

Bernfeld P. Amylases,  $\alpha$  and  $\beta$ . *Meth. Enzymology* 1:149-58, 1955.  
[Tufts University School of Medicine, Boston, MA]

This paper reviewed the state of the art of amylases in the early 1950s. It described their assay, purification, and properties, with particular emphasis on work of the Geneva (Switzerland) laboratory, where alpha-amylases from four different sources had just been obtained in crystalline form. [The SCI® indicates that this paper has been cited in over 1,165 publications.]

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During the late 1930s, research at the Chemistry Department of the University of Geneva (Switzerland), under the late Kurt H. Meyer, was directed toward macromolecular natural compounds, such as cellulose, starch, rubber, and silk. Having received a doctoral degree in chemistry a few years earlier, I found myself in charge of the group working on starch. End-group determinations of the two starch polysaccharides, amylose and amylopectin, pointed toward a linear structure of the former and a branched structure of the latter component.<sup>1</sup> It appeared desirable to confirm and expand our findings, resulting from applying chemical and physicochemical methods, by the use of biochemical techniques. Thus, research on amylases was initiated at Geneva.

All participating researchers, having been trained in organic chemistry, were accustomed to working only with many times distilled and/or recrystallized substances. Because of our work with high polymers, which did not lend themselves to such treatment, we were

already branded as "dirty" chemists. Nevertheless, amylose and amylopectin had been extensively fractionated and purified by us, and many of the subfractions carefully characterized. These precious products had now to be incubated with crude extracts of animal or plant tissues, because such was, with a few exceptions, the state of enzymes at that time. This was too much! And thus started the program of purifying amylases at the Geneva laboratory.

Fewer than a dozen enzymes had then been reported crystallized. By today's standards, the available methods for enzyme purification were primitive and rudimentary. Yet, within about 10 years, alpha-amylases from four different sources had been obtained in crystalline form and had been well characterized at the Geneva laboratory. These were the amylases from hog pancreas, human saliva, human pancreas, and from *Bacillus subtilis*. Credit is due to Edmond H. Fischer, Alfred Staub, François Duckert, and Maria Fuld, respectively, for their contributions. The results of these enzyme studies were all published in French.<sup>2-5</sup> When I came to the US in 1949, I found that the work on amylases by the Geneva group was little known in this country, and I decided to write two review articles on that general subject matter,<sup>6</sup> the second one being the paper under discussion. (For a more recent review on amylases see reference 7.)

Why has one of these papers become a "most-cited item in its field"? Amylases are intriguing enzymes, occurring in many different sources, their mode of action and their substrates being unconventional. Most of the publications on amylases from the Geneva laboratory had been in French and, hence, not easily accessible to many. Most important of all, the above-cited paper appeared in one of the foremost publications in the field, *Methods in Enzymology*.

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2. Meyer K H, Fischer E H & Bernfeld P. Sur les enzymes amylolytiques (I). L'isolement de l' $\alpha$ -amylase de pancréas. *Helv. Chim. Acta* 30:64-78, 1947.
3. Meyer K H, Fischer E H, Staub A & Bernfeld P. Sur les enzymes amylolytiques X. Isolement et cristallisation de l' $\alpha$ -amylase de salive humaine. *Helv. Chim. Acta* 31:2158-64, 1948.
4. Fischer E H, Duckert F & Bernfeld P. Isolement et cristallisation de l' $\alpha$ -amylase de pancréas humain. Sur les enzymes amylolytiques XIV. *Helv. Chim. Acta* 33:1060-4, 1950.
5. Meyer K H, Fuld M & Bernfeld P. Purification et cristallisation de l' $\alpha$ -amylase de bactérie. *Experientia* 3:411-12, 1947.
6. Bernfeld P. Enzymes of starch degradation and synthesis. *Advan. Enzymol.* 12:379-428, 1951. (Cited 235 times since 1955.)
7. French D. Amylases: enzymatic mechanisms. (Hollaender A, Rabson R, Rogers P, San Pietro A, Valentine R & Wolfe R, eds.) *Trends in the biology of fermentations for fuels and chemicals*. New York: Plenum, 1981. p. 151-82.