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FIBRINOGEN CONCENTRATION IN BLOOD PLASMA OF NEWBORN PIGLETS

Plasma fibrinogen levels may be useful in the assessment of newborn piglet status, It is important, however, to know the based on age levels of this protein. The aim of this study was to compare levels of plasma fibrinogen in healthy newborn piglet and piglet with low viability score and natural occurring sepsis.

Keywords: *newborn piglets, fibrinogen, acute phase protein, sepsis, DIC*

Introduction

It is well known that synthesis and catabolism of plasma proteins are affected by multiple factors. Serum levels of certain liversynthesized proteins changes by at least 25% in response to injury, infection, inflammation, stress or diet change. These are so called acute phase proteins (APPS). Changes in APPS can be used to monitor a systemic response to a physiological insult. The most widely used indicator for the presence of inflammation in medical practice is CRP. It is because of its ability to change rapidly with changing conditions. In Poland, CRP is not generally tested in swine practice because of limited availability and high costs.

Inflammation and coagulation are triggered by a number of different mediators. These multiple factors promote amplification of the coagulation responses when a large or persistent stimulus is present. Some proteins of the coagulation cascade whose concentration in serum varies in response to disease include alpha antitrypsin, antithrombin III, Protein C, and fibrinogen. Fibrinogen, while of primary importance as a coagulation protein, is also an acute-phase protein reactant [6, 10]. Its level rises slowly in response to the inflammatory stimulus. Increased fibrinogen levels may also be seen in non-inflammatory processes e.g. with pregnancy and in women taking oral contraceptives. Despite this, measurement of fibrinogen levels may be useful in screening for infectious diseases and in monitoring disease activity [9]. According to data presented by Shalm, in healthy pigs 2-3 months of age, plasma fibrinogen concentration ranges from 2,0 to 4,0g/l [7]. It has been demonstrated that concentration of fibrinogen greater than 10g/l in adult pig indicates inflammation.

The aim of this study was to compare levels of plasma fibrinogen in healthy piglet and piglet with low viability score and natural occurring sepsis.

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Materials and methods

34 newborn piglets from 4 PenArLan sows were included in the study. The animals were examined on the breeding farm near Lublin (Poland). The pigs were divided into 3 groups. The first group (K) – consisted of 6 piglets from one littermate. The sow in group K was inseminated using semen collected from boar X. Viability rate in this group was ≥ 8. The other two groups: A and B consisted respectively of 13 and 15 piglets with viability rate ≤ 6, from 4 littermates. Sows were inseminated using semen collected from boar Y.

Viability score of each piglets was assigned according to the method described by Randall [8] (Table 1).

Table 1.

Viability score (from Zaleski, 1993)

Score	0	1	2
Heart rate	Absent	<120/min	> 120/min
Onset of respiration	Absent	After 15 s	Before 15 s
Muscle tone	Flaccid	Poor	Good
Color	Pale	Cyanotic	Pink
Attempts to stand	None in 5 min	1 to 5 min	Before 1 min

Blood samples were obtained from piglet in group A and K at day 1, approximately 30 min. after birth and from piglet in group B on the day 2 (24hours after birth). Three ml of blood was collected from each pig by jugular venipuncture. One ml of blood sample was carefully discharged in a 1 ml test tube containing EDTA, while the remaining blood sample were carefully discharged in another test-tube containing 0.15 ml 3.8% trisodium citrate solution.

The samples collected from the piglets were utilized to analyze morphology of blood cells and for the estimation of fibrinogen.

Basic hematological parameters (RBC, WBC and PLT count, hematocrit, hemoglobin concentration and MCV, MCH, MCHC) were measured by a Exigo (Boule Medical AB) analyzer. EDTA anticoagulated venous whole blood was also used to prepare a blood film stained with MGG. Plasma fibrinogen concentration was determined in duplicate by a coagulation assay (Bio-Ksel, Toruc, Polska).

The Statistically significant difference between the mean values in the groups was evaluated by students ‘t’ test. The difference in mean values of two groups was regarded as statistically significant if the P value was less than 0.05 and it was taken as highly significant if P value was less than 0.01

Results.

Viability scores were moderate-to-severe approximately 3 units lower in the piglets from group A and B than in control piglets (K – 8,9, A – 5,7 B – 6,5). The piglets from A and B group were weak, cyanotic or pale, exhibit dyspnoea and prolonged bleeding from the umbilical cord. The piglet mean mortality rate in litters from group A

and B has been very high. It was 60% in A group and near 90% in B. There was no death in the control (K) group observed. In 2 blood samples from group A and 10 from group B schistocytes were found in blood smears. In 7 cases presence of bacteria was demonstrated by direct examination of plain peripherical blood smears.

Table 2. shows comparison of hematological parameters of piglet. Mean values almost all of them were significantly different between all three groups except the WBC and platelets count. Piglet from groups A and B were anemic. Mean values of hemoglobin concentration, hematocrit and RBC count were significantly lower in groups A and B than in K group. Parameters listed above were lowest in piglet from B group.

Table 2.

Hematological parameters of examined piglets.

