MCPD and glycidyl esters in the food chain

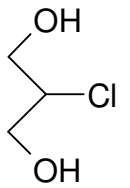
Kerstin Knott

Chiron AS, Trondheim, Norway

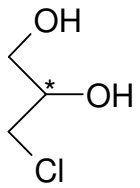
Content

- What exactly are MCPD and glycidyl esters?
- History of 3-MCPD esters and glycidyl esters in foodstuffs
- Formation of 3-MCPD and related compounds
- Toxicology
- Simultaneous Monitoring of 2-MCPD, 3-MCPD and glycidyl esters
- Challenges

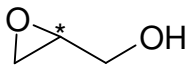
Free MCPD (monochloropropanediol) and glycidol



2-MCPD

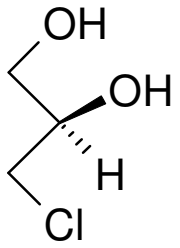


3-MCPD

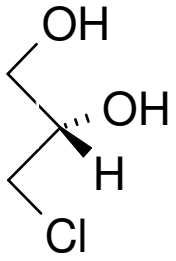


Glycidol

Chirality:



(*R*)-(-)-3-MCPD



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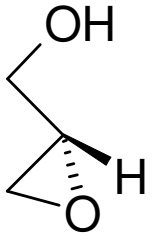
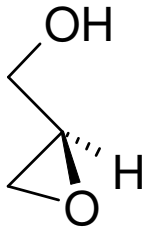
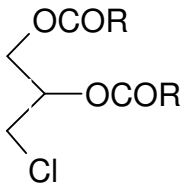


(*S*)-(-)-Glycidol

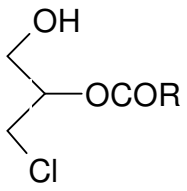


(*R*)-(+)-Glycidol

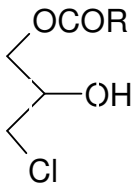
Bound MCPD and glycidol



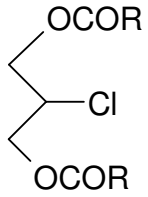
3-MCPD diester



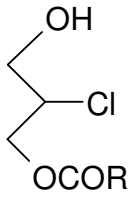
3-MCPD 2-monoester



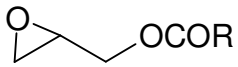
3-MCPD 1-monoester



2-MCPD diester



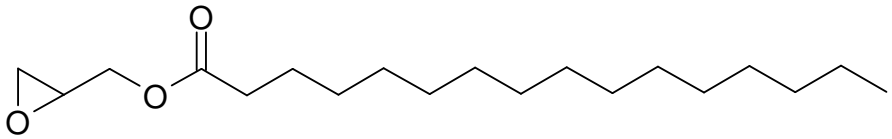
2-MCPD monoester



Glycidyl ester

R = fatty acid

e.g. palmitic acid



History and general background

- 3-MCPD is known since 1978 as food processing contaminant in hydrolyzed vegetable proteins (HVP).
- 2004: Data published on the ocurrence of 3-MCPD fatty acid esters in various types of processed food.
- 2008: Glycidyl esters were detected in palm oil.
- 3-MCPD and glycidyl ester are not found in virgin oils.
- EC recommends the monitoring of the presence of MCPD, MCPD-esters and glycidyl esters in vegetable oils and fats.

History and general background

- Toxicology still under evaluation.

- European Food Safety Authority (EFSA) Panel on Contaminants in the Food Chain (CONTAM) : 100 % metabolism of bound 3-MCPD to free 3-MCPD in humans.

- Commission Regulation (EC) 1881/2006:
  - TDI 2 µg/kg bodyweight per day of bound 3-MCPD.
  - MRL of 3-MCPD in HVP 20 µg/kg.

- Preferred method for the determination of ester bound MCPD and glycidol is published by The American Oil Chemists' Society (AOCS).

Important questions?

- What is the toxicological assessment of 3-MCPD and glycidyl esters?
- What is the mechanism of formation?
- How is it possible to reduce the esters during processing without impairing the quality of the product?
- Do we have robust methods of determining the esters?

