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This Week's Citation Classic™

Woods K R & Wang K T. Separation of dansyl-amino acids by polyamide layer chromatography. *Biochim. Biophys. Acta* **133**:369-70, 1967.
[New York Blood Center, New York, NY and Dept. Chemistry, National Taiwan University, Taipei, Taiwan]

The paper describes polyamide separation of dansyl derivatives. The terse, simple instructions were apparently easy to follow reproducibly in many laboratories, establishing the method as a standard analytical technique. [The *SCI*® indicates that this paper has been cited in over 1,505 publications since 1967.]

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"I have not met Kung-Tsung Wang to this day. He became my pen-pal collaborator for a short while and made a significant contribution to our repertoire of methods for unraveling protein structure.

"During 1966, I was privileged to serve the Rockefeller Foundation on a brief scientific mission to India and took the opportunity en route to visit Brian Hartley's laboratory in Cambridge where intensely fluorescent dansyl derivatives were being separated and identified on filter paper by high-voltage electrophoresis.[1] The wattage requirement was sufficient for an electrocution; thus, the apparatus and its lethal power supply had to be secured in a special limited-access room. The British are so heroic! I lacked the courage to duplicate that apparatus in my laboratory.

"After returning from India intending to find a safer way to separate dansyl-amino acids, I noticed an article by Wang *et al.* describing use of nylon-coated Mylar films for separating dinitrophenyl amino acids.[2] I was curious and wrote to Wang suggesting that his substrate should have just the right properties for partition chromatography of dansyl-amino acids. He promptly replied with an offer not to be refused. Explaining that his department could not afford dansyl-reference standards or reagents to synthesize them, he proposed an exchange. I went to the balance table; tipped a few crystals of each of my reference standards onto weighing papers; folded, labeled, and sealed them with Scotch tape; and sent them to Wang in an ordinary airmail envelope. Soon I received a return letter with a 2.5 × 15 cm. polyamide-Mylar strip enclosed. Many separation experiments were performed using that strip, rinsing away the dansyl compounds with ammonia or acetone between each trial. We quickly confirmed that solvent systems later to be described in our publication enabled consistently comparable separations both in Taipei and New York.

"And now for the rest of the story. About four years ago, I received a telephone call from the late Stanford Moore, an infrequent though hardly unique occasion. 'Woods?' he paused awaiting confirmation that indeed it was I, 'I have just returned from Taiwan. Have you ever met K.T. Wang?' 'No,' I replied, 'I have not.' 'I looked him up,' said Moore, 'and he's quite an undaunted scientist.' Moore went on to tell me he had learned that when Wang was a graduate student and came up with the idea of making polyamide laminates for chromatographic separations, it was Wang's professor who contributed the only readily available polyamide—his best white nylon shirt! Wang dissolved the shirt in formic acid (presumably having removed the buttons) and went to work coating window glass to make his first polyamide layers.[3]

"Now if it should happen that readers of this account have enjoyed the precision and simplicity of using polyamide layers for separating dansyl-amino acids, you will share my admiration for Wang's resourcefulness and appreciate the generosity of his professor who literally, for the sake of Wang's experiments, gave him the shirt off his back!

"Why do I think this publication has been so frequently cited? It is brief, fewer words than this commentary, and the technique is elegant for its simplicity, providing a rapid and sensitive means for separating dansylated products of proteins and peptides and identifying their N-terminal amino acids.

"An example of its persistence as a standard technique appeared as recently as Vãradi and Patthy's report from Budapest delineating a tripeptide segment of fibrinogen responsible for plasminogen binding.[4] The technique provided convenient help in sorting out essential enzymic fragments derived from fibrinogen."

1. **Gray W R & Hartley B S.** A fluorescent end-group reagent for proteins and peptides. *Biochemical J.* **89**:59P, 1963. (Cited 640 times.)
2. **Wang K T, Huang J M K & Wang J S Y.** Polyamide layer chromatography of dinitrophenyl amino acids. *J. Chromatography* **22**:362-8. 1966.
3. **Wang K T, Wang J S Y & Lin A L.** Polyamide thin layer chromatography. *J. Chin. Chem. Soc. Ser. II* **8**:241-50. 1961.
4. **Vãradi A & Patthy L.** $\beta(Leu_{121}\text{-}Lys_{122})$ segment of fibrinogen is in a region essential for plasminogen binding by fibrin fragment E. *Biochemistry—USA* **23**:2108-12. 1984.