

# Test Chromatogram

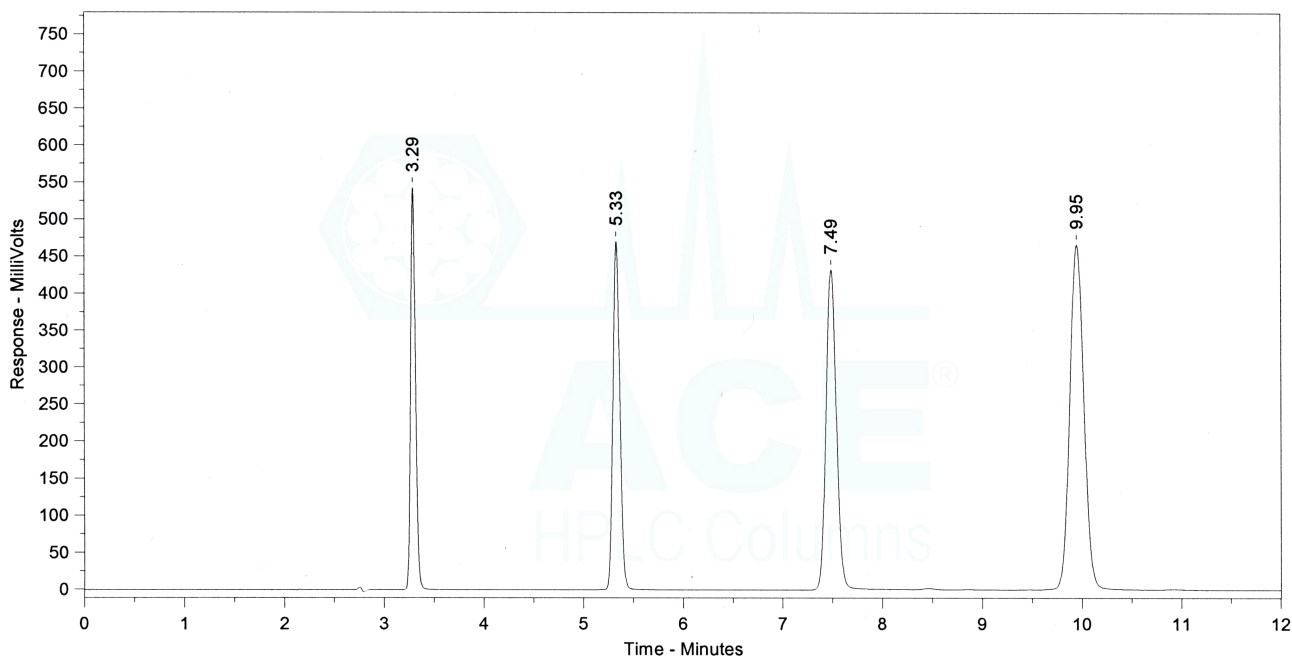


## COLUMN DETAILS:

Catalogue Number: ACE-121-2546      Serial Number: A96000  
Packing: ACE 5 C18      Length: 25cm  
Batch: V11-5080      Internal Diameter: 4.6mm

## EVALUATION CONDITIONS:

Mobile Phase: MeOH:H2O (85:15)      Storage Solvent: Mobile Phase  
Flow Rate: 1.0 ml/min      Wavelength: 254nm  
Pressure: 1400psi      Temperature: 22°C



## TEST RESULTS:

Peak	Identity	$t_r$ (minutes)	Efficiency		Asymmetry Calculations	
			$N_{0.5}$	%	$As_{0.1}$	USP Tailing Factor
1	Dimethyl Phthalate	3.29	21700	94	1.26	1.16
2	Toluene	5.33	28300	97	1.15	1.09
3	Biphenyl	7.49	25400	96	1.07	1.05
4	Phenanthrene	9.95	23900	99	1.07	1.04

Please see overleaf for care and use, asymmetry and efficiency information.  
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# Care and Use of ACE® HPLC COLUMNS

Every ACE® HPLC column is individually manufactured and validated to exceed stringent specifications. The following measures will enhance its performance and lifetime.

