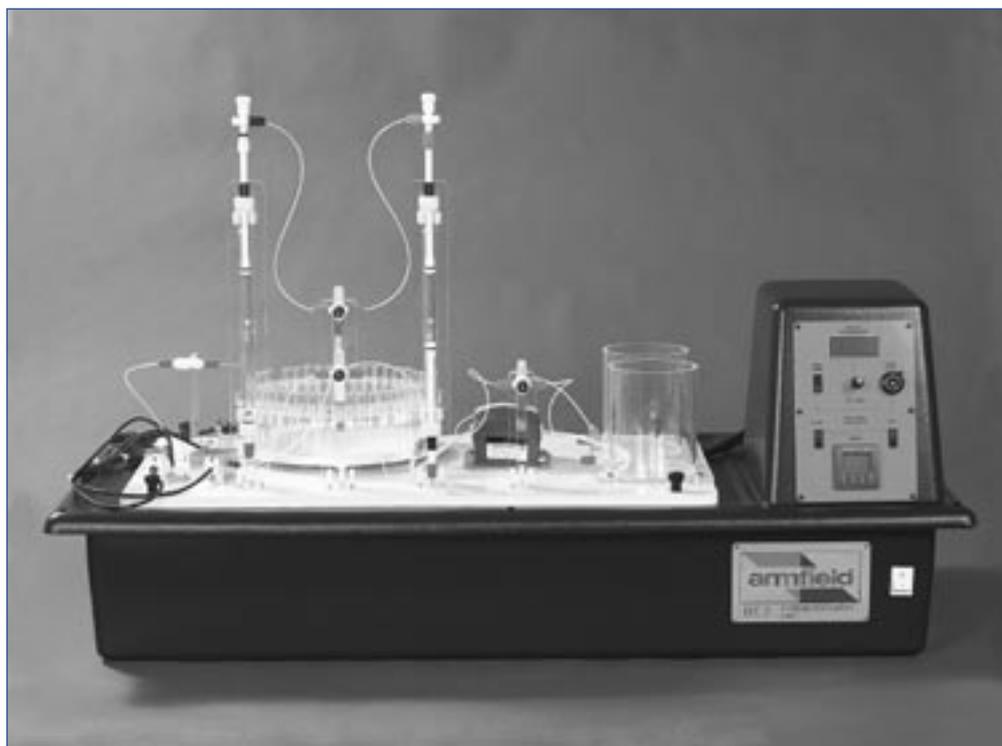




armfield

CHROMATOGRAPHY UNIT



BE2
issue 1

Chromatography is a very widely used separation process in the chemical and biochemical engineering fields. It is a highly selective process capable of separating components of closely similar physical and chemical properties. It is important both at processing and process analysis scales.

The BE2 system demonstrates an industrially important type of chromatography, namely low pressure liquid chromatography of which there are several varieties including ion-exchange, size-exclusion, hydrophobic interaction and affinity chromatography. Each of these types uses a different method of separation and respectively these are: particle charge, particle size and shape, molecule hydrophobicity, and biological specificity. The BE2 system includes detailed experimental procedures for size exclusion chromatography particularly in relation to the separation of proteins and dyes. However, it may be used to investigate other types of low pressure liquid chromatography.

Size exclusion chromatography uses porous beads to separate particles of different sizes. The size range of the pores in the beads defines whether particles are totally excluded, partially excluded or whether they have full entry into all beads. Particles that are totally excluded have the shortest residence time in the column whereas particles that have full entry into the beads have the longest residence time. Partially excluded particles have residence times between the two. On this basis particles of different sizes are separated.

