# Investigation of the formation of 3-MCPD from mono and di-esters of its fatty acids

Area of research interest: Chemical hazards in food and feed
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Conducted by: Premier Analytical Services (with FERA and ICT)
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## Background

Chloropropanols and their fatty acid esters (also known as chloroesters), are contaminants formed during the processing and manufacture of certain foods and ingredients. The formation of chloroesters may be widespread in processed foods derived from cereals, coffee, fish, meat, potatoes, nuts and refined oils. Chloroesters may occur in many ingredients that are processed by heat and the subsequent release of the chemical 3-MCPD from these materials during processing and storage needs to be considered.

The presence of chloropropanols, chloroesters and the breakdown product, 3-MCPD, in foods is of concern because toxicological studies suggest they might pose a risk to human health.

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## Research Approach

These projects aimed to increase our understanding of the occurrence of 3-MCPD fatty acid esters in food.

A literature review was carried out and a report compiled summarising the theoretical and practical aspects of 3-MCPD occurrence in food. Following the review, labelled and non-labelled fatty acid esters, including mono- and diacylglycerols 3-MCPD esters and glycidol esters were synthesised in the laboratory.

An analytical method was developed and validated that enabled the determination of esters of 3-MCPD and related chemicals such as glycidol esters. This method was used to quantify the relative amounts of 3-MCPD, 3-MCPD esters (mono- and diesters) and glycidol esters in food.

The formation, decomposition and mitigation of 3-MCPD esters was studied in a model system using pure reference chemicals. The effects of processing and storage on the relative amounts of 3-MCPD, 3-MCPD esters (mono- and diesters) and glycidol esters in food was also studied and compared to the model system.

The projects enabled the our organisation to identify how contamination of food by 3-MCPD and its esters could be minimised in order to reduce potential risks to human health.

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# Results

1. **Summary**

The results of new research into the formation of chloropropanediol (MCPD) esters and related compounds in both model systems and foods are presented covering four main work areas:

- development and extension of analytical methods for bound forms of MCPD and glycidol (GE)
- occurrence in target foods
- model system studies with pure reference chemicals
- a study of the effects of food processing on the relative amounts of GE and free and bound forms of MCPD in target foodstuffs

### 1.1 Key findings

1.1.1 Development and extension of analytical methods:

- A method was developed to quantify amounts of bound mono- and diesters of MCPD in foods using solid phase extraction to separate mono- and diesters with GC/MS/MS detection.
- A method was developed and validated to quantify individual GE in fats extracted from foods using gel permeation chromatography (GPC) clean up followed by HPLC coupled to either a quadrupole (LC-MS/MS) or Time of Flight mass spectrometer (LC-TOF-MS).
- Extraction of fats from infant formulae with cold solvents was incomplete.
- The use of pressurised liquid extraction at effective temperatures, ie 125°C, appeared to generate GE, even from virgin olive oil.

### 1.1.2 Occurrence in target foods

1.1.2.1 Non-cereal foods (cheeses, salami, cooking oils, potato products):

- For all samples, other than the refined fats and oils, amounts of bound GE and 3-MCPD levels in the whole food were low.
- Consumer exposure to GEs and bound 3-MCPD is likely to be dominated by occurrence in refined vegetable oils.

### 1.1.2.2 Cereal products

- Amounts of bound MCPD measured in a bakery fat and a retail soft dough biscuit were 1,285 and 632 ?g/kg and consistent with amounts reported previously.
- Application of the developed method for bound mono- and diesters of MCPD showed that mono-esters of MCPD accounted for 15.7% and 9.4% of the total bound MCPD in the bakery fat and biscuit respectively.

### 1.1.3 Model system studies with pure chemicals

- A model system was developed to simultaneously measure the kinetics of formation and decomposition of 3-MCPD and glycidol esters (GE) from the starting materials tripalmitin, dipalmitin, monopalmitin, 3-MCPD dipalmitate, 3-MCPD monopalmtitate and glycidyl palmitate.
- Triacylglycerol (TAG) did not act as a direct precursor of 3-MCPD esters and glycidyl esters.

eg tripalmitin must first be hydrolysed to di- and tripalmitin.

- The extent of hydrolysis increased with increasing temperature and led to the formation of 3-MCPD esters and esters of glycidol, but in significantly lower amount than from monopalmitin directly.
- In models with tripalmitin, amounts of 3-MCPD esters and GE were not directly influenced by chloride ions in the investigated range of 0.1-1%.
- In models with monopalmitin, the amount of 3-MCPD esters and glycidol esters formed showed a linear dependence on the concentration of chloride ions.
- In studies with enzymes, both nonspecific and specific lipases did not change the concentration of 3-MCPD esters present in fats under the conditions of enzyme-catalyzed transesterification, ie typical of those used in the production of structural fats for the manufacture of margarines and spreads.

**1.1.4 Effects of processing**

1.1.4.1 Non-cereal foods (cheeses, salami, cooking oils, potato products)

- There was no evidence of substantial changes in GE or bound MCPD concentrations during cooking.

1.1.4.2 Cereal products

- Under conditions simulating the baking of a cereal product (e.g. short dough biscuits) concentrations of added (labelled) and native (present in fats) bound MCPD remained unchanged.
- Under these conditions, some formation and decay of free non-labelled MCPD was observed presumably from precursors present in the recipe ingredients (eg glycerol, acylglycerols, salt)
- In cereal dough containing a commercial lipase, free MCPD isomers were readily released from added (labelled) 3-MCPD esters.

**1.1.5 Recommendations for future work**

1.1.5.1 Processing:

- Studies into the formation of GE under mild temperature conditions including the effect of moisture content, pH, time etc.

1.1.5.2 Analysis:

- Develop / extend methods (direct and indirect) for the analysis of GE with particular reference to:
  - fats extracted from food products
  - matrices where lipids may bind to proteins, such as cheese and infant formulae

Research report

## England, Northern Ireland and Wales

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