These two papers describe a simple density gradient centrifugation technique for separation of lymphocytes and granulocytes from human blood. [The SCI® indicates that these papers have been cited in more than 580 and 910 publications, respectively.]

### Methods of Separation

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These two papers are both true offspring of my thesis,1 a section of which also became a Citation Classic.2 The 1974 paper was written to make a long story short. It was hardly encouraging that my thesis (109 pages) could be condensed to 4-6 pages, with a striking gain of precision. The 1976 paper is a short version that also represents an extension of the work, including some modifications and additional applications.

As the purpose of a Citation Classic commentary is to write with a personal touch, let me then at least give myself some credit for the development of a very simple technique for separation of lymphocytes and granulocytes from blood—not really spectacular, but probably the right technique at the right time. And, of course it had no negative effect on my self-esteem to find that it became widely used.

The two papers represent methodological work. My intention was to develop rapidly a cell separation technique that could serve as a basis for functional studies of leukocyte subgroups. It took several years! Much to my surprise, even cell separation, boring as it may seem, became interesting. Clearly, I had found my niche, in which the centrifuge and the microscope were indispensable partners. Maybe I had one special qualification for this work, a certain feeling of thrill and well-being at the microscope. And I certainly got the opportunity to enjoy this feeling. Nevertheless, after several hours, when my eyes were glistening with tears, it often became a matter of persistence rather than pleasure.

The work was based on some classical laws of motion and osmosis, applied to cells. I was particularly fascinated by how promptly the cells responded, by swelling or shrinking to small osmotic variations. This led to changes of volume and density in different cells, giving rise to different sedimentation patterns. A 2-3 percent osmotic change often had a significant impact on the flow of cells through the separation fluid.

For the human aspect, two episodes spring to mind. A young colleague in Moscow was anxious to know whether I had developed the Isopaque-Ficoll technique, which I could not deny. He then waved his cap, saying, “I am so happy. I use it every day.” There and then, I felt a flare of joy, as it appeared, for a moment, I had made a person happy.

Another story relates to the number of commercial products based on my recipes. My laboratory roommate in Norway met a professor in Portugal, and my name was mentioned. “He must be a rich man,” she said. I admit I have had my rewards, but not in terms of money.

As time goes by, more sophisticated techniques appear. Still, my old-fashioned centrifugation method serves the purpose, and it has proved to be a good starting point for many of the newer methods. And, as before, I am here in my little niche, fiddling with the gradient medium osmolality to possibly further improve separation techniques applied to blood from different species. And, it sometimes works.3,4 In the end, the key word is density—of cells and medium. As this British chap I met recently said: “Boyum[?]—oh, the density man.” Maybe that’s the name one leaves behind.

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