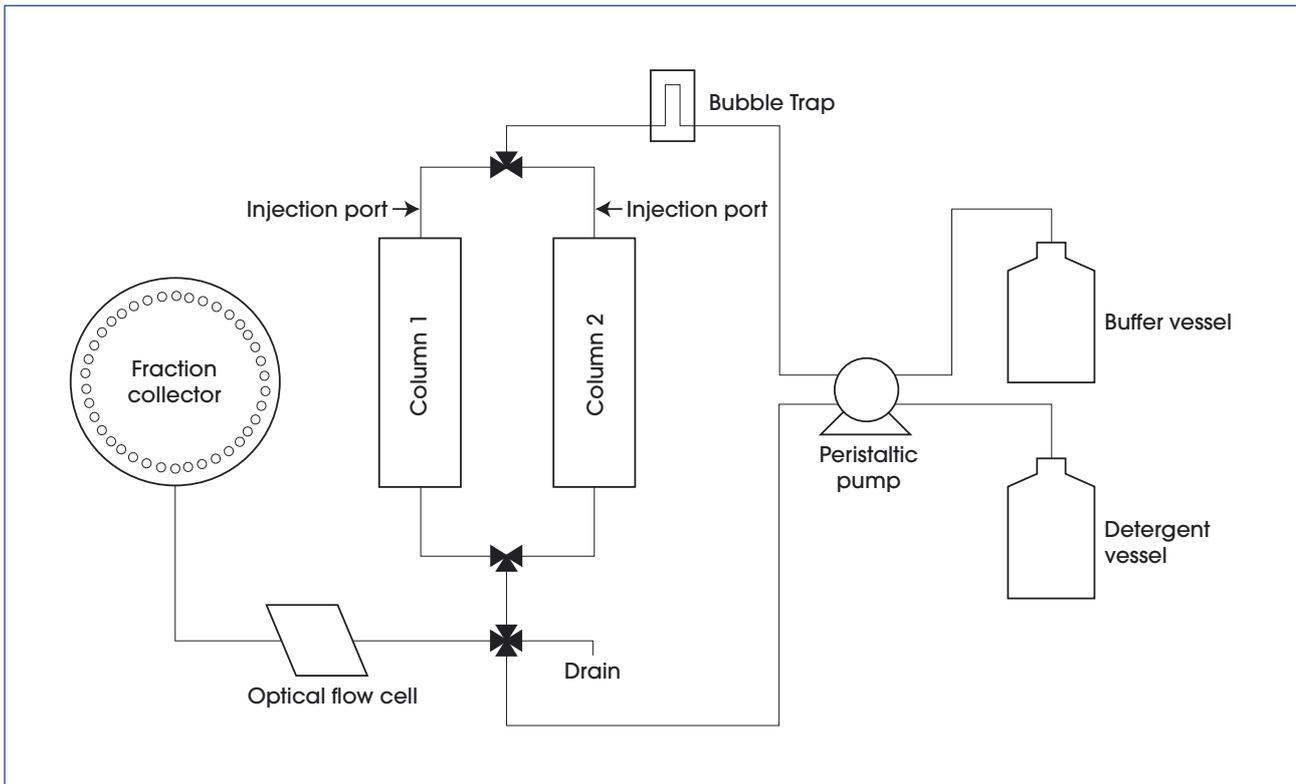
Biochemical Engineering



BE

INSTRUCTIONAL CAPABILITIES

- *Understanding the principles and practices of liquid chromatography including:*
 - *Column packing*
 - *Sample application*
 - *Column development*
 - *Analysis and fraction collection*
- *Understanding the factors affecting separation performance*
- *Understanding protein concentration measurement through UV assays*



BE2 - Process Schematic

DETAILED CAPABILITIES

Detailed teaching exercises are included to demonstrate to students the important aspects of size exclusion (gel filtration) chromatography. Information provided includes details of suitable gel matrices, buffers, proteins etc and in-depth experimental protocols. Although experiments relate to the size-exclusion method alone, the system is suitable for use with other low pressure liquid chromatography systems such as ion-exchange, affinity and hydrophobic interaction. The system includes a UV flow cell for on-line measurement of protein concentration. Level detection is 0.1 mg/ml of protein (bovine serum albumin).

- *Understanding the protocols of running low pressure liquid chromatography systems*
- *Visual demonstration of size exclusion chromatography using dyes*
- *Determination of the effect of flow rate and column length on product separation and concentration*
- *Calibration of the column in terms of protein molecular weight and determination of the molecular weight of an unknown protein*