Formation of chloropropanediols and glycidyl esters

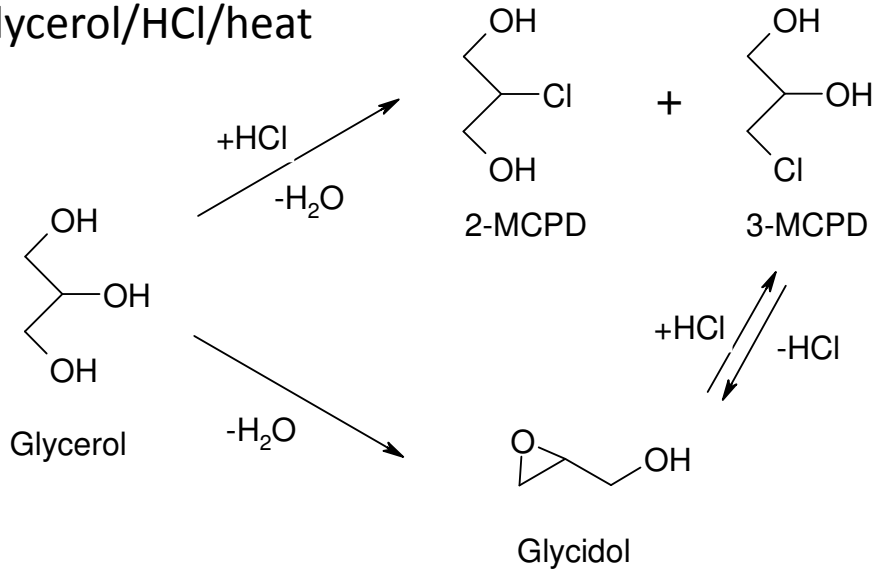
Mechanism of formation:

- Processing food (e.g. deodorization step of edible oil)
- Domestic preparation (grilled cheese, toasted bread)
- Food-contact materials
  (polyamidoamine-epichlorohydrin)

Formation of chloropropanediols and glycidyl esters

Potential precursors:

- Lipids/HCl/heat
- Glycerol/HCl/heat



Formation of chloropropanediols and glycidyl esters

Potential precursors:

- Lipids/HCl/heat
- Glycerol/HCl/heat
- Allyl alkohol/HOCl
- Carbohydrates/HCl (from residual lipids?)
- Natural occurring 3-MCPD esters (in goat milk)

"Goats in mountains" by Taken byfir0002 | flagstaffotos.com.auCanon 20D + Tamron 28-75mm f/2.8 - Own work. Licensed under GFDL 1.2 via Wikimedia Commons – http://commons.wikimedia.org/wiki/File:Goats_in_mountains.jpg#mediaviewer/File:Goats_in_mountains.jpg

Production of edible vegetable oil

http://www.turismo.intoscana.it/allthingstuscany/tuscanycious/files/2010/06/DSCF2524.JPG

Extraction

Crude edible oil

Degumming
Neutralisation

Bleaching

Deodorization

Refined edible oil

Deodorization step

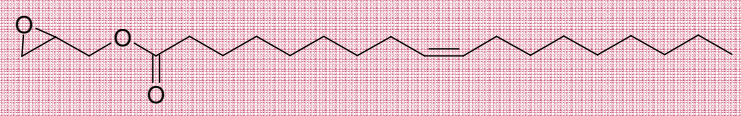
Mono, di- and triacylglycerides

Deodorization step >200°C

Deodorization step >200°C

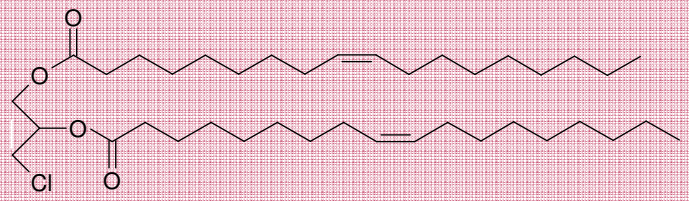
- Natural HCl sources like NaCl, FeClx and chlorinated phytosphingosides
- Acidic hydrolysis with mineral acids

Glycidylester



Glycidyl oleate
C18:1 (9-cis)
9671.21

2- and 3-MCPD mono- and di-esters



3-MCPD-1,2-diooleate
C18:1 (9-cis)
8970.39

Modified from Kuhlmann J., «Analysis & occurrence of bound glycidol and MCPD in oil containing food», presentation at the 103rd AOCS annual meeting and expo, 2012.

