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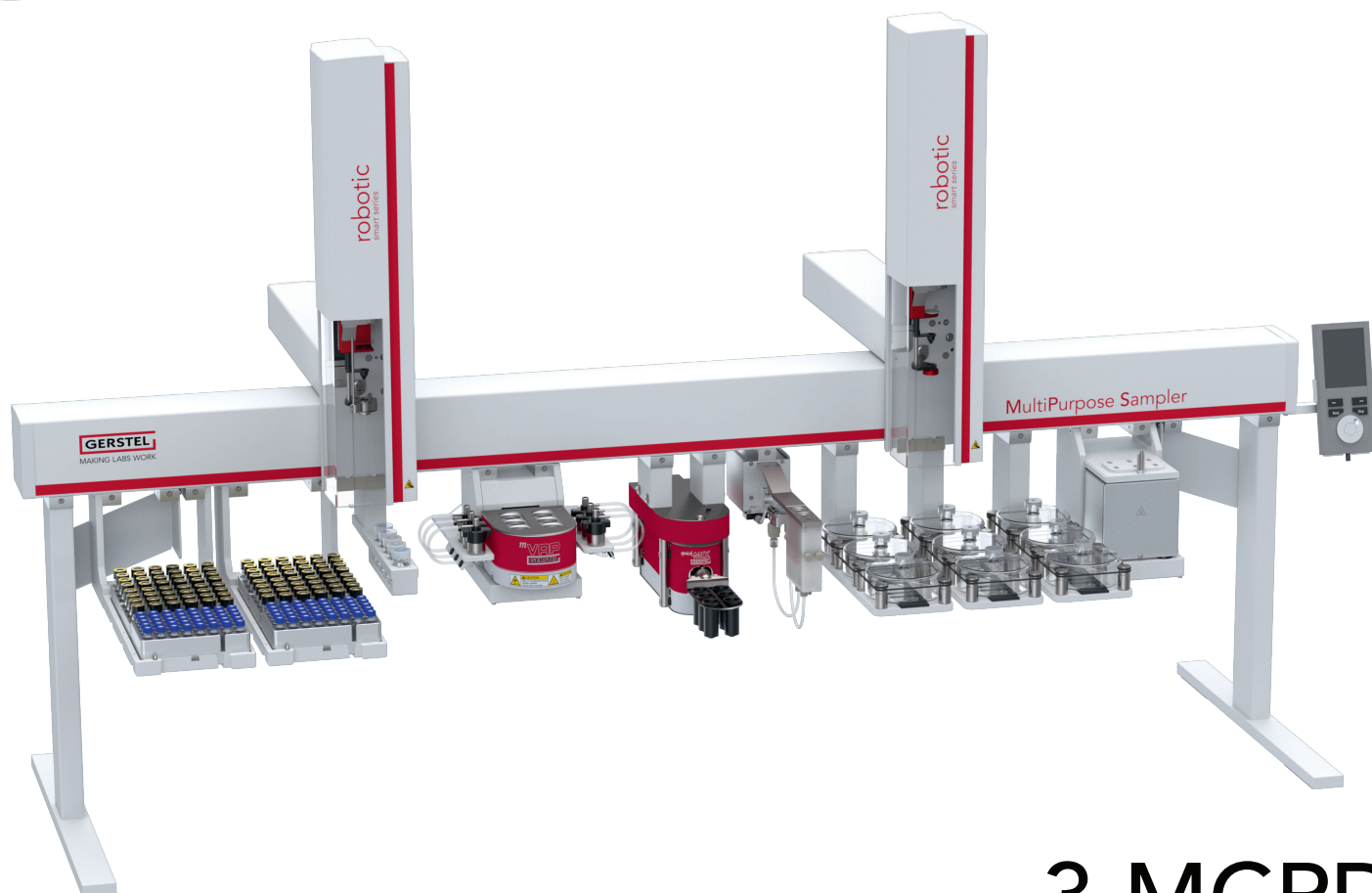
3-MCPD
Sample Prep Solution

Efficient Sample Preparation for the determination of 3-MCPD, 2-MCPD and Glycidol in edible oil and fat

Automated systems available for:

ISO 18363-1:2015 - AOCS Cd 29c-13
ISO 18363-4:2021 (Zwagerman - Overman)
ISO 18363-3:2017 - AOCS Cd 29a-13

Automated 3-MCPD Analysis

From Sample Prep to GC-MS analysis report

3-Monochloropropanediol (3-MCPD) esters and glycidyl esters (GE) are formed as by-products during refining of edible oils. In the human body, they react to form the known toxins 3-monochloropropanediol (3-MCPD) and glycidol. The European Union has imposed stringent regulations, specifying maximum allowable concentrations of these compounds in food products. In the oil matrix, the compounds are partially present in their ester form. The analysis requires transesterification and derivatization.

