Table 38 hR_f ($R_f \times 100$) Values of Enantiomers of DL-Amino Acids Resolved on Plates Impregnated with (1*R*,3*R*,5*R*)-2-Azabicyclo[3,3,0]octan-3-Carboxylic Acid

DL-Amino acid	Solvent system, acetonitrile–methanol–water	Pure L	hR_f Value from DL mixture	
			L	D
Arginine	7:6:3	13	13	6
Histidine	7:6:2	25	25	18
	3:1:1[a]	36	36	11
Lysine	10:5:2	31	31	15
Leucine	10:4:3	21	21	11
Valine[b]	10:5:2	50	50	38
Serine	7:1:1[a]	18	18	06
Tryptophan	8:1:1[a]	62	63	38

Time: 30–35 min; solvent front, 8.5 cm; detection, ninhydrin (0.2% in acetone); temp., 26 ± 2°C.
[a]Time: 25–30 minutes for 10 cm run (from Ref. 156a).
[b]At 20 ± 2°C (from Ref. 165d).

A new chiral reagent, NSP-Cl, was synthesized and used to derivatize amino acids, and the resulting diastereomers were resolved by TLC (166). Chiralplate with MeCN–MeOH–H_2O (4:1:1) as mobile phase was used to evaluate reaction products in the synthesis of modified Phe and Tyr derivatives (167) and also to separate aspartame and its precursor stereoisomers (168). Tryptophans and substituted tryptophans were separated on cellulose layers developed with copper sulfate solutions (169) when excess Cu^{2+} ions decreased the chiral discrimination of the system, and with aqueous α-cyclodextrin (1–10%) plus NaCl solutions (0.1, 0.5, 1.0 M) when the best results were obtained with aqueous 4% α-cyclodextrin–1 M NaCl solution (170), comparable to Chiralplate. It was observed that chiral effects are essentially additive (for cellulose and α-cyclodextrin) and there is a strong temperature dependence for the chiral separations.

Enantiomeric RP-TLC separations of amino acids and derivatives were obtained with α- and β-cyclodextrins (171), hydroxypropyl-β-cyclodextrin (172), and bovine serum albumin (173–176) in the mobile phase. Chiral monohalo-*S*-triazines were used for the TLC resolution of DL-amino acids (177). Racemic dinitropyridyl, dinitrophenyl, and dinitrobenzoyl amino acids were separated on RP-TLC plates developed with aqueous organic mobile phases containing bovine serum albumin as a chiral agent (178).

VII. QUANTIFICATION

Thin-layer chromatography is supplemented with spectrophotometric methods for the quantification of amino acids and their PTH and DNP derivatives.

A. Amino Acids*

1. Materials

Materials consist of standard solutions of amino acids, acetate buffer (4 M, pH 5.5), ethanol (50%), methyl Cellosolve (ethylene glycol monomethyl ether), and ninhydrin reagent (0.9 g of ninhydrin and 0.12 g of hydrantin dissolved in 30 mL of methyl Cellosolve and 10 mL of acetate buffer, prepared fresh).

*Based on Ref. 180.