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Application News

Gas Chromatograph Mass Spectrometer GCMS-QP2020 NX

Screening of Ethylene Glycol and Diethylene Glycol in Medicinal Syrup by GCMS with FASST mode (Part 1 – as per Indonesian BPOM Method)

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User Benefits

- GCMS-QP2020 NX delivers high scan speed capabilities for screening analysis
- Fast Automated Scan/SIM Type (FASST) mode enables consecutive collection of scan and single ion monitoring (SIM) data
- Accurately identify and quantify both EG and DEG in medicinal syrup with a single injection
- Superior reliability and reproducibility of the results obtained

■ Introduction

Chemical contamination in pharmaceutical products can lead to fatal consequences. In Indonesia, for example, at least 195 deaths among children have been reported relating to ethylene glycol (EG) and diethylene glycol (DEG) contamination in medicinal syrup [1]. In West Africa, Gambia, 70 child deaths were also suspected to be caused by contaminated medicinal syrup [2]. This has caught the attention of the World Health Organization (WHO), hence leading to the issuance of a global alert on this issue.

Propylene glycol, glycerol, and sorbitol are commonly used in medicinal syrup as excipients. Their presence helps improve the solubility of the active ingredients during formulation. They also function as thickeners and sweeteners impacting the taste of the medication. These raw materials are easily contaminated with toxic ethylene glycol (EG) and diethylene glycol (DEG). Over the past few decades, numerous contamination incidents of medicinal syrup with EG and DEG have been reported [3]. Accidental ingestion of EG and DEG may result in abdominal pain, vomiting, diarrhea, inability to pass urine, headache, altered mental state, and kidney injury which leads to death.

In this article, we will examine the usage of the Shimadzu GCMS-QP2020 NX to identify and quantify EG and DEG in medicinal syrup. The analysis will be demonstrated using the Fast Automated Scan/SIM Type (FASST) mode, which enables consecutive operation of scan mode and selected ion monitoring (SIM) mode for accurate qualification and quantitation within a single injection.

Figure 1. Shimadzu GCMS-QP2020 NX with AOC-20i+s Plus

■ Measurement Conditions

The analysis was performed using Shimadzu GCMS-QP2020 NX and AOC-20i+s Plus autosampler (**Figure 1**). Details of the analytical conditions were depicted in **Table 1**, in accordance with the method from the Indonesian national food and drug agency (BPOM) with slight modifications [4]. As the recommended method has minimum sample preparation, modifications were made to enhance the performance and robustness of the system. A capillary column with a 5 m integrated guard column was used to enhance the setup capability to handle a wide range of complex matrices without the risk of losing its performance. Due to the superior sensitivity of GCMS-QP2020 NX, a higher split ratio of 20:1 was used to minimize system contamination from the sample matrix. Event times for Scan and SIM were reduced to 0.2 and 0.1 sec, respectively, to increase the number of data points for better peak shape and integration.

Table 1. GCMS Parameters

Flow Control Mode	Constant Flow	
Column Flow Rate	0.65 mL/min	
Injection Mode	Split (Split Ratio = 20)	
Injection Port Temp.	250 °C	
Injection Volume	1 µL	
Carrier Gas	Helium	
Column	SH-PolarWax column with 5 m integrated guard column (30 m long, 0.25 mm I.D., 0.25 µm film thickness) [P/N: 227-36360-01]	
Column Oven Temp. Program	Initial Temp 100 °C (hold for 1 min) - Increase to 130 °C with a rate of 10 °C/min (hold 7 min) - Increase to 240 °C with a rate of 20 °C/min (hold 3 min) - Increase to 250 °C with a rate of 20 °C/min (hold 3 min)	
Ion Source Temp.	230 °C	
Interface Temp.	240 °C	
Acquisition Mode	FASST (Scan/SIM)	
Event Time (sec)	Q3 Scan: 0.2 Q3 SIM: 0.1	
Scan *m/z* Range	29 to 400 amu	
SIM Ions	EG:	31 (target ion) 33 and 62 (reference ions)
	DEG:	45 (target ion) 75 and 31 (reference ions)

■ Sample Preparation

1000 ppm calibration stock solution preparation

EG and DEG were purchased from TCI, Japan. Standard solutions of EG and DEG in methanol were prepared by dissolving 100 mg of each in separate 100 mL volumetric flasks. To improve dissolution, sonicate EG and DEG with 50 mL methanol (MeOH) before topping up to the 100 mL mark. The standard solutions were subsequently used for the preparation of a series of various concentrations of calibration standard solutions in 5 mL volumetric flasks in accordance with **Table 2**.

Table 2. Preparation of EG and DEG calibration plots in 5 mL volumetric flasks

Level	Ethylene Glycol		Diethylene Glycol	
	Conc /ppm	Amount from 1000 ppm stock/ µL	Conc /ppm	Amount from 1000 ppm stock/ µL
1	6	30	12	60
2	8	40	16	80
3	10	50	20	100
4	12	60	24	120
5	14	70	28	140

Medicinal syrup sample preparation

A blank medicinal sample solution was prepared by transferring 10 mL of the medicinal syrup sample into a 100 mL volumetric flask. To improve dissolution, the medicinal syrup in 50 mL of methanol was sonicated for 5 minutes before topping it up to the mark. The diluted mixture was then filtered with a 0.45 µm PTFE membrane filter. A portion of the blank sample solution was subsequently spiked with 6 ppm of EG and 12 ppm of DEG for repeatability study.

