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This Week's Citation Classic

Laurell C B. Quantitative estimation of proteins by electrophoresis in agarose gel containing antibodies. *Anal. Biochem.* **15:**45-52, 1966.
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The paper describes a method for rapid quantitative analysis of proteins with a charge differing from that of the bulk of the immunoglobulins. The method utilizes the difference between the rate of electrophoretic migration of proteins and of their antibody complexes in agarose gel. [The *SCI*® indicates that this paper has been cited over 1,640 times since 1966.]

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"Quantitation of proteins by electrophoresis into agarose containing the corresponding antibodies has received the nickname rocket' (immuno) electrophoresis because the precipitates formed when the migrating protein antigens are precipitated by the antibodies resemble upright rockets. The method was later more thoroughly described under the name of electroimmunoassay, whereas electroimmunodiffusion is more frequently used in the US. The original method was invented to minimize the effect of diffusion! The historical background is as follows.

"I had found a slight difference in mean electrophoretic mobility of α_1-antitrypsin in normal plasma and in plasma from patients with α_1-antitrypsin deficiency. I assumed that the deficiency cases had a slightly abnormal protein in low concentration or that their α_1-antitrypsin was in complexed form. I wanted to demonstrate the occurrence of molecules with normal and retarded mobility in plasma from heterozygotcs with α_1-antitrypsin deficiency having only half the normal amount of α_1-antitrypsm in their plasma.

"On immunoelectrophoresis, the precipitation bows with rabbit anti-α_1-antitrypsin were too extended to allow any conclusions about one or two precipitation maxima to be drawn. Our agar gel electrophoresis had high resolution, but development of immuno precipitates by diffusion — the second step in immunoelectrophoresis — caused blurring. Therefore, I invented (antigen-antibody) crossed immunoelectrophoresis. Here the diffusion step of immunoelectrophoresis was exchanged for an electrophoresis of the separate proteins into a gel with mainly stationary antibodies. The precipitation of proteins in an electric field by antibodies was rapid enough to give precipitates with peak heights related to the amount of antigen applied.

"Tristram Freeman from Mill Hill visited us and became enthusiastic about crossed immunoelectrophoresis. He announced the following spring that he had developed a quantitative method which he wanted to present in June.[2] Our old collaborator, J. F Heremans, reported simultaneously that he had developed an immunochemical method for determination of plasma proteins together with Mancini.[3] Most plasma proteins had been isolated around 1960, creating an urgent need for fast methods of quantitative specific protein estimation. My experience from crossed immunoelectrophoresis suggested the logical development of a one-dimensional technique as a variant of crossed immunoelectrophoresis. Both the techniques could be used as alternatives to Mancini-Heremans single radial immunodiffusion.

"Of these methods, the 'Mancini' technique has been most used because of its simplicity and because the immunoplates are commercially available. Both will slowly lose in popularity and nephelometric methods will spread for a while, but radioimmunorockets' will probably have a good future in the protein concentration range of 5-0.1 mg/l Variants of crossed immunoelectrophoresis will probably spread more, because of its capacity to reveal antigenic microheterogeneity, indicate complex formation, and unmask partial antigenic identity."

1. **Laurell C B,** ed. Elcctrophorctic and electro-immunochemical analysis of proteins. *Scand. J. Clin Invest Suppl 124* **29**:l-36. 1972.
2. **Clark H G M & Freeman I**. A quantitative immuno-electrophoresis method (Laurell electrophoresis) (Peeters H.ed.) *Protides of the biological fluids*. Amsterdam: Flsevier. 1967. p. 503-9.
3. **Mancini G. Carbonara A () & Heremans J F.** immunochemical quantitation of single radial immunodiffusion. Immunochemistry **2**:235-54, 1965.