CC/NUMBER 38
SEPTEMBER 23, 1985

This Week's Citation Classic®

Osserman E F & Lawlor D P. Serum and urinary lysozyme (muramidase) in monocytic and monomyelocytic leukemia. *J. Exp. Med.* **124**:921-52, 1966.
[Department of Medicine, Columbia University College of Physicians & Surgeons, and Francis Delafield Hospital, New York, NY]

This paper describes the finding of markedly elevated concentrations of the enzyme lysozyme in the serum and urine of patients with monocytic and monomyelocytic leukemia. In addition to its clinical importance, this finding permitted extensive biochemical and physicochemical studies of human lysozyme. [The *SCI*® indicates that this paper has been cited in over 825 publications since 1966.]

Elliott F. Osserman
Department of Medicine
Institute of Cancer Research
College of Physicians & Surgeons
Columbia University
New York, NY 10032

November 19, 1984

For many years, we had investigated the immunoglobulin and Bence-Jones protein abnormalities in patients with multiple myeloma, a neoplastic disease of plasma cells. One of our patients had multiple myeloma and was excreting a kappa Bence-Jones protein in his urine. With chemotherapy, it was possible to suppress the abnormal plasma cells and virtually eliminate the production of Bence-Jones protein in the patient. Unfortunately, however, after 18 months of treatment, the patient suddenly developed monomyelocytic leukemia, which is a recognized complication of myeloma after long-term chemotherapy. At this point, we restudied his urine and confirmed that there was no Bence-Jones protein. However, electrophoresis of the urine proteins demonstrated a markedly basic, cationic protein (CP) in the far "post-gamma" mobility range, an abnormality that neither we nor anyone else had previously seen.

Within a few weeks, we had determined that virtually all patients with monocytic and monomyelocytic leukemia had this protein in their urine and it, therefore, was a specific biochemical marker for this group of leukemias. Over the next several months, we determined the molecular properties of the CP protein. Because of the circumstances of the first case, we were convinced that this was another "piece" of immunoglobulin, and we attempted to prove this—obviously with thoroughly negative results.

The "breakthrough" came when I finally accepted the fact that CP was *not* an immunoglobulin fragment and cleared my mind of this wrong preconception. Having done this, I went back to the initial and obvious fact that monocytes are precursors of macrophages and produce a wide variety of enzymes, e.g., ribonuclease, cathepsin, proteases, glucuronidase, elastase, and also lysozyme. We made a list of these enzymes and prepared to assay our purified protein for these specific activities. Several of the assays were set up in our own laboratory and others were carried out by friends who were working on macrophage functions. Among these was Zanvil Cohn of Rockefeller University, whose lab was set up for doing lysozyme assays. A few days after he received our sample, he called and said, "Elliott, I don't know what else you have in your sample, but there certainly is an enormous amount of lysozyme activity." Indeed, since our protein had been purified to homogeneity, it was clear that this was the enzyme itself. That day was probably the happiest and most exhilarating in my research career because it was obvious that we had identified a phenomenon with extremely wide and potentially important implications.

Over the next several months, we completed the basic biochemical, immunochemical, and clinical studies that were all assembled and published in this paper along with a description of a new assay method for quantifying lysozyme. This paper, which ran to over 30 pages in the *Journal of Experimental Medicine*, could easily have been divided into three or four separate publications, but it was my conviction that the information would be more readily accessible as a single publication. Happily, this proved to be true. Our finding made possible a host of subsequent studies of human lysozyme including the amino acid sequence determination that was carried out by my associate, Robert Canfield,[1] and crystallization, which we achieved.[2] The paper also permitted the crystallographic studies of lysozyme by Colin Blake and his associates at Oxford.[3] A very large number of other investigations have been and continue to be carried out on human lysozyme,[4] and this is the most gratifying and rewarding consequence of our work.

1. **Canfield R E, Kammerman S, Sobek J H & Morgan F J.** Primary structure of lysozymes from man and goose. *Nature* **232**:16-17, 1971. (Cited 115 times.)
2. **Osserman E F.** Crystallization of human lysozyme. *Science* **155**:1536-7, 1967. (Cited 20 times.)
3. **Blake C C F & Swan I D A.** X-ray analysis of structure of human lysozyme at 6 ångström resolution. *Nature* **232**:12-15, 1971. (Cited 60 times.)
4. **Osserman E F, Canfield R E & Beychok S**, eds. *Lysozyme*. New York: Academic Press, 1974. 641 p.

