

Commission Regulation (EC) No 273/2008 of 5 March 2008 laying down detailed rules for the application of Council Regulation (EC) No 1255/1999 as regards methods for the analysis and quality evaluation of milk and milk products (repealed)

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  - 8.1. ....
  - 8.2. ....
  - 8.3. ....
  - 8.4. ....
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- 2. TERMS AND DEFINITION
- 3. PRINCIPLE
- 4. REAGENTS
  - 4.1. ....
  - 4.2. ....
  - 4.3. ....
  - 4.4. ....
- 5. APPARATUS
  - 5.1. ....
  - 5.2. ....
  - 5.3. ....
  - 5.4. ....
  - 5.5. ....
  - 5.6. ....
  - 5.7. ....
    - 5.7.1. ....
    - 5.7.2. ....
      - 5.7.2.1. ....
      - 5.7.2.2. ....
  - 5.8. ....
- 6. SAMPLING
- 7. PROCEDURE
  - 7.1. Preparation of the test sample and test portion
    - 7.1.1. Butter-oil, Butter
      - 7.1.1.1. ....
      - 7.1.1.2. ....
      - 7.1.1.3. ....
    - 7.1.2. Cream
      - 7.1.2.1. ....
      - 7.1.2.2. ....
      - 7.1.2.3. ....
      - 7.1.2.4. ....
      - 7.1.2.5. ....
      - 7.1.2.6. ....
  - 7.2. Preparation of the calibration standards
    - 7.2.1. ....
    - 7.2.2. ....
    - 7.2.3. ....
    - 7.2.4. ....
    - 7.2.5. ....
  - 7.3. Chromatographic determination
    - 7.3.1. ....

**Changes to legislation:** There are currently no known outstanding effects for the Commission Regulation (EC) No 273/2008 (repealed). (See end of Document for details)

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- 7.3.2. ....
- 7.3.3. ....
- 8. CALCULATION OF RESULTS
  - 8.1. Calibration
    - 8.1.1. ....
    - 8.1.2. ....
    - 8.1.3. ....
  - 8.2. Test samples
- 9. PRECISION
  - 9.1. Repeatability
  - 9.2. Reproducibility
- 10. TOLERANCE LIMITS: LOWER LIMITS (CASE OF INSUFFICIENT QUANTITIES)
  - 10.1. ....
  - 10.2. Butter and concentrated butter
    - 10.2.1. ....
    - 10.2.2. ....
  - 10.3. Cream
    - 10.3.1. ....
    - 10.3.2. ....
- 11. TOLERANCE LIMITS: UPPER LIMITS (CASE OF EXCEEDING QUANTITY BY MORE...)
  - 11.1. ....
  - 11.2. Butter and concentrated butter
    - 11.2.1. ....
  - 11.3. Cream
    - 11.3.1. ....
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  - Tests for outliers:
  - Precision parameters
  - Figure 1
    - Experimental results: Sample A
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### (Article 5)

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- 1. SCOPE AND FIELD OF APPLICATION
- 2. PRINCIPLE
- 3. APPARATUS
  - 3.1. ....
  - 3.2. ....
  - 3.3. ....
  - 3.4. ....
  - 3.5. ....
  - 3.6. ....

3.7.	.....
4.	<b>REAGENTS</b>
4.1.	.....
4.2.	.....
4.3.	.....
4.4.	.....
4.5.	.....
4.5.1.	Vanillin stock solution (= 500 µg/ml)
4.5.2.	Vanillin standard solution (= 10 µg/ml)
4.5.3.	.....
4.5.4.	.....
4.5.5.	.....
4.5.6.	HPLC mobile phase
5.	<b>PROCEDURE</b>
5.1.	Preparation of the test sample
5.1.1.	Butter
5.1.2.	Concentrated butter
5.1.3.	Cream
5.2.	Preparation of the test solution
5.3.	Calibration
5.4.	Determination by HPLC
6.	<b>CALCULATION OF THE RESULTS</b>
7.	<b>ACCURACY OF THE METHOD</b>
7.1.	Repeatability (r)
7.2.	Reproducibility (R)
8.	<b>TOLERANCE LIMITS</b>
8.1.	.....
8.2.	.....
8.2.1.	.....
8.2.2.	.....
8.3.	.....
8.3.1.	.....
8.3.2.	.....
9.	<b>NOTES</b>
9.1.	.....
9.2.	.....
9.3.	.....
9.4.	.....

