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Hematological Profiles of Green Sea Turtle (*Chelonia mydas*) Rehabilitated at Turtle Conservation and Education Center, Serangan Island, Denpasar, Bali

(PROFIL HEMATOLOGI PENYU HIJAU (CHELONIA MYDAS) YANG DIREHABILITASI DI TURTLE CONSERVATION AND EDUCATION CENTER PULAU SERANGAN, DENPASAR, BALI)

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ABSTRACT

Threat of extinction results in the increasing sea turtle conservation efforts through scientific research in recent years. During rehabilitation process, the non-invasive blood analysis is considered to be highly valuable to determine sea turtle's health. The objective of this study was to assess the hematological profiles of green sea turtles (*Chelonia mydas*) under rehabilitation at Turtle Conservation and Education Center (TCEC) Serangan Island, Denpasar, Bali. Three mililiters of peripheral blood from healthy green sea turtles were collected from cervical dorsal sinuses and kept in heparinized vacutainer tube before proceeded with hematological evaluation. PCV was determined by microhematocrit method. Diff count and thrombocyte percentage were determined using Giemsa-stained blood smear under the microscope. RBC and WBC total count were determined using hemocytometer. Hemoglobin (Hb) value was determined using Sahli method. Each hematological values were then analyzed using descriptive analysis. The correlation between body condition index (BCI) and hematological values were analyzed using Spearman's rank correlation test. The mean values evaluated was 7.28 g/dL in Hb, 31.35% in PCV, 0.67 x 10^6/µL in total RBC count, 479.37 fl in MCV, 111.38 pg in MCH, 23.42 g/dL in MCHC, 4.01 x 10^3/µL in total WBC count, 1.20 x 10^3/µL in lymphocyte, 0.21 x 10^3/µL in monocyte, 0.10 x 10^3/µL in eosinophil, 2.48 x 10^3/µL in heterophil and 0.01 x 10^3/µL in basophil. No significant correlation was found between the BCI and hematological profiles evaluated.

Keywords: Green Sea Turtle (*Chelonia mydas*); hematological profiles, BCI

ABSTRAK

Status kelestarian penyu yang terancam punah mendorong meningkatnya usaha konservasi penyu melalui penelitian ilmiah dalam beberapa tahun terakhir. Dalam kegiatan rehabilitasi, pemeriksaan darah yang bersifat non-invasif dianggap sangat berguna dalam menentukan status kesehatan penyu. Penelitian ini bertujuan untuk mengetahui profil hematologi penyu hijau *(Chelonia mydas)* yang direhabilitasi di *Turtle Conservation and Education Center* (TCEC) Serangan. Sebanyak 3 mL darah perifer penyu hijau yang diambil dari sinus servikalis dorsalis disimpan dalam tabung heparin untuk dilanjutkan dengan analisis hematologi. Penentuan nilai PCV dilakukan dengan metode mikrohematokrit. Persentase diferensial leukosit dan trombosit ditentukan dengan pembuatan apusan darah dengan pengecatan Giemsa yang kemudian diamati di bawah mikroskop. Nilai total eritrosit dan leukosit dihitung menggunakan hemositometer. Kadar hemoglobin (Hb) diukur menggunakan Hb Sahli. Hasil penelitian kemudian dianalisis secara kuantitatif menggunakan analisis deskriptif. Hubungan antara indeks kondisi tubuh/

body condition index (BCI) dengan profil hematologi dari hasil pemeriksaan dianalisis dengan uji korelasi Spearman. Hasil penelitian menunjukkan nilai rata-rata kadar Hb 7,28 g/dL, nilai PCV 31,35%, nilai MCV 479,37 fl, nilai MCH sebesar 111,38 pg, nilai MCHC 23,42 g/dL, nilai total eritrosit 0,67 x 10^6/µL, kadar trombosit 16,56 x 10^3/µL, nilai total leukosit 4,01 x 10^3/µL, kadar limfosit 1,20 x 10^3/µL, kadar monosit 0,21 x 10^3/µL, kadar eosinofil 0,10 x 10^3/µL, kadar heterofil 2,48 x 10^3/µL serta kadar basofil 0,01 x 10^3/µL. Tidak ditemukan korelasi yang signifikan antara indeks kondisi tubuh/*body condition index* (BCI) dengan profil hematologi sampel yang diteliti.

