MICROSOLV

Aqueous Normal Phase defined & compared to Reversed Phase & Normal Phase definitions HPLC - Tech Information

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Understanding ANP, Reversed Phase, and Normal Phase Chromatography

If you're new to HPLC (High Performance Liquid Chromatography) or somewhat experienced, understanding the differences between **Normal Phase (NP)**, **Reversed Phase (RP)**, and **Aqueous Normal Phase (ANP)** chromatography can help you choose the right method for your analysis.

🔹 Normal Phase (NP) Chromatography

- **How it works:** The column is polar, and the mobile phase is non-polar.
- **Retention behavior:** Polar compounds stay on the column longer (more retention) when the mobile phase is **less polar** (e.g., 100% hexane).
- **Use case:** Best for separating polar compounds using non-polar solvents.

🔹 Reversed Phase (RP) Chromatography

- **How it works:** The column is non-polar, and the mobile phase is polar (usually water mixed with an organic solvent like acetonitrile or methanol).
- **Retention behavior:** Non-Polar compounds are retained longer as the mobile phase becomes **more polar** (e.g., increasing water content).
- **Use case:** Most common HPLC method, ideal for a wide range of compounds.

🔹 Aqueous Normal Phase (ANP) Chromatography

- **How it works:** ANP uses **reversed phase solvents** (water and acetonitrile) but behaves like **normal phase** chromatography.
- **Mobile phase:** Typically high in acetonitrile (up to 98%) with a small amount of water (2%) and sometimes an acid or base.
- **Retention behavior:** Polar Compounds are retained longer in **high acetonitrile** (low water) conditions. As you increase the water content, retention decreases.
- **Use case:** Great for polar compounds that don’t retain well in reversed phase but are difficult to work with in traditional normal phase.

🧪 Summary

Method	Column Type	Mobile Phae Polarity	Retention Increases with...	Targets
Normal Phase	Polar	Non-Polar	Less polar mobile phase	Polar Compounds
Reversed Phase	Non-Polar	Polar	More polar mobile phase	Non-Polar Compounds

ANP	Polar-like	Reversed Phase Solvents	Less polar mobile phase	Polar compounds

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🔗 **Want to learn more?**
Check out these resources for deeper explanations:

- Why Aqueous Normal Phase Methods are better than HILIC
- Wikipedia: Aqueous Normal Phase Chromatography

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Attachment:

Using Aqueous Normal Phase White Paper 0.2 Mb Download File

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