## ANNEX VII

### (Article 5)

#### DETERMINING THE ETHYL ESTER OF BETA-APO-8'-CAROTENIC ACID IN CONCENTRATED BUTTER...

1.	<b>SCOPE AND FIELD OF APPLICATION</b>
2.	<b>PRINCIPLE</b>
3.	<b>APPARATUS</b>
3.1.	.....
3.2.	.....
3.3.	.....
3.4.	.....

- 3.5. ....
- 3.6. ....
- 4. REAGENTS
  - 4.1. ....
    - 4.1.1. ....
    - 4.1.2. ....
  - 4.2. ....
  - 4.3. ....
  - 4.4. ....
  - 4.5. ....
- 5. PROCEDURE
  - 5.1. Preparation of the test sample
    - 5.1.1. Concentrated butter
    - 5.1.2. Butter
  - 5.2. Determination
  - 5.3. Calibration
- 6. CALCULATION OF THE RESULTS
  - 6.1. ....
- 7. ACCURACY OF THE METHOD
  - 7.1. Repeatability
    - 7.1.1. Butter analysis
    - 7.1.2. Concentrated butter analysis
  - 7.2. Reproducibility
    - 7.2.1. Butter analysis
  - 7.3. Concentrated butter analysis
  - 7.4. Source of precision data
- 8. TOLERANCE LIMITS
  - 8.1. ....
  - 8.2. Butter
    - 8.2.1. ....
    - 8.2.2. ....
  - 8.3. Concentrated butter
    - 8.3.1. ....

## ANNEX VIII

### (Article 5)

#### DETERMINING SITOSTEROL OR STIGMASTEROL IN BUTTER OR CONCENTRATED BUTTER BY...

- 1. SCOPE AND FIELD OF APPLICATION
- 2. PRINCIPLE
- 3. APPARATUS
  - 3.1. ....
  - 3.2. ....
  - 3.3. ....
  - 3.4. ....
  - 3.5. ....
  - 3.6. ....
  - 3.7. ....
  - 3.8. ....
    - 3.8.1. ....

	3.8.2.	.....
	3.8.3.	.....
	3.8.4.	.....
	3.9.	.....
	3.10.	.....
4.	REAGENTS	
	4.1.	.....
	4.2.	.....
	4.3.	.....
	4.3.1.	.....
	4.3.1.1.	.....
	4.3.1.2.	.....
	4.4.	.....
	4.5.	.....
	4.6.	.....
	4.7.	.....
	4.8.	.....
	4.8.1.	.....
	4.9.	.....
	4.9.1.	.....
	4.10.	.....
5.	METHOD	
	5.1.	Preparation of standard solutions for chromatography
	5.1.1.	.....
	5.1.2.	.....
	5.2.	Preparation of the unsaponifiables
	5.2.1.	.....
	5.2.2.	.....
	5.2.3.	.....
	5.3.	Preparation of trimethyl silyl ethers
	5.3.1.	.....
	5.3.2.	.....
	5.4.	Gas-chromatographic analysis
	5.4.1.	Choice of operating conditions
	5.4.2.	Analytical procedure
6.	CALCULATION OF RESULTS	
	6.1.	.....
7.	ACCURACY OF THE METHOD	
	7.1.	Butter
	7.1.1.	Repeatability
	7.1.1.1.	Stigmasterol
	7.1.1.2.	Sitosterol
	7.1.2.	Reproducibility
	7.1.2.1.	Stigmasterol
	7.1.2.2.	Sitosterol
	7.1.3.	Source of precision data
	7.2.	Concentrated butter
	7.2.1.	Repeatability
	7.2.1.1.	Stigmasterol
	7.2.1.2.	Sitosterol
	7.2.2.	Reproducibility
	7.2.2.1.	Stigmasterol
	7.2.2.2.	Sitosterol

- 7.2.3. Source of precision data
- 8. TOLERANCE LIMITS
  - 8.1. ....
  - 8.2. Butter
    - 8.2.1. Stigmasterol
      - 8.2.1.1. ....
      - 8.2.1.2. ....
    - 8.2.2. Sitosterol
      - 8.2.2.1. ....
      - 8.2.2.2. ....
  - 8.3. Concentrated butter
    - 8.3.1. Stigmasterol
      - 8.3.1.1. ....
      - 8.3.1.2. ....
    - 8.3.2. Sitosterol
      - 8.3.2.1. ....
      - 8.3.2.2. ....