Kata-kata kunci: penyu hijau (*Chelonia mydas*); profil hematologi; BCI.

INTRODUCTION

Six out of all seven species of sea turtles in the world are found in the waters of Indonesia. Sea turtle is among the list of protected animals according to Government Regulation (PP) No. 7 1999 on Preserving Flora and Fauna Species Species (Dermawan *et al*., 2009). Being among the IUCN's Red List of Threatened Species results in the increasing sea turtle conservation efforts through scientific research in recent years. Thus, the result can be the basis in developing conservation plans and managing diseases, and also play a role in preventing the decline of its populations in the wild (Orós *et al*., 2010).

Sea turtle has a unique anatomy. This can be an obstacle in making diagnosis and assessing its health using common method. Therefore, the non-invasive blood analysis is considered to be very valuable to determine sea turtle's health (Perpiñán, 2017). Hematological analysis becomes a main tool of diagnostic method especially to detect the presence of such as blood parasite and other infectious agents by looking at the elevation or decreasing of certain blood parameters (Reséndiz *et al*., 2018).

Information regarding hematological profiles of green sea turtle was already reported from different parts of the world, including North Carolina (Anderson *et al*., 2011) and Taiwan (Fong *et al*., 2010). However, until now there has been no publication related to research on hematological profiles of sea turtles—either the green sea turtles or other species—in Indonesia. Environmental and psychological conditions, such as season, type of foods available and stress level, can affect hematological values (Anderson *et al*., 2011). Turtle Conservation and Education Center (TCEC) in Serangan is one of five sea turtle conservation sites in Bali that are still actively rehabilitating sea turtles of which mostly originated from Bali and Java waters. By knowing the base value of hematological profiles of green sea turtle under rehabilitation at TCEC Serangan, it is expected to be used as a reference in determining the health status of green sea turtles originating from waters around Bali, Indonesia.

RESEARCH METHODS

Sample collection and examination were carried out at the Turtle Conservation and Education Center (TCEC) Serangan, Bali. The time required for this research is ± 4 weeks, around March-April 2019.

Morphometric Measurement, BCI Determination and Physical Examination

Health status and age classification of green sea turtles were determined by body condition index (BCI) and morphometric measurements. The measured morphometry including straight carapace length (SCL) and curved carapace length (CCL). Straight carapace length and body weight were then used to calculate BCI using the formula described by Bjorndal (2000). The health status were considered as normal if the BCI value ≥ 1 (Norton and Wyneken, 2015). The age was classified using CCL value, where turtles with CCL ≥ 85 cm were categorized as adult, CCL 84 - 70 cm were categorized as subadult and CCL 69 - 36 cm were categorized as juvenile (Hamann *et al*., 2006; Samour *et al*., 1998). Next, turtles were inspected to see the presence of epibionts, tumors, open wounds or movement abnormalities (Arthur *et al*., 2008; Fong *et al*., 2010).

Blood Sample Collection

Green sea turtles under rehabilitation period for 11 to 14 days had their blood samples taken a few hours before being released back into the sea, when the turtles are considered to be clinically healthy. Approximately 3 mL of peripheral blood were taken from 1 juvenile, 2 subadult, and 2 adult turtles after 11 to 14 days of rehabilitation period and 4 juvenile and 1 subadult turtles after several months to years of rehabilitation period at TCEC Serangan. Turtles are positioned in ventral recumbency with the head and neck maintained extended. A 5 mL syringe with a 22G needle was inserted into the dorsal cervical sinus at a 45° angle. To avoid the occurrence of hemolysis, the needle was removed from the syringe before the blood was transferred to the tube, given the size of the erythrocytes that made it easy to lyse (Saggese, 2009). Blood stored in heparinized vacutainer 3 mL.

