Polyamine analysis using the Biochrom 30 Amino Acid Analyser

Polyamines are a class of biogenic amines which are of biological importance in both eukaryotic and prokaryotic cells. Polyamine analysis has applications in a wide range of areas including food science, clinical science, and cancer research.

● Food science

Biogenic amines are normally produced in foods by decarboxylation of free amino acids by bacteria, amination of ketones or aldehydes, or enzymatic hydrolysis of nitrogen-containing compounds.
In food microbiology they have often been related to spoilage and fermentation processes. Natural polyamines such as putrescine, spermidine and spermine and biogenic amines like histamine can both be detected in food products but they have different effects: while polyamines appear to be essential, biogenic amines are mostly detrimental and can generally be considered as markers of protein degradation. For example, while ingestion of food containing small amounts of histamine has little effect on humans, in large doses histamine can cause food poisoning and become toxic. According to the 91/493/EEC directive the average level of histamine in fish should not exceed 100 mg/kg.

● Cancer Research

Polyamines play an important role in cell proliferation, cell growth, and synthesis of protein and nucleic acids. They have been implicated in regulating many development processes including DNA biosynthesis and cell division. Research groups have shown a growing interest in understanding polyamine metabolism in correlation with cancer research, as some cancer cells have shown increased levels of polyamines when compared to that of normal cells. Measurement of polyamines in biological fluids and tissues levels could have the potential to be useful for the diagnosis of disorders, monitoring of patients and cancer therapy.

Using the Biochrom 30 amino acid analyser, a wide range of samples can be analysed for their polyamine content. These samples include foods, plant extracts, cell and tissue extracts, serum, plasma, CSF and hydrolysed urine.
The polyamine system consists of a specially formulated buffer and a 10 cm polyamine column (see ordering information). This system can be used in conjunction with sodium chemistry.

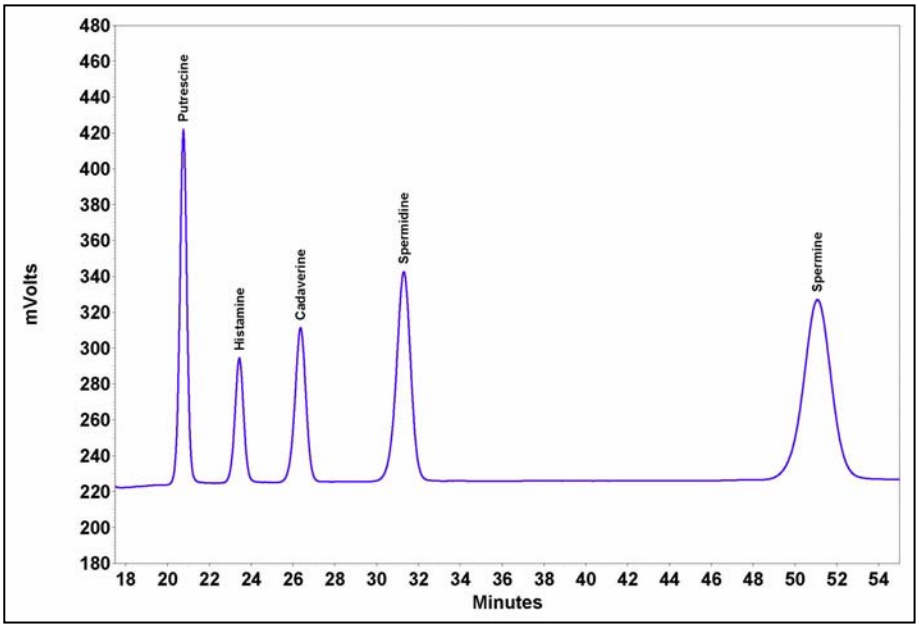

Figure 1: Polyamine standard (5 nmol/20 µL) run on the Biochrom 30 Amino acid analyser

Customer case study

The effect of dietary methionine on polyamine homeostasis in the mouse is being investigated by Dr. Warren Kruger at Fox Chase Cancer Center in Philadelphia, PA, USA.

Spermidine and spermine are formed from putrescine and decarboxylated S-adenosylmethionine, a metabolite of the methionine cycle. Mouse liver lysates from animals fed either normal chow or methionine-deficient chow were analyzed for polyamines using a Biochrom 30 Amino Acid Analyzer with a polyamine column supplied by Biochrom.

Spermidine and spermine are formed from putrescine and decarboxylated S-adenosylmethionine, a metabolite of the methionine cycle. As shown in the figure, the depletion of dietary methionine resulted in increase of putrescine, decrease of spermine, and no change of spermidine .