Automated analysis systems are available for the following methods:

- ISO 18363-1, AOCS Cd 29c-13, and DGF C-VI 18 (10), which are practically identical, based on indirect determination of glycidol. These are the most widely used methods world-wide. The analysis system is available as SamplePrep Solution with all method parameters (GERSTEL AppNote 191)

- ISO 18363-4:2021, known as the Zwagerman-Overman method, offers analysis in one GC/MS-run. The derivatized analytes are determined directly. There is no need for evaporation (GERSTEL AppNote 239)

- ISO 18363-3, AOCS Cd 29a-13, known as the "Unilever" method, requires a 16h reaction period at 40 °C (GERSTEL AppNote 217)

The GERSTEL MultiPurpose Sampler (MPS) is available as WorkStation independent of the analysis system or as integrated part of a complete automated analysis system that also performs introduction of the prepared extract followed by GC-MS(/MS) determination. Backflush options are used to shorten the analysis time for best possible throughput and to remove derivatization reagent for extended system stability.

To prepare edible oils for consumption, a refining process is frequently required. During this process, 3-MCPD-, 2-MCPD-, and glycidyl fatty acid esters can be formed. A reduction in their levels is typically achieved by optimizing the refining process conditions.

Degumming Dewaxing → Bleaching → Deodorization →

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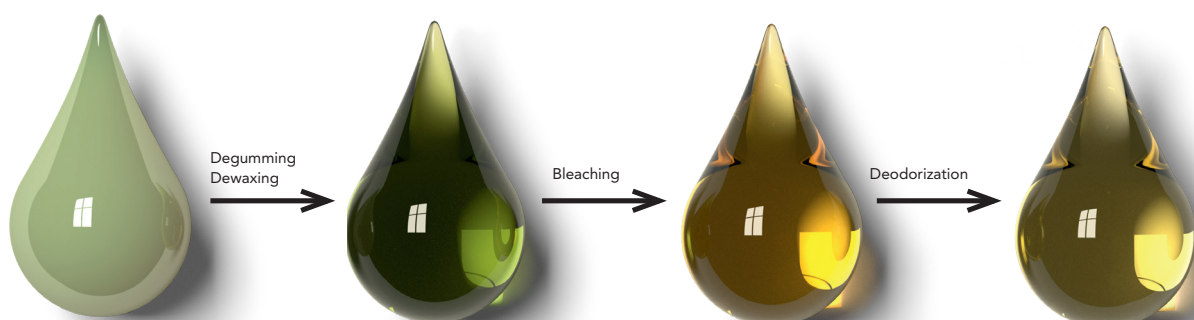
# 3-MCPD Sample Prep Solution for ISO 18363-1 / AOCS Cd 29c-13

Manual

- Weigh a 100 mg sample into a vial
- Fill a second vial with sodium sulfate drying agent (drying vial) - optional

Automated by the MPS:

- Add MTBE to the sample
- Add ISTD solution and mix, or melt and mix (solids)
- Add MeOH/NaOH mixture
- Agitate and incubate
- Add acidic NaCl solution (Assay A)
- Add acidic NaBr solution (Assay B)
- Add n-hexane for matrix extraction
- Agitate and incubate
- Discard hexane phase
- Repeat extraction with n-hexane twice
- Perform multiple analyte extractions using MTBE/Ethylacetate 3:2 (v/v), transfer the organic phases to the drying vial
- Add phenylboronic acid solution
- Evaporate to dryness and derivatize in the mVAP at 50 °C and subambient pressure
- Take up the derivatives in isooctane
- Introduction to GC/MS(/MS) if integrated with sampler.

One manual step is required followed by the long list of steps shown to the left prescribed in the unified DGF C-VI 18 (10) method. These are all performed automatically by the GERSTEL MultiPurpose Sampler (MPS). Depending on the instrument configuration, introduction of the prepared extract to the GC/MS system can be included.
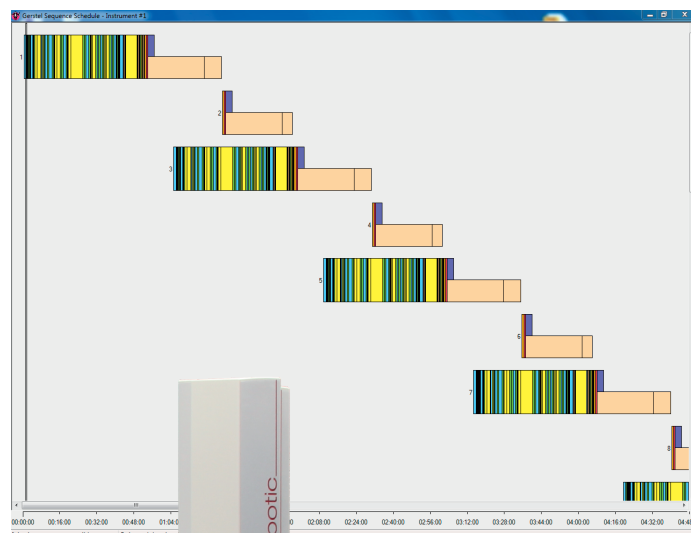