## ANNEX IX

(Article 6)

## REFERENCE METHOD FOR THE DETECTION OF COWS' MILK AND CASEINATE...

- 1. SCOPE
- 2. FIELD OF APPLICATION
- 3. PRINCIPLE OF THE METHOD
  - 3.1. ....
  - 3.2. ....
  - 3.3. ....
  - 3.4. ....
- 4. REAGENTS
  - Isoelectric focusing
    - 4.1. Reagents for production of the urea containing polyacrylamide gels
      - 4.1.1. Stock gel solution
      - 4.1.2. Gel solution
      - 4.1.3. Catalyst solutions
        - 4.1.3.1. ....
        - 4.1.3.2. ....
    - 4.2. Contact fluid
    - 4.3. Anode solution
    - 4.4. Cathode solution
  - Sample preparation
    - 4.5. Reagents for protein isolation
      - 4.5.1. ....
      - 4.5.2. ....
      - 4.5.3. ....
    - 4.6. Protein dissolving buffer
    - 4.7. Reagents for plasmin cleavage of caseins
      - 4.7.1. Ammonium carbonate buffer
      - 4.7.2. ....



- 4.7.3.  $\epsilon$ -Aminocaproic acid solution for enzyme inhibition
- 4.8. Standards
  - 4.8.1. ....
  - 4.8.2. Preparation of laboratory interim-standards of buffalos' renneted milk containing 0 %...
- Reagents for protein staining
- 4.9. Fixative
- 4.10. Destaining solution
- 4.11. Staining solutions
  - 4.11.1. Staining solution (stock solution 1)
  - 4.11.2. Staining solution (stock solution 2)
  - 4.11.3. Staining solution (working solution)
- 5. EQUIPMENT
  - 5.1. ....
  - 5.2. ....
  - 5.3. ....
  - 5.4. ....
  - 5.5. ....
  - 5.6. ....
  - 5.7. ....
  - 5.8. ....
  - 5.9. ....
  - 5.10. ....
  - 5.11. ....
  - 5.12. ....
  - 5.13. ....
  - 5.14. ....
  - 5.15. ....
  - 5.16. ....
  - 5.17. ....
  - 5.18. ....
  - 5.19. ....
- 6. PROCEDURE
  - 6.1. Sample preparation
    - 6.1.1. Isolation of caseins
    - 6.1.2. Plasmin cleavage of  $\beta$ -caseins to intensify  $\gamma$ -caseins
  - 6.2. Preparation of the urea containing polyacrylamide gels
  - 6.3. Isoelectric focusing
    - Conditions for isoelectric focusing:
    - 6.3.1. Gel size  $265 \times 125 \times 0,25$  mm
    - 6.3.2. Example of a voltage programme for an automatic electrophoresis device...
  - 6.4. Protein staining
    - 6.4.1. Protein fixation
    - 6.4.2. Washing and staining the gel plate
    - 6.4.3. Destaining the gel plate
- 7. EVALUATION
  - 7.1. Visual estimation
  - 7.2. Densitometric estimation
- 8. REFERENCES

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(Article 7)

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1. PREPARATION OF SAMPLES
2. PROCEDURE
3. RESULTS

## ANNEX XI

(Article 8)

## DETERMINATION OF LACTOSE IN COMPOUND FEEDINGSTUFFS

1. SCOPE AND FIELD OF APPLICATION
2. REFERENCE
3. DEFINITION
4. PRINCIPLE
5. REAGENTS
  - 5.1. General
  - 5.2. Lactose
  - 5.3. Concentrated Biggs/Szijarto solution
  - 5.4. Diluted Biggs/Szijarto solution
  - 5.5. Preparation of HPLC grade water
6. APPARATUS
  - 6.1. HPLC ion exchange resin column
  - 6.2. Guard column
  - 6.3. Column oven
  - 6.4. HPLC pump
  - 6.5. HPLC injection device
  - 6.6. HPLC detector
  - 6.7. Integrator
  - 6.8. Water purification unit
  - 6.9. Solvent filtration unit
  - 6.10. Analytical balance
  - 6.11. Water bath
  - 6.12. Centrifuge
  - 6.13. Volumetric flask 50 mL
  - 6.14. Volumetric flask 100 mL
  - 6.15. Graduated pipette
7. SAMPLING
8. PREPARATION OF LACTOSE STANDARD SOLUTION
  - 8.1. Standard 1
  - 8.2. Standard 2
9. PREPARATION OF THE TEST SAMPLE
  - 9.1. Reconstitution of the sample
  - 9.2. Sample treatment
10. HPLC DETERMINATION
  - 10.1. Preliminary preparation of HPLC
    - 10.1.1. Installation of the column and pre-column
    - 10.1.2. Detector and initial flow
    - 10.1.3. Column oven and final flow-rate

- 10.1.4. Integration
- 10.1.5. Guard column test
- 10.2. Running standards
- 10.3. Running samples
- 11. CALCULATION AND EXPRESSION OF THE RESULTS
  - 11.1. Calibration
  - 11.2. Samples
- 12. PRECISION
  - 12.1. Repeatability
  - 12.2. Reproducibility
- 13. REFERENCES

## ANNEX XII

### (Article 9)

#### DETECTION OF RENNET WHEY IN SKIMMED-MILK POWDER FOR PUBLIC STORAGE...

- 1. SCOPE AND FIELD OF APPLICATION
- 2. REFERENCE
- 3. DEFINITION
- 4. PRINCIPLE
- 5. REAGENTS
  - 5.1. Trichloroacetic acid solution
  - 5.2. Eluent solution, pH 6,0
  - 5.3. Flushing solvent
  - 5.4. Standard samples
    - 5.4.1. ....
    - 5.4.2. ....
- 6. APPARATUS
  - 6.1. ....
  - 6.2. ....
  - 6.3. ....
  - 6.4. ....
  - 6.5. ....
  - 6.6. ....
  - 6.7. ....
  - 6.8. ....
  - 6.9. ....
  - 6.10. ....
  - 6.11. ....
    - 6.11.1. ....
    - 6.11.2. ....
    - 6.11.3. ....
    - 6.11.4. ....
    - 6.11.5. ....
    - 6.11.6. ....
- 7. SAMPLING
  - 7.1. ....
  - 7.2. ....
- 8. PROCEDURE
  - 8.1. Preparation of the test sample

**Changes to legislation:** There are currently no known outstanding effects for the Commission Regulation (EC) No 273/2008 (repealed). (See end of Document for details)

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- 8.2. Test portion
- 8.3. Removal of fat and proteins
  - 8.3.1. ....
  - 8.3.2. ....
  - 8.3.3. ....
- 8.4. Chromatographic determination
  - 8.4.1. ....
  - 8.4.2. ....
- 8.5. Calibration
  - 8.5.1. ....
  - 8.5.2. ....
  - 8.5.3. ....
- 9. EXPRESSION OF RESULTS
  - 9.1. Method of calculation and formulae
    - 9.1.1. Calculation of the response factors R:
    - 9.1.2. Calculation of the relative area of the peaks in the...
    - 9.1.3. Calculation of the relative retention time of peak III in...
    - 9.1.4. Experiments have shown that there is a linear relation between...
  - 9.2. Calculation of the percentage of rennet whey powder in the...
  - 9.3. Accuracy of the procedure
    - 9.3.1. Repeatability
    - 9.3.2. Reproducibility
  - 9.4. Interpretation
    - 9.4.1. ....
    - 9.4.2. ....
    - 9.4.3. ....
      - 9.4.3.1. ....
      - 9.4.3.2. ....

