Measurement of Sorbic Acid Content of Mayonnaise

The method is based on the NMKL (Nordic Committee of Food Analyses) standard method No. 124(87)

SCOPE: Measurement of Benzoic and Sorbic acid content. The measurement procedure has been validated for the following matrices: juice, margarine, ketchup and mayonnaise.

PRINCIPLE: The preservatives are extracted using methanol and water. The samples are filtered and the resulting solution is injected into HPLC. Supelco C18 column is used. UV-Vis detector at 235 nm is used. The limit of quantification is 5 mg/kg.

Model Equation:

{ The main equation }

$C = (A_{sample} - b_0) / b_1 * L / R;$

{ Chromatographic measurements

Peak areas are used as the basis for quantification.

We assume that the uncertainty of chromatographic peak areas has 3 components (using the sample peak as example):

- Repeatability (contained in A_{sample_rep});

- Uncertainty due to detector drift (A_{sample_drift})

- Uncertainty due to imperfection of peak integration (A_{sample_integr})

The A_{sample_rep} carries the value and the repeatability uncertainty. The other components carry the respective uncertainty component only, their value is 0.

}

{ Sample peak area }

$A_{sample} = A_{sample\_rep}+A_{sample\_drift}+A_{sample\_integr};$

{ Recovery

Recovery has been determined from spiking studies. $f_{inhomog}$ takes into account the possible different behaviour of the spike relative to the native analyte.

}

$R = R_0 * f_{inhomog};$

{ Peak areas of the calibration standard solutions }

$A_1 = A_{1rep} + A_{1drift} + A_{1integr};$

$A_2 = A_{2rep} + A_{2drift} + A_{2integr};$

$A_3 = A_{3rep} + A_{3drift} + A_{3integr};$

$A_4 = A_{4rep} + A_{4drift} + A_{4integr}$;

$A_5 = A_{5rep} + A_{5drift} + A_{5integr}$;

{ Linear regression equations }

$\Sigma AC = C_1*A_1 + C_2*A_2 + C_3*A_3 + C_4*A_4 + C_5*A_5$;

$AvgC=(C_1+C_2+C_3+C_4+C_5)/n$;

$AvgA=(A_1+A_2+A_3+A_4+A_5)/n$;

$\Sigma CC=C_1*C_1+C_2*C_2+C_3*C_3+C_4*C_4+C_5*C_5$;

$b_1=(\Sigma AC-n*AvgC*AvgA)/(\Sigma CC-n*AvgC*AvgC)$;

$b_0=AvgA-b_1*AvgC$

List of Quantities:

Quantity	Unit	Definition
C	mg/kg	Sorbic acid content of the sample
A_{sample}	AU	Peak area of the sample solution
b_0	AU	Intercept of the calibration line
b_1	AU*l/mg	Slope of the calibration line
L	l/kg	Dilution factor
R	unitless	Recovery
A_{sample_rep}	AU	Sample solution peak area together with its repeatability uncertainty
A_{sample_drift}	AU	Drift uncertainty component of the sample peak area
A_{sample_integr}	AU	Integration uncertainty component of the sample peak area
R_0	unitless	The value of R and its uncertainty component that is due to the imperfection of the sample preparation procedure
$f_{inhomog}$	unitless	Inhomogeneity factor that takes into account that that as the result of the spiking
A_1	AU	Peak area of the 1. calibration standard solution
A_{1rep}	AU	alue and the repeatability uncertainty component of the peak area of the 1. calibration standard solution
A_{1drift}	AU	Drift uncertainty component of the peak area of the 1. calibration standard solution
$A_{1integr}$	AU	Integration uncertainty component of the peak area of the 1. calibration standard solution
A_2	AU	Peak area of the 2. calibration standard solution
A_{2rep}	AU	alue and the repeatability uncertainty component of the peak area of the 2. calibration standard solution

