

Rapid Analysis of Lysine for Feedstuff Applications



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Lysine is an essential amino acid which means that it cannot be synthesized in the body at a sufficient rate to meet the physiological needs of the body, and therefore must be supplied by the diet. Lysine is one of the most important essential amino acids: it is a building block of protein and an important component in amino acid balance. It is a limiting amino acid in cereal grains and in some vegetable protein sources, therefore requiring synthetic supplementing to meet the animals needs. Lysine is a structural element of proteins; it is a vital component of the enzymes and is present in almost all the tissues of the animal.

Because lysine is involved in a large number of metabolic processes it has a fundamental role in animal nutrition and animal health. Improvement in animal performance through better amino acid balance, better carcass quality, and prevention of lysine deficiency are some of the benefits associated with the right lysine content in the animal diet. For example, for piglets and pigs the use of synthetic lysine allows an increase in feed consumption and body weight gain; and an improvement in feed conversion and in nitrogen retention. Particularly with pigs, its use allows an improvement of the carcass quality. Using the Biochrom 30 buffer system, lysine can be accurately quantified in less than 10 minutes to enable adjustments to the formulation of feedstuff.

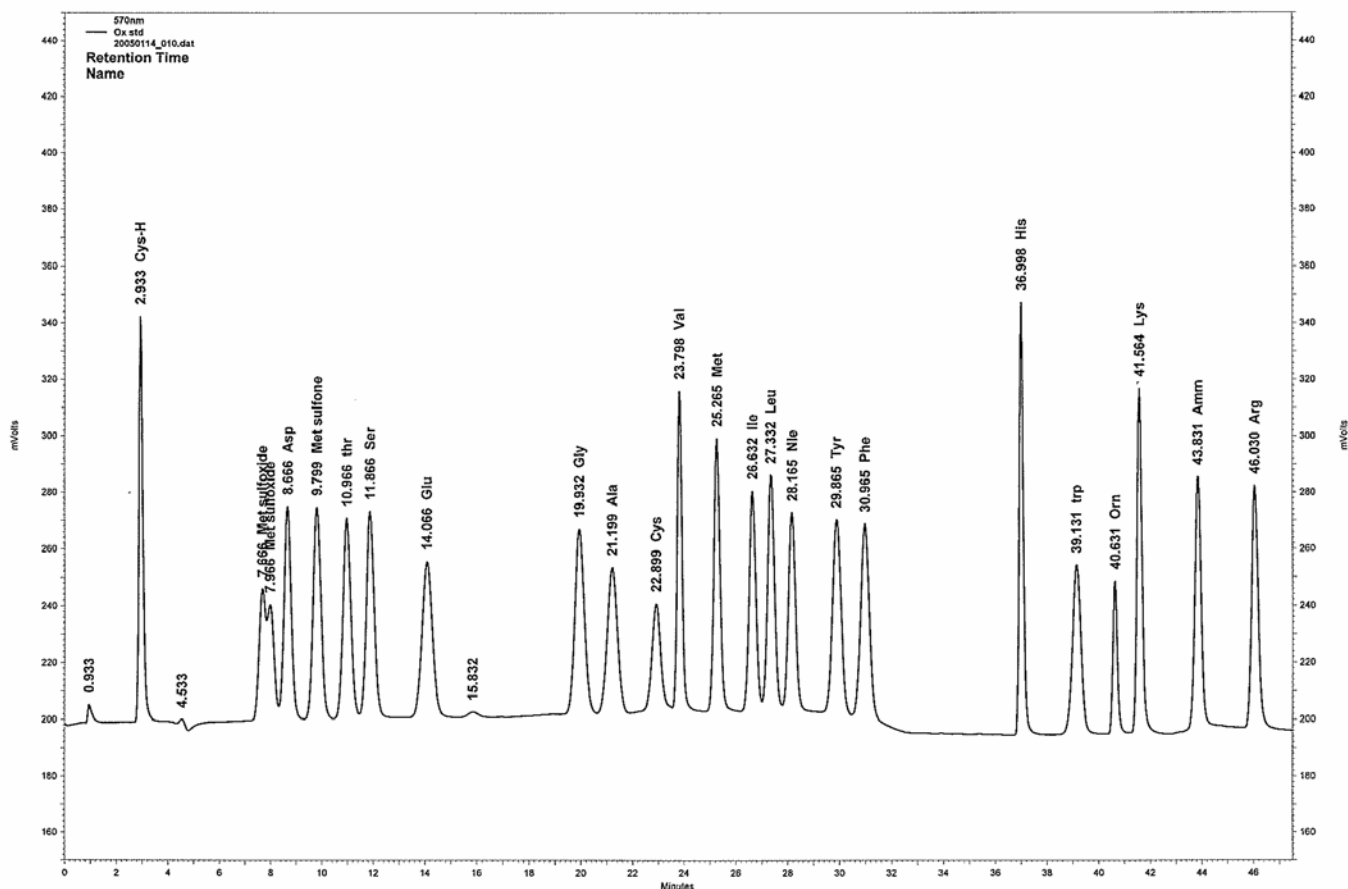


Figure 1: Standard chromatogram obtained with the full program (Lysine retention time : 41.564 min)

