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T. S. OKADA

Institute of Animal Genetics, University of Edinburgh (on leave of absence from the Zoological Institute, Faculty of Science, University of Kyoto, Japan), December 12, 1958.

Zusammenfassung

Aus 8 Tage alten Hühnerembryonen wurden durch Verdauung der intrazellulären Matrix des differenzierten Knorpelgewebes dissoziierte Chondrozyten gewonnen. Diese Chondrozyten schlossen sich wieder zu Kolonien lose gepackter sphärischer Zellen zusammen. Durch Regeneration der interzellulären Matrix bildete sich in den Organkulturen solcher Reaggregate die ursprüngliche Knorpel-Architektur wieder aus.

Normal Count and Physiological Variability of Rabbit Blood Basophils¹

Rabbit blood contains an exceptionally great number of basophil granulocytes. Basophil countings from rabbit blood smears have been performed by various authors, yielding highly varying results. Using a direct chamber counting method², the present study of the basophil count in rabbit blood under physiological variations was undertaken.

¹ Aided by grants from the Danish Rheumatism Association and Eli Lilly and Co., Indianapolis, Ind., U.S.A., to Dr. ASBOE-HANSEN.

² J. E. MOORE and G. W. JAMES, *Proc. Soc. exp. Biol. Med.* 82, 601 (1953).

Methods. 159 six-months-old white rabbits of both sexes and of known homogeneous breed were used. They were separately caged, at a temperature of $20^{\circ} \pm 1^{\circ}\text{C}$, fed on oats, hay, and sugar beets. The animals were carefully handled and trained to accommodate in blood-sampling boxes. For blood sampling, the ear was tapped lightly by finger to stimulate blood circulation, and a prick with a 14-gauge needle or a small incision in one of the ear veins usually secured a free flow of blood drops. After discarding the first drop, 20 μl of blood was drawn into a standard blood pipette, and quickly transferred into a glass tube containing 180 μl of a modified Moore & James toluidine blue solution³. The tube was shaken until complete hemolysis of the red cells. A solution of 0.05% toluidine blue in 50% watery propylene glycol was used in few cases. The diluted blood was then transferred by capillarity to Fuchs-Rosenthal counting chambers (0.2 mm deep) and the cells allowed to settle. Counting slides were kept in a covered Petri dish floored with a damp filter paper to avoid evaporation. Two chambers of 16 mm² area each were counted for basophils, and two big squares of 1 mm² area each for obtaining total leucocyte counts. A binocular microscope with $\times 10$ oculars and a $\times 10$ achromatic plane objective was used; a blue filter contrasted the meta-chromatically stained basophils against the faintly blue-stained leucocytes of different types.

Results. Figure 1 presents distribution curves, as well as mean values \pm standard error, standard deviations and range of 9 a.m. basophil counts (absolute and differential) of the first blood samples ever taken from all rabbits under study. The curves are more or less positively skewed⁴.

A very slight variation in the morning count from day to day was encountered (Table). A study of diurnal variation in the number of basophils in 58 male rabbits, however, revealed a statistically significant ($P < 0.001$) increase in late afternoon (3 p.m.) as compared to the comparatively low morning (9 a.m.) count (Fig. 2).

³ A.-W. A. BOSEILA, *Proc. Soc. exp. Biol. Med.* 98, 184 (1958).

⁴ The advice and help of Miss HANNE BACKHAUSEN with statistical analysis are gratefully acknowledged.