Medicinal syrup samples (Sample A and Sample B) were purchased commercially and were prepared in a similar fashion. However, they were scaled down proportionally using 10 mL volumetric flasks instead. A portion of Sample A was spiked with both 4 ppm of EG and 20 ppm of DEG and similarly, a portion of Sample B was spiked with 20 ppm of DEG for recovery studies.

■ Results and Discussion

Setting up the FASST screening method

Calibration plots were obtained by spiking various concentrations of EG (6 to 14 ppm) and DEG (12 to 28 ppm) in methanol and analyzing them using FASST mode. **Figures 2a-d** demonstrated that the linearity plots obtained from two separate preparations have linear fits with R^2 of at least 0.9985 for both EG and DEG. The blank medicinal syrup and its spiked solutions (6 ppm EG and 12 ppm DEG) were analyzed subsequently. For the blank control medicinal syrup, DEG was not detected (**Figure 3b**), and only a negligible amount of EG (not quantifiable) was present (**Figure 3a**), thus the endogenous amount of EG was assumed to have an insignificant contribution to the results.

The results of the repeated injections (n=10) of the spiked blank medicinal syrup are summarized in **Table 3**. Concentration repeatability (%RSD) of 1% and 2% were obtained for 6 ppm EG and 12 ppm DEG, respectively, demonstrating the high degree of precision of the entire setup. Decent %Recovery values were also obtained for both EG (118% to 123%) and DEG (103% to 109%). The quantifications of 6 ppm EG (**Figure 3c**) and 12 ppm DEG (**Figure 3d**) were successfully demonstrated and established as the limits of quantification (LOQ), with a signal-to-noise (S/N) ratio above 200.

Figure 4a-b show the total ion chromatogram (TIC) scan profile and the SIM mass chromatogram (MC) profile for the target ions of EG (m/z 31) and DEG (m/z 45) of the level 5 calibration standard (**Table 2**). Based on the TIC profiles, two prominent peaks at the retention time of 8.817 mins (**Figure 4a**) and 15.374 mins (**Figure 4b**) were observed and that corresponded to EG and DEG, respectively.