## Column Installation

**System dead volume:** Always ensure that the injection valve and flow cell introduce the smallest possible dead volume. Connections between injector, column and detector should be kept as short as possible, preferably using tubing with an internal diameter of 0.010" (0.25mm) or less.

**Column connection:** For optimum performance, the tubing connecting the column to injector and detector must abut the internal shoulder of the fitting. The use of PEEK fingertight fittings is recommended for such connections. The direction of flow is marked on the column.

**Mechanical damage:** Protect the column from mechanical shock. Dropping a column can impair its performance.

**Equilibration:** Flush onto the desired mobile phase (ensure that precipitation does not occur). Ensure that the column is fully equilibrated prior to starting an analysis.

**Performance testing:** It is recommended that the performance of columns is tested on arrival and periodically during use. Performance parameters are as defined below.

**Guard cartridges:** The use of a guard cartridge is strongly recommended to prevent both inlet frit blockage and irreversible sample adsorption onto the top of the column. Guard cartridges are available for all ACE® columns

## Operational Guidelines

**HPLC solvents:** Use only HPLC grade solvents and freshly prepared aqueous buffer solutions to minimise bacterial growth. A slip-on pump filter will remove extraneous particles.

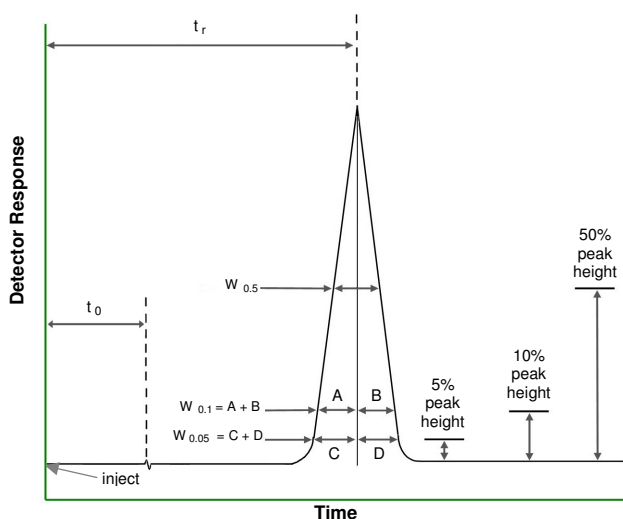
**Mobile phase pH:** To ensure maximum column lifetime, a pH range of 2.0 - 8.0 should be maintained. When operating at high pH, the use of an organic buffer is recommended.

**Pressure:** Exposure to rapid changes in pressure or to pressures >4000psi (275 bar) may reduce column lifetime.

**Temperature:** Exposure to temperatures >60°C may reduce column lifetime.

**Column Warranty:** All columns are warranted to meet the specifications stated on the Test Chromatogram and any Batch Validation documents supplied with the column. Removal of an end fitting to replace a frit or top-up the packing material should be regarded as a last resort to prolonging column lifetime. Removal of the column end fittings will automatically invalidate the column warranty.

**Storage:** Wash out any buffer (ensure that precipitation does not occur) and flush onto the storage solvent defined overleaf. Replace the end-stops to prevent the packing bed drying out and store in a cool area.



### Performance

At 50% peak height

$$N_{0.5} = 5.54 (t_r / W_{0.5})^2$$

At 10% peak height

$$N_{0.1} = 18.55 (t_r / W_{0.1})^2$$

### Peak Shape

Overlap to a perfect Gaussian peak

$$\% = N_{0.1} / N_{0.5}$$

Peak asymmetry (measured at 10% peak height)

$$As_{0.1} = B/A$$

US Pharmacopeia Tailing Factor (measured at 5% peak height)

$$T = W_{0.05} / 2C$$

### Selectivity

Capacity factor

$$k' = (t_r - t_0) / t_0$$

**Safety and Disposal:** This column contains amorphous silica which may be hazardous to health if the column is unpacked and the silica allowed to dry. The silica presents no hazard whilst contained within the column. When the column has reached the end of its life, dispose of it in a similar manner to the samples that have been injected onto it.