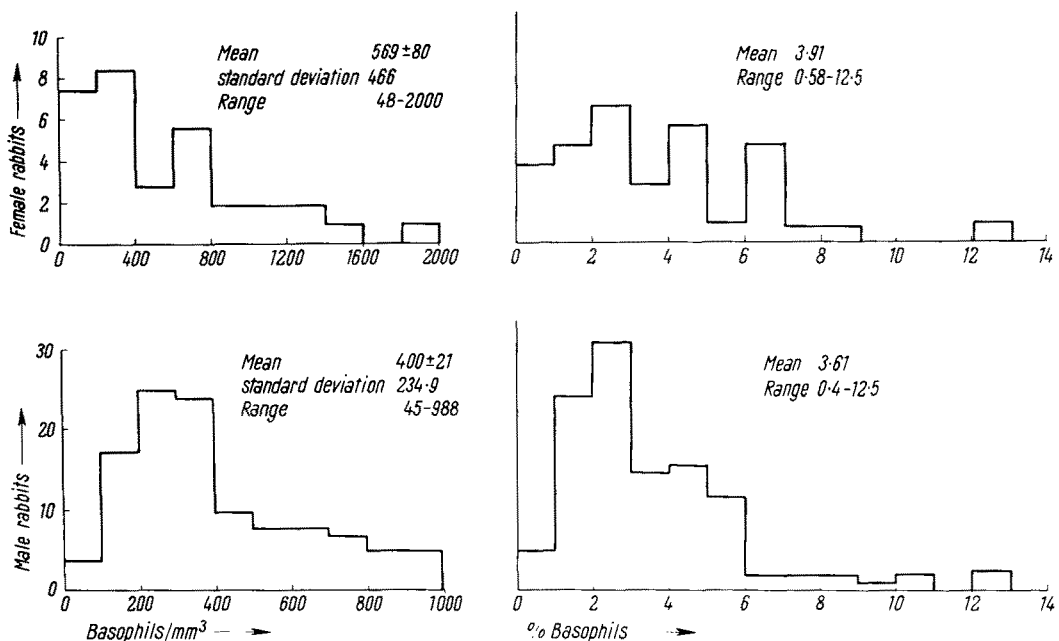


Fig. 1.—Distribution curves of first absolute and relative basophil leucocyte morning (9 a.m.) counts ever taken from 116 male and 43 female adult rabbits. Mean \pm standard error, standard deviation, and range are indicated.

The average of 1142 basophil counts at different times of day was $516.4 \pm 10.8/\mu\text{l}$ blood (st. dev. ± 366). The mean of 426 basophil counts of females was 545.6 ± 19.3 (st. dev. ± 399), which is insignificantly higher than the mean of 716 basophil counts of male rabbits, showing 499 ± 12.9 (st. dev. ± 344).

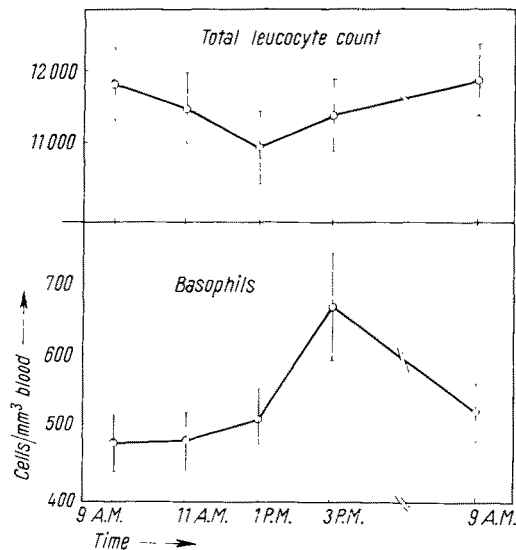


Fig. 2.—Diurnal variation of blood basophil and total leucocyte counts for 58 male adult rabbits (\bar{x} : mean \pm standard error).

During pregnancy, 6 females demonstrated a clear tendency towards a fall in basophil count, most pronounced on the day of delivery and the third day after delivery (Fig. 3).