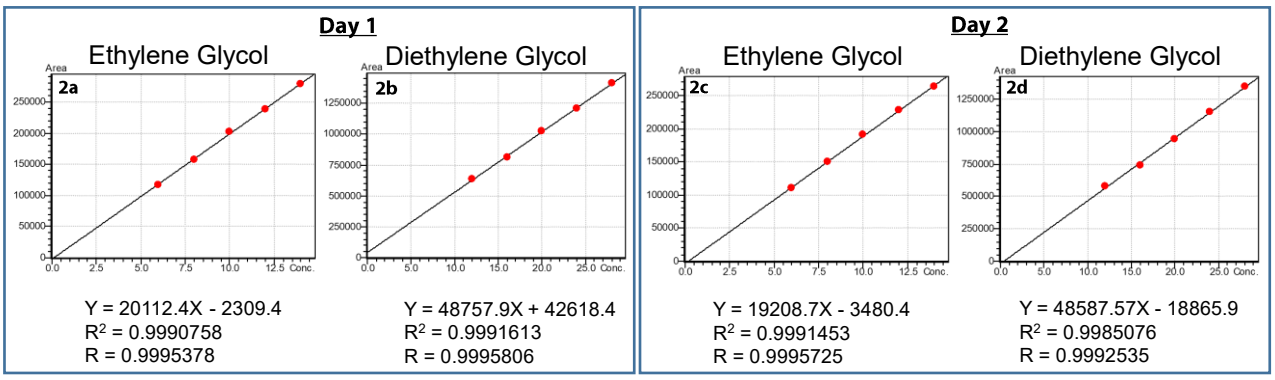

Figure 2a-d. Calibration plots of EG and DEG obtained on separate preparations

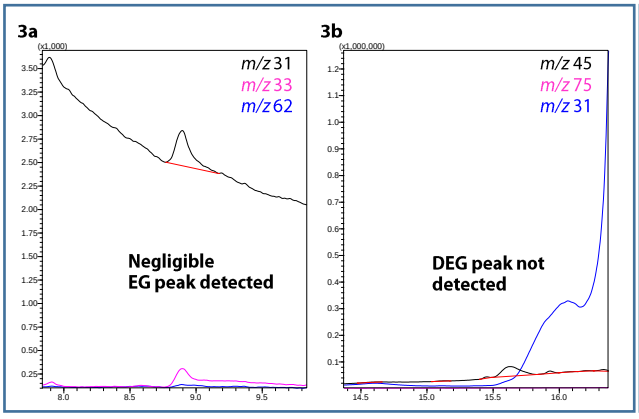

Figure 3a-b. SIM Mass Chromatogram (MC) of blank medicinal syrup

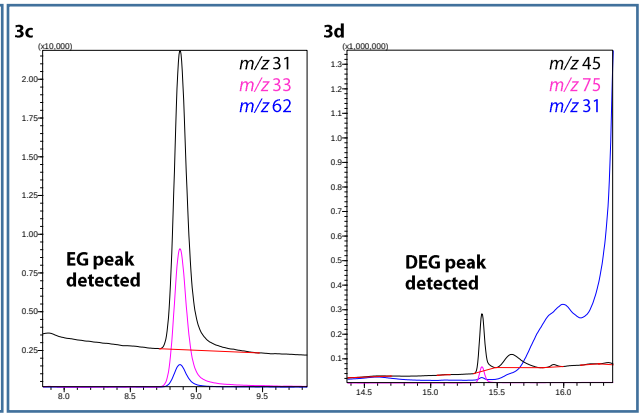

Figure 3c-d. SIM Mass Chromatogram (MC) of spiked (6 ppm EG and 12 ppm DEG) blank medicinal syrup

Table 3. Repeatability (*n*=10) and recovery of the spiked blank medicinal syrup with 6 ppm EG and 12 ppm DEG

Injection	EG detected /ppm	DEG detected /ppm	%Recovery EG	%Recovery DEG
1	7.08709	13.07627	118%	109%
2	7.24933	12.75458	121%	106%
3	7.40988	12.76441	123%	106%
4	7.30253	12.45931	122%	104%
5	7.32590	12.47512	122%	104%
6	7.32913	12.57976	122%	105%
7	7.28901	12.60053	121%	105%
8	7.30924	12.54618	122%	105%
9	7.18782	12.31182	120%	103%
10	7.21388	12.38494	120%	103%
Std. Dev.	0.09001963	0.22249365		
Average	7.270381	12.595292		
%RSD	1.24%	1.77%		

As indicated by the BPOM method, the SIM profiles were used for quantitation. EG was quantitated with the target ion of *m/z* 31 and qualified with the reference ions of *m/z* 33 and 62. On the other hand, DEG was quantitated with the target ion of *m/z* 45 and qualified with reference ions of *m/z* 75 and 31. **Figures 5a-b** depict the mass spectrum profiles of identified EG and DEG, respectively, in spiked medicinal syrup, with high similarity indices of 90 and 93 when matched against the NIST 2020 mass spectral library.

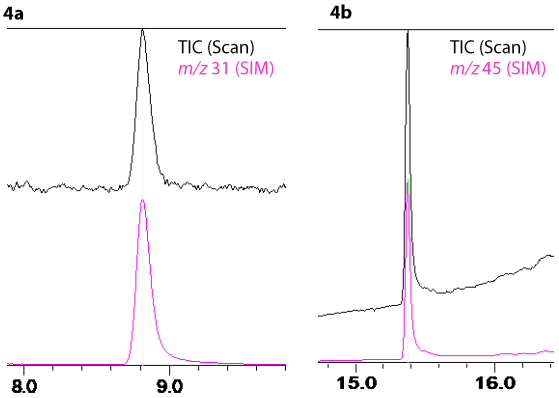

Figure 4a-b. Calibration standard level 5's Total Ion Chromatogram (TIC) scan profile and SIM target ion profiles of EG and DEG

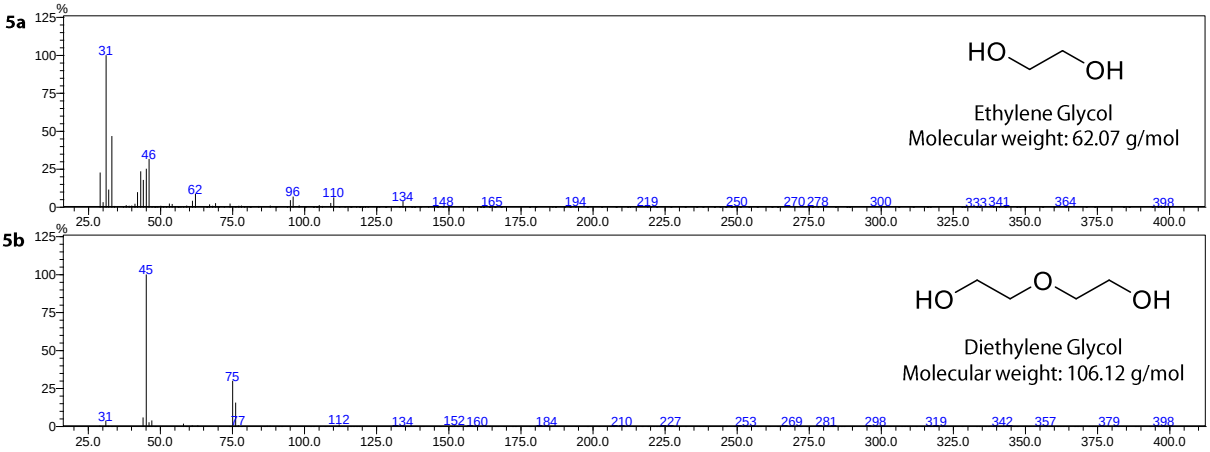

Figure 5a-b. Mass spectrum profiles of 6 ppm EG and 12 ppm DEG detected in spiked medicinal syrup

Screening of commercially purchased medicinal syrup

Commercially purchased Sample A and Sample B medicinal syrup underwent the same sample treatment as the blank medicinal syrup control. Briefly, 1 mL of the neat sample was transferred to a 10 mL volumetric flask containing 5 mL of MeOH. The diluted mixture was then sonicated for 5 minutes and topped up to the mark. The diluted mixture was then filtered through a 0.45 µm PTFE membrane filter. The filtered sample was then used for GCMS analysis directly.