DESCRIPTION

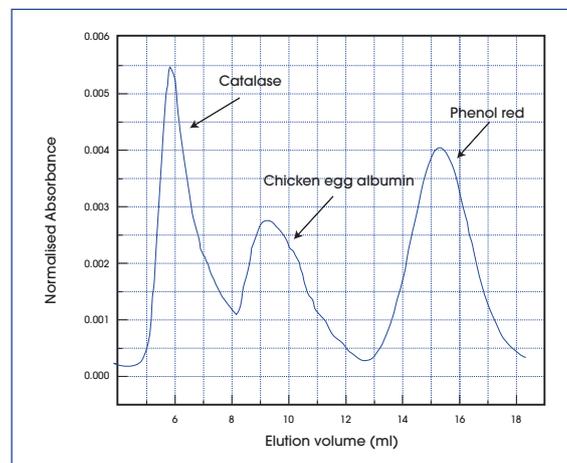
BE2 consists of a bench-top unit onto which are mounted two 250mm long, 10mm diameter borosilicate glass chromatography columns. Both are equipped with adjustable end pieces to allow different bed heights to be investigated. On the top of each column are septum injectors to allow introduction of a sample onto the column surface. The columns are fed by a 3 channel peristaltic pump of an 8 roller design in order to give smooth flow. The pump has variable speed drive and a wide range of tube bores can be used thus giving a very wide flow rate range. Switching valves allow the operator to easily select the desired column for operation. A bubble trap is fitted before the inlet to the columns to prevent air from entering. The outlet from the column passes to a UV optical flow cell for on-line measurement of sample elution. Liquid exiting the flow cell can either be led to drain or collected using the fraction collector. The fraction collector, which has capacity for forty-eight 3 ml test tubes, is timer controlled.

It is essential that all buffers and samples introduced to the column are filtered and degassed in order to protect the column. Equipment required for this is standard laboratory equipment but these are available as an option (BE2-1) if required. BE2-1 consists of a membrane filter holder, vacuum flask, water vacuum pump and membrane filters. The sample filtration system consists of a syringe and disposable membrane filters. Sample injection syringes and needles are also included.

The methods required for liquid chromatography do take some practice in order to achieve reproducible results. Therefore, for users with no experience in this area it is recommended that training is supplied.

Educational Software and Data Logging.

Electronic outputs from the sensors are available for data logging and analysis. The Armfield data logger interfaces between the BE2 and the user's computer via a USB port. The associated software provides a mimic diagram with current sensor values displayed. Also incorporated in the software are graph plotting facilities, full instructions on equipment set-up and experimental methods, related theory and full help texts.



Elution curve of catalase, chicken egg albumin & phenol red

ORDERING SPECIFICATION

- **A bench top unit comprising a vacuum formed ABS plastic plinth with integral electrical console.**
- **Two borosilicate glass chromatography columns (250mm long and 10mm internal diameter) fitted with septum injectors and fed by a 3-channel peristaltic pump.**
- **Syringe and needles for sample injection.**
- **Fraction collector.**
- **Analysis using a UV optical flow cell.**
- **Protection devices for all electrical circuits.**
- **Two displays: Optical absorbance, Timer display for fraction collector.**
- **Sensor signals are routed to the USB port for connection to a PC.**
- **Comprehensive instruction manual with detailed procedures and laboratory teaching exercises.**

OPTIONAL ACCESSORIES

BE2-1:

Buffer filtration and degassing system consisting of membrane filter holder, vacuum flask, water vacuum pump and membrane filters. Sample filtration system consisting of syringe and disposable membrane filters. Sample injection syringes and needles.

ESSENTIAL ACCESSORIES

Software requires a computer running Windows 98, 2000 or XP with a USB port. (Computer not supplied by Armfield).

CHEMICALS

Chemicals supplied are: size exclusion gel (Sephadex G-100), blue dextran and phenol red. Proteins and Vit B12 used in the experiments are not supplied but may be obtained from chemical suppliers.

SERVICES REQUIRED

Single phase mains electrical supply:

BE2-A: 220/240/1ph/50Hz at 10A

BE2-B: 120V/1ph/60Hz at 15A

BE2-G: 220V/1ph/60Hz at 10A

OVERALL DIMENSIONS

Height: 0.70m

Width: 1.10m

Depth: 0.50m

SHIPPING SPECIFICATION

Volume: 0.44m³

Gross weight: 36kg

COMPLEMENTARY EQUIPMENT

BE1: Batch enzyme reactor

CEU: Catalytic reactors

UOP12: Filtration Unit

W8: Anaerobic digester

W11: Aerobic digester

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