

STUDIES ON THE MOBILITY OF AMINO ACIDS IN SOILS BY SOIL THIN LAYER CHROMATOGRAPHY

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The mobility of some cyclic and aliphatic amino acids in terms of R_F value in two soils of diverse nature were determined by soil thin layer chromatography. The influence of particle size, exchangeable cations and pH on the movement was studied. The behaviour, wherever marked, is explained on the basis of adsorption and some other characteristics of the soils and amino acids.

INTRODUCTION

AMINO ACIDS play a vital role in biochemical processes (Theng 1974). They can be adsorbed on clays forming clay-amino acid complexes (Talibudeen 1955). Their products such as indoles (Waksman 1952) act as growth promoting substances and hormones in plants. In view of the importance of amino acids and their relation to leaching of soil $\text{NO}_3\text{-N}$, it was considered appropriate to examine the mobility of some amino acids in a group of two diverse soils. The effect of certain variables on mobilities was also determined. Soil thin layer chromatography (Helling & Turner 1968; Singhal *et al.* 1977; and Singhal & Bansal 1978) was used to measure the mobility.

MATERIALS AND METHODS

The soils selected for the investigation were an illitic alkaline saline silt loam from Aligarh (Sand = 33%, Silt = 63%, Clay = 3%, CEC = 16.3 meq/100 g, pH = 9.2 and E.C. = 5.0 mmhos cm^{-1}) and a red kaolinitic acidic sandy loam from Bangalore (Sand = 70%, Silt = 17%, Clay = 12%, CEC = 3.5 meq/100 g, pH = 5.7 and E.C. = 1.2 mmhos cm^{-1}).

Among the materials the soils with particle sizes ≤ 212 , ≤ 150 and ≤ 75 micron respectively were used as static phase. For an examination of the effect of cations on the mobility the ≤ 150 micron natural and Na and K soils were used as static phase. For a study of the pH effect, unbuffered distilled water and water adjusted to pH 2 and 12 respectively, were used as developers.

For the measurement of mobility in terms of R_F values, a slurry of prewashed soil samples was prepared in distilled water. The slurries (30 g soil + 60 ml water per plate) were applied on glass plates (20 × 20 cm) to a thickness of 0.5 mm with the help of an applicator and then air dried at room temperature. Five μl and 15 μl of 0.01M amino acid solutions per spot were applied with a micropipette on triplicate soil TLC plates. The plates were developed in closed chambers, using distilled water as the developing solvent.

After air drying visualization of the mobility was done with 0.2% ninhydrin in *n*-butanol (*w/v*) as a detection reagent (Hais & Macek 1963). "Frontal R_F " value was then measured directly by the distance moved by the amino acid in mm (Helling 1971). Some typical chromatograms are given in Fig. 1. The results are given in Tables I-III.

TABLE I
Effect of particle size on the R_F value of amino acids in natural soil

Amino acid	R_F value in Aligarh silt loam			R_F value in Bangalore sandy loam		
	< 212 micron	< 150 micron	< 75 micron	< 212 micron	< 150 micron	< 75 micron
Glycine	0.77	0.98	0.91	0.44	0.58	0.28
Alanine	0.95	0.97	1.00	0.75	0.45	0.72
Valine	0.98	1.00	1.00	0.85	0.71	0.78
Leucine	0.93	1.00	0.86	0.83	0.66	0.81
Phenyl-alanine	0.90	1.00	0.98	0.72	0.72	0.63
Tyrosine	0.82	0.93	0.93	0.35	0.27	0.40
Tryptophan	0.74	0.89	1.00	0.48	0.30	0.66

TABLE II
Effect of cations on the R_F value of amino acids in soils at < 150 microns and natural pH

Amino acid	R_F value in Aligarh silt loam			R_F value in Bangalore sandy loam		
	Natural-soil	Na-soil	K-soil	Natural soil	Na-soil	K-soil
Glycine	0.98	0.91	0.98	0.58	0.35	0.42
Alanine	0.97	1.00	1.00	0.45	0.54	0.31
Valine	1.00	1.00	1.00	0.71	0.60	0.59
Leucine	1.00	1.00	1.00	0.66	1.00	0.84
Phenyl-alanine	1.00	1.00	1.00	0.72	0.96	0.75
Tyrosine	0.93	0.93	0.94	0.27	0.26	0.25
Tryptophan	0.89	0.86	0.98	0.30	0.47	0.29

