Life of Guinea Pig Circulating Erythrocyte and its Relation to Erythrocyte Population of Bone Marrow.* (24923)

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In the course of quantitative studies of blood formation in the guinea pig(1,2,3) it became necessary to determine the life of the circulating red cell. The literature contains data on life span of red cells in 2 closely related forms, namely rat and rabbit, but no information about the guinea pig. In the rat Berlin and Lotz(4), after feeding glycine-2-C^{14}, estimated that the mean life span of the normal red cell was 64 days in the female and 68 days in the male. In the rabbit Neuberger and Niven(5), using glycine labeled with N^{15}, concluded that the mean life of the normal erythrocyte was about 65-70 days. In the present work we report efforts to determine the life of the circulating red cell in normal guinea pig by autoradiographic technic.

Technic. The work has been performed on 3 adult healthy male guinea pigs, of mixed strain, weighing approximately 400 g each. Each animal was given a single intraperitoneal injection of high specific activity† $Fe^{59}Cl_3$ (0.75 μc/g body weight), and blood smears were then made with blood obtained from ear vein from 12 hours to 8 days afterwards. The smears were fixed in methyl alcohol, then stained with haemalum and eosin. Coated radioautographs(6) were prepared using Eastman Kodak NTB-3 emulsion. The smears were finally stained differentially in 0.5% Giemsa solution at pH 6.4 for 12-15 hours at 5°C. Staining with either Giemsa or methylene blue before development is unsatisfactory, since methylene blue effects histochemical reduction of the emulsion(7). Preliminary staining with haemalum and eosin does not give rise to histochemical reduction and appears to improve subsequent Giemsa staining of cells through the emulsion.

Results. Radioautographs made of blood smears for the first several days after iron administration, provide for a clear separation between newly formed labeled erythrocytes and older non-labeled cells (Fig. 1). These labeled cells show a steady increase with time and at 7 days comprise 8.4% of the total (Fig. 2). Seven days, during which the rise is linear, is assumed to be the period in which there is enough Fe^{59} to label effectively all newly formed cells entering the blood stream. The radioautographs made at 8 or more days after Fe^{59} administration were not satisfactory for determining percent of labeled cells. This was due to reduced Fe^{59} content of cells entering the blood stream and thus the failure of single cells to produce distinct radioautographs. In the normal animal the level of circulating red cells represents a balance between newly formed cells and those undergoing destruction. If so, then in experimental animals in which 1.2% of newly formed red cells are daily entering the blood, the life of circulating red cells may be presumed to be $\frac{100}{1.2}$ *e.g.*, 83 days. This would make the life of guinea pig red cells appreciably shorter than that of the red cell in man, but longer than in the rat (64-68 days) or rabbit (65-70 days). It would be interesting to see whether application of other methods, to which reference has already been made, would confirm the figure thus obtained.

The significance of these data relates to the study of quantitative relationships between red cell populations of bone marrow and blood. Thus if one takes the data obtained in the Dunklin-Hartley strain of guinea pig(3,8) and assumes that the figures there given are applicable to our animals of mixed strain, then total circulating red cells are $157{,}766 \times 10^6$. If circulating red cells have a life of 83 days, then the daily requirement for newly formed red cells would be $1{,}900 \times 10^6$. The total nucleated red cell population of bone marrow is $2{,}616 \times 10^6$(3). This should

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† 1.3 mc/mg.

be quite capable of meeting the daily need for new red cells if the nucleated red cells of marrow have a mean doubling time of 33 hours. This is rather longer than the time arrived at in the previous calculation(3), based upon an estimated life of 70 days for circulating red cells, when it was concluded that the need of blood for new red cells would be met if the nucleated red cells of marrow had an overall doubling time of 28 hours.

The figure now obtained for the life of the circulating red cell, namely 83 days, makes the task of marrow rather less onerous than it would be on the basis of a life of 70 days. It should perhaps be noted that there is not a large reticulocyte reserve in guinea pig marrow(3) and therefore a need for additional red cells must in the main be met by increased formation of new cells. The extent to which the normal erythrogenic capacity is already taxed to its utmost is an important factor in considering the ability of marrow to cope with increased red cell formation.

Summary. Three male guinea pigs of mixed strain, and weighing approximately 400 g were given 0.75 μc of $Fe^{59}Cl_3$/g body weight and percentage of labeled cells appearing in blood was noted over 8 days. For the first 7 days there was a linear increase in percentage of labeled erythrocytes. On the assumption that this represents the normal rate of new red cell formation, it is calculated that the circulating red cell in the guinea pig has a life of 83 days.

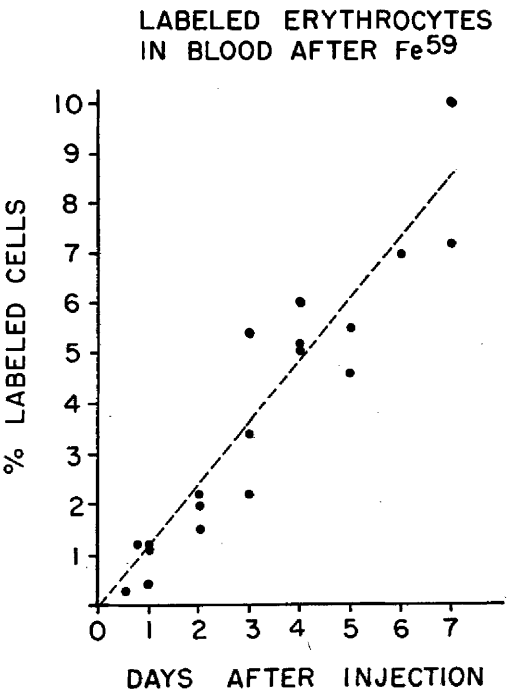

FIG. 2. Scattergram showing percentage of labeled erythrocytes in radioautographs of blood smears from individual animals at intervals after Fe^{59} administration.

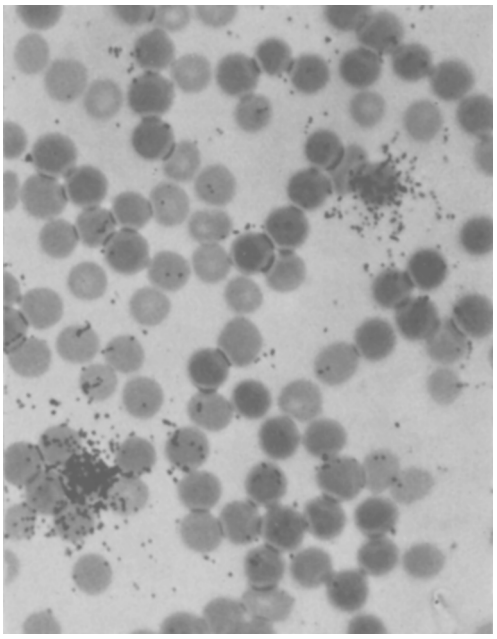

FIG. 1. Radioautograph of erythrocytes from blood smear made 24 hr after administering Fe^{59}. Two erythrocytes evidence characteristic labeling. 17 days' exposure. ×729.

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