

Third Commission Directive of 27 April 1972 establishing Community methods of analysis for the official control of feedingstuffs (72/199/EEC) (repealed)

Article 1
Article 2
Article 3
Article 4

ANNEX I

1. DETERMINATION OF STARCH

- 1. Purpose and scope
- 2. Principle
- 3. Reagents
 - 3.1.
 - 3.2.
 - 3.3.
 - 3.4.
 - 3.5.
- 4. Apparatus
 - 4.1.
 - 4.2.
- 5. Procedure
 - 5.1. Preparation of the sample
 - 5.2. Determination of the total optical rotation (P or S) (see...
 - 5.3. Determination of the optical rotation (P' or S') of substances...
- 6. Calculation of results
 - 6.1. Measurement by polarimeter
 - 6.2. Measurement by saccharimeter
 - 6.3. Repeatability
- 7. Observations
 - 7.1.
 - 7.2.
 - 7.3. Following feed materials, in case they are present in significant...

2. DETERMINATION OF CRUDE PROTEIN

- 1. Purpose and scope
- 2. Principle
- 3. Reagents
 - 3.1.
 - 3.2.
 - 3.3.
 - 3.4.
 - 3.5.
 - 3.6.
 - 3.7.

*Status: EU Directives are being published on this site to aid cross referencing from UK legislation. After
IP completion day (31 December 2020 11pm) no further amendments will be applied to this version.*

- 3.8
 - 3.9
 - 3.10
 - 3.11
 - 3.12
 - 3.13
 - 4. Apparatus
 - 5. Procedure
 - 5.1 Digestion
 - 5.2 Distillation
 - 5.3 Titration
 - 5.4 Blank test
 - 6. Calculation of results
 - 7. Verification of the method
 - 7.1 Repeatability
 - 7.2 Accuracy
 - 8. Observations
 - 8.1
 - 8.2
 - 8.3
3. DETERMINATION OF CRUDE PROTEIN DISSOLVED BY PEPSIN AND HYDROCHLORIC ACID...
- 1. Purpose and scope
 - 2. Principle
 - 3. Reagents
 - 3.1
 - 3.2
 - 3.3
 - 3.4
 - 3.5
 - 3.6
 - 4. Apparatus
 - 4.1
 - 4.2
 - 5. Procedure
 - 5.1 Preparation of solution (see observation 7.2)
 - 5.2 Digestion
 - 5.3 Distillation and titration
 - 5.4 Blank test
 - 6. Calculation of results
 - Repeatability
 - 7. Observations
 - 7.1
 - 7.2
4. ESTIMATION OF PEPSIN ACTIVITY
- 1. Purpose and scope
 - 2. Principle
 - 3. Reagents
 - 3.1
 - 3.2
 - 3.3

	3.4
	3.5
	3.6
	3.7
	3.8
4.	Apparatus	
	4.1
	4.2
	4.3
	4.4
5.	Procedure	
	5.1	Preparation of the solution (see observation 7.1)
	5.2	Hydrolysis
	5.3	Development of coloration and measurement of optical density
	5.4	Blank test
	5.5	Calibration curve
6.	Calculation of results	
7.	Observations	
	7.1
	7.2
5.	DETERMINATION OF FREE AND GOSSYPOL	
	1.	Purpose and scope
	2.	Principle
	3.	Reagents
	3.1
	3.2
	3.3
	3.4
	3.5
	3.6
	4.	Apparatus
	4.1
	4.2
	5.	Procedure
	5.1	Test sample
	5.2	Determination of free gossypol
	5.3	Determination of total gossypol
	6.	Calculation of results
	6.1	From the specific optical density
	6.2	From a calibration curve
		6.2.1 Free gossypol
		6.2.2 Total gossypol
	6.3	Repeatability

ANNEX II

1.	DETECTION AND IDENTIFICATION OF ANTIBIOTICS OF THE TETRACYCLINE GROUP
	1. Purpose and scope
	2. Principle

*Status: EU Directives are being published on this site to aid cross referencing from UK legislation. After
IP completion day (31 December 2020 11pm) no further amendments will be applied to this version.*

3. Reagents and culture medium
 - 3.1 Buffer solution, pH 3.5
 - 3.2 Phosphate buffer solution, pH 5.5
 - 3.3
 - 3.4
 - 3.5
 - 3.6
 - 3.7
 - 3.8
 - 3.9 Micro-organism: *B. cereus* ATCC No 11.778
 - 3.10 Culture medium
 - 3.11
 4. Apparatus
 - 4.1
 - 4.2
 - 4.3
 - 4.4
 - 4.5
 5. Standard solutions
 - 5.1 Stock solutions
 - 5.2 Reference solutions for detections by UV light
 - 5.3 Reference solutions for detection by bioautography
 6. Extraction
 7. Detection and identification
 - 7.1 Chromatography
 - 7.2 Detection by UV light
 - 7.3 Detection by bioautography
 - 7.4 Identification
2. DETERMINATION OF CHLORTETRACYCLINE, OXYTETRACYCLINE AND TETRACYCLINE
- A. BY DIFFUSION ON AGAR
1. Purpose and scope
 2. Principle
 3. Micro-organism: *B. cereus*, ATCC No 11.778
 - 3.1 Maintenance of the parent strain
 - 3.2 Preparation of the spore suspension
 4. Culture media and reagents
 - 4.1 Basic medium for the determination
 - 4.2 Phosphate buffer solution, pH 5.5
 - 4.3
 - 4.4 Phosphate buffer solution, pH 8
 - 4.5
 - 4.6
 - 4.7
 - 4.8
 - 4.9
 - 4.10
 - 4.11
 5. Standard solutions
 - 5.1 Chlortetracycline
 - 5.2 Oxytetracycline
 - 5.3 Tetracycline

6. Extraction
 - 6.1 Contents of 50 ppm or less
 - 6.2 Contents greater than 50 ppm
 - 6.2.1 Chlortetracycline
 - 6.2.2 Oxytetracycline and tetracycline
 7. Determination method
 - 7.1 Inoculation of the culture medium
 - 7.2 Preparation of the trays
 - 7.3 Incubation
 8. Evaluation
 9. Repeatability
- B. BY TURBIDIMETRY
1. Purpose and scope
 2. Principle
 3. Micro-organism: *Staphylococcus aureus* K 141
 - 3.1 Maintenance of the parent strain
 - 3.2 Preparation of the inoculum
 4. Culture media and reagents
 - 4.1 Basic medium for the determination
 - 4.2 Phosphate buffer solution, pH 4·5
 - 4.3
 - 4.4
 - 4.5
 - 4.6
 - 4.7
 5. Standard solution
 6. Extraction
 - 6.1 Chlortetracycline
 - 6.2 Oxytetracycline and tetracycline
 7. Determination method
 - 7.1 Preparation of the standard series and of the extract
 - 7.1.1 Chlortetracycline
 - 7.1.2 Oxytetracycline and tetracycline
 - 7.2 Inoculation of the culture medium
 - 7.3 Seeding
 - 7.4 Incubation
 - 7.5 Measurement of growth
 8. Calculation of results
 9. Repeatability
3. DETERMINATION OF OLEANDOMYCIN
1. Purpose and scope
 2. Principle
 3. Micro-organism: *B. cereus* K 250 TR (resistant to tetracyclines)
 - 3.1 Maintenance of the parent strain
 - 3.2 Preparation of the spore suspension
 4. Culture media and reagents
 - 4.1 Medium for maintenance of the parent strain
 - 4.2 Basic medium for the determination
 - 4.3
 - 4.4
 - 4.5
 - 4.6

- 4.7 Extraction solution
 - 4.8
 - 5. Standard solution
 - 6. Extraction
 - 7. Determination method
 - 7.1 Inoculation of the culture medium
 - 7.2 Preparation of the trays
 - 7.3 Incubation
 - 8. Evaluation
 - 9. Repeatability
4. DETERMINATION OF TYLOSIN
- 1. Purpose and scope
 - 2. Principle
 - 3. Micro-organism: *Sarcina lutea* ATCC No 9341
 - 3.1 Maintenance of the parent strain
 - 3.2 Preparation of the bacteria suspension
 - 4. Culture media and reagents
 - 4.1 Basic medium for the determination
 - 4.2 Phosphate buffer solution, pH 8
 - 4.3 Phosphate buffer solution, pH 7
 - 4.4
 - 4.5
 - 4.6
 - 4.7
 - 4.8
 - 5. Standard solutions
 - 6. Extraction
 - 7. Determination method
 - 7.1 Inoculation of the culture medium
 - 7.2 Preparation of the trays
 - 7.3 Incubation
 - 8. Evaluation
 - 9. Repeatability
5. DETERMINATION OF VIRGINIAMYCIN
- 1. Purpose and scope
 - 2. Principle
 - 3. Micro-organism: *Micrococcus luteus* ATCC 9341 (NCTC 8340, NCIB 8553)
 - 3.1. Maintenance of stock culture
 - 3.2. Preparation of the bacterial suspension
 - 4. Culture media and reagents
 - 4.1. Culture and assay medium
 - 4.2. Phosphate buffer, pH 6
 - 4.3.
 - 4.4.
 - 4.5.
 - 4.6.
 - 4.7.
 - 5. Standard solutions
 - 6. Preparation of the extract and assay solutions
 - 6.1. Extraction
 - 6.1.1. Products with a virginiamycin content up to 100 mg/kg

- 6.1.2. Products with a virginiamycin content greater than 100 mg/kg
- 6.2. Assay solutions
- 7. Assay procedure
 - 7.1. Inoculation of the assay medium
 - 7.2. Preparation of the plates
 - 7.3. Incubation
- 8. Evaluation
- 9. Repeatability