## ANNEX XIII

(Article 9)

### DETERMINING RENNET WHEY SOLIDS IN SKIMMED-MILK POWDER AND THE MIXTURES...

1. PURPOSE: DETECTING THE ADDITION OF RENNET WHEY SOLIDS TO:
2. REFERENCES: INTERNATIONAL STANDARD ISO 707
3. DEFINITION
4. PRINCIPLE
5. REAGENTS
  - 5.1. Trichloroacetic acid solution
  - 5.2. Eluents A and B
  - 5.3. Conservation of the column
  - 5.4. Standard samples
    - 5.4.1. ....
    - 5.4.2. ....
    - 5.4.3. ....
6. APPARATUS

6.1.	.....
6.2.	.....
6.3.	.....
6.4.	.....
6.5.	.....
6.6.	.....
6.7.	.....
6.8.	.....
6.9.	.....
6.10.	.....
6.11.	.....
6.11.1.	.....
6.11.2.	.....
6.11.3.	.....
6.11.4.	.....
6.11.5.	.....
6.11.6.	.....
7.	SAMPLING
7.1.	.....
7.2.	.....
8.	PROCEDURE
8.1.	Preparation of the test sample
8.2.	Test portion
8.3.	Removal of fat and proteins
8.3.1.	.....
8.3.2.	.....
8.3.3.	.....
8.4.	Chromatographic determination
8.4.1.	.....
8.4.2.	.....
8.4.3.	.....
8.4.4.	.....
8.5.	Calibration
8.5.1.	.....
8.5.2.	.....
8.5.3.	.....
9.	EXPRESSION OF RESULTS
9.1.	Method of calculation and formulae
9.1.1.	Calculation of the response factor R:
9.2.	Calculation of the percentage of rennet whey powder in the...
9.3.	Accuracy of the procedure
9.3.1.	Repeatability
9.3.2.	Reproducibility
9.3.3.	Linearity
9.4.	Interpretation
Table 1 Ni –4,6 standard	

## ANNEX XIV

## (Article 10)

## SKIMMED-MILK POWDER: QUANTITATIVE DETERMINATION OF PHOSPHATIDYLSERINE AND PHOSPHATIDYLETHANOLAMINE

1. PURPOSE AND FIELD OF APPLICATION
2. DEFINITION
3. PRINCIPLE OF THE METHOD
4. REAGENTS
  - 4.1. Standard material: PEDP, at least 99 % pure
  - 4.2. Reagents for standard sample and test sample preparation
    - 4.2.1. ....
    - 4.2.2. ....
    - 4.2.3. ....
  - 4.3. Reagents for o-phthaldialdehyde derivatisation
    - 4.3.1. ....
    - 4.3.2. ....
    - 4.3.3. ....
    - 4.3.4. ....
  - 4.4. HPLC elution solvents
    - 4.4.1. ....
    - 4.4.2. ....
    - 4.4.3. ....
    - 4.4.4. ....
    - 4.4.5. ....
    - 4.4.6. ....
    - 4.4.7. ....
5. APPARATUS
  - 5.1. ....
  - 5.2. ....
  - 5.3. ....
  - 5.4. ....
  - 5.5. ....
  - 5.6. ....
  - 5.7. ....
  - 5.8. ....
  - 5.9. ....
  - 5.10. ....
  - 5.11. ....
  - 5.12. ....
  - 5.13. ....
    - 5.13.1. ....
    - 5.13.2. ....
    - 5.13.3. ....
    - 5.13.4. ....
    - 5.13.5. ....
    - 5.13.6. ....
6. SAMPLING
7. PROCEDURE
  - 7.1. Preparation of the internal standard solution
    - 7.1.1. ....
    - 7.1.2. ....

- 7.2. Preparation of the test sample solution
  - 7.2.1. ....
  - 7.2.2. ....
  - 7.2.3. ....
- 7.3. Preparation of the external standard solution
  - 7.3.1. ....
  - 7.3.2. ....
- 7.4. Preparation of the derivatising reagent
- 7.5. Determination by HPLC
  - 7.5.1. Elution solvents (4.4)
  - 7.5.2. Suggested eluting gradient:
  - 7.5.3. ....
  - 7.5.4. Column equilibration
  - 7.5.5. ....
  - 7.5.6. Perform the sequence of the chromatographic analyses keeping constant the...
- 7.6. Integration mode
  - 7.6.1. PEDP peak
  - 7.6.2. Tryptamine peak
  - 7.6.3. PS and PE peaks groups
- 8. CALCULATION AND EXPRESSION OF RESULTS
- 9. ACCURACY OF THE METHOD
  - 9.1. Repeatability
  - 9.2. Reproducibility
- 10. REFERENCES
  - 10.1. ....