Hematological Analysis

Determination of PCV values was carried out by the microhematocrit method that was centrifuged using Nesco DSC-100MH-1 12,000 rpm for 6 minutes. The results were read using a hematocrit reader (Reséndiz *et al*., 2018). Differential leukocytes and platelets were calculated by making blood smears using Giemsa staining. Blood smears were examined using a microscope with magnification of 1000 x. The types of leukocytes were counted to the number 100 (Perpiñán, 2017). The platelet count was performed by counting the cells in a hundred erythrocytes. The percentage of platelets obtained then multiplied by the number of erythrocytes collected using hemocytometer. Hemoglobin levels and erythrocyte count were determined using Sahli hemoglobinometer (OneMed) as described by Tosunoglu *et al.* (2011). Erythrocyte counts were performed using hemocytometer (Assistent). Blood with anticoagulants which had been diluted 200 x in Hayem solution was inserted into Neubauer counting chamber (Tosunoglu *et al*., 2011) and the cells counted under microscope with magnification of 45 x according to Tsai *et al*. (2001). Erythrocyte indices evaluated including mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC). MCV was calculated using the formula as described by Mahaffey *et al*. (2003). Same with erythrocytes, leukocyte counts were also performed using hemocytometer. Blood with 20 x anticoagulants diluted in Rees-Ecker solution (Perpiñán, 2000; Mitchell and Tully, 2009) then inserted into Neubauer counting chamber. This counting chamber is examined under a microscope with a magnification of 40 x according to Tsai *et al*. (2001).

Statistical Analysis

The hematological profiles of the three sample groups divided by age-juvenile, subadult and adult—were analyzed quantitatively using descriptive analysis, the results of which were presented in mean±standard deviation and range. The correlation between body condition index (BCI) and hematological profiles collected was then analyzed using Spearman's rank correlation test (Sampurna and Nindhia, 2017) with the interpretation of the coefficients as perfectly correlated (r_s = 1), very strong (0.75 <r_s ≤ 0.99), strong (0.5 <r_s ≤ 0.75), sufficient (0.25 <r_s ≤ 0.5), very weak (0 <r_s ≤ 0.25) and no correlation (r_s = 0). Correlation with sig. <0.05 was categorized as significant (Sarwono, 2013). All data was processed in the IBM® SPSS® Statistics program ver. 25.0.

Ethics Statement

Research permits were granted by Animal Ethics Committees Veterinary Medicine, Udayana University (1592/UN14.2.9/PD/2019). These cover the sampling, collection and processing of blood samples.

RESULTS AND DISCUSSION

Results

The 10 green sea turtles sampled in this study had BCI values ranged from 1.02 to 1.47. The physical examination showed no presence of epibiont, tumor, open wound or any movement abnormality. Five juveniles that were subjected in the study had weight ranged from 5.2 to 18.3 kg, SCL from 33.5 to 48.3 cm, CCL from 36.5 to 54 cm. Three subadults that were subjected to the study had weight ranged from 36.5 to 57.7 kg, SCL from 71 to 76.5 cm, CCL from 71 to 83 cm. The two adults that were subjected to the study respectively had weight 97 and 100.9 kg, SCL 93.2 and 90.5 cm, CCL 97 and 95 cm.

Hematological profiles of green sea turtles rehabilitated at TCEC Serangan (n=10) resulted from this study are presented in Table 1. The average value of total erythrocyte calculated from all samples was 0.67 x 10^6/µL, with the average hemoglobin level was 7.28 g/dL, PCV 31.35%, and erythrocyte indices including MCV 479.37 fl, MCH 111.38 pg, and MCHC 23.42 g/dL. The average value of total leucocyte was 4.01 x 10^3/µL, separately lymphocyte level was

1.21 x $10^3/\mu L$, monocyte 0.21 x $10^3/\mu L$, eosinophil 0.10 x $10^3/\mu L$, heterophil level which had the highest leukocyte proportion was 2.48 x $10^3/\mu L$, and the lowest leukocyte proportion calculated was basophil (0.01 x $10^3/\mu L$).

Hematological profiles of juvenile green sea turtle rehabilitated at TCEC Serangan (n=5) are presented in Table 2. Hematological profiles of subadult green sea turtle rehabilitated at TCEC Serangan Island (n=3) are presented in Table 3.