Amount of EG and DEG detected in Sample A and Sample B

Table 4 and **Table 5** summarize the concentration results obtained. For Sample A, 7.18 ppm of EG (avg *n*=4) was detected. Thus, the sum of EG and DEG present in Sample A was equivalent to 0.07 mg/mL after considering the dilution factor. For Sample B, 10.20 ppm of EG (avg *n*=4) was detected. Meanwhile, a trace amount of DEG (below LOQ) was detected, which explained the poor inter-day concentration reproducibility for DEG in this sample. Thus, the sum of the concentration of EG and DEG present in Sample B after taking into consideration of the dilution factor was ~0.11 mg/mL.

This experiment demonstrated Shimadzu GCMS-QP2020 NX superior performance in terms of repeatability and sensitivity for detecting EG and DEG in contaminated medicinal syrup.

High repeatability observed

A high degree of reproducibility is observed when the concentration of the analyte of interest falls within the calibration plot range. Within the day, repeatability %RSD between 0.01% to 1.95% was observed. The high degree of precision in the results observed in **Table 4** and **Table 5** were in strong agreement with the data in **Table 3**. Good intermediate precision across different days was observed as well, ranging between 2.22% to 3.92%.

%Recovery of the experiment

Due to the lack of a blank control matrix for Sample A and Sample B, the %Recovery was determined by spiking into the Sample A and Sample B syrup directly. Sample A syrup was spiked with 4 ppm of EG and 20 ppm of DEG, while 20 ppm of DEG was spiked into Sample B syrup. To avoid exceeding the level in the calibration plot, Sample B was not spiked with EG.

The additional amount of spiked analyte detected in the individual sample was calculated by taking the difference between the result obtained for the spiked sample with respect to the average reading of the unspiked sample that day. As the level of endogenous DEG in Sample B was below LOQ, the basal level of DEG contribution will be assumed to be negligible. **Table 6** summarizes the result of the spiking experiment and its %Recovery.

Table 4. Summarized result of EG and DEG detected in Sample A syrup

Day	EG detected /ppm	Avg EG detected /ppm (n=2)	%RSD	Day	DEG detected /ppm	Avg DEG detected /ppm (n=2)	%RSD	(EG+DEG)/ (mg/mL)
1	7.17694	7.277225	1.95%	1	ND	ND	ND	0.07
1	7.37751			1	ND			0.07
2	6.98670	7.084415	1.95%	2	ND	ND	ND	0.07
2	7.18213			2	ND			0.07
Avg EG *n*=4	7.18082			**Avg DEG *n*=4**	ND		**Avg Sum *n*=4**	0.07
Intermediate Precision	2.22%			**Intermediate Precision**	ND			

EG detected
DEG not detected in Sample A

Abbreviation used: avg = average, ND = Not Detected

Table 5. Summarized result of EG and DEG detected in Sample B syrup

Day	EG detected /ppm	Avg EG detected /ppm (n=2)	%RSD	Day	DEG detected /ppm	Avg DEG detected /ppm (n=2)	%RSD	(EG+DEG)/ (mg/mL)
1	10.54466	10.54558	0.01%	1	0.51743	0.53152	3.75%	0.11
1	10.54650			1	0.54561			0.11
2	9.79652	9.857925	0.88%	2	1.48060	1.49213	1.09%	0.11
2	9.91933			2	1.50365			0.11
Avg EG *n*=4	10.20175			**Avg DEG *n*=4**	1.0118225		**Avg Sum *n*=4**	0.11
Intermediate Precision	3.92%			**Intermediate Precision**	54.83%			

EG detected
DEG detected, but below LOQ in Sample B

Abbreviations used: avg = average

Table 6. Summarized result of the spiked EG and DEG detected in Sample A and spiked DEG detected in Sample B

Sample A				Sample B	
Amount of spiked EG detected/ppm	**%Recovery**	**Amount of spiked DEG detected/ppm**	**%Recovery**	**Amount of spiked DEG detected/ppm**	**%Recovery**
4.405055	110%	23.93676	120%	23.24588	116%
4.705645	118%	24.05775	120%	23.23355	116%
3.144905	79%	24.74546	124%	24.36432	122%
3.225340	81%	25.07093	125%	22.95413	115%

In Sample A syrup, the %Recovery for EG was observed to be between 79% to 118% and 120% to 125% for DEG. For Sample B syrup, the %Recovery for DEG was observed to be between 115% to 122%. The slight deviation from the ideal accuracy was attributed to the observable matrix effect. This is within expectation, as with this approach, the sample undergoes minimum sample preparation before the injection into the GCMS system. A matrix-matched calibration curve is expected to improve the performance, and this will be explored in part 2 of this application news.

