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Turtle Blood Cell Morphology¹

JUDITH M. HEADY and T. EDWIN ROGERS²

Abstract. Blood cells from Pseudemys elegans, P. troostii, Emys blandingii, and Chrysemys bellii bellii were studied. Smears were stained with Wright's stain and washed with citrate buffer and double distilled water. The RBC were typical. White cells, in order of frequency of occurrence, were large acidophil granulocytes or eosinophils, 26-41%; small acidophils, 22-26%; neutrophil granulocytes, 19-32%; lymphocytes, 11-22%; and monocytes, 0.3%. Thrombocytes were not measured or counted. Data on cell sizes are given for P. elegans.

Until recently no thorough studies had been made on turtle blood cells since that of Charipper (1932). Measurements of cells and hematocrit values were reported by Gaumer and Goodnight (1957). In 1961, Taylor and Kaplan published a study of the blood cells of pseudemyid turtles. Many of the data given below confirm the observations of Taylor and Kaplan.

MATERIALS AND METHODS

Four species of turtles were used: *Pseudemys elegans*, *P. troostii*, *Emys blandingii*, and *Chrysemys bellii bellii*. Cell measurements given refer only to *P. elegans*. These turtles were shipped from Wisconsin and were kept in the laboratory at a relatively constant temperature of about 25°C. Blood from fifteen turtles was used.

Anesthesia was accomplished with 0.5-2.0 ml Nembutal given intraperitoneally or rectally. Blood was taken from the aorta or carotid or subclavian arteries. Smears were made using the cover slip technique. These were dried, stained two minutes with Wright's stain, buffered for three minutes with citrate buffer (pH 6.7), and then washed with double distilled water. Permanent slides were made with xylol and permount.

Results

The RBC were the typical elliptical, nucleated cells. The nucleus stained dark purple, occasionally with a light bluish cast, and had visible chromatin clumps. The cytoplasm was light red, and there was a distinct cell boundary. A few granules could be seen. Some RBC had clear cytoplasm, were more nearly round

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than elliptical, and were smaller. These may possibly have been immature cells.

The white cells, in order of frequency, were large acidophil granulocytes or eosinophils, 26-41%; small acidophils, 22-26%; neutrophil granulocytes, 19-32%; lymphocytes, 11-22%; and monocytes, 0-3%. Thrombocytes were not included. It is evident that cell distribution varied greatly; only the small acidophils remained fairly constant.



Figure 1. A, eosinophil; B, small acidophil; C, D. neutrophils; E, lymphocytes; F, thrombocyte. (X1200)

The eosinophil typically was circular with a relatively small, slightly elliptical nucleus (Fig. 1A). The nucleus was medium to dark purple and was located peripherally with the inside

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edge covered by granules. The small, compact, circular red granules covered any cell surface and gave an uneven edge and rough appearance to the cells.

The small acidophil was irregularly circular with a pale blue to medium purple nucleus (Fig. 1B). The round nucleus was surrounded by the feathery red, blue, and purple granules of the cytoplasm. The nucleus was centered and covered about onethird of the visible area. The cytoplasm presented a homogeneous blue background, but the granules gave a rough appearance to the cell boundaries.

The neutrophil was irregularly circular with an elliptical nucleus which was frequently larger at one end or kidney-shaped (Fig. 1C and 1D). The nucleus stained a light purple, showed visible chromatin clumping, and covered one-third to one-half of the visible area. It was located between the center and the edge of the cell. The small blue granules with red in some areas gave the cytoplasm a mottled appearance. The cell edges were even and showed a distinct boundary.

The lymphocyte was round and the dark purple nucleus was circular and large (Fig. 1E). The cytoplasm was clear-to-bluish ring around the nucleus. No granules were visible. A smooth cell boundary was apparent. A few slightly elliptical lymphocytes were seen.

The monocytes were few in number but were easily distinguished. The round nucleus stained a medium purple and was centrally located. It covered about two-thirds of the visible area. The cytoplasm sometimes had a mottled blue appearance, but no granules were present.

The thrombocyte nucleus stained a very dark purple, and many times the clear cytoplasmic projections were difficult to detect (Fig. 1F). Often these cells were fragmented and occurred in clusters, making measurements or counts infeasible.

DISCUSSION

The RBC were typical of the lower vertebrates. The measurements agreed generally with those in the literature. The mean length of the RBC of P. elegans was 18.5μ and the mean width was 10.5μ (Table 1). In Spector (1956), the dimensions for the box turtle were 19.0 μ x 9.0 μ . Taylor and Kaplan (1961) reported dimensions of 20.5μ x 12.7μ for pseudemyid turtles. Their measurements were made on unstained cells suspended in isotonic saline solution. Sayles (1949) gave the length as 20.0μ . Gaumer and Goodnight (1957), who also used Wright's stain and pseudemyid turtles, gave the size as $20.125 \mu \times 10.625 \mu$.

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	Table 1.	able 1. Blood cell sizes in <i>Pseudemys elegans</i> .				
Cells		Numbers of cells	Size Ranges (microns)	Mean	Standard Error	
RBC: length width Eosinophils Small acidophils Neutrophils Lymphocytes Monocytes		100 100 153 120 113 91 4	$\begin{array}{c} 16.5\text{-}22.5\\ 9.0\text{-}13.5\\ 9.0\text{-}18.0\\ 7.5\text{-}15.0\\ 10.5\text{-}16.5\\ 6.0\text{-}15.0\\ 10.5\text{-}13.5\end{array}$	18.5 10.5 13.6 10.4 13.5 9.5	$\begin{array}{c} \pm 0.46 \\ \pm 0.54 \\ \pm 0.63 \\ \pm 0.67 \\ \pm 0.65 \\ \pm 0.74 \end{array}$	

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Until the study by Taylor and Kaplan in 1961, there were few descriptions of turtle white blood cells. Their descriptions of these cells were generally in agreement with ours except in the interpretation of the small acidophils. The cells which we called small acidophils because of their resemblance to the small acidophils of fish were described by Taylor and Kaplan as basophils. Their use of a different stain, as well as the unstained suspensions, made accurate comparisons of these white cells difficult.

The eosinophils and neutrophils were more numerous in our counts than in theirs, but their descriptions emphasized more the red-to-purple cast found in the cytoplasm. The lymphocytes were easily identified and occurred in nearly equal percentages in both studies. All the cells were monolobed, but a bi-lobed neutrophil was reported by Ryerson (1943). The monocyte occurred with greater frequency in their studies, but in some instances comprised as few as 2.0% of the cells. The thrombocytes seemed large compared with our observations, but since we made no measurements, this can not be verified.

Our cell measurements were larger in about the same ratio for each type of white cell. This probably was because the cells tend to flatten in a stained smear.

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