Use of 8-quinolinol as a developing reagent for amino acids.

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Ninhydrin has been used as an effective developer for the identification of amino acids since 1910.[1] Improvements in the procedure were made by addition of metal ions such as zinc,[2] copper,[3] cadmium,[4] strontium[5] etc. These metal ions form a complex which give a bathochromic shift and is relatively stable. The chromatogram sprayed with ninhydrin containing one of these ions retain the coloured spots for upto six weeks[5] where as ninhydrin colour is stable for about one hour. Several modification[6] for selective identification of amino acids have also been made and addition of bases like collidine[7] and cyclohexylamine[8] in ninhydrin impart specific colours. Smith etal[9] have described the production of orange red spot on using 8-quinolinol-hypobromite for developing arginine and other guanidine derivatives. However, no further investigation of its reactions with amino acids is reported elsewhere.

The identification of amino acids in nano-mole quantity has been possible by flourometric reactions. This is 10-100 time more sensitive than the assay with ninhydrin. Fluescin, was used by Roth[10] for this assay, and DNS-Cl[11] for coupling with the amino acids and identified as flourscent amino acids spots. Stein etal[12] used flourescamine for the identification of amino acids at pico mole level.

In this communication, we are reporting the use of 8-quinolinol as a spraying reagent in the identification of amino acid on paper and TLC. The detection limit of amino acid complex with this reagent is as low as 10^{-10}M. The spraying reagent is made up from following:-

8-quinolinol,	200mg
Acetic acid	40ml
Pyridine	1.5ml
De-ionized water	60ml

(Better results were obtained when freshly prepared solutions were used).

The chromatogram after drying was sprayed with the reagent and dried in an oven at 60° for 5-10 minutes. Green-yellow spots were obtained for all the protein amino acids except Phe, Pro, Val and Arg, under U.V. (308 and 240 nm) were identified as which dark orange spots. However when chromatogram was sprayed with 1% NH_4OH and dried as earlier these amino acid also exhibited dull-green yellow flourescent spots.

The reaction mechanism of 8-quinolinol, and its use for a quantitative flourescence developer is under study and shall be reported later.

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