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2	Screening the Reproductive Tract of Dairy Cattle for Pathogenic Micros	2019
3	Human Chorionic Gonadotropin (hCG) from Urine of Pregnant Women to Manipulate in vivo Ovulation and Pregnancy of Madura Cows	2019
4	Anti Early Embryonic Protein (EEP) for Pregnancy Test by Microtiter Strip in Dairy Cows	2019
5	The Effect of Feeding High Level of Protein on Reproductive Performance of Bali Starling.	2019
6	Antisperm Antibody in Repeat Breeder Friesian Holstein Cows at KPSP Setia Kawan Nongkojajar, Tutur District, Pasuruan, Indonesia.	2019
7	Diagnosis of Single and Twin Pregnancy, and Early Embryo Mortality Through Progesterone Level Test on Local Does.	2019
8	Improvement of Pregnancy Rate in Bali Cows with the Combination of Equine Chorionic Gonadotropine (eCG) from Local Pregnant Mare with PGF2α.	2019
9	Progesterone Profile of Dairy Cows which Experienced the Failure of Pregnancy to Artifical Insemination (AI).	2019
10	Effect of Heat Shock Protein (HSP) in Post Thaw Baluran Bull Semen	2018
11	Potency of Mycotoxin Binders on MDA Level, Expressions of Caspase 9 and Caspase 3 in The Uterus of Mice Exposed to Zearalenone	2017

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12	Polymorphism of Growth Hormone Gene in The Artificial Insemination Result of Madura Cattle with Limousin Semen as a Reference for Genetic Selection	2018
13	Implementation of fotogrametry techniques as body mass estimation of indo-pacific bottle nose dolphin (Tursiops aduncus) in bali dolphin lodge	2020
14	Uji Sensitivitas Kebuntingan Sapi Perah Menggunakan Pregnancy Specific Protein B (PSPB) Microtiter Strip dan Progesteron sebagai Gold Standard	2007
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16	Efek Pemberian L-Arginin Terhadap Gambaran Histologi Jumlah Spermatosit Primer pada Mencit (Mus musculus) Setelah Terpapar Suhu Panas	2019
17	Anti Prolactine Overcomes Heat Stress on Laying Hen.	2008
18	Unnatural Forced Moulting in The Laying Hen as Cause of Zoonosis from Salmonella Enteritidis	2009
19	Case Study: Dystocia on Beef Cattle in Kunir Regency of Lumajang District, East Java, Indonesia in 2015 and 2016	2017
20	Teratogenic Effect of Congenital Toxoplasmosis in Chicken Embryo	2017

Adapun penelitian tersebut layak dilakukan, meskipun belum ada ***Ethical Clearence*** karena menggunakan hewan coba yang minimal dan menghasilkan output yang sangat baik.

Demikian surat keterangan ini kami buat untuk dapat dipergunakan sebagai persyaratan pengususlan Jabatan Fungsional **Guru Besar**

Surabaya, 3 April 2023
Wakil Dekan III,

Prof. Dr. Mustofa Helmi Effendi, drh., DTAPH
NIP 196201151988031002

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inflammatory response and immune response, as well as, reduces apoptosis further. RBM-MSC is also accomplishes the stimulation of VEGF, HGF, G-SCF, which play a role in the healing process of injury (Martin, 2005).

Summary

Based on the results of this study, the impact of RBM-MSC therapy on suppressing the inflammatory response, necrosis and apoptosis effects of induction of carbon black on the placenta at a dose of $1x10^6$ cells / 0.1 ml has not been seen so that RBM-MSC therapy needs to be done before exposure (as preventive therapy) and takes more than 14 days after administration of therapy to get optimal results.

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Indian Vet. J., December 2019, 96 (12) : 27 - 30

Progesterone Profile of Dairy Cows which Experienced the Failure of Pregnancy to Artificial Insemination (AI)

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Abstract

The aim of study was to know the profile of progesterone in milk samples of dairy cows that experienced after pregnancy failure AI. Milk samples of fifteen AI failure dairy cows were taken on days 0, 7, 14, 21 and 28 (day 0 = estrus). Progesterone analysis was performed by ELISA method. On day 7^{th} and 14^{th}progesterone concentrations has increased in all cows, on 21^{th} day the levels were decreased in 3 cows and they returned to estrous, while in 12 cows the progesterone levels were high.