Table 1. Hematological profiles of green sea turtle *(Chelonia mydas)* rehabilitated at TCEC Serangan

Variables	Units	All Samples (n=10)		Reference
		Mean ± SD	Range	
Hb	g/dL	7.28 ± 1.52	4.80-10.20	5.30-12.40[a]
PCV	%	31.35 ± 3.39	27.50-36.00	28.00-42.25[b]
MCV	fL	479.37 ± 70.51	387.10-625.00	400.00-1429.00[c]
MCH	pg	111.38 ± 24.66	66.67-145.71	71.90-400.00[b]
MCHC	g/dL	23.42 ± 5.50	15.00-36.43	20.00-45.50[a]
RBC	$10^6/\mu L$	0.67 ± 0.13	0.44-0.93	0.24-0.68[a]
T	$10^3/\mu L$	16.56 ± 4.14	10.80-21.90	13.30-24.90[b]
WBC	$10^3/\mu L$	4.01 ± 0.77	2.70-5.05	1.76-22.40[d]
Lymphocyte	$10^3/\mu L$	1.21 ± 0.39	0.60-1.58	0.94-4.34[e]
Monocyte	$10^3/\mu L$	0.21 ± 0.17	0.07-0.62	0.18-2.92[d]
Eosinophil	$10^3/\mu L$	0.10 ± 0.04	0.03-0.13	0.00-0.48[e]
Heterophil	$10^3/\mu L$	2.48 ± 0.57	1.81-3.44	1.57-15.72[e]
Basophil	$10^3/\mu L$	0.01 ± 0.02	0.00-0.05	0.00-1.94[e]

Hb = hemoglobin, PCV = packed cell volume, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, T = thrombocyte, WBC = white blood cell, SD = standard deviation.
[a]Samour *et al.* (1998), [b]Reséndiz *et al.* (2018), [c]Aguirre *et al.* (1995), [d]Lewbart *et al.* (2014), [e]Suarez-Yana *et al.* (2016)

Table 2. Hematological profiles of juvenile green sea turtle *(Chelonia mydas)* rehabilitated at TCEC Serangan

Variables	Units	Juvenile (n=5)	
		Mean ± SD	Range
Hb	g/dL	7.16 ± 1.09	6.20-8.80
PCV	%	31.90 ± 3.73	27.50-36.00
MCV	fL	529.81 ± 61.04	469.70-625.00
MCH	pg	118.86 ± 16.73	95.89-140.91
MCHC	g/dL	22.45 ± 2.11	19.72-24.52
RBC	$10^6/\mu L$	0.61 ± 0.12	0.44-0.73
T	$10^3/\mu L$	15.96 ± 4.99	10.80-21.90
WBC	$10^3/\mu L$	3.96 ± 0.93	2.70-5.05
Lymphocyte	$10^3/\mu L$	1.17 ± 0.44	0.60-1.52
Monocyte	$10^3/\mu L$	0.12 ± 0.06	0.07-0.22
Eosinophil	$10^3/\mu L$	0.08 ± 0.05	0.03-0.13
Heterophil	$10^3/\mu L$	2.57 ± 0.78	1.81-3.44
Basophil	$10^3/\mu L$	0.02 ± 0.02	0.00-0.04

Hb = hemoglobin, PCV = packed cell volume, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, T = thrombocyte, WBC = white blood cell, SD = standard deviation.

Hematological profiles of adult green sea turtle rehabilitated at TCEC Serangan (n=3) are presented in Table 4.

Correlation test result between BCI and the hematological profile of green sea turtles rehabilitated at TCEC Serangan are presented in Table 5. There was a weak positive correlation between BCI and MCH, MCHC, eosinophils and heterophils ($0 < r_s \leq 0.25$). Sufficient positive correlation between BCI and hemoglobin levels,

Table 3. Hematological profiles of subadult green sea turtle *(Chelonia mydas)* rehabilitated at TCEC Serangan