FASST advantage in complex matrices

Figure 6a-b and **Figure 7a-c** depict the TIC and MC of Sample A and Sample B samples, respectively, using FASST mode. As shown, EG was detected in Sample A, and both EG and DEG were detected in Sample B. In Sample A, it was observed that the retention time of the EG peak shifted from 8.760 min to 9.025 min (**Figure 6a-b**). This was attributed to the huge earlier eluting peak (**Figure 6a**) in the Sample A matrix, which overloaded the column. As the FASST analysis mode was used, EG was still successfully identified with high confidence even though its expected retention time has shifted. Using the scan mode data from FASST analysis mode, the huge interfering peak that affected the elution of EG was identified as propylene glycol, with a high similarity index of 97 when matched against the NIST 2020 library. The advantage of FASST mode analysis in identifying unknown interfering peaks while maintaining the sensitivity of target compounds was well-demonstrated in this experiment.

Calculation and tips

The concentration of the sample is inferred from the linear calibration plot equation obtained in the form of:

$$y = ax + c$$

where x = amount of EG or DEG in ppm
y = area obtained from GCMS
a = slope of the calibration plot
c = intercept

The sum amount of EG+DEG (mg/mL) for medicinal syrup is calculated using the following formula:

$$(EG+DEG) = x \cdot F/1000$$

where x = sum of EG and DEG in ppm
F = dilution factor

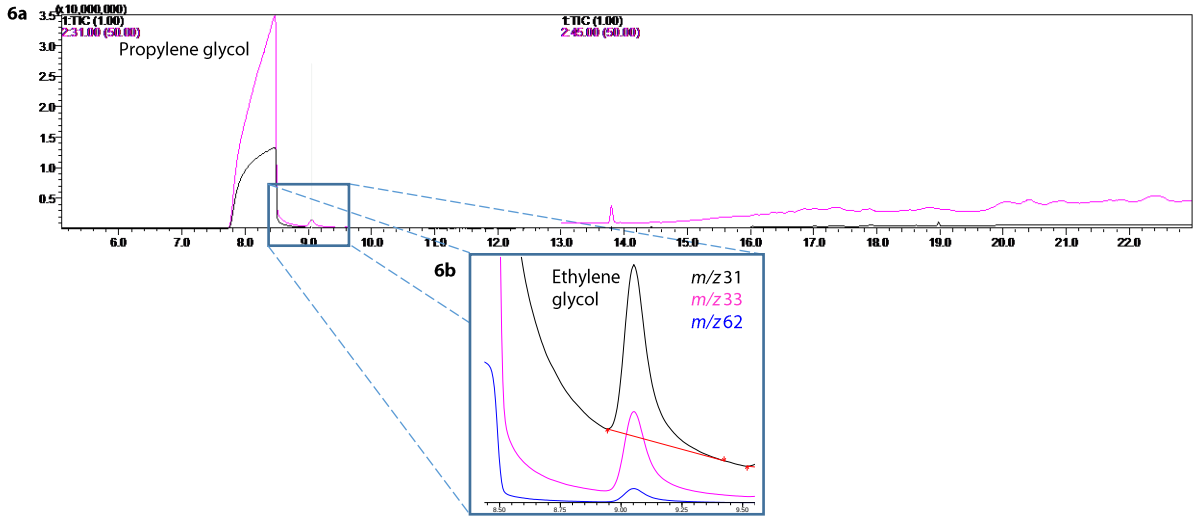

Figure 6a-b. TIC and MC (EG and DEG quantifier ion) profile of Sample A, and its zoom section (SIM profile) where EG was detected

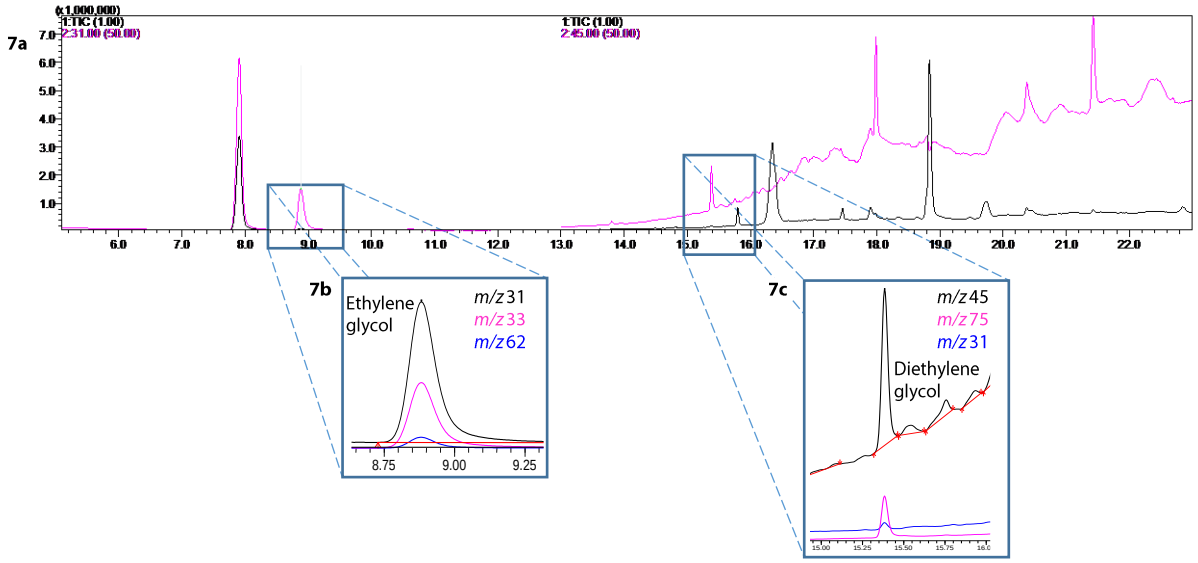

Figure 7a-c. TIC and MC (EG and DEG quantifier ion) profile of Sample B, and its zoom section (SIM profile) where EG and DEG were detected

