistent and uniform results using the reference sera's which were applied at each used plate (data not shown). The differing results by the use of different methods implies that, when is looked at the IgG concentration of porcine colostrum, the method of determination has to be taken into account and associated benchmarks should be used. Also the rapid decrease in colostrum protein content by the lowering of immunoglobulins, stresses out the importance of a uniform colostrum sampling protocol.

Recent scientific publications related to colostrum production of sows show that there is increasing interest in this field of research. The availability of new techniques like the immunocrit method will lead to further insights in the dynamics around colostrum production and uptake by piglets. This knowledge can be translated in best practices around farrowing, to reduce pre weaning mortality and in this way give further contributions to a sustainable and profitable pig industry.

Conclusion

The increase in litter size the recent years results in lighter birth weight piglets. Piglets with a low birth weight are at a high risk to die before weaning. To reduce the risk of pre weaning mortality colostrum uptake plays a pivotal role. To increase colostrum production of the sow, a negative energy balance at the end of gestation should be prevented. A homogenous distribution of the sows body condition is also important for maximizing colostrum production and piglet vitality. However, this can be challenging with group housing systems without the possibility of individual feeding of the sows. From the piglets perspective, sufficient colostrum uptake is needed, both in quantity as in quality. Piglets under the weight of 1100 grams are extra vulnerable, as well as the offspring of gilts. Management practices at the farm should focus on a quick birth process and a fast access to the udder by piglets and is correlated with pre weaning mortality and piglets' performance. The presented data gives good West European benchmarks for checking a sow farms' colostrum management practices and can help in improving productivity.

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