solvent system, these techniques can be suitable for separation of mono- and disaccharides or for the separation of higher oligosaccharides and maltodextrins. The separation can sometimes be improved by the use of additives such as boric acid or boronic acid (64,65), buffer solutions (66), or inorganic salts (14).

Some natural sugar derivatives need special separation conditions. Uronic acids, for instance, are best separated by using solvent systems that contain acetic, phosphoric, or hydrochloric acid (50,51), and some amino sugars and their derivatives can be satisfactorily separated with solvent systems containing ammonia.

Glycolipids are typically resolved using solvent mixtures that contain organic solvents such as chloroform or hexane, alcohols such as methanol or isopropanol, and water. A frequently used separation system for neutral glycolipids is chloroform–methanol–water (65:25:4) and, for the charged gangliosides, chloroform–methanol–water (60:35:8).

C. Mobile-Phase Additives

The selectivity of separation of some closely related carbohydrates can be modified by mobile-phase additives. Typical additives include boric acid, phenylboronic acid, and 2-aminoethyl diphenyl borinate (NST) (64,65,67,68). The reaction of polyhydroxy compounds with boric acid or boronic acids has been used for derivatization and separation of carbohydrates and other compounds containing vicinal diols by using chromatographic and electrophoretic techniques (7a,11b,69). The mechanism of reaction is a complex between *cis*-diol moieties and borate or boronate groups. It has been demonstrated that the borate ion, rather than boric acid, is complexed by the polyol (70,71). The reaction is pH-dependent, and the optimum conditions are usually at pH >8.0. In a pH ranging from 8 to 12, aqueous borate solutions contain tetrahydroxyborate ions and also more highly condensed polyanions such as triborate and tetraborate. Equilibrium between the different species depends on the pH and the total borate concentration. The migration of the resulting complexes of sugars and boric or boronic acids on thin layers is dependent on their polarity. With solvent systems containing boric acid, the migration of some sugars is inhibited, whereas certain sugars have increased R_f values when separated by TLC using mobile phases containing phenylboronic acid (65). Furanoses more readily form complexes or esters than sugars in the pyranose form. Fructose (α-D-fructofuranose) reacts with boric acid or phenylboronic acid at weak acidic pH. This reaction has been exploited to enhance the selectivity of separation of glucose and fructose on silica gel and cellulose thin layers (64,65,67,68) (Figs. 2 and 3). Separation is dependent on the polarity of the additive, its concentration (Fig. 4), pH, and the composition of the buffer. The concentration of the additive also influences the shape of the spots. When additives result in spots that are elongated, for example, separation can be improved by reducing the concentration of additive, by developing the plate at a subambient temperature, or by performing the separation with a more appropriate buffer.

IV. DETECTION

Carbohydrates show very low ultraviolet (UV) absorption. They can be satisfactorily visualized and evaluated on TLC plates only after suitable derivatization. The majority of chemical derivatization procedures are based on the reductive properties of carbohydrates. Reductive amination of sugars in the presence of an acid is a typical example. Methods based on reductive amination require an aldehydic reducing carbon on the saccharide that reacts with the amino group of the chromophore or fluorophore.

A. Prechromatographic Derivatization

Prechromatographic derivatization of carbohydrates is popular in elution chromatographic techniques (7a,7b), but is rarely used in planar chromatography because of the time-consuming derivatization reactions of individual samples compared to the relatively simple postchromatographic derivatization of the whole plate, the limited number of suitable reagents, and the relatively high detection limits, which are comparable to those obtained by postchromatographic derivatization.