For very viscous samples, it might not be practical to transfer the desired volume accurately. In such instance, the density of the syrup sample should be determined first, then the amount of sample transferred should be weighed precisely. The dilution factor of should be calculated as followed:

$$F = \rho \cdot Vt/Wt$$

where F = dilution factor
ρ = density of medicinal syrup (g/mL)
Vt = total volume during dilution (mL)
Wt = Weight of medicinal syrup (g)

If the sample viscosity did not pose difficulty in drawing the exact desired volume, the dilution factor should be calculated as below:

$$F = Vt/V1$$

where F = dilution factor
Vt = total volume during dilution (mL)
V1 = Volume of medicinal syrup (mL)

■ Conclusion

To prevent any future risk of mass poisoning tragedies due to the ingestion of contaminated medicinal syrup, stringent quality control should be advocated. In this application news, Shimadzu GCMS-QP2020 NX was demonstrated to provide sensitive, precise, and robust detection of EG and DEG in medicinal syrup in two different sample matrices with minimum sample preparation, as per the Indonesian BPOM method.

The limitation of the approach is that as minimal sample preparation is used, and due to matrix effect only decent %Recovery is obtained. While this method is appropriate for quick testing in labs for large variety of samples with different sample matrices, it might not meet the stricter accuracy requirement for quality control labs in pharmaceutical industry. A separate improvised method will be developed to address the said problem using matrix-matched calibration curve in Part 2 of this application news.

■ References

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3. Barr, D.B., et al, Identification and Quantification of Diethylene Glycol in pharmaceuticals implicated in poisoning epidemics: An historical laboratory perspective. 2007. *J. Anal. Toxicol.*, **31**, pp 209-303.

4. Metode Analysis Uji Cemaran Etilen Glikol (EG) dan Dietilen Glikol, Badan Pengawas Obat dan Makanan, 24 Oct 2022.

04-AD-0283-EN First Edition: Dec. 2022

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Application News

Gas Chromatograph Mass Spectrometer GCMS-QP2020 NX

Screening of Ethylene Glycol and Diethylene Glycol in Medicinal Syrup by GCMS with FASST mode (Part 2 – Improved Method for QC Testing)

Chia Chee Geng, Cynthia Melanie Lahey, Chua Chun Kiang, and Jackie Jackie
Shimadzu (Asia Pacific) Pte Ltd

User Benefits

- GCMS-QP2020 NX delivers high scan speed capabilities for screening analysis
- Fast Automated Scan/SIM Type (FASST) mode enables consecutive collection of scan and single ion monitoring (SIM) data
- Accurately identify and quantify both EG and DEG in medicinal syrup with a single injection
- Superior reliability, reproducibility and selectivity of the results obtained
- Matrix-matched calibration greatly improves %Recovery

■ Introduction

In Part 1 of the application news, we have successfully demonstrated GCMS-QP2020 NX superior capability in screening of ethylene glycol (EG) and diethylene glycol (DEG) in medicinal syrup, using the recommended method by Indonesian BPOM [1]. The method requires minimum sample preparation, making it suitable for laboratories in relevant authorities to screen medicinal syrup from different brands easily, thus making it possible to make rapid assessment on the safety of the medicinal syrup on the market. However, notable matrix effect has been observed, thus affecting the %Recovery of the method. As a result, this approach might not be capable of meeting the stringent QC testing requirements in pharmaceutical industry. Part 2 of the application news intends to address this gap.

To improve the accuracy of the method, our team decided to modify the BPOM method slightly using matrix-matched calibration plot. This slight modification greatly enhances the method selectivity, enabling accurate quantitation of EG and DEG in the presence of complex sample matrices.

In this article, we will be using the exact setup as Part 1. We will examine the usage of the Shimadzu GCMS-QP2020 NX to identify and quantify EG and DEG in medicinal syrup. The analysis will be demonstrated using the Fast Automated Scan/SIM Type (FASST) mode, which enables consecutive operation of scan mode and selected ion monitoring (SIM) mode for accurate qualification and quantitation within a single injection.

Figure 1. Shimadzu GCMS-QP2020 NX with AOC-20i+s Plus

■ Measurement Conditions

The analysis was performed using Shimadzu GCMS-QP2020 NX and AOC-20i+s Plus autosampler (**Figure 1**). Details of the analytical conditions were depicted in **Table 1**, in accordance with the method from the Indonesian national food and drug agency (BPOM) with slight modifications [2]. As the recommended method has minimum sample preparation, modifications were made to enhance the performance and robustness of the system. A capillary column with a 5 m integrated guard column was used to enhance the setup capability to handle a wide range of complex matrices without the risk of losing its performance. Due to the superior sensitivity of GCMS-QP2020 NX, a higher split ratio of 20:1 was used to minimize system contamination from the sample matrix. Event times for Scan and SIM were reduced to 0.2 and 0.1 sec, respectively, to increase the number of data points for better peak shape and integration.