Variables	Units	Subadult (n=3)	
		Mean ± SD	Range
Hb	g/dL	6.33 ± 1.50	4.80-8.80
PCV	%	32.00 ± 4.00	28.00-36.00
MCV	fL	427.72 ± 35.36	387.10-625.00
MCH	pg	84.59 ± 18.29	66.67-40.91
MCHC	g/dL	19.84 ± 4.24	15.00-24.52
RBC	$10^6/\mu L$	0.76 ± 0.16	0.62-0.73
T	$10^3/\mu L$	19.60 ± 1.73	18.60-21.90
WBC	$10^3/\mu L$	3.88 ± 0.80	3.30-5.05
Lymphocyte	$10^3/\mu L$	1.17 ± 0.47	0.66-1.52
Monocyte	$10^3/\mu L$	0.31 ± 0.28	0.07-0.22
Eosinophil	$10^3/\mu L$	0.11 ± 0.02	0.10-0.13
Heterophil	$10^3/\mu L$	2.29 ± 0.32	1.92-3.44
Basophil	$10^3/\mu L$	0.00 ± 0.00	0.00-0.04

Hb = hemoglobin, PCV = packed cell volume, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, T = thrombocyte, WBC = white blood cell, SD = standard deviation.

Table 4. Hematological profiles of adult green sea turtle (*Chelonia mydas*) rehabilitated at TCEC Serangan Island

Variables	Units	Adult (n=3)	
		Mean ± SD	Range
Hb	g/dL	9.00 ± 1.70	7.80-10.20
PCV	%	29.00 ± 1.41	28.00-30.00
MCV	fL	430.77 ± 43.52	400.00-461.54
MCH	pg	132.86 ± 18.18	120.00-145.71
MCHC	g/dL	31.22 ± 7.38	26.00-36.43
RBC	$10^6/\mu L$	0.68 ± 0.04	0.65-0.70
T	$10^3/\mu L$	13.50 ± 0.71	13.30-24.90
WBC	$10^3/\mu L$	4.30 ± 0.64	3.85-4.75
Lymphocyte	$10^3/\mu L$	1.35 ± 0.32	1.12-1.57
Monocyte	$10^3/\mu L$	0.28 ± 0.05	0.24-0.31
Eosinophil	$10^3/\mu L$	0.11 ± 0.01	0.10-0.12
Heterophil	$10^3/\mu L$	2.56 ± 0.35	2.31-2.80
Basophil	$10^3/\mu L$	0.03 ± 0.04	0.00-0.05

Hb = hemoglobin, PCV = packed cell volume, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, T = thrombocyte, WBC = white blood cell, SD = standard deviation.

Table 5. Correlation between body condition index (BCI) and hematological profiles of green sea turtle (*Chelonia mydas*) (n=10) rehabilitated at TCEC Serangan Island

Parameters	Correlation Analysis	BCI
BCI	Correlation Coefficient	1.000
	Sig. (2-tailed)	
	N	10
Hb	Correlation Coefficient	0.263
	Sig. (2-tailed)	0.463
	N	10
PCV	Correlation Coefficient	0.307
	Sig. (2-tailed)	0.389
	N	10
MCV	Correlation Coefficient	0.283
	Sig. (2-tailed)	0.428
	N	10
MCH	Correlation Coefficient	0.246
	Sig. (2-tailed)	0.493
	N	10
MCHC	Correlation Coefficient	0.025
	Sig. (2-tailed)	0.946
	N	10
RBC	Correlation Coefficient	0.332
	Sig. (2-tailed)	0.348
	N	10
T	Correlation Coefficient	0.412
	Sig. (2-tailed)	0.237
	N	10
WBC	Correlation Coefficient	-0.302
	Sig. (2-tailed)	0.397
	N	10
Lymphocyte	Correlation Coefficient	-0.357
	Sig. (2-tailed)	0.311
	N	10
Monocyte	Correlation Coefficient	-0.525
	Sig. (2-tailed)	0.119
	N	10
Eosinophil	Correlation Coefficient	0.057
	Sig. (2-tailed)	0.876
	N	10
Heterophil	Correlation Coefficient	0.056
	Sig. (2-tailed)	0.879
	N	10
Basophil	Correlation Coefficient	0.525
	Sig. (2-tailed)	0.119
	N	10

PCV, MCV, total erythrocytes and platelets, and negative correlation with total WBC and lymphocyte, was found ($0.25 < r_s \leq 0.5$). Strong positive correlation ($0.5 < r_s \leq 0.75$) between BCI and basophil levels, and negative correlation between BCI and monocyte levels, was determined. No significant correlation was found between BCI and the hematological profile of the sample studied (sig.> 0.05).

DISCUSSION

Animals' clinical status can be determined by various methods. Body condition index (BCI) is used to estimate the nutritional status of various species by obtaining body length and weight values (Jakob *et al.*, 1996). Subjectively, BCI can be interpreted into four visual assessments, where BCI that is less than 1.00 is categorized as emaciated, normal if it ranges from 1.00 to 1.10, robust if it ranges from 1.11 to 1.20, and very good if it values more than 1.20 (Norton and Wyneken, 2015). The results of the physical examination together with the determination of BCI showed that the object of this study was clinically healthy.

Blood drawing on adult turtles through the dorsal cervical sinuses in this study required several trials. In addition to the sizes, difficulties were also present in maintaining neck extension, since they were very strong (Samour *et al.*, 1998). Blood drawing through the external jugular vein was an alternative to the juveniles, which are in the direction of 1 o'clock assuming the 12 o'clock as the most dorsal part of the neck (Mader, 2006).

Hematological examination plays a role in detecting several conditions that affect blood cells (Campbell, 2006), and is considered to be diagnostic in detecting the presence of agents or damage that directly affects blood cells (Mahaffey *et al*. 2003). Conditions that might be seen from hematological examination including anemia, leukocytosis, inflammation, the presence of blood parasites, blood cell formation disorders, dehydration, and hemostatic changes (Campbell, 2006; Meyer and Harvey, 2004). Hematological profile of green sea turtle rehabilitated at TCEC Serangan resulted from this study is within the range of hematological values that have been previously reported (Lewbart *et al*., 2014; Anderson *et al*., 2016; Suarez-Yana *et al*., 2016; Reséndiz *et al*., 2018). The limited number of objects in this study makes the comparison of hematological profiles between the three age groups—juvenile, subadult and adult—from this study become relative. The rehabilitation period that was considerably short in one juvenile and each of the two subadult and adult turtles raised the possibility that they had not reached their best condition during blood sampling.

The mean hemoglobin levels found in this study are similar to the results reported by Samour *et al*. (1998) in green sea turtles in Saudi Arabia, where they found a correlation between age and hemoglobin levels. The highest hemoglobin level was found in adult, followed by juvenile and the lowest belongs to subadult group. Mean and range of PCV from this study are similar to the one conducted by Reséndiz *et al*. (2018) in subadult green sea turtles in Mexico. According to Bolten and Bjorndal (1992), PCV does not correlated significantly with the sex or body size of wild green sea turtles. In contrast to the research previously done by Wood and Ebanks (1984) and Frair (1977) on green sea turtles in captivity, which showed a significant correlation between body size and PCV value. Also supported by research conducted by Fong *et al*. (2010), the results of this study indicated that PCV range in juveniles is lower than the adults, with the highest value belonged to the subadults. Beside the different method that was used, the high average value of the total erythrocytes in this study may be due to the small number of objects used in this study, so that it was not enough to show an average value similar to previous studies. However, the average value of total RBC from this study is still within the range previously stated by Samour *et al*. (1998). Increasing values of the three parameters PCV, hemoglobin and total RBC can indicate dehydration or transfer of fluid concentration to visceral organs. The decrease in these three parameters may indicate anemia.

Determined hemoglobin and erythrocyte levels means erythrocyte indices can also be determined (Mahaffey *et al*. 2003). Mean MCV value conducted in this study are within the range of values reported by Aguirre *et al*. (1995). The average MCHC value of the subadult group in this study is similar to the results of a study conducted by Reséndiz *et al*. (2018). Overall, MCH is not generally used as an indicator in detecting anemia if it is not accompanied by an increased or decreased MCV and MCHC levels (Mahaffey *et al*. 2003). Referring to the results of the study by Samour *et al*. (1998), mean value of MCHC in juvenile group from this study is within the range of the previously reported from subadult group, whereas the mean value in adult group from this study are within the range of values in the same age group previously reported. This might occur due to inadequate number of samples that might not enough for the average value conducted to represent the value in each age group. Determination of MCV value is useful to determine the type of anemia based on the average size of erythrocytes, namely microcytic, normocytic, or macrocytic anemia. The MCHC value is useful for determining the type of anemia based on hemoglobin levels in one erythrocyte, namely hypochromic, hyper-chromic, or normochromic anemia.