## ANNEX XV

(Article 11)

### DETECTION OF ANTIMICROBIAL RESIDUES IN SKIMMED MILK POWDER

Positive results are to be interpreted as follows:

## ANNEX XVI

(Article 12)

### QUANTITATIVE DETERMINATION OF SKIMMED-MILK POWDER IN COMPOUND FEEDINGSTUFFS BY ENZYMATIC...

- 1. PURPOSE
- 2. SCOPE
- 3. PRINCIPLE OF THE METHOD
  - 3.1. ....
  - 3.2. ....
  - 3.3. ....
- 4. REAGENTS
  - 4.1. ....
  - 4.2. ....
  - 4.3. ....
  - 4.4. ....
  - 4.5. ....

	4.6.	.....
5.	APPARATUS	
	5.1.	.....
	5.2.	.....
	5.3.	.....
	5.4.	.....
	5.5.	.....
	5.6.	.....
	5.7.	.....
	5.8.	.....
	5.9.	.....
	5.10.	.....
	5.11.	.....
	5.12.	.....
6.	PROCEDURE	
	6.1.	Preparation of the sample
	6.2.	Dissolving of milk powder and separation of the insoluble residue...
	6.2.1.	.....
	6.2.2.	.....
	6.2.3.	.....
	6.2.4.	.....
	6.3.	Coagulation of casein with the enzymes of rennet
	6.3.1.	.....
	6.3.2.	.....
	6.3.3.	.....
	6.3.4.	.....
	6.4.	Determination of casein nitrogen
	6.4.1.	.....
7.	BLANK TEST	
	7.1.	.....
	7.2.	.....
8.	EXPRESSION OF RESULTS	
	8.1.	.....
9.	ACCURACY OF THE METHOD	
	9.1.	Repeatability
	9.2.	Reproducibility
10.	OBSERVATIONS	
	10.1.	.....
	10.2.	.....
	10.3.	.....
	10.4.	.....

## ANNEX XVII

(Article 13)

### DETECTION OF STARCH IN SKIMMED-MILK POWDER, DENATURED MILK POWDER AND...

1.	SCOPE	
2.	PRINCIPLE	
3.	REAGENTS	
	3.1.	.....



- 4. APPARATUS
  - 4.1. ....
  - 4.2. ....
  - 4.3. ....
- 5. PROCEDURE
- 6. EXPRESSION OF RESULTS
- 7. REMARKS

## ANNEX XVIII

(Article 14)

### DETERMINATION OF MOISTURE CONTENT IN DRIED CREAM

- 1. SCOPE
- 2. TERMS AND DEFINITIONS
- 3. PRINCIPLE
- 4. APPARATUS
  - 4.1. ....
  - 4.2. ....
  - 4.3. ....
  - 4.4. ....
  - 4.5. ....
- 5. SAMPLING
- 6. PREPARATION OF TEST SAMPLE
- 7. PROCEDURE
  - 7.1. Preparation of the dish
    - 7.1.1. ....
    - 7.1.2. ....
  - 7.2. Test portion
  - 7.3. Determination
    - 7.3.1. ....
    - 7.3.2. ....
    - 7.3.3. ....
    - 7.3.4. ....
- 8. CALCULATION AND EXPRESSION OF RESULTS
  - 8.1. Calculation
- 9. PRECISION
  - 9.1. Repeatability
  - 9.2. Reproducibility
- 10. TEST REPORT

## ANNEX XIX

(Article 15)

### DETERMINATION OF MOISTURE IN ACID BUTTERMILK POWDER

- 1. SCOPE
- 2. PRINCIPLE
- 3. APPARATUS
  - 3.1. ....
  - 3.2. ....
  - 3.3. ....