Quantity	Unit	Definition
A_{2drift}	AU	Drift uncertainty component of the peak area of the 2. calibration standard solution
$A_{2integr}$	AU	Integration uncertainty component of the peak area of the 2. calibration standard solution
A_3	AU	Peak area of the 3. calibration standard solution
A_{3rep}	AU	alue and the repeatability uncertainty component of the peak area of the 3. calibration standard solution
A_{3drift}	AU	Drift uncertainty component of the peak area of the 3. calibration standard solution
$A_{3integr}$	AU	Integration uncertainty component of the peak area of the 3. calibration standard solution
A_4	AU	Peak area of the 4. calibration standard solution
A_{4rep}	AU	alue and the repeatability uncertainty component of the peak area of the 4. calibration standard solution
A_{4drift}	AU	Drift uncertainty component of the peak area of the 4. calibration standard solution
$A_{4integr}$	AU	Integration uncertainty component of the peak area of the 4. calibration standard solution
A_5	AU	Peak area of the 5. calibration standard solution
A_{5rep}	AU	alue and the repeatability uncertainty component of the peak area of the 5. calibration standard solution
A_{5drift}	AU	Drift uncertainty component of the peak area of the 5. calibration standard solution
$A_{5integr}$	AU	Integration uncertainty component of the peak area of the 5. calibration standard solution
ΣAC	AU*mg/l	Interim quantity for regression statistics analysis
C_1	mg/l	Concentration of the 1. calibration standard solution
C_2	mg/l	Concentration of the 2. calibration standard solution
C_3	mg/l	Concentration of the 3. calibration standard solution
C_4	mg/l	Concentration of the 4. calibration standard solution
C_5	mg/l	Concentration of the 5. calibration standard solution
AvgC	mg/l	Interim quantity for regression statistics calculation
n	unitless	Number of points on the calibration line
AvgA	AU	Interim quantity for regression statistics calculation
ΣCC	mg^2/l^2	Interim quantity for regression statistics calculation

C: Result

A_{sample}: Interim Result

b_0: Interim Result

b_1: Interim Result

L: Type B rectangular distribution
Value: 20 l/kg
Halfwidth of Limits: 0.2 l/kg

R: Interim Result

A_{sample_rep}: Type B rectangular distribution
Value: 200 AU
Halfwidth of Limits: 2 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{sample_drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

A_{sample_integr}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 4 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

R_0: Type B rectangular distribution
Value: 0.93 unitless
Halfwidth of Limits: 0.07 unitless

The uncertainty of R_0 is essentially the uncertainty of sample preparation. As such it also incorporates a large part of the overall reproducibility of the method.

$f_{inhomog}$: Type B rectangular distribution
Value: 1 unitless
Halfwidth of Limits: 0.03 unitless

Uncertainty of the inhomogenity factor takes into account that as the result of the spiking procedure the analyte added does not have the same conditions in the sample as the analyte already present in the sample.

A_1: Interim Result

A_{1rep}: Type B rectangular distribution
Value: 50 AU
Halfwidth of Limits: 0.5 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{1drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: .5 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

$A_{1integr}$: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

A_2: Interim Result

A_{2rep}: Type B rectangular distribution
Value: 100 AU
Halfwidth of Limits: 1 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{2drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 1 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

$A_{2integr}$: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

A_3: Interim Result

A_{3rep}: Type B rectangular distribution
Value: 250 AU
Halfwidth of Limits: 2.5 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{3drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2.5 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

$A_{3integr}$: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

A_4: Interim Result

A_{4rep}: Type B rectangular distribution
Value: 500 AU
Halfwidth of Limits: 5 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{4drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 5 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

$A_{4integr}$: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

A_5: Interim Result

A_{5rep}: Type B rectangular distribution
Value: 750 AU
Halfwidth of Limits: 7.5 AU

The repeatability uncertainty of the sample peak area has been estimated as ca ± 1% of the peak area

A_{5drift}: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 7.5 AU

The uncertainty due to detector drift has been estimated as not more than 1% of the peak area

$A_{5integr}$: Type B rectangular distribution
Value: 0 AU
Halfwidth of Limits: 2 AU

This uncertainty is estimated to be higher with sample than with calibration solutions: ca ± 6 area units (as a first approximation, not dependent on the area)

ΣAC: Interim Result

C_1: Type B normal distribution
Value: 5 mg/l
Expanded Uncertainty: 0.05 mg/l
Coverage Factor: 1

C_2: Type B normal distribution
Value: 10 mg/l
Expanded Uncertainty: 0.1 mg/l
Coverage Factor: 1