TABLE III
Effect of pH on the R_F value of amino acids

Amino acid	R_F value in Aligarh silt loam			R_F value in Bangalore sandy loam		
	pH-2	pH-7	pH-12	pH-2	pH-7	pH-12
Glycine	0.96	0.98	0.88	0.40	0.58	0.35
Alanine	0.96	0.97	1.00	0.84	0.45	0.93
Valine	0.97	1.00	1.00	0.85	0.71	0.68
Leucine	1.00	1.00	1.00	0.79	0.66	0.71
Phenyl-alanine	1.00	1.00	1.00	0.79	0.72	0.70
Tyrosine	0.88	0.93	0.70	0.47	0.27	0.35
Tryptophan	0.90	0.89	0.94	0.74	0.30	0.60

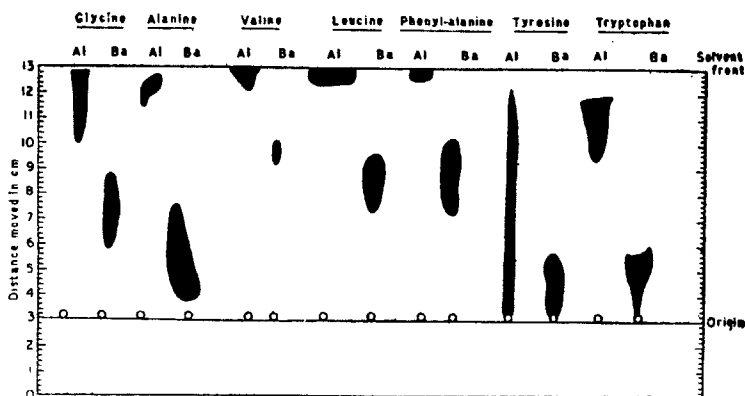


Fig. 1. Mobility of amino acids in untreated alkaline, saline silt loam from Aligarh and the red, acidic sandy loam from Bangalore. (Al = Aligarh, Ba = Bangalore).

RESULTS AND DISCUSSION

An examination of the R_F values of the cyclic as well as aliphatic amino acids (Tables I, II & III and Fig. 1) showed that the mobility of all the amino acids at different particle sizes, exchangeable cations and pH were higher in the illitic alkaline silt loam from Aligarh than in the kaolinitic acidic sandy loam from Bangalore. The difference in the texture, pH and composition of the two soils under study resulted in a difference in the adsorptive capacity, capillarity and permeability of the two soils. Thus the lower adsorptive capacity of Aligarh soil caused by saturation of its sites due to salinity caused the amino acids to move faster in it while the complexation of the amino acids with ferruginous matter of the Bangalore loam caused them to become less mobile in the red loam.

The R_F of the cyclic amino acids at all pH values, exchangeable cations and particle sizes of the red loam followed the order phenyl alanine > tryptophan > tyrosine. This order was the same as the solubility of the amino acids in water. The nature of the functional group in these amino acids also appeared to control the R_F value e.g., the presence of OH group in tyrosine resulted in hydrogen bonding causing a resistance in its movement and hence a fall in R_F . Similarly the presence of NH group in tryptophan caused it to move less faster than phenyl alanine. This order of mobility was, however, not followed so regularly by the above amino acids in Aligarh soil.

A study of the effect of particle size (Table I) did not yield a definite correlation between particle size of the soils and the R_F values of the amino acids.

An examination of Tables I and II further showed that in red sandy loam, the order of R_F values of the amino acids was in accordance with an increase in their molecular weight and pK_b . The former fact was in accordance with the observations of Hais and Macek (1963). No such correlation could be established in case of Aligarh silty loam.

An examination of the effect of the change of cation of the soil on the mobility of amino acids (Table II) showed that the R_F values of alanine, valine, leucine and

phenyl alanine were the same in both the Na-Soil as well as K-Soil from Aligarh. However, glycine, tyrosine and tryptophan exhibited higher mobilities in K-Soil from Aligarh. On the other hand in Bangalore loam, except for glycine the R_F values of the other amino acids were higher in Na-Soil than in K-Soil. Thus while Na-saturation increased the mobility of most of the amino acids in Bangalore loam, it was K-saturation which brought about such an effect in case of some amino acids in Aligarh loam.

An examination of the pH of the ascending developer (Table III) indicated that while the lower pH ($= 2$) of the developer favoured an increase in R_F in red sandy soil, it was either the higher or the neutral pH (12 or 7) which caused this effect in Aligarh soil. Dissolution of ferruginous matter at lower pH in red loam and of organic matter at higher pH in saline loam decreased the adsorptive capacity of the soils bringing about respective increases in the R_F value of the amino acids in the two soils.

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