

# GC/MS Analysis under GMP — Highly Sensitive Analysis for Impurities in Nucleic Acids

In the quality test of nucleic acid medicines, plenty of samples can not be used for analysis, unlike the case of small molecule drugs. Especially in the analysis for impurities, highly sensitive analysis is needed to perform the test using limited amount of valuable samples. In this report, the residual solvents contained in a commercially available nucleic acid sample were measured by highly sensitive headspace-GC/MS.

## 1. Quality Test Items for Nucleic Acid Medicines

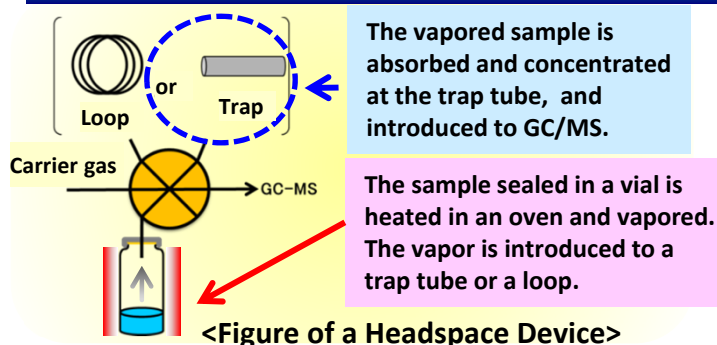
Requirement of guideline*	Available analysis at TRC
Specification: Test procedures	
(1) Appearance	Observation
(2) Identification	UHPLC, UV spectrophotometer
(3) Purity	
1. Oligonucleotide related substance	UHPLC, Capillary electrophoresis
2. Organic small molecular impurities	LC-MS/MS, GC-MS, MS, NMR, UHPLC
3. Residual solvents	GC-MS, GC
4. Inorganic impurities	ICP-OES, ICP-MS
(5) Assay	UHPLC, UV spectrophotometer
(6) Biological activity	Cell-based assay, ELISA

\*Quoted from "Matters to be considered for assurance and evaluation of the quality of nucleic acid medicines" issued by MHLW, Japan on September 27, 2018



SOP: Operation and maintenance of headspace-GC/MS (SOP No. NME18500)

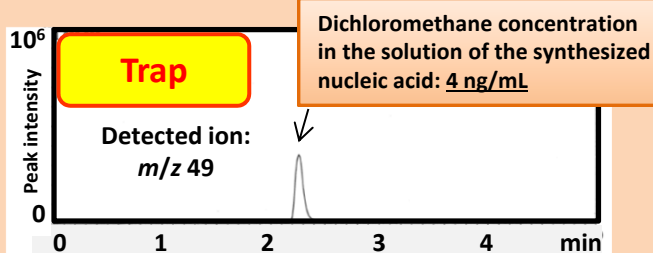
## 2. Measurement of Residual Solvents in the Nucleic Acid by Headspace-GC/MS



Improvement of detection capability for **dichloromethane** (a typical solvent used for nucleic acid synthesis)

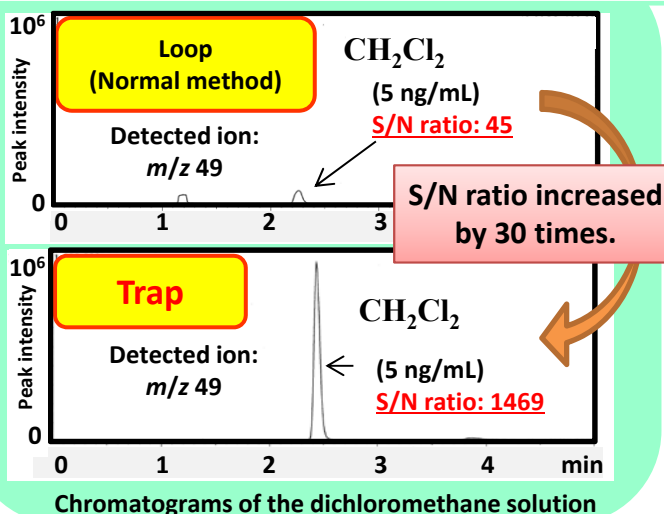
Residual **dichloromethane** in **artificially synthesized nucleic acid** was measured by the **trap** mode.

Sample: **Artificially synthesized nucleic acid**  
(C-Myc antisense DNA, sulfurized)  
Sequence: **CACGTTGAGGGGCAT**  
Sample concentration: **1 mg/mL**



Chromatogram of the artificially synthesized nucleic acid by the trap mode

A trace amount of dichloromethane (**4ppm** for the synthesized nucleic acid) was detected.



- S/N ratio of dichloromethane increased by 30 times in measurement by trap mode headspace-GC/MS.
- Quantitation of dichloromethane was performed using very small amount (1 mg) of the nucleic acid.