

EARLY PREGNANCY DIAGNOSIS OF COW WITH PSP – B LEVELS IDENTIFICATION

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ABSTRACT

Delay information of pregnancy will less efficient reproductive performance of cow, so it is needed an earlier pregnancy detect method. The presence of Pregnancy Specific Protein B (PSP – B) in blood of cow since early pregnant, can be used for it. Aim of research to obtain data of PSP – B levels in Ongole Crossing cow. Research done for 180 days at research stall of Beef Cattle Research Station and for 30 days at Veterinary Medicine Faculty in Airlangga University; using 20 heads cow who are not pregnant and normal its follicle development, also 2 heads bulls. Observed 24 hours/day during 120 days for mating events, every cow who mated is collected its blood sample on 2th, 4th, 8th, 16th and 30th days after mated. Analysis blood serum using ELISA method and only carried out from pregnant cow. Parameters : levels of PSP – B and progesterone in a few days pregnant. The data presented descriptively. The results showed : PSP – B and progesterone levels at pregnant age : 2 days = 1.25 and 0.54 ng/ml; 4 days = 2.32 and 0.60 ng/ml; 8 days = 4.84 and 1.44 ng/ml; 16 days = 9.48 and 1.76 ng/ml; also 30 days = 8.24 and 2.12 ng/ml. Conclusion: identification of early pregnancy for local beef cows, through PSP – B levels in blood, can be performed on 2th day after occurrence of fertilization.

Keywords : Ongole Crossing cow, PSP – B, early pregnant diagnosis.

ABSTRAK

Informasi terjadinya kebuntingan yang terlambat akan menurunkan efisiensi performans reproduksi sapi induk, sehingga diperlukan adanya metode untuk mendeteksi kebuntingan secara dini. Keberadaan Protein B Spesifik Kebuntingan (PSP – B) di darah sapi induk sejak awal umur kebuntingan, dapat digunakan untuk mengidentifikasi kebuntingan dini tersebut. Penelitian ini bertujuan mendapatkan data kadar PSP – B pada sapi Peranakan Ongole (PO) induk. Penelitian berlangsung selama 180 hari di kandang percobaan Loka Penelitian Sapi Potong dan selama 30 hari di Fakultas Kedokteran Hewan Universitas Airlangga, menggunakan 20 sapi PO induk yang tidak bunting dan perkembangan folikelnya normal, serta dua sapi PO pejantan. Dilakukan pengamatan terjadinya perkawinan selama 120 hari; setiap hari selama 24 jam; kemudian setiap sapi induk yang dikawini di ambil sampel darahnya pada hari ke 2, 4, 8, 16 dan 30 dari perkawinannya. Analisis sarum darah menggunakan metode ELISA dan hanya dilakukan terhadap sapi yang terbukti bunting. Parameter yang diamati : kadar PSP – B dan hormon progesteron pada beberapa hari umur kebuntingan. Data disajikan secara deskriptif. Hasil penelitian menunjukkan : kadar PSP – B dan progesteron pada umur kebuntingan : 2 hari = 1,25 dan 0,54 ng/ml; 4 hari = 2,32 dan 0,60 ng/ml; 8 hari = 4,84 dan 1,44 ng/ml; 16 hari = 9,48 dan 1,76 ng/ml; serta 30 hari = 8,24 dan 2,12 ng/ml. Disimpulkan : identifikasi kebuntingan secara dini pada induk sapi potong lokal, melalui kadar PSP – B darah, dapat mulai dilakukan pada hari kedua setelah terjadinya kebuntingan.

Kata kunci : PO induk; PSP – B, diagnosis kebuntingan lebih dini.

INTRODUCTION

A cow who did not have reproductive process can still to normal life, but it have less/not efficiently maintained. Efforts to improve to reproductive performance cow will be very important part in the effort to increase population of cattle, if followed by ability to detect early pregnancy.

Delay aware of a pregnancy at a cow, will become a negative impact on intake of nutrient deficiency and pregnancy safety threat that already exists, so that is one factor that can cause of beef cattle reproductive performance cow becomes less efficient (Lamaster *et al.*, 2001). Therefore, ability to be able to identify the earlier occurrence of pregnancy in a cow, is expected to a positive impact avoidance of feed nutrients intake that do not enough to physiological needs of pregnant cows, as well as avoiding a young pregnant

cows to be mated again. The aim of this research was to obtain baseline data about presence (levels) of specific protein B in early pregnant cows.

