

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

PEPTIDE CALIBRATION KIT

from Capillary Electrophoresis

Product Number PEP-CE

TECHNICAL BULLETIN

Product Description

Capillary Electrophoresis has become a powerful analytical separation technique for analysis of peptides in solution. The Sigma Peptide Calibration Kit is designed to provide reagents and conditions for peptide separations, purity determination for synthetic peptides, calibrations of capillary electrophoresis instruments, and tests for the performance of capillaries.

Reagents

<u>Item</u>	Product No.	<u>Amount</u>
Peptide Standards	P 2693	1 Vial
Peptide Separation Buffer	P 2188	50 ml
Washing Solution	W 2629	100 ml

Procedure

Peptide Standards

- 1. Add 50 μ l of the Peptide Separation Buffer (Product No. P 2188) and 450 μ l of deionized water to the vial of Peptide Standards (Product No. P 2693).
- 2. Vortex the vial for a few seconds and the solution of Peptide Standards is ready to use.
- 3. Aliquot the unused portion and immediately freeze at –20 °C or below.

Unknown Peptide Samples

- Prepare each peptide sample at a concentration of 50-100 μg/mg in a 10-fold dilution of Peptide Separation Buffer (Product No. P 2188).
- 2. Vortex for a few seconds and the sample is ready to use.

 Aliquot the unused portion and immediately freeze at -20 °C or below.

Running Conditions

- Peptide Separation Buffer (Product No. P 2188) and Washing Solution (Product No. W 2629) should be degassed before use.
- 2. Follow manufacturer's instructions to assemble the capillary column to be used.
- For an electrophoretic load, use 10 kV for 8 seconds or for a pressure load, use 20 psi/second.
- Run the solution of Peptide Standards at 10 kV constant voltage, positive to negative polarity, for 15 minutes.
- 5. Detection system: UV, 200 nm
- 6. Column size: 25 μm x 24 cm, coated
- 7. Before each sample application, perform the following purge cycles:
 - 60 seconds with deionized water
 - 60 seconds with Washing Solution
 - 60 seconds with deionized water
 - 60 seconds with Peptide Separation Buffer

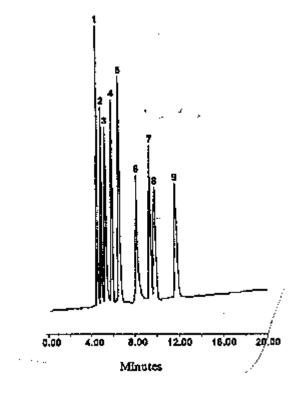
After the completion of the run, perform the following purge cycles:

- 60 seconds with deionized water
- 60 seconds with Washing Solution
- 60 seconds with deionized water
- 3 minutes with dry nitrogen gas

Figure 1 A Typical Electropherogram of Peptide Standards on BioRad BioFocus 3000 system. Peptide Standards contain approximately 25 μg of each of the following 9 peptides:



- 2. Bradykinin Fragment 1-5
- 3. Substance P
- 4. [Arg⁸]-Vasopressin
- 5. Luteinizing Hormone Releasing Hormone
- 6. Bombesin
- 7. Leucine Enkephalin
- 8. Methionine Enkephalin
- 9. Oxytocin



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