Metabolism in vivo and toxicology

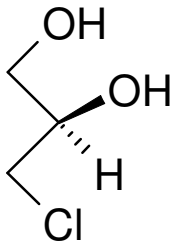
- Little toxicological data available on MCPD and glycidyl esters

- Main toxicological concern: potential to release free 3-MCPD and glycidol during digestion

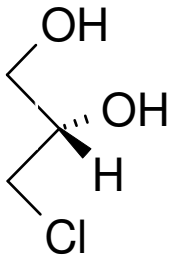
- Toxicology data on free MCPD and glycidol:
  - 3-MCPD: non-genotoxic carcinogen (IARC group 2A).
  - 2-MCPD: little data available.
  - Glycidol: genotoxic carcinogen (IARC group 2A).

- "Comparison between 3-MCPD and its palmitic esters in a 90-day toxicological study", E. Barocelli et al., University of Parma, 2011.

Biological activity of (*S*)- and (*R*)-stereoisomer of 3-MCPD are different:



(*R*)-(-)-3-MCPD



(*S*)-(+)-3-MCPD

Detrimental effect on kidneys

Antifertility activity

Analysis methods

Direct method by analysing all intact and individual MCPD and glycidyl esters

- LC-MS

- Oil containing 3 different fatty acids:
  MCPD mono-ester: 9
  MCPD-diester: 15
  Glycidyl ester: 3

→ total: 27 potential different analytes

- Detailed information about all individual contaminants

Modified from Kuhlmann J., «Analysis & occurrence of bound glycidol and MCPD in oil containing food», presentation at the 103rd AOCS annual meeting and expo, 2012.

Analysis methods

Indirect method by analysing free MCPD and glycidol

- Oil containing 3 different fatty acids: 3 different analytes, 2- MCPD, 3-MCPD and glycidol

- GC-MS:
  - enzymatic or acidic hydrolysation of esters
  - derivatisation

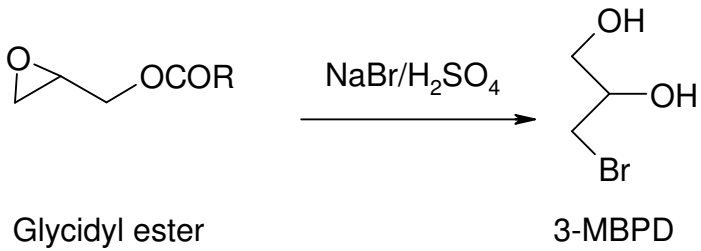
- Internal standards

- AOCS Official Method Cd 29a-13, Cd 29b-13 and Cd 29c-13: simultaneous monitoring of 2- MCPD, 3-MCPD and glycidyl esters in edible oils and fats

AOAC Official Method Cd 29a-13

Test sample preparation (I):

- oil sample, THF, internal standards, vortex mixer
- incubation: aqu. NaBr/H2SO4, 50°C, 15 min
- aqu. NaHCO3
- addition of n-heptane and separation of layers
- evaporation to dryness, dissolve in THF

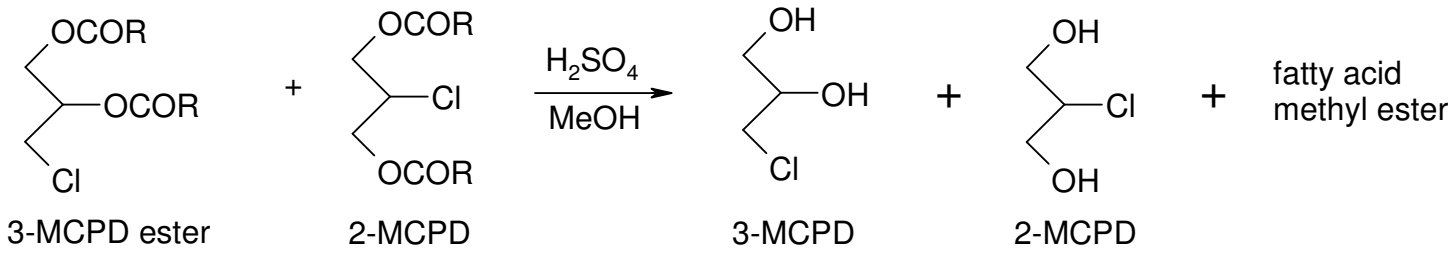


Glycidyl ester 3-MBPD

AOAC Official Method Cd 29a-13

Test sample preparation (II):

