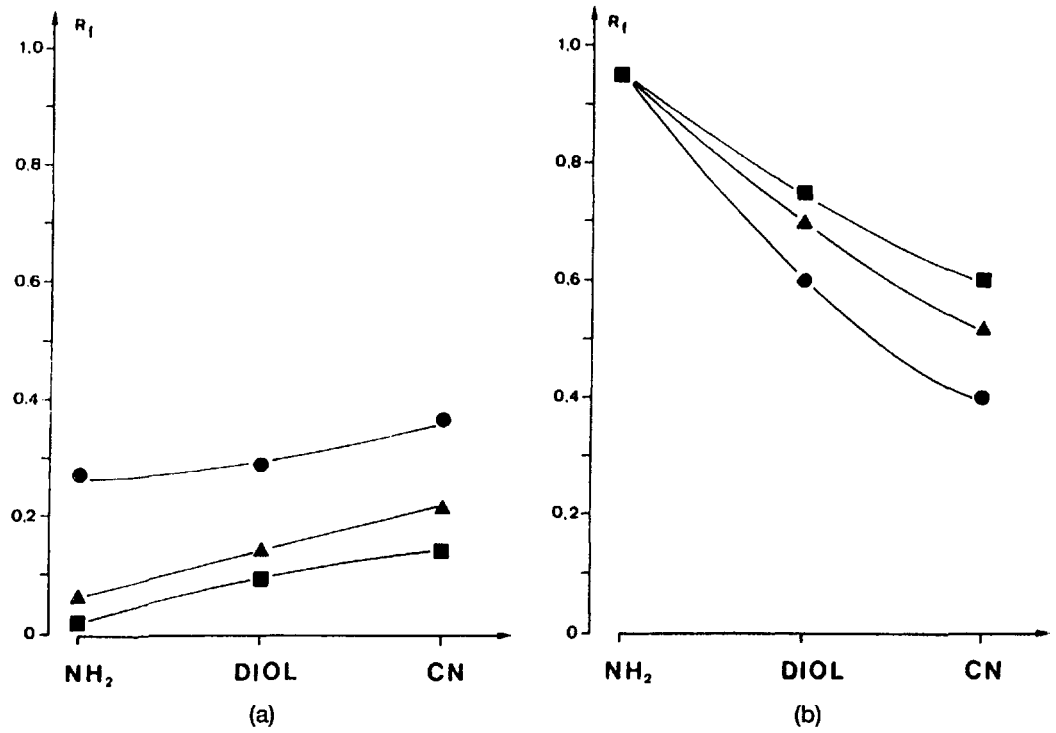

Figure 9 The influence of different hydrophilic modifications on R_f values of steroids. Plates: HPTLC precoated plate NH_2 F254s, diol F254s, CN F254s. Eluents: (a) Normal-phase system, petroleum ether (40–60°C)–acetone (80:20). (b) Reversed-phase system, acetone–water (60:40). Migration distance 7 cm. Normal chamber without saturation. Compounds: (■) Cortisone; (▲) corticosterone; (●) cortexone. Detection at 254 nm.

3. Sorbents and Precoated Layers for Ion-Exchange Chromatography

The ion-exchange mechanism has only minor importance in TLC. With the advent of genetic engineering, the possibility of a re-evaluation of this mode seems possible. At present, silica gels, celluloses, and organic polymers are used as matrices for functional groups suitable for performing ion-exchange separations.

a. Amino-Modified Precoated Silica Layers. The amino-modified precoated layer discussed in Section III.A.2 is not only suitable in normal-phase and RP chromatography, it can also act as a weakly basic anion exchanger. In this special case, the functional groups of the stationary phase, present in the $—NH_3^+$ form, show interactions that are different in strength with differently charged anions. Therefore, it is possible to influence the intensity of retention in a definite way by varying the concentration of an added salt, i.e., varying the ionic strength of the mobile phase.

A typical example of the use of an NH_2-modified precoated layer in ion-exchange chromatography is shown in Fig. 10. In this example, adenosine triphosphate (ATP) with a charge of -4 has the greatest retention (lowest R_f value). Adenosine diphosphate (ADP) (-3) and adenosine monophosphate (AMP) (-2) show increasingly higher R_f values. The noncharged nucleoside adenosine is eluted with the solvent front. Further areas of application besides the nucleotides include, e.g., carboxylic and sulfonic acids (171).

b. Modified Celluloses. Cellulose, described in Section I.D, was the first sorbent in thin-layer chromatography to be used for ion-exchange mechanisms after suitable modifications or impregnations (172). Functional groups used for chemical modification of celluloses are the following: