FLOW CYTOMETRIC QUANTIFICATION OF PERIPHERAL BLOOD RETICULOCYTES AFTER RHODAMINE 123 STAINING

D. Grunwald, J. Prudhomme, P. Laroque, and G. Frelat
Commissariat a l'Energie Atomique, IPSN, DPS/SPE,
BP 6, F92265 Fontenay aux Roses, Cedex, France

ABSTRACT

The counting of the rare peripheral blood reticulocytes, which are immature red blood cells, can be used as an indicator of radiation exposure. However, the current microscopic method cannot be envisaged on large scale, and more rapid and precise techniques are needed.

Flow cytometry could be one of them. Indeed, some results have been still obtained in reticulocyte quantification by fluorescent measurement, after staining with acridine orange, pyronin Y, thioflavine and cyanin dye DIOC1(3). Nevertheless, the fluorochromes used have several drawbacks such as: difficulty in handling, apparatus specificity, low quantum yield, narrow Stroke's shifts, and no commercial availability.

We have set up a flow cytometric method (ATC 3000, ODAM-Bruker), using rhodamine 123, a common fluorescent probe for mitochondrion. The results obtained in reticulocyte detection in human and rat blood, showed close agreement between the manual method using brilliant cresyl blue, and the flow cytometric one. Indeed, the presence of mitochrondrions have been demonstrated by electronic microscopy, and allow to suggest a new operational definition of the reticulocyte.

Although further work is needed, this new method, incomparably more rapid and precise, seems promising for the determination of reticulocyte frequencies after irradiation exposure, especiall in case of mass examination.