Pregnancy early detection in cattle can be done with identification of substances that are formed since beginning of pregnancy (known as early pregnancy factor (EPF), which it detect specific substances contained in blood and milk as "Bovine Pregnancy Associated Glycoprotein" or bPAG (Hafez (2000), also through identification of non specific substances in blood, urine or milk during pregnancy such as progesterone, estrone sulfate (Samik and Safitri, 2010). Aulanni'am , *et al.* (2004) states, a specific antigen that produced from insulating EPF and classified as a specific substance of bPAG , it was Protein B or often called Pregnancy Specific Protein B (PSP-B)

Protein B is one of the specific substance in form of a glycoprotein, it is resulted and secreted into blood by binukleat tropoblast cells of placenta stem (Green *et al.*, 2000), is secreted into blood began less than one day after fertilization, its level increases in next few days, then to decline and disappear after 21 to 30 days from fertilization, so its presence/levels in bovine blood has been used as a means of identifying early pregnancy (DuPlants, 2000) .

RESEARCH METHODS

This research was conducted for 180 days at an experimental stall of Beef Cattle Research Station (BCReS) and for 30 days at Faculty of Veterinary Medicine, Airlangga University. The research is began with prepare 20 heads of Ongole Crossing (OC) cow which its rectal palpation results indicate that not pregnant and normal its follicle development, also with select 2 heads of OC bull who have been accustomed to natural mating. Livestock are maintained in two stalls with comparison of cows : bull is 10 : 1 (Figure 1).

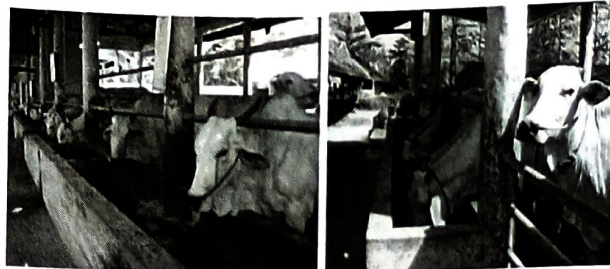


Figure 1 . Cattles material and its stall of this research

Research in experimental stall of BCReS consists of 120 days for mating cows and blood samples collecting, also 60 days for get enough of old pregnant to can be rectal palpation, in order to obtain certainty and old pregnant cows.

Blood sampling technique is every cows that have been known of its time and date mating (resulted of observation during 24 hours/day for 120 days), blood samples were taken immediately on the first day (as a control) and then day 2th, 4th, 8th, 16th and 30th after mating ; the section in jugular vein ; as much as 8 ml ; and using tubes without anti-coagulant. Samples were immediately stored at temperatures 5^o C for about 60 minutes, then centrifuge with 4000 rpm speed for 15 minutes. Serum is formed in upper layer are transferred to evendoff and stored in freezer for waiting to be analyzed (Figure 2). Blood serum samples that will be analysis of its protein B and progesterone levels at the end of the study, are only from cows that found positive pregnant.

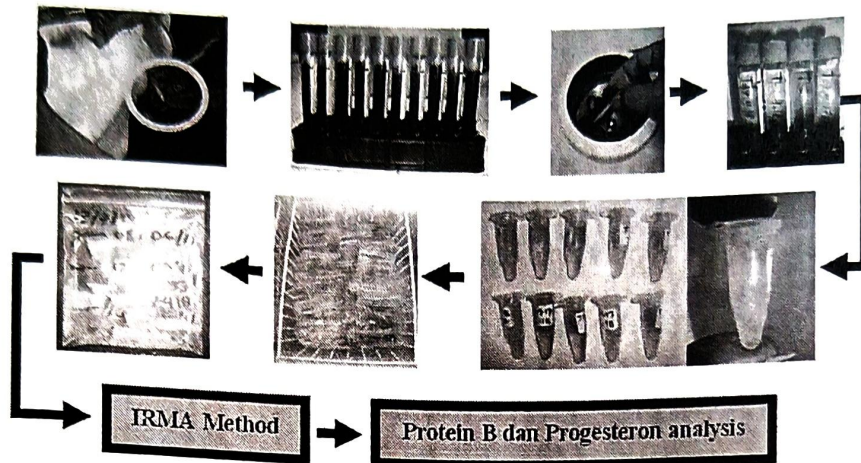


Figure 2. Blood collecting and process of making blood serum samples

Analysis of protein level using kit B CUSABIO from China and progesterone level using kit GB corp from Taiwan. Analysis of hormone progesterone is needed to ensure that cattles who analysis her protein B level is positive and has remained pregnant.

During this study, cows were given ration that containing crude protein 9 – 10 %, crude fiber \leq 22 % and TDN 55 – 60 %, ad libitum control giving (dry matter more than 3% of its body weight cattle). Observation of ration nutrient consumption is done during 2x24 hours/every month.

Parameters were observed :

- a. time and body condition score (BCS) cows at mating
- b. protein B and progesterone levels in some pregnant old
- c. ration nutrients consumption .

