Guanidinoacetic acid as feed additive – Determination of Guanidinoacetic acid in the pure substance, CreAmino™, and feed by ion chromatography

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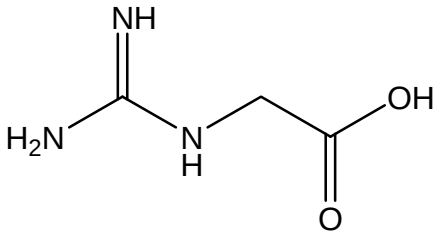
1 Scope

This test method is applicable to the determination of guanidinoacetic acid (N-(Aminoiminomethyl)-glycine, CAS Registry No.: 352-97-6),



in the pure substance and CreAmino™ as well as in broiler and turkey feed supplemented with 100 – 5000 mg/kg of the active ingredient.

2 Principle

Guanidinoacetic acid and CreAmino™ samples are dissolved in water and injected for analysis. In case of feed samples guanidinoacetic acid is extracted into deionised water using an ultrasonic bath. The resulting suspension is filtered and injected for analysis. Guanidinoacetic acid is separated by ion chromatography and determined by UV detection. The calculations are performed with an external standard.

3 Reagents

- Water, deionised
- Guanidinoacetic acid, purity: 99.84 %, Degussa AG

4 Apparatus

4.1 Preparation of the sample

- Membrane filter Cellulose nitrate, 0.45 µm, Sartorius
- Analytical Balance, readability 0.01 mg
- Ultrasonic bath
- Standard laboratory equipment

4.2 Chromatography

- Ion chromatograph with UV/VIS-Detector, Dionex Series DX500 consisting of:
  Gradient pump GP50
  Chromatographic module LC30
  UV/VIS-Detector AD25
  Auto sampler, Abimed Series 234
- Computer (PC) with control and calculation software Dionex, Chromeleon 6.60

5 Procedure

5.1 Preparation of the sample solution

5.1.1 Guanidinoacetic acid and CreAmino™

Weigh 50 ± 5 mg of the sample into a 1000 mL volumetric flask, dissolve in water with the help of an ultrasonic bath, and make up to volume with water.

5.1.2 Feed samples

Weigh 10 ± 1 g of the sample (pellets are grinded) into a 500 ml volumetric flask and add 350 – 400 mL of water. Apply the ultrasonic bath 4 times 5 minutes and shake and mix well in between. Make up to volume with deionised water. Wait for a few minutes until the sample settled down. Filter through a 0.45 µm membrane filter and inject for analysis.

5.2 Determination by ion chromatography

Column	Hypersil Hypercarb 4.6 x 100, Thermo 35007-104630 (1st column) and Aminopac PA1, Dionex P/N 37022 (2nd column) with pre column Dionex 37022
Mobile phase	water, deionised, filtered and vacuum degassed
Flow	1.0 mL/min
Injection volume	50 µL

Detection	UV at 200 nm
Column temperature	30 °C
Retention time	approx. 12 min

6 Calibration

Preparation of the standard solution:

6.1 Pure Substance and CreAmino™

Weigh 50 ± 5 mg of the guanidinoacetic acid reference item into a 1000 mL volumetric flask, dissolve in water with the help of an ultrasonic bath and make up to volume with water.

6.2 Feed

Weigh 10 ± 2 mg of the guanidinoacetic acid reference item into a 1000 mL volumetric flask, dissolve in water with the help of an ultrasonic bath and make up to volume with water.

7 Calculation

Calculation is done by the following formula:

$$\text{Cont} = \frac{A_S \cdot c_{St} \cdot P_{St}}{A_{St} \cdot c_S}$$

Cont	= content of guanidinoacetic acid in the sample [%]
A_S	= peak area of sample signal
A_{St}	= peak area of standard signal
c_S	= concentration of sample solution [mg/L]
c_{St}	= concentration of standard solution [mg/L]
P_{St}	= purity of standard [%]

Each sample is determined twice. The result is the mean of two determinations.