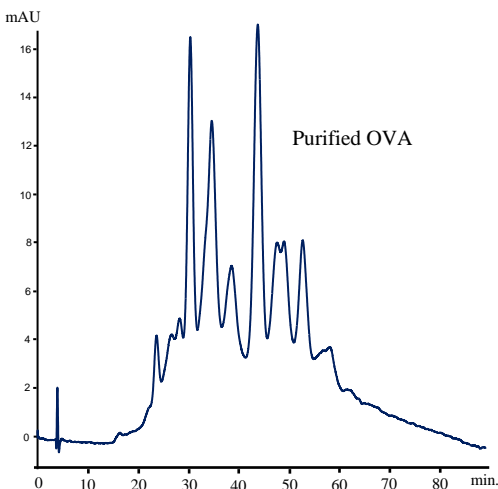


APPLICATION NOTE

Comparison of STYROS™ HPA Anion Exchanger with STYROS™ 3R Reversed Phase In the Assessment of Commercial OVA.

The following 2 chromatograms show the profile variations of the same sample of commercially purified OVA on both a weak anion exchanger and a reversed phase.



Chromatogram 1

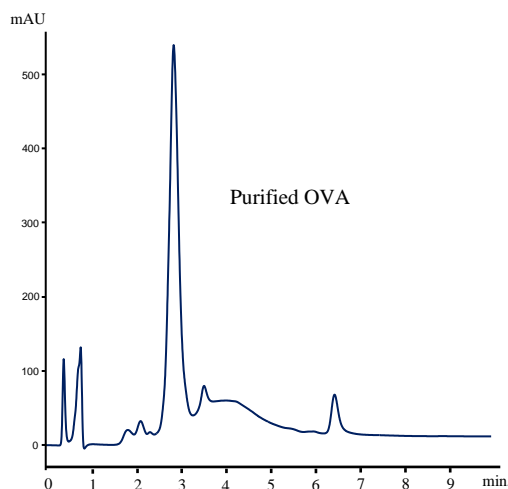
Commercial OVA on **STYROS™ HPA/XH** 4.6 x 300 mm at 1 ml/min (360 cm/hr)

Table 1. Operating parameters.

HPLC System.	Agilent 1100 with thermostatted column compartment and quaternary pump.
Columns	STYROS™ HPA/XH 4.6 X 300 mm (4.98 ml)
Mobile phase.	A: 20 mM Tris, pH=8.2 B: A + 1 M NaCl, pH= 8.2
Flow rates	1 ml/min (360 cm/hr of linear velocity)
Gradient	6 to 35 % B in 18 cv
Temperature	30°C
Detection	280 nm
Pressure Drop	18 bar (261 psi)
Sample:	Commercial OVA (sold as 98 % pure)

The weak anion exchanger with polyamine coating can interact with the charges of the protein in addition to the hydrophobic patches.

It is therefore able to move far beyond the characterization capabilities of gel electrophoresis or reversed phase media.



Chromatogram 2

Commercial OVA on **STYROS™ 3R/XH** 4.6 x 50 mm at 1 ml/min (360 cm/hr)

Table 2. Operating parameters.

HPLC System.	Agilent 1100 with thermostatted column compartment and quaternary pump.
Columns	STYROS™ 3R/XH 4.6 X 50 mm (0.83 ml)
Mobile phase.	A: H2O B: ACN
Flow rates	1 ml/min (360 cm/hr of linear velocity)
Gradient	20 to 85 % B in 6.25 cv
Temperature	30°C
Detection	214 nm
Pressure Drop	10 bar (145 psi)
Sample:	Commercial OVA (sold as 98 % pure)

It is important to be aware of the detection characteristics of the process by which the chromatic run has been carried however the fact remains that the assessment of purity needs to be looked from different angles.

Application Note 86 provides a good baseline of what is to be expected if one were to isolate Albumin from the egg white in a pure and unaltered form.