- 3.4. ....
- 3.5. ....
- 4. PROCEDURE
- 5. CALCULATION
- 6. PRECISION
  - 6.1. Repeatability limit
  - 6.2. Reproducibility limit
  - 6.3. Source of precision data

## ANNEX XX

(Article 16)

### REFERENCE METHOD FOR THE DETERMINATION OF MILK FAT PURITY BY...

- 1. SCOPE AND FIELD OF APPLICATION
- 2. DEFINITION
- 3. PRINCIPLE OF THE METHOD
- 4. REAGENTS
  - 4.1. ....
  - 4.2. ....
    - 4.2.1. ....
    - 4.2.2. ....
  - 4.3. ....
  - 4.4. ....
  - 4.5. ....
  - 4.6. ....
  - 4.7. ....
- 5. APPARATUS
  - 5.1. High-temperature gas chromatograph
  - 5.2. Chromatography column
    - 5.2.1. Packed column
    - 5.2.2. Capillary column
  - 5.3. ....
  - 5.4. ....
  - 5.5. ....
  - 5.6. ....
  - 5.7. ....
  - 5.8. ....
  - 5.9. ....
  - 5.10. ....
  - 5.11. ....
  - 5.12. ....
  - 5.13. ....
  - 5.14. ....
  - 5.15. ....
  - 5.16. ....
- 6. SAMPLING
- 7. PROCEDURE
  - 7.1. Preparation of test samples
    - 7.1.1. Isolation from butter or butteroil
    - 7.1.2. Extraction according to the Röse–Gottlieb gravimetric method

- 7.1.3. Extraction from milk using silica gel columns
- 7.2. Preparation of sample solution
- 7.3. Chromatographic triglyceride determination
  - 7.3.1. Baseline drift
  - 7.3.2. Injection technique
    - 7.3.2.1. Packed column
    - 7.3.2.2. Capillary column
  - 7.3.3. Calibration
    - 7.3.3.1. General
    - 7.3.3.2. Commercial milk fat standard
    - 7.3.3.3. Laboratory milk fat standard
  - 7.3.4. Chromatographic conditions
    - 7.3.4.1. Packed column
    - 7.3.4.2. Capillary column
- 8. INTEGRATION, EVALUATION AND CONTROL OF THE ANALYTICAL PERFORMANCE
- 9. CALCULATION AND EXPRESSION OF RESULTS
  - 9.1. Triglyceride composition
    - 9.1.1. Calculation
    - 9.1.2. Expression of test results
  - 9.2. S-values
    - 9.2.1. Calculation
      - 9.2.1.1. ....
      - 9.2.1.2. ....
      - 9.2.1.3. ....
      - 9.2.1.4. ....
      - 9.2.1.5. ....
      - 9.2.1.6. ....
    - 9.2.2. Expression of test results
  - 9.3. Detection of foreign fat
- 10. PRECISION
  - 10.1. Interlaboratory test
  - 10.2. Repeatability
  - 10.3. Reproducibility
- 11. UNCERTAINTY OF MEASUREMENT
- 12. TEST REPORT

## ANNEX A

(normative)

### PREPARATION OF THE PACKED COLUMN

#### A.1 REAGENTS AND APPARATUS

- A.1.1 .....
- A.1.2 .....
- A.1.3 .....
- A.1.4 .....
- A.1.5 .....
- A.1.6 .....
- A.1.6.1 .....
- A.1.6.2 .....
- A.1.6.3 .....

**Changes to legislation:** There are currently no known outstanding effects for the Commission Regulation (EC) No 273/2008 (repealed). (See end of Document for details)

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- A.1.6.4 . . . . .
- A.1.6.5 . . . . .
- A.1.6.6 . . . . .
- A.1.6.7 . . . . .
- A.2 SILANIZATION (DEACTIVATION OF THE GLASS SURFACE)
- A.3 FILLING
- A.4 CONDITIONING

ANNEX B  
(informative)

- QUANTIFICATION OF THE FOREIGN FAT CONTENT
- B.1 GENERAL
  - B.2 CALCULATION
  - B.3 EXPRESSION OF TEST RESULTS
  - Bibliography

ANNEX XXI  
(Article 18)

- PROCEDURE APPLICABLE WHEN THE RESULTS OF AN ANALYSIS ARE DISPUTED...
- 1. . . . .
  - 2. . . . .
  - 3. . . . .
  - 4. . . . .

ANNEX XXII  
CORRELATION TABLE

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**Changes to legislation:**

There are currently no known outstanding effects for the Commission Regulation (EC) No 273/2008 (repealed).