

Aqueous Normal Phase ANP HPLC what is it – FAQ

Aqueous Normal Phase chromatography (ANP) is a chromatographic technique which uses the mobile phase region between Reversed Phase chromatography (RP) and organic Normal Phase chromatography (NP) used mainly to separate polar compounds such as acids, bases and peptides..

ANP methods use a slightly hydrophobic stationary phase such as silica hydride (*available as Cogent TYPE-C™ HPLC columns*) a mobile phase of 98% organic solvent (*such as acetonitrile*) with 2% water and 0.5% formic acid, TFA or similar acids or bases such as ammonium acetate, ammonium formate, as the “**B solvent**” and HPLC grade water with the same acid or base as the “**A solvent**” in a gradient.

In the presence of a high concentration of the B component the hydrophobic stationary phase will adsorb negatively charged hydroxyl ions from the water component of the mobile phase creating a negatively charged column surface environment. Ionized polar compounds will either be attracted to the negative charge or will compete with the negative charge on the surface and be retained. As the water content increases the polarity of the mobile phase increases and the polar compounds are more attracted to the water than to the hydrophobic, charged surface of the silica.

This mode of chromatography has many advantages to chromatographers including run to run **precision**, extremely fast equilibration, unique selectivity and long column life time.

Note: all Cogent TYPE-C Silica™ can be used with ANP, Reversed Phase or Normal Phase methods and can transition between mode to mode with little hysteresis. Depending on the bonded phase of these columns, some are more suited for ANP than others and some more suited for RP or a combination of both.



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