Table 1. GCMS Parameters

Flow Control Mode	Constant Flow	
Flow Rate	0.65 mL/min	
Injection Mode	Split (Split ratio = 20)	
Injection Port Temp.	250 °C	
Injection Volume	1 µL	
Carrier Gas	Helium	
Column	SH-PolarWax column with 5 m integrated guard column (30 m long, 0.25 mm I.D., 0.25 µm film thickness) [P/N: 227-36360-01]	
Column Oven Temp. Program	Initial Temp 100 °C (hold for 1 min) - Increase to 130 °C with a rate of 10 °C/min (hold 7 min) - Increase to 240 °C with a rate of 20 °C/min (hold 3 min) - Increase to 250 °C with a rate of 20 °C/min (hold 3 min)	
Ion Source Temp.	230 °C	
Interface Temp.	240 °C	
Acquisition Mode	FASST (Scan/SIM)	
Event Time (sec)	Q3 Scan: 0.2 Q3 SIM: 0.1	
Scan *m/z* Range	29 to 400 amu	
SIM Ions	EG:	31 (target ion) 33 and 62 (reference ions)
	DEG:	45 (target ion) 75 and 31 (reference ions)

■ Sample Preparation

Medicinal syrup sample preparation

A medicinal sample solution was prepared by transferring 10 mL of the medicinal syrup sample into a 100 mL volumetric flask. To improve dissolution, the medicinal syrup in 50 mL of methanol was sonicated for 5 minutes before topping it up to the mark. The diluted mixture was then filtered with a 0.45 µm PTFE membrane filter. 1 µL filtered sample was then analyzed using GCMS, and only negligible amount of EG was detected, and DEG was not being detected [1]. Thus, this medicinal sample solution was used as a blank sample because endogenous level of EG will be assumed to have negligible contribution to the experimental results. Subsequently, this filtered blank medicinal syrup sample was used for the preparation of the matrix-matched calibration plot (**Table 2**), and in parallel, a separate preparation of spiked samples at corresponding to Level 1 (LOQ) and level 3 of the calibration plot was prepared.

Matrix-matched calibration plot preparation

EG and DEG were purchased from TCI, Japan. Standard solutions of EG and DEG in methanol were prepared by dissolving 100 mg of each in separate 100 mL volumetric flasks. To improve dissolution, sonicate EG and DEG with 50 mL methanol (MeOH) before topping up to the 100 mL mark (1000 ppm standard solution). The 1000 ppm standard solutions were subsequently used for the preparation of a series of various concentrations of calibration standard solutions in 5 mL volumetric flasks in accordance with **Table 2**, topped to the mark with the filtered blank medicinal syrup sample.

Table 2. Preparation of EG and DEG calibration plots in 5 mL volumetric flasks

Level	Ethylene Glycol		Diethylene Glycol	
	Conc /ppm	Amount from 1000 ppm stock/ µL	Conc /ppm	Amount from 1000 ppm stock/ µL
1	6	30	12	60
2	8	40	16	80
3	10	50	20	100
4	12	60	24	120
5	14	70	28	140

■ Results and Discussion

Matrix-matched calibration plot

Matrix-matched calibration plots were obtained by spiking various concentrations of EG (6 to 14 ppm) and DEG (12 to 28 ppm) in the blank medicinal syrup and analyzed them using FASST mode. **Figures 2a** to **2d** demonstrate the linearity of the calibration plot from two separate preparations, having linear fits with R^2 of at least 0.999, for both EG and DEG. The high degree of similarity between the equations of the linearity plot from the two different preparations reflects the robustness and reliability of the method used.

Like Part 1 of the application news, in accordance with the BPOM method, the SIM profiles were used for quantitation. EG was quantitated with the target ion of *m/z* 31 and qualified with the reference ions of *m/z* 33 and 62. On the other hand, DEG was quantitated with the target ion of *m/z* 45 and qualified with reference ions of *m/z* 75 and 31.

%Recovery at levels 1 and 3

Table 3 summarized the result for the %Recovery of the spiked samples using the conventional non-matrix-matched standards