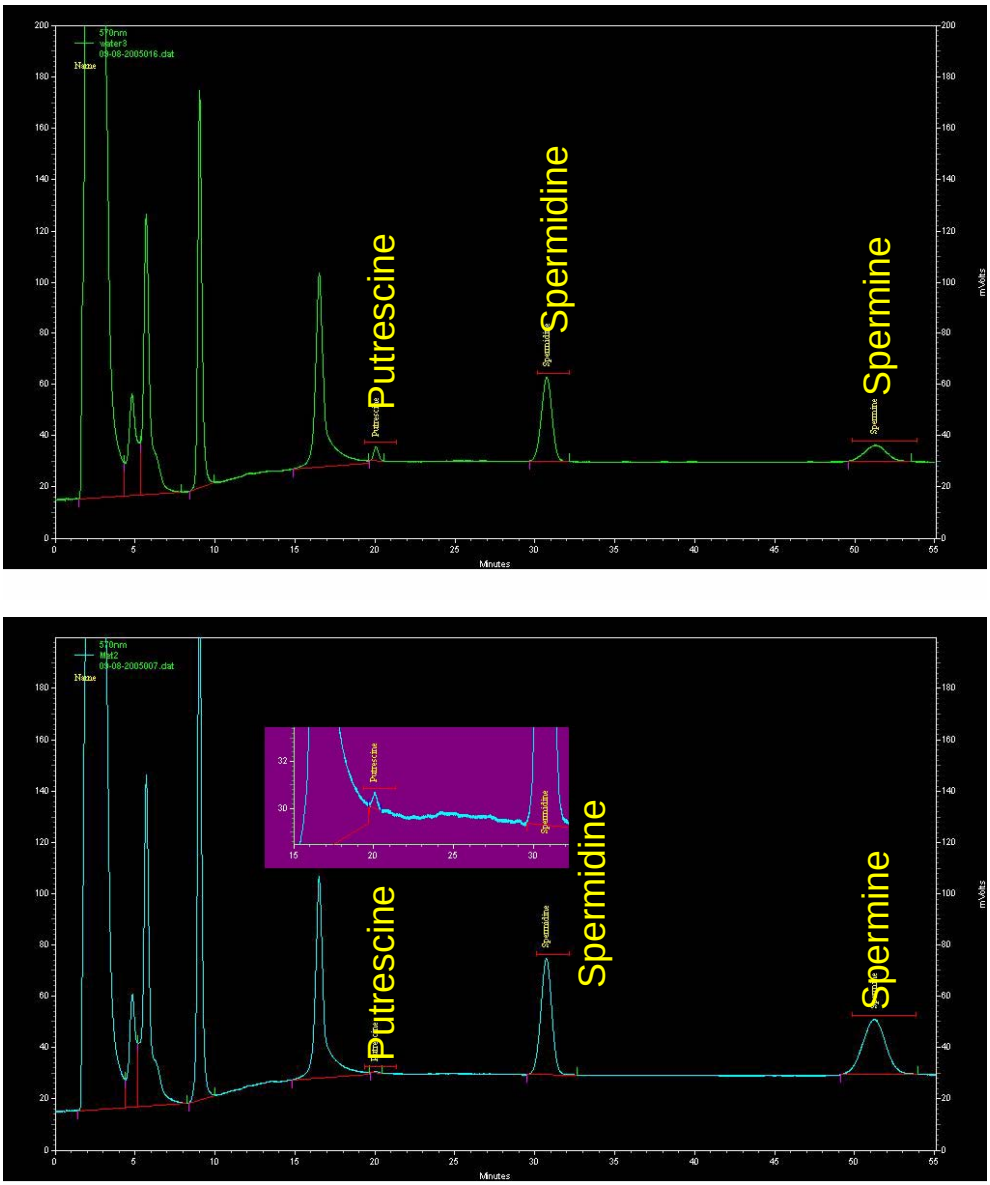

Figure 2: Chromatogram of mouse liver lysates

The analytical performances of the Biochrom 30 polyamine method are summarised in the table below.

Compound	Detection limit (pmol)	Quantitation limit (pmol)	Linearity : Regression coefficient r^2	%CV (5nmol / 20 µL)
Putrescine	**24**	**79**	**0.999755**	**1.0**
Histamine	**13**	**42**	**0.999727**	**1.5**
Cadaverine	**14**	**46**	**0.999913**	**1.1**
Spermidine	**11**	**37**	**0.999862**	**1.3**
Spermine	**15**	**49**	**0.999871**	**1.2**

Table 1: Analytical performance

For most compounds the detection limit was better than 25 pmol on column and the quantitation limit was typically better than 50 pmol for a 20 µL injection (80 pmol for putrescine). All the compounds showed a linear response up to a minimum of 5 nmol on column. The repeatability on concentrations was typically better than 1.5% for 5 nmol on column (20 µL injection).

Conclusion

The Biochrom 30 Amino Acid Analyser has the flexibility to be used for polyamine analysis, which is useful for labs that require both amino acid analysis and polyamine analysis.

This technique also offers the advantage of requiring no method development.

Ordering information

Part number	Description	Price
80-2037-74	Polyamine Buffer	enquire
80-2104-19	Polyamine column	enquire
80-2038-09	Sodium citrate buffer pH6.5	enquire
80-2037-57	Sodium hydroxide	enquire
80-2118-30	Ultra Ninhydrin Reagent Kit	enquire