Analysis of data : the data obtained are presented descriptively

RESULTS AND DISCUSSION

Time and body condition score cows at mating

Data of time and BCS of cows at mating that produced pregnancy, in Table 1. The data shows that 19 of 20 cows had been mated and successful pregnant. To be suspected, it related with BCS of cattle that supported estrous cycles is normal and fertile. BCS of not pregnant cow who is too low (according to Barry, *et al.* (2007) under 5.5), tends to cause abnormalities of ovulation so cows are not able to show signs of her estrus Putro, 2007). Kunkle and Sand (2003) provide guidance, cows who will be mated, expected to have BCS between 5.5 to 6.5 (on a scale of 1 to 9).

Protein B and progesterone levels in some pregnant old

The data of analysis results for protein B and progesterone levels in some pregnant old, showed in Table 2 and in Figure 3. The data in Table 2 or Figure 3 shows that :

- a. at 2 days pregnant old, protein B presence/levels can already be detected. Protein B levels appear higher and peaked at about 16 days of pregnant old, and then tended to decline in about 30 days pregnant old. This results are identical to statement of DuPlants (2000), that is protein B has been produced by zygote begin no more than 24 hours since fertilization, its levels will continue to rise in next few days, than will decreases and disappears after 21 to 30 days of fertilization. DuPlants not mention how much protein levels in several early mating old in cattle, while resulted of this study can inform it.

Table 1. Time and BCS cows at mating

Cows number	Time and BCS cows at mating			
	dates	Time	Body weight (kg)	BCS (1 – 9)
01	17 april	12.47	379	6,0
02	4 april	14.00 and 20.15	374	5,5
03	28 april	08.15 and 10.00	379	5,5
04	07 april	05.45	376	6,0
05	06 mei	9.20	380	6,0
06	14 april	13.00 and 14.40	383	6,5
07	02 april	19.15 and 23.20	380	6,5
08	28 april	14.50 and 16.35	381	6,0
09	24 april	18.30	384	5,5
10	16 mei	07.35	385	6,5
11	13 april	16.30	382	5,5
12	07 mei	08.30 and 10.40	384	6,0
13	12 mei	6.30	385	6,0
14	13 april	10.30 and 11.00	381	5,5
15	11 mei	23.05 and 24.45	383	6,0
16	06 april	11.30 and 12.45	379	6,0
17	12 mei	16.30	383	6,0
18	23 april	18.30	382	6,0
19	09 mei	20.10 and 22.20	384	5,5
20	--	Never mating	382	5,5

Table 2. Protein B and progesterone levels in some pregnant old (ng/ml)

Hormon	Pregnant old (days)				
	2	4	8	16	30
Protein B	1.25 ± 0.75	2.32 ± 1.21	4.84 ± 3.67	9.48 ± 7.45	8.24 ± 7.64
Progesterone	0.54 ± 0.47	0.60 ± 0.43	1.44 ± 0.64	1.76 ± 0.56	2.12 ± 0.69

b . based on protein B content in above, effort toward identification of early pregnancy in cattle by identifying its presence/level of protein B, can be performed after the 2th days until 30th days from mating.

c. progesterone hormone levels continue to rise slowly appear until about 30 days pregnant. Increased levels of progesterone is one indicator, that cows that are analyzed its content protein B was experiencing pregnancy. Hawsky (2008) explained, progesterone is produced by *corpus luteum* (CL), so that cow is pregnant after ovulation, its CL will produce progesterone with higher and higher levels to suppress *hypothalamus*, and *anterior pituitary* inhibits release of LH and FSH (Leslie, 2003) ; prevent subsequent ovulation by inhibiting development and maturation of *follicles* (Britt, 2008), also prepare *uterine endometrium* for implantation and care of early pregnancy (Atkins *et al.*, 2010).



Gambar 3. Protein B and progesterone level in some pregnant old

Ration nutrients consumption

Data of ration nutrients consumption observed, are listed in Table 3. Appears that giving dry matter ration more than 3% of body weight was not able to entirely consumed by cattle. However, when seen BCS cow, ration nutrients intake is enough to cows, so it does not interfere with pregnancy.

Table 3 . Average of ration nutrients consumption of cow

observation	Ration nutrients consumption (kg/head/day)		
	Dry matter	PK	TDN
1 th month	10.76 ± 2.11	1.04 ± 0.20	6.03 ± 1.08
2 th month	10.91 ± 1.47	1.04 ± 0.18	6.00 ± 0.77
3 th month	11.26 ± 1.39	1.10 ± 0.11	6.42 ± 0.69

CONCLUSION

Identification of early pregnancy on local cattle through presence protein B levels in blood cattle, can be performed on the second day after fertilization and formed zygote/embryo.

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