Key words : Dairy cow, Progesterone, Estrous cycle, Artificial insemination.

AI Failure can be an economically important problem on dairy farms (Canu *et al*., 2010), due to increased costs for mating, long calving intervals, rejects of cow and fewer birth of calves per year (Rustamadji *et al*., 2007).The reproductive disorder is caused by imbalance of progesterone and estrogen. The concentration of progesterone in the blood and milk can be determined to assess the animals infertile, estrous

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Table I. Milk Progesterone Concentrations in Dairy Cows on days 0, 7, 14, 21, 28of estrous Cycle

	Progesterone Concentration (ng/mL)				
No. of Cows	**D 0**	**D 7**	**D 14**	**D 21**	**D 28**
1	2.923	4.718	7.265	3.023	5.326*
2	3.122	5.898	7.138	3.028	4.102*
3	2.855	4.307	8.12	15.81	16.521**
4	3.148	5.551	8.879	15.341	11.815***
5	2.453	4.301	7.622	2.677	4.237*
6	3.202	5.379	9.381	16.024	13.181***
7	3.019	5.417	8.992	15.821	12.193***
8	2.971	5.209	9.095	15.722	11.892***
9	3.305	5.281	9.198	15.38	11.913***
10	3.199	5.362	8.683	15.653	13.00***
11	3.258	4.583	8.216	15.483	17.028**
12	3.348	5.189	8.86	16.256	17.803**
13	3.325	5.382	8.897	15.89	12.574***
14	3.099	5.324	8.989	15.206	13.011***
15	2.989	5.541	9.079	15.153	12.796***

Note:
Cow * = Failure of fertilization(on 21thand 28thday the levels progesterone were decreased)
Cow ** = Pregnant(on 21th and 28th day the levels progesterone were still increased)
Cow *** = Early Embryonic Death(on 21th the levels progester one were increased, but on 28th day were declined)

and pregnant state, so that it can be used for estrous detection and other pathological conditions (Samik and Safitri, 2019).

Materials and Methods

Milk samples were taken on days 0, 7, 14, 21 and 28 (day 0 = estrous) in the morning from the 15 AI failure cows aged 3.5 to 5.5 years with the normal breeding cycle. The samples were kept into the ice box at 4^0C, and transferred into the freezer. Before analyze the samples were thawed to room temperature. Furthermore 6 mL of each sample was taken and centrifuged at 3500 rpm for 15 minutes to separate fat and skim milk. One ml of skim milk was transferred into microtube. Hormonal assay were performed using the sandwich ELISA method. The data obtained were analyzed for ANOVA using Statistical Package Progams Social Sciences Software (SPSS).

Results and Discussion

Milk progesterone hormone profile determined on day 0 (estrous), 7, 14, 21 and 28 in 15 dairy cattle aged 3.5- 5.5 year with the failure of AI is shown in Table I.

The progesterone hormone concentration on the day 0 (estrous) of 15 dairy cows were consistent (2.2 - 3.5 ng/mL) the results of some researchers suggested that the animals were in estrous (Hoffman *et al*., 1983).

The hormone concentration on the day 7 of 15 dairy cow explained earlier observation that during the cycle, progesterone concentrations can be detected on the third day (Valdez *et al*., 2005) which increases until day eight of estrous and continue to increase until 21st day of pregnancy cycle (McDonald, 2000). Which concurs the findings of Frestantie (2017) that, the formation of the corpus luteum has occurred after ovulation, so the hormone progesterone secretion begins.

The high hormone concentration on the day 14 of 15 cow explain the luteal phase, the hormone progesterone inhibit gonadotropin hormone secretion, that is Folicle Stimulating Hormone (FSH) and Luteinizing Hormone

(LH) from the anterior pituitary gland. These barriers prevent further development of successive follicles and the hormone estrogen is not produced, so the animals do not show symptoms of estrous (Hafez and Hafez, 2013).