GERSTEL mVAP

The GERSTEL mVAP evaporates solvent and excess derivatization reagent from up to 6 samples in one batch under controlled vacuum, temperature, and agitation speed for lower limits of determination and improved system stability. Removing residual derivatization reagent by GC column backflush, further improves GC-MS(/MS) system stability and shortens analysis time.
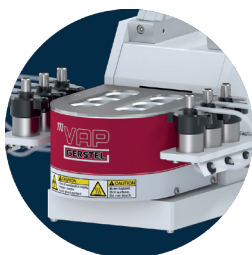
GERSTEL quickMIX

GERSTEL QuickMIX performs extremely efficient mixing for extraction of up to 6 samples in one batch with mixing power comparable to vortex mixers.
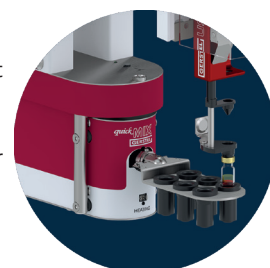
For **method ISO 18363-4:2021** (Zwagerman/Overman), GERSTEL AppNote 239 provides a detailed overview of the automated analysis system performance. Two internal standards compensate for even minor deviations in chemical reactions and in sample preparation, such as, for example, an overestimation of glycidol when large amounts of 3-MCPD are present. Some system details:

- A triple quadrupole GC-MS/MS is used, analytes are determined in MRM Mode
- The method including all sample preparation steps is performed by a GERSTEL MPS directly coupled to a GC-MS/MS system for a fully automated workflow
- A high-power quickMIX module performs efficient vortex-like mixing during extraction
- A cooled vial tray performs accurate temperature control during transesterification
- A fast wash station is integrated in the automated system, eliminating carry over
- A backflush system prevents high-boiling matrix residue and reagent from reaching the analytical column and the MS/MS system, ensuring long-term system stability
- The only manual step that remains is to weigh in the sample and transfer it to the MPS tray
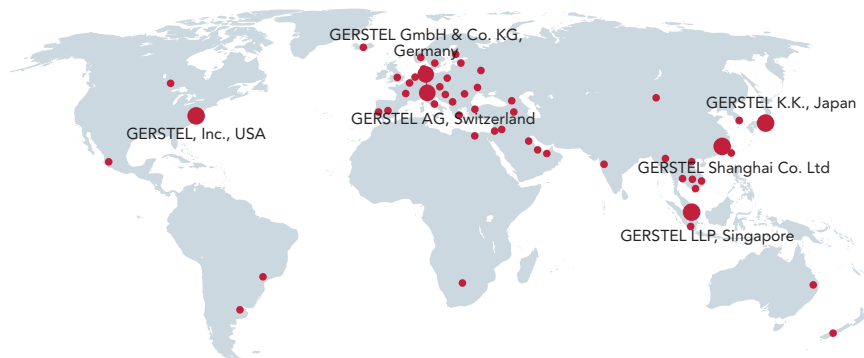
Successful analysis of round robin samples has demonstrated the high quality of the automated sample preparation process, the method, and the analysis system. RSDs were between 0.1 and 10 % for all analytes in different matrices, with only a few values above 5 %. The required Limit of Quantitation of 0.1 mg/kg was reached, lower LOQs are possible using an evaporation module (ᵐVAP). Chromatograms are easy to interpret, the GC-MS/MS largely eliminates nontarget signal. Automation enables 24/7 operation and priority samples are easily inserted into the running sequence.

For **method AOCS Cd 29a-13 / DIN EN ISO 18363-3:2017**, AppNote 217 provides a detailed overview. The main difference from the differential method is the 16 h sample preparation. Just one analysis run per sample is required, analytes are released in multiple steps and determined by GC-MS. The automated sample preparation greatly simplifies the analysis and excess derivatization reagent is removed for long-term GC-MS stability and accuracy. Depending on the type of oil, a concentration step enables even GC-MSD systems to reach the required limits of detection.

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