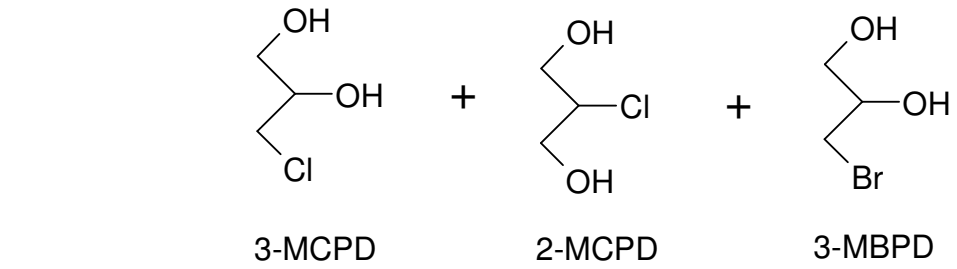
- H2SO4/methanol, 40°C, 16 h, then aqu. NaHCO3,
- evap. to dryness and extraction of fatty acid methyl esters with n-heptane



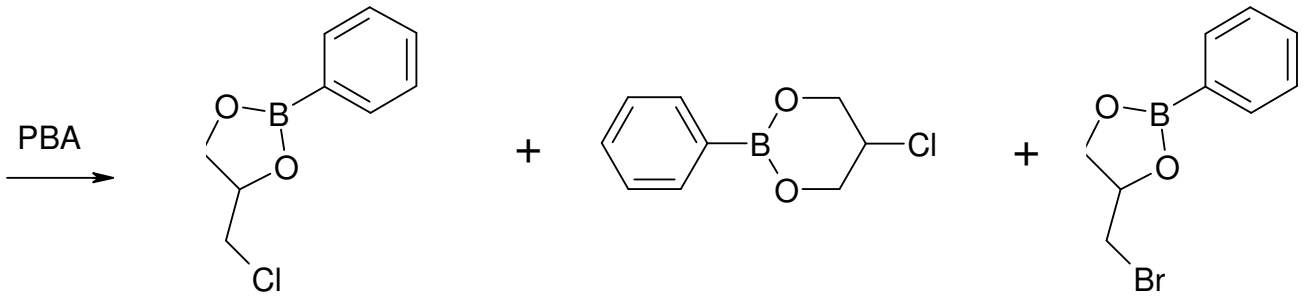
AOAC Official Method Cd 29a-13

Test sample preparation (III):

- phenylboronic acid, ultrasonic bath, 5 min
- extration of phenylboronic derivatives of 2- and 3-MCPD and 3-MBPD with n-heptane