The mean platelet count from this study is within the range described by Reséndiz *et al*. (2018) in subadult green sea turtles from Mexico. Some conditions such as thrombosis, bleeding, immune system disorders and infectious diseases can show changes in platelet levels in the blood (Mahaffey *et al*. 2003). In reptiles, morphological changes in platelets usually come as polymorphic nuclei and can be related to the presence of serious inflammatory diseases (Campbell and Ellis, 2007).

Most studies on chelonian hematological profiles use Natt and Herrick method to calculate total leukocyte and erythrocytes (Mahaffey *et al*. 2003). It is not easy to obtain Natt Herrick's solution in Indonesia, making this study used another alternative solutions, separately leukocytes were calculated by diluting blood in Rees-Ecker's solution and erythrocytes in

Hayem's solution. In calculating total leukocytes, solutions with the principle as to lyse cell membranes such as Turk are not appropriate to be used in chelonian blood, because erythrocytes can remain intact or lyse and leave the cell nuclei that may confuse the calculation (Kerr, 2002). The average leukocyte count resulted in this study are within the range of values reported by Lewbart *et al.* (2014) and Suarez-Yana *et al.* (2016). Similar to the one conducted by Samour *et al.* (1998), subadult green sea turtles have a higher total leukocyte count than other age groups. The mean differential leukocyte percentage from this study is within the range reported by Anderson *et al.* (2016) on green sea turtles in North Carolina. Lymphocyte, monocyte, eosinophil, heterophil and basophil levels from this study are within the range of values delivered by Anderson *et al.* (2016), Lewbart *et al.* (2014) and Suarez-Yana *et al.* (2016). Change levels of leukocytes—heterophilia, eosinophilia, basophilia, lymphocytosis, lymphopenia and monocytosis—can be associated with stress, parasitic and virus infections, ecdysis, inflammation or bacterial infections, fibropapiloma tumors to lack of nutrients (Campbell, 2004).

No significant correlation was found between BCI and the hematological profiles evaluated. This result is similar with the previous study by Espinoza-Romo *et al.* (2018) of olive ridley (*Lepidochelys olivacea*) sea turtles in Mexico. The positive correlation between PCV and carapace size previously reported by Frair (1977) and Stacy *et al.* (2018) is said to relate to the size of erythrocytes. Although not significant, a positive correlation between BCI and all erythrocyte examination parameters was also found in this study. A negative correlation that was found between BCI and three parameters of leukocyte—total leukocyte counts, lymphocyte and monocyte levels—can be assumed if green sea turtle experiences overall leukocytosis or lymphocytosis and monocytosis, the BCI might shown to be below the normal range (BCI <1.0).

CONCLUSION

Mean values of hematological profiles of green sea turtles rehabilitated at TCEC Serangan is 0.67 x 10^6/µL in RBC value, with Hb concentration of 7.28 g/dL, PCV 31.35%, and RBC indices including MCV, MCH, and MCHC respectively are 479.37 fl, 111.38 pg and 23.42 g/dL. Mean value of total leukocyte obtained from this study is 4.01 x 10^3/µL, with an average of 1.20 x 10^3/µL in lymphocyte levels, 0.21 x 10^3/µL in monocytes, 0.10 x 10^3/µL in eosinophils, 2.48 x 10^3/µL in heterophils and 0.01 x 10^3/µL in basophils. The mean platelet level is 16.56 x 10^3/µL. No significant correlation was found between BCI and the hematological profiles conducted.

SUGGESTION

Based on the reported study, this is the first time a research on hematological profiles of green sea turtles in Bali, Indonesia, ever conducted. The results obtained from this study are expected to be used as a reference in determining the health status of green sea turtles, especially during rehabilitation process in Bali rehabilitation centers.

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