		WBC [10^9/l]	RBC [10^{12}/l]	HCT [%]	HGB [g/dl]	MCV [fl]	MCH [pg]	MCHC [g/dl]	PLT [10^9/l]
Group K	mean	11,78	4,63[1,2,3]	28,18[1,2,3]	9,86[1,2,3]	60,8[1]	21,34[2]	35,08[1]	196,60
	SD	4,86	0,34	2,28	0,74	2,24	1,19	1,17	56,55
	max	18,60	4,95	30,70	11,00	64,20	23,00	36,40	286,00
	min	5,70	4,21	25,30	9,20	58,00	20,10	33,30	152,00
Group A	mean	8,35	3,82[1,2,3]	23,35[1,2,3]	7,67[1,2,3]	61,90[2]	20,34	32,88[3]	222,77
	SD	4,91	0,48	2,33	0,91	4,43	1,43	1,39	87,06
	max	20,10	4,39	25,70	8,80	70,90	22,40	35,20	378,00
	min	3,40	2,90	17,80	5,70	53,70	17,70	29,90	83,00
GroupB	mean	7,61	2,80[1,2,3]	15,63[1,2,3]	5,47[1,2,3]	55,1[1,2]	18,41[3]	35,47[3]	202,20
	SD	3,00	0,93	5,71	1,85	3,12	4,78	1,66	63,81
	max	13,50	4,69	27,50	9,20	60,20	21,50	38,80	311,00
	min	1,70	1,35	6,90	2,60	50,90	1,50	33,40	77,00

1 – significantly important differences between group A and K ($p \leq 0,05$)

2 – significantly important differences between group B and K ($p \leq 0,05$)

3 – significantly important differences between group B and A ($p \leq 0,05$)

The levels of fibrinogen of 6 piglets from K group were extremely low (Table 3). Values of this parameter were highest in group A. In group B fibrinogen levels were found to be within the expected range of normal adult pigs.

Table 3.

Fibrinogen concentration in blood plasma of examined piglets.

	group K				**group A**				**group B**			
	mean	SD	max	min	mean	SD	max	min	mean	SD	max	min
Fibrinogen [g/l]	0,64	0,18	0,92	0,47	4,01	2,15	6,88	0,39	1,64	1,11	3,80	0,48

Discussion

The hemostatic system in the pig is thought to be very similar to that of human beings [1]. Compared to adult, the values of nearly all factors involved in the hemostatic pathway are deficient at birth [1, 2, 3]. Unfortunately, to our knowledge, there are only very few literature data available concerning the fibrinogen concentration in newborn pigs plasma [2, 3]. The results of our study suggest that concentration of swine plasma protein appears to be age dependent and normal neonatal ranges for fibrinogen levels are different from those for older individuals. Our results are in agreement with the data presented by Petroianu [3]. Concentration of this protein in fetal individuals at 110 day of gestation and in newborn piglet of Goettingen race could be even 10 fold lover than in juvenile pigs. Fibrinogen levels increased with age, probably because the prevalence and severity of illness and its associated inflammatory state increased with age. The age based levels of coagulation factors and the age based normal values for coagulation tests, result in a major challenge to veterinarians [5].

In the light of these data the concentration of the fibrinogen in group A and even in group B was considered to be very high. The reason for such high levels is not clear. However, one of them may be intrauterine infection. Early onset sepsis syndrome is associated with acquisition of microorganisms from the mother. Transplacental infection or an ascending infection from the cervix may be caused by organisms that colonize in the sow genitourinary tract [4]. In our study bacteria were demonstrated by direct examination of plain peripheral blood smears in 7 cases. Almost all newborns developed symptoms and signs of sepsis. During acute-phase conditions, such as sepsis the fibrinogen level rise slowly within 24 hours. At list 17 piglet probably developed disseminated intravascular coagulation (DIC). The presence of shistocytes supports such diagnosis but it was not proven in further investigation. An elevated fibrinogen level among septic patients could lead to a misdiagnosis. As an acute phase reactant, fibrinogen might not decrease until DIC is severe. In our study the fibrinogen concentration in group B was high, but significantly lower than in A group. However fibrinogen is consumed in the process of intravascular coagulation, nevertheless, the concentration of this protein remains still high. Therefore the age based levels of fibrinogen are very important for proper diagnosis.

Conclusion

1. The concentration of the fibrinogen in newborn normal pigs at the first day of life is very low.
2. Our results suggested that plasma fibrinogen levels may be useful in the assessment of newborn piglet status, but it is important to know the age based levels of this protein.

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Summary

Inflammation and coagulation are triggered by a number of different mediators. Fibrinogen, while of primary importance as a coagulation protein, is also an acute-phase protein reactant. According to data presented by Shalm, in healthy pigs 2-3 months of age, plasma fibrinogen concentration ranges from 2,0 to 4,0g/l. Concentration of this protein in fetal individuals at 110 day of gestation and in newborn piglets of Goettingen race could be even 10 fold lower than in juvenile pigs. During acute-phase conditions, such as sepsis, the fibrinogen level rise slowly within 24 hours. The aim of this study was to compare levels of plasma fibrinogen in healthy piglet and piglet with low viability score and natural occurring sepsis. Our results are in agreement with the data presented erlier by Petroianu concerning basal levels of fibrinogen in Goettingen race piglets and suggested that plasma fibrinogen levels may be useful in the assessment of newborn piglet status, but it is important to know the based on age levels of this protein.