Experimental conditions

Instrument: Biochrom 30 amino acid analyser using post column Ninhydrin derivatisation and photometric detection at 570 nm

Column: Sodium High Performance 200 x 4.6 mm (80-2104-15)

Buffers: Sodium Buffer A pH 3.25
Sodium Buffer B pH 3.50
Sodium Buffer C pH 4.40
Sodium Buffer D pH 9.23

Ninhydrin flowrate: 20 ml/hr
Buffer flowrate: 35 ml/hr

Sample prep: Prepared according to the EC official method.

Programme:

| <u>No.</u> | <u>Time</u> | <u>Temp</u> | <u>Buffer</u> | <u>Pump</u> | <u>Nin</u> | <u>Rec</u> | <u>Commands</u> |
|------------|-------------|-------------|---------------|-------------|------------|------------|-----------------|
| 1 | 01:00 | 54°C | 4 | 35.0ml/h | ON | ON | |
| 2 | 00:00 | 54°C | 4 | 35.0ml/h | ON | ON | Reset |
| 3 | 01:00 | 54°C | 4 | 35.0ml/h | ON | ON | Load |
| 4 | 04:00 | 54°C | 4 | 35.0ml/h | ON | ON | |
| 5 | 04:00 | 94°C | 6 | 35.0ml/h | ON | ON | |
| 6 | 02:00 | 94°C | 4 | 35.0ml/h | ON | ON | |
| 7 | 06:00 | 54°C | 4 | 35.0ml/h | OFF | OFF | |
| 8 | 02:00 | 54°C | 4 | 35.0ml/h | ON | OFF | |

