

APPLICATION NOTE

Quality Control of Proteins from Different Lots of the Same Supplier.

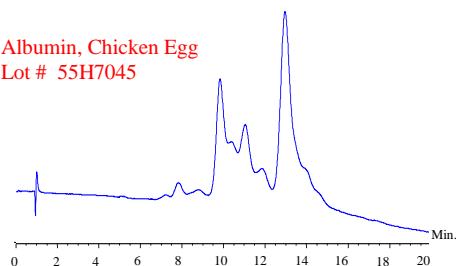
The QC manager is always looking for a fast, reliable method to check the incoming protein batches for purity, impurities, and lot to lot variation –critical information for end-users.

HPLC is routinely used as a powerful tool to make such characterization.

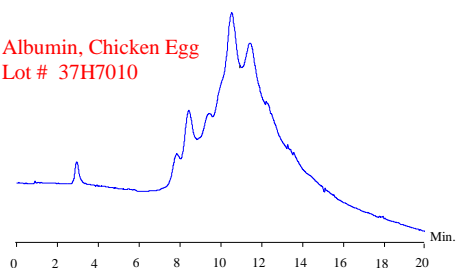
The high resolution offered by ion exchange chromatography of proteins could be used to detect and quantitate even the slightest change in composition across batches or lots of proteins.

The chromatograms below were each performed on a preparation of chicken egg albumin described by the supplier as “99% pure by Agarose electrophoresis”.

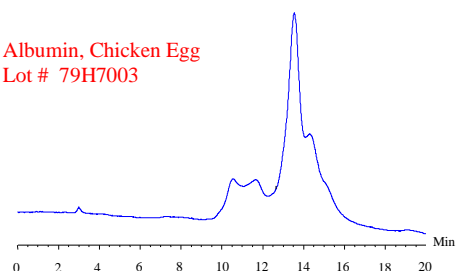
Albumin, Chicken Egg
Lot # 55H7045



Albumin, Chicken Egg
Lot # 37H7010



Albumin, Chicken Egg
Lot # 79H7003



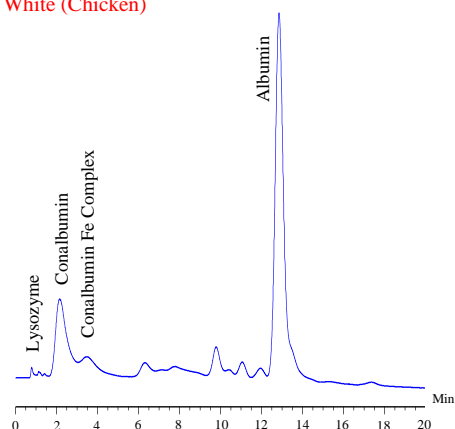
To minimize the use of sample proteins and also generate the least amount of waste, the tests were run on a narrow bore **STYROS™ HQ/NB** as indicated in table 1.

Table 1. Operating Parameters.

HPLC System.	HP 1100
Column	STYROS™ HQ/NB 150x2.1mm
Mobile Phase	A: 20 mM Tris, pH = 8.2 B: A + 1 M NaCl
Flow rate	0.5 ml/min (870 cm/hr)
Gradient	7 to 30% B in 18 Column Volume
Temperature	30°C
Detection	280 nm
Injection volume	2 µl
Samples	OVA (5mg/ml), Egg white (chicken)

Use of the same system, column, and conditions to analyze a freshly constituted preparation of egg-white (diluted 1-to 9 in buffer A) produced the chromatogram below:

Egg White (Chicken)



The striking differences across the four preparations demonstrate a not uncommon degree of variation as well as the high resolution of protein mixes provided by the **STYROS™ HQ/NB** stationary phase.

STYROS™ HQ/NB is a high capacity quaternary amine surface. Its hard gel polymer composition and fully pervious internal structure provide high resolution at high linear velocities.

STYROS™ HQ/NB tolerates pressures of 5,000 psi without any loss of integrity, and can therefore be re-equilibrated within a few minutes at a flow rate of 2-3 ml/min.

Just as **STYROS™ HQ/NB** cleanly separates egg albumin from other constituents in a complex mix, it offers sensitive and rapid assessment of purity and/or lot-to-lot variation for any protein of interest, whether purchased or prepared in-house.