C_3: Type B normal distribution
Value: 25 mg/l
Expanded Uncertainty: 0.25 mg/l
Coverage Factor: 1

C_4: Type B normal distribution
Value: 50 mg/l
Expanded Uncertainty: 0.5 mg/l
Coverage Factor: 1

C_5: Type B normal distribution
Value: 75 mg/l
Expanded Uncertainty: 0.75 mg/l
Coverage Factor: 1

AvgC: Interim Result

n: Constant
Value: 5 unitless

AvgA: Interim Result

ΣCC: Interim Result

Uncertainty Budget:

Quantity	Value	Standard Uncertainty	Distribution	Sensitivity Coefficient	Uncertainty Contribution	Index
L	20.000 l/kg	0.115 l/kg	rectangular	22	2.5 mg/kg	1.3 %
A_{sample_rep}	200.00 AU	1.15 AU	rectangular	2.2	2.5 mg/kg	1.3 %
A_{sample_drift}	0.0 AU	1.15 AU	rectangular	2.2	2.5 mg/kg	1.3 %
A_{sample_integr}	0.0 AU	2.31 AU	rectangular	2.2	5.0 mg/kg	5.4 %
R_0	0.9300 unitless	0.0404 unitless	rectangular	-460	-19 mg/kg	76.5 %
$f_{inhomog}$	1.0000 unitless	0.0173 unitless	rectangular	-430	-7.5 mg/kg	12.1 %
A_{1rep}	50.000 AU	0.289 AU	rectangular	-0.66	-0.19 mg/kg	0.0 %
A_{1drift}	0.0 AU	0.289 AU	rectangular	-0.66	-0.19 mg/kg	0.0 %
$A_{1integr}$	0.0 AU	1.15 AU	rectangular	-0.66	-0.76 mg/kg	0.1 %
A_{2rep}	100.000 AU	0.577 AU	rectangular	-0.62	-0.36 mg/kg	0.0 %
A_{2drift}	0.0 AU	0.577 AU	rectangular	-0.62	-0.36 mg/kg	0.0 %
$A_{2integr}$	0.0 AU	1.15 AU	rectangular	-0.62	-0.71 mg/kg	0.1 %
A_{3rep}	250.00 AU	1.44 AU	rectangular	-0.50	-0.71 mg/kg	0.1 %
A_{3drift}	0.0 AU	1.44 AU	rectangular	-0.50	-0.71 mg/kg	0.1 %
$A_{3integr}$	0.0 AU	1.15 AU	rectangular	-0.50	-0.57 mg/kg	0.0 %
A_{4rep}	500.00 AU	2.89 AU	rectangular	-0.29	-0.84 mg/kg	0.2 %
A_{4drift}	0.0 AU	2.89 AU	rectangular	-0.29	-0.84 mg/kg	0.2 %
$A_{4integr}$	0.0 AU	1.15 AU	rectangular	-0.29	-0.34 mg/kg	0.0 %
A_{5rep}	750.00 AU	4.33 AU	rectangular	-0.088	-0.38 mg/kg	0.0 %
A_{5drift}	0.0 AU	4.33 AU	rectangular	-0.088	-0.38 mg/kg	0.0 %
$A_{5integr}$	0.0 AU	1.15 AU	rectangular	-0.088	-0.10 mg/kg	0.0 %
C_1	5.0000 mg/l	0.0500 mg/l	normal	6.6	0.33 mg/kg	0.0 %
C_2	10.000 mg/l	0.100 mg/l	normal	6.2	0.62 mg/kg	0.0 %
C_3	25.000 mg/l	0.250 mg/l	normal	5.0	1.2 mg/kg	0.3 %
C_4	50.000 mg/l	0.500 mg/l	normal	2.9	1.5 mg/kg	0.5 %
C_5	75.000 mg/l	0.750 mg/l	normal	0.88	0.66 mg/kg	0.0 %
n	5.0 unitless					
C	430.1 mg/kg	21.4 mg/kg				

Result: Quantity: C
Value: 430 mg/kg
Expanded Uncertainty: ±43 mg/kg
Coverage Factor: 2.00
Coverage: manual