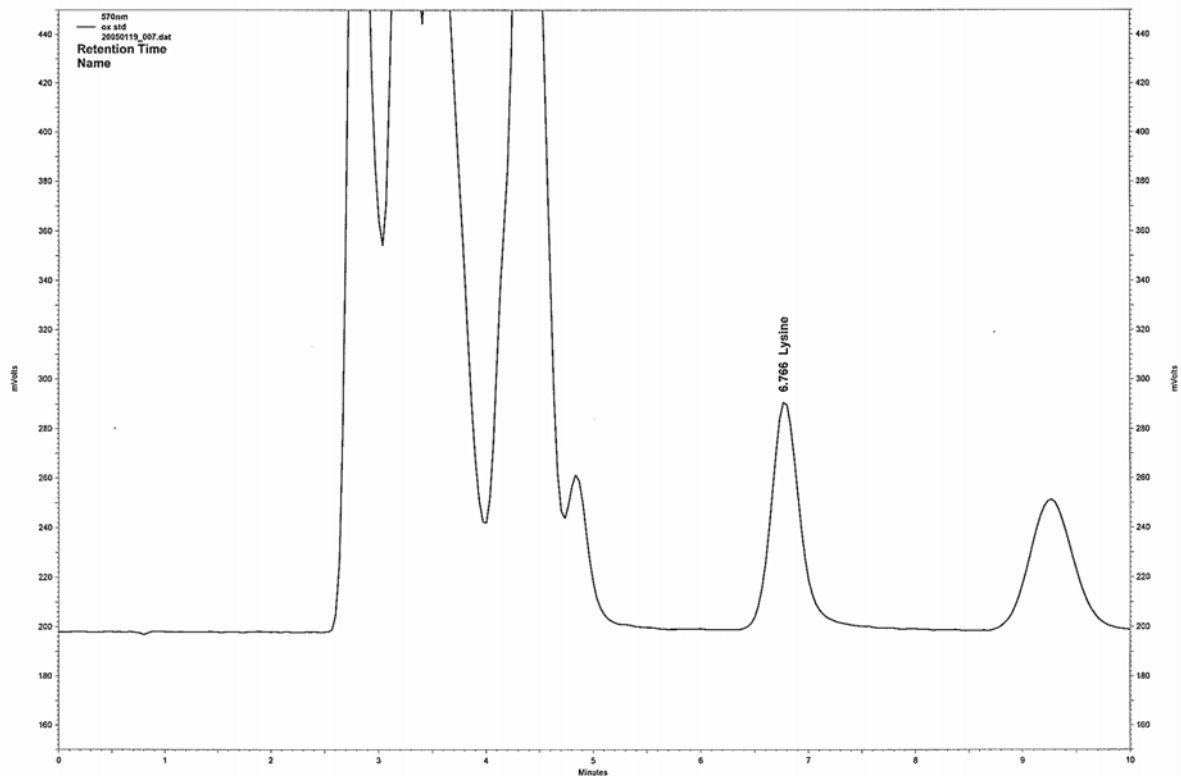


Figure 2: Standard chromatogram obtained with short program (Lysine retention time: 6.766 min)

| Samples (hydrolysates) | Lysine concentration (nM/20µL) Short program | Lysine (% of raw material) | |
|---------------------------|--|----------------------------|--------------|
| | | Biochrom | External Lab |
| Feedstuff Sample A | 1.877 | 1.14 | 1.13 |
| Feedstuff Sample B | 1.368 | 0.83 | 0.86 |
| Pig feed Sample | 1.745 | 1.06 | 1.05 |
| Piglet Feed Control | 2.120 | 1.29 | 1.28 |

Table 1: Results obtained with the short program on hydrolysate samples

| | |
|-----------------------------------|-------------------------------|
| Run time (injection to injection) | 20 min |
| Intermediate precision | 0.9% (5 nmol/20 µL injection) |
| Detection limit | 14 pM/20 µL |
| Quantification limit | 48 pM/20 µL |

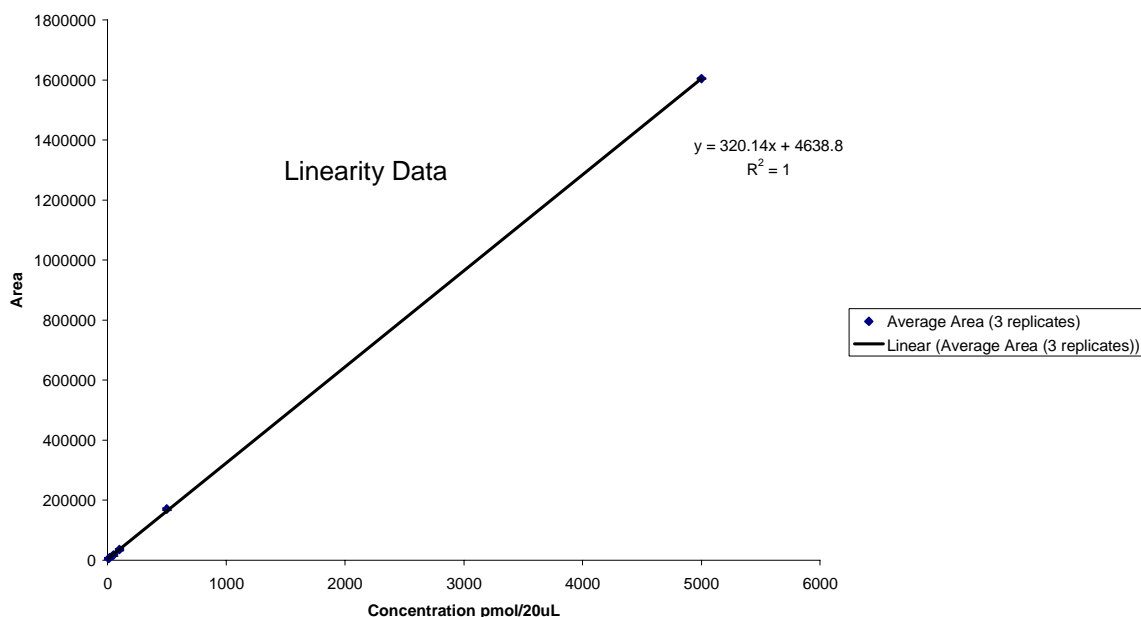


Table 2 : Analytical performance

Conclusion

The results obtained with the short program showed a very good correlation with the results obtained with the full program, as well as with the results obtained when tested by an external laboratory. The program gave good analytical performance with good repeatability and low detection and quantification limits.

The flexibility of the Biochrom 30 allows this program to be used as a complement of the full program or on its own if only lysine quantification is required. For lysine analysis alone, more than 70 analyses can be performed per day, making it a very useful tool for busy quality control laboratories.