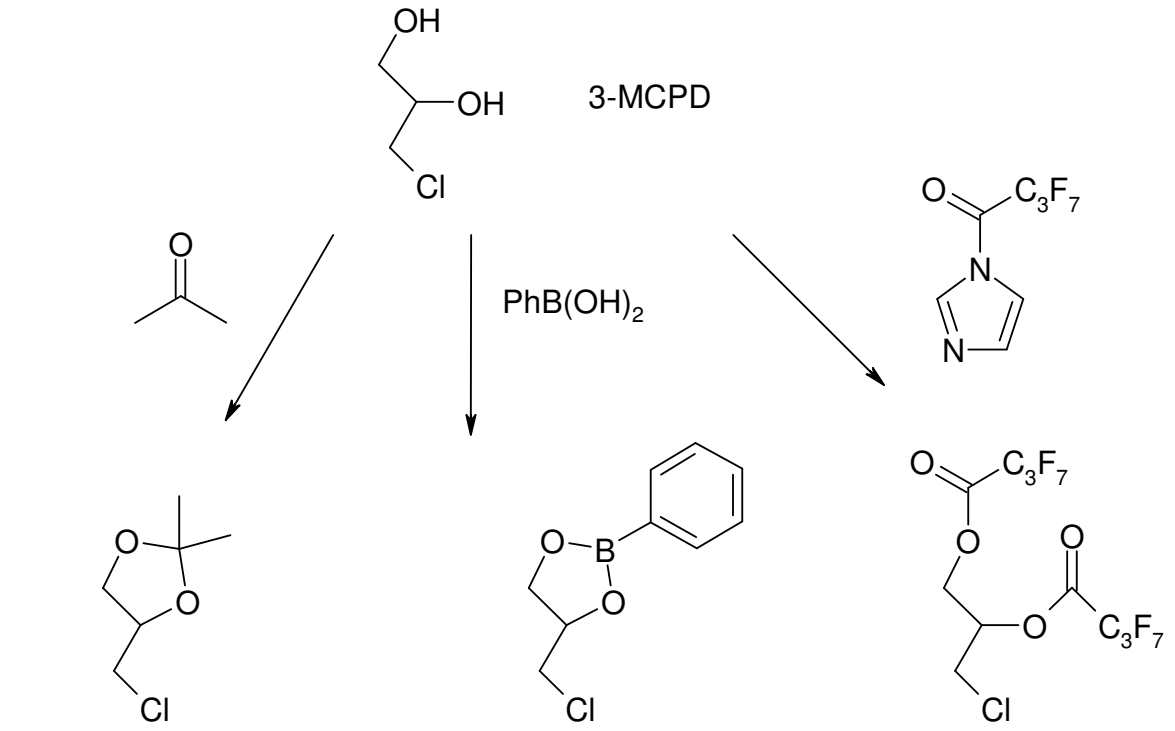


3-MCPD 2-MCPD 3-MBPD



3-MCPD-PBA 2-MCPD-PBA 3-MBPD-PBA

Dervatization of 3-MCPD prior to GC-MS analysis



3-MCPD acetonide

3-MCPD
phenylboronic acid ester

3-MCPD-
diheptafluorobutyric ester

Chiron standards

2-MCPD diesters:

Chiron Cat. No.	Name	Abbreviation	Concentration
10523.35-K-T	2-Chloro-1,3-propanediol-dipalmitate, PP-2-MCPD	2-MCPD-di16:0	1000µg/mL in toluene
10559.39-K-T	2-Chloro-1,3-propanediol-disterarate	2-MCPD-di16:0	1000µg/mL in toluene

3-MCPD diesters:

Chiron Cat. No.	Name	Abbreviation	Concentration
8967.35-100-T	3-Chloro-1,2-propandiol-dipalmitate, PP-3-MCPD	3-MCPD-di16:0	100µg/mL in toluene
8967.35-K-T	3-Chloro-1,2-propandiol-dipalmitate, PP-3-MCPD	3-MCPD-di16:0	1000µg/mL in toluene
8968.35-100-T	3-Chloro-1,2-propandiol-dipalmitoleate	3-MCPD-di16:1	100µg/mL in toluene
8969.39-100-T	3-Chloro-1,2-propandiol-distearate	3-MCPD-di18:0	100µg/mL in toluene
8970.39-100-T	3-Chloro-1,2-propandiol-dioleate	3-MCPD-di18:1	100µg/mL in toluene
8971.39-100-ME	3-Chloro-1,2-propandiol-dilinoleate	3-MCPD-di18:2	100µg/mL in methanol
8972.43-100-T	3-Chloro-1,2-propandiol-diarachidate	3-MCPD-di20:0	100µg/mL in toluene
8973.43-100-T	3-Chloro-1,2-propandiol-digadolenate	3-MCPD-di20:1	100µg/mL in toluene
8974.47-100-T	3-Chloro-1,2-propandiol-dibehenate	3-MCPD-di22:0	100µg/mL in toluene
8975.47-100-T	3-Chloro-1,2-propandiol-dierucidate	3-MCPD-di22:1	100µg/mL in toluene

Chiron standards

3-MCPD 1-monoesters:

Chiron Cat. No.	Name	Abbreviation	Concentration
8949.19-100-T	3-Chloro-1,2-propandiol-1-monopalmitate	3-MCPD-1-16:0	100µg/mL in toluene
8950.19-100-T	3-Chloro-1,2-propandiol-1-monopalmitoleate	3-MCPD-1-16:1 (9-cis)	100µg/mL in toluene
8951.21-100-T	3-Chloro-1,2-propandiol-1-monostearate	3-MCPD-1-18:0	100µg/mL in toluene
8952.21-100-T	3-Chloro-1,2-propandiol-1-monooleate	3-MCPD-1-18:1 (9-cis)	100µg/mL in toluene
8953.21-100-T	3-Chloro-1,2-propandiol-1-monolinoleate	3-MCPD-1-18:2 (9,12-dicis)	100µg/mL in toluene
8954.23-100-T	3-Chloro-1,2-propandiol-1-monoarachidate	3-MCPD-1-20:0	100µg/mL in toluene
8955.23-100-T	3-Chloro-1,2-propandiol-1-monogadolenate	3-MCPD-1-20:1 (11-cis)	100µg/mL in toluene
8956.25-100-T	3-Chloro-1,2-propandiol-1-monobehenate	3-MCPD-1-22:0	100µg/mL in toluene
8957.25-100-T	3-Chloro-1,2-propandiol-1-monoerucidate	3-MCPD-1-22:1 (13-cis)	100µg/mL in toluene