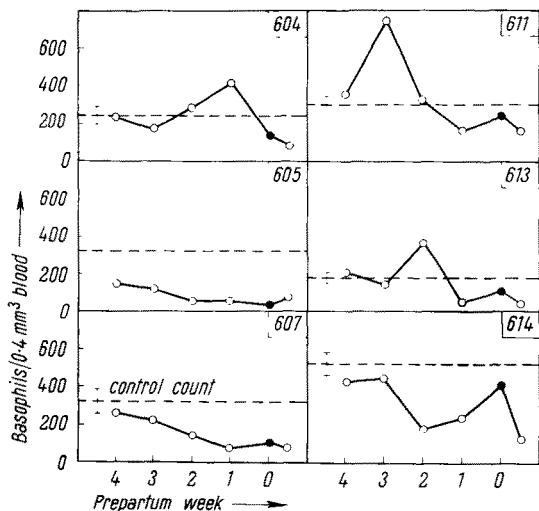


Fig. 3.—Blood basophil count variations for 6 rabbits during pregnancy (o: counts before or after delivery, •: count on day of delivery; ---: mean of 4 control counts taken on 4 successive days before copulation with standard error indicated by vertical line).

Discussion. By indirect counting methods, the number of basophils in rabbit blood ranged from 0–950 cells/ μl blood, with differential counts of 0–12%⁵. Direct counts of the present study, however, revealed a minimum count of 45 basophils/ μl blood, corresponding to a differential

Variations in the morning basophil count in rabbit blood (counts per mm^3 blood, at 9 a.m.) on successive days.

Day	Males			Females		
	No. of counts	Mean \pm standard error	Standard deviation \pm	No. of counts	Mean \pm standard error	Standard deviation \pm
1	128	475 ± 23.6	267	43	552 ± 72.0	472
2	128	528 ± 20.5	232	43	453 ± 49.9	327
3	70	422 ± 34.0	284	43	524 ± 46.1	302
4	58	428 ± 35.8	272			
5	50	422 ± 45.0	318			
Total	434	469 ± 12.9	269	129	509 ± 32.9	374

count of 0.4%. Although a wide individual variation in the counts was observed, they differed but slightly from day to day under standard conditions. The higher level of basophils in the female group and the afternoon rise in both counts are in accordance with reported findings in normal humans⁶. Even the decrease observed during pregnancy agrees with the decreasing tendency reported in pregnant women. An influence of adrenocortical as well as female sex hormones on the number of circulating basophils has been suggested previously⁷, and may also be responsible for these variations.

A.-W. A. BOSEILA⁸

Connective Tissue Research Laboratory, University Institute of Medical Anatomy, Copenhagen (Denmark), January 19, 1959.

Zusammenfassung

Direkte Zählung von basophilen Leukozyten 195 männlicher und weiblicher Kaninchen in einer Blutkammer ergab eine Durchschnittszahl von 516 pro mm^3 Blut. Eine signifikante Zunahme von Basophilen findet nachmittags statt. Während der Schwangerschaft wurde eine Abnahme der Basophilen beobachtet.

⁶ A.-W. A. BOSEILA, Acta med. scand. 163 (1959). – Y. OSADA, Bull. Inst. publ. Health 5, 5 (1956).

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⁸ On leave from the Histology Department, Faculty of Medicine, Cairo University, Egypt.

The Embryonic Origin of the Intrinsic Limb Musculature in Amphibia, Salientia

The mesodermal cells that develop into the intrinsic limb muscles, in tetrapods, are generally considered to be exclusively somatopleural in origin^{1–6}. MILAIRE⁷ has re-

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² S. R. DETWILER, J. exp. Zool. 31, 117 (1920).

³ V. HAMBURGER, J. exp. Zool. 71, 379 (1938).

⁴ W. H. LEWIS, Anat. Rec. 4, 183 (1910).

⁵ W. H. LEWIS, Development of the Muscular System in Human Embryology (Lippincott Co., Philadelphia 1910), Chapter 12.

⁶ J. W. SAUNDERS, Anat. Rec. 100, 756 (1948).

⁷ J. MILAIRE, Arch. Biol. 68, 429 (1957).

⁵ E. C. ALBRITTON, Standard Values in Blood (Saunders, Philadelphia 1953), p. 53. – N. A. MICHELS, Downey's Handbook of Hematology, vol. I (Hoeber, New York 1938), p. 334.