The higher hormone concentration on the day 21 explains that the concentration of progesterone in pregnant dairy cow rises in line with positive pregnancy period. Pemayun and Budiasa (2014) suggested that concentration progesterone above 15 ng / ml can be taken as positive. This study showed that 12 dairy cows had 15 ng /mL and above, while three cow that returned to the cycle probably due to the failure of fertilization (Table I). Increased concentrations of progesterone occur gradually from day 4 to a peak on day 14 after estrous, while decreased concentrations of progesterone begin to occur after day 14 and approach concentration during pregnancy from the day 20. The up and down fluctuations of progesterone are related to the development of corpus luteum during the estrous cycle (Frastantie *et al*., *loc. cit*). The decrease in progesterone concentration is due to the luteolytic properties of endogenous Prostaglandin F2α (PGF2α).

The hormone concentration on the day 28 in 3 cows out of 15 cows has shown a concentration of progesterone increase above 15 ng/mL which is comparable to the reports of Pemayun and Budiasa (*loc cit*). The high concentration of progesterone during pregnancy due to the corpus luteum still serves to stimulate the endometrial cells to produce uterine milk which is the initial nutrient for the embryo before implantation (Hafez and Hafez, *loc cit*). The corpus luteum will be maintained during pregnancy so that the concentration of the hormone progesterone in milk remains high (Drajat, 2002). The formation of the placental membrane begins to develop at 15-17 days after fertilization which is the period of recognition of pregnancy which prevents the release of prostaglandin αF2 so thus preventing the regression of the corpus and maintenance level during progesterone pregnancy (Hafez and Hafez, *loc cit*). Besides, 9 cows with concentration below 15 ng/mL (Table II) shown early embryonic death. In animals that failed to become pregnant, the concentration of progesterone will decrease due to regression of the corpus luteum on the 18-24 days after estrous (Drajat, *loc cit*).

One of the factor affecting early embryonic death is contaminated insemination gun or plastic sheeth and fungal infested feed. The AI in the field without supporting infrastructure and poor sanitation conditions leads to bacterial and fungal infections. The presence of bacteria in the uterus may result in mild endometrial infections or subclinical endometritis (Madyawati *et al*., 2019). Gautam *et al.* (2010) stated that the failure of AI of cattle in Indonesia subclinical bacterial and fungal infections.

Summary

On 28thday the levels were high and indicated pregnancy while in 9 other cows progesterone levels declined due to embryonic death.

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Characterization and Production of Polyclonal Antibody Anti Excretory Secretory Protein of *Blastocystis* sp

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Abstract

This study aims to produce and characterize polyclonal antibody anti excretory secretory (ES) protein of *Blastocystis* sp. ES protein profile was analyzed using SDS-PAGE and used to immunize 2 rabbits. Rabbit's serum were analyzed using indirect ELISA and Western Blot. The result showed that molecular weight of ES protein of *Blastocystis* sp was 40 and 50 kDa and the protein was immunogenic. Both ES protein and antibody anti ES of *Blastocystis* sp can be promoted as diagnostic kit.

Key words : *Blastocystis* sp, Excretory Secretory Protein, Polyclonal Antibody.

Blastocystis sp is a protozoan parasite that widely prevalent in many countries and it causes gastrointestinal symptoms such as diarrhea, nausea, vomiting, abdominal pain, irritable bowel syndrome, and urticaria (Ajjampur *and* Tan., 2016). This parasite can infect human (Roberts *et al.,* 2014) and various animals like amphibian, reptiles, bird, and mammals (Alfellani *et al.*, 2013).

Excretory secretory (ES) protein of parasites have an important role as virulence factor, it can affect and control host immune system during infection and it can be used as a biomarker to detect the presence of parasite and status of the infection in infectious disease (Gomez *et al.,* 2015). This study aims to characterize and produce polyclonal antibody anti ES protein of *Blastocystis* sp.

Materials and Methods

Blastocystis sp was isolated from dunges of beef cattle in Bangkalan, Madura. The sample was cultured using yeast extract media (Mohammed *et al.,* 2015). The growing culture was confirmed with PCR examination using primers b11400 FORC (5`-GGA ATC CTC TTA GAG GGA CAC TAT ACA T-3`) and b11710 REVC (5`-TTA CTA AAA TCC AAA GTG TTC ATC GGA C-3) from Badparva *et al* (2014).

Blastocystis sp was harvested from media and centrifuged with 10.000 rpm for 10 minutes. Pellets was washed 3 times with PBS and

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SUBJECT INDEX

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