Labelled MCPD-esters:

Chiron Cat. No.	Name	Abbreviation	Concentration
8981.19-100-T	3-Chloro-1,2-propandiol-1-monopalmitate-d5	3-MCPD-1-16:0-d5	100µg/mL in toluene
8976.21-100-T	3-Chloro-1,2-propandiol-1-monostearate-d5	3-MCPD-1-18:0-d5	100µg/mL in toluene
8977.25-100-T	3-Chloro-1,2-propandiol-1-monobehenate-d5	3-MCPD-1-22:0-d5	100µg/mL in toluene
8982.35-100-T	3-Chloro-1,2-propandiol-dipalmitate-d5, PP-3-MCPD-d5	3-MCPD-di16:0-d5	100µg/mL in toluene
8982.35-K-T	3-Chloro-1,2-propandiol-dipalmitate-d5, PP-3-MCPD-d5	3-MCPD-di16:0-d5	1000µg/mL in toluene
8978.39-100-T	3-Chloro-1,2-propandiol-distearate-d5	3-MCPD-di18:0-d5	100µg/mL in toluene
8978.39-K-T	3-Chloro-1,2-propandiol-distearate-d5	3-MCPD-di18:0-d5	1000µg/mL in toluene
8979.47-100-T	3-Chloro-1,2-propandiol-dibehenate-d5	3-MCPD-di22:0-d5	100µg/mL in toluene
8979.47-K-T	3-Chloro-1,2-propandiol-dibehenate-d5	3-MCPD-di22:0-d5	1000µg/mL in toluene

Chiron standards

Glycidyl fatty acid esters:

Chiron Cat. No.	Name	Abbreviation	Concentration
9674.19-100-T	Glycidyl palmitate, Gly-P	glycidyl C16:0	100µg/mL
9896.19-100-T	Glycidyl palmitoleate	glycidyl C16:1 (9-cis)	100µg/mL
9899.21-100-T	Glycidyl stearate	glycidyl C18:0	100µg/mL
9671.21-100-T	Glycidyl oleate	glycidyl C18:1 (9-cis)	100µg/mL
9673.21-100-T	Glycidyl linoleate	glycidyl C18:2 (9-cis, 12-cis)	100µg/mL
9672.21-100-T	Glycidyl linolenate	glycidyl C18:3 (9-cis, 12-cis, 15-cis)	100µg/mL
9897.23-100-T	Glycidyl arachidate	glycidyl C20:0	100µg/mL
9900.23-100-T	Glycidyl gondolenate	glycidyl C20:1 (11-cis)	100µg/mL
9898.25-100-T	Glycidyl behenate	glycidyl C22:0	100µg/mL

Labelled glycidyl esters:

Chiron Cat. No.	Name	Abbreviation	Concentration
10524.19-K-T	Glycidyl palmitate-d5, Gly-P-d5	glycidyl C16:0-d5	1000µg/mL
9924.19-K-T	Glycidyl palmitate-d31, Gly-P-d31	glycidyl C16:0-d31	1000µg/mL
9924.19-10MG	Glycidyl palmitate-d31, Gly-P-d31	glycidyl C16:0-d31	neat

Labelled and native glycidol:

Chiron Cat. No.	Name	Abbreviation	Concentration
9714.3-10MG	(+/-)-Glycidol		neat
10551.3-10MG	(+/-)-Glycidol-1,1,2,3,3-d5		neat

Challenges

- Different levels of free and bound MCPD and glycidol in various foodstuffs is not fully understood
- Food processing: lower level without impairing quality of the product?
- Health effects?
- Dietary sources?

Thank you for your kind attention!

Literature:

AOCS Official Method Cd 29a-13: 2- and 3-MCPD Fatty Acid Esters and Glycidol Fatty Acid Esters in Edible Oils and Fats by Transesterification.

Commission Regulation (EC) 1881/2006

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