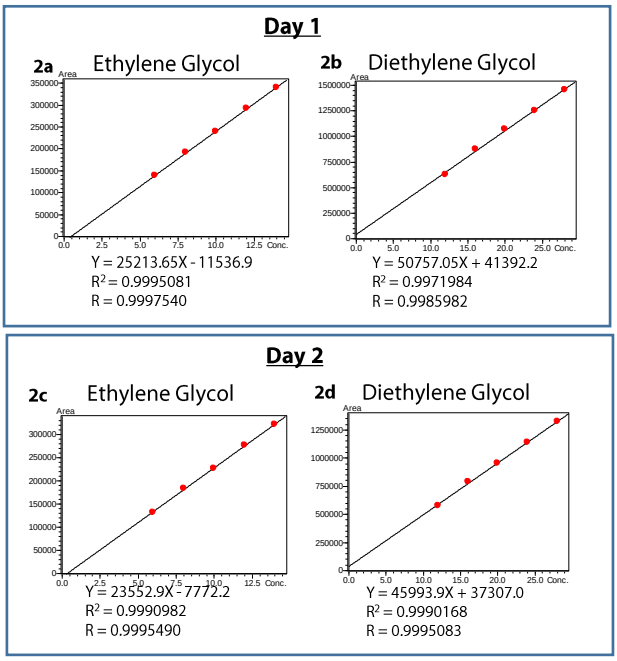

Figure 2a-d. Matrix-matched calibration plots of EG and DEG obtained on separate preparations

calibration plot approach vs matrix-matched calibration plot approach, at level 1 (LOQ) and level 3 concentration levels of EG (6 and 10ppm) and DEG (12 and 20 ppm). For the conventional non-matrix-matched calibration plot approach, %Recovery for EG ranges from 118% to 120%, and 102% to 110% for DEG. Significant positive bias is observed upon attempting to quantify EG and DEG in the spiked sample. This biasness was corrected when using the matrix-matched calibration approach. For the same set of data, using the matrix-matched calibration approach, the %Recovery for EG obtained ranges from 99% to 102%, and 98% to 105% for DEG. The results obtained were also in strong agreement with **Figure 2a** to **2d**, whereby the equations of the separate preparation of the matrix-matched calibration plots on different days shares high degree of similarity.

High degree of precision observed

The high degree of precision observed in **Table 3** is in strong agreement with the observation in Part 1 of the application news. The concentration %RSD (**Table 3**) of the results matrix-match calibration plots obtained were very similar to those of non-matrix-matched. Using the matrix-matched calibration plots, the concentration %RSD obtained ranged from 0.38% to 2.90%. This demonstrates that modifying the method has negligible effect on the precision of the data.

Improving selectivity and accuracy of measurement

As mentioned above, after the modification using the matrix-matched calibration plot to process the data, the %Recovery of the results approaches the ideal value of 100% (**Table 3**). Our team has thus successfully improved the selectivity of the setup that enables us to quantify EG and DEG with a better accuracy. This approach is suitable for QC lab in pharmaceutical industry with previously released batches of uncontaminated finished product that has negligible amount of EG and DEG presence (suitable for use as blank).

The limitation of this approach is the requirement of a corresponding blank matrix (with negligible amount of EG and DEG). In cases where a lab needs to test a diverse range of samples made up of various matrices, such as regulatory agencies or testing laboratories, the approach presented in Part 1 of the application note is more practical [1].

Table 3. Comparison of results using conventional non-matrix-matched calibration plot (spiked in MeOH) with matrix-matched calibration plot approach.

		Non-Matrix-Matched Calibration plot				Matrix Matched Calibration plot			
EG spiked /ppm	DEG spiked /ppm	EG detected /ppm	DEG detected /ppm	%Recovery EG	%Recovery DEG	EG detected /ppm	DEG detected /ppm	%Recovery EG	%Recovery DEG
6	12	7.20433	12.9938	120%	108%	6.11272	12.50618	102%	104%
6	12	7.20901	13.15353	120%	110%	6.11644	12.65962	102%	105%
6	12	7.08610	12.80426	118%	107%	6.01840	12.32410	100%	103%
6	12	7.06675	12.29578	118%	102%	6.00297	11.83565	100%	99%
	%RSD	1.06%	2.91%		%RSD	1.00%	2.90%		
10	20	11.94484	20.35343	119%	102%	9.89412	19.57594	99%	98%
10	20	11.92798	20.42981	119%	102%	9.88068	19.64931	99%	98%
10	20	12.01733	21.17994	120%	106%	9.95195	20.36990	100%	102%
10	20	12.01584	21.47227	120%	107%	9.95076	20.65070	100%	103%
	%RSD	0.39%	2.65%		%RSD	0.38%	2.65%		

Calculation

In this application news, the %Recovery and precision calculations were based on the concentrations of the sample inferred from the linear calibration plot equation, and therefore the final concentrations of EG and DEG in the medicinal syrup were not shown.

Refer to Part 1 of the application news [1] for tips for performing concentration calculations.

■ Conclusion

As a follow-up to Part 1 of the application news, our team showed how we were able to improve the method's selectivity using the matrix-matched calibration plot approach. As a result, we were able to satisfy the demanding QC standards of the pharmaceutical industry for the release testing of finished goods. The highly precise results obtained in this Part 2 is in strong agreement with the observation made in Part 1 of this application news.

Both Parts 1 and 2 of the application news demonstrated that Shimadzu GCMS-QP2020 NX can provide accurate, sensitive, precise, and robust detection of EG and DEG in medicinal syrup with minimum sample preparation. The developed methods are suitable for meeting the stringent requirements of relevant authorities, testing labs, or pharmaceutical quality control.

■ References

1. Chia, C. G. et al, Screening of Ethylene Glycol and Diethylene Glycol in Medicinal Syrup by GCMS with FASST mode (Part 1 – as per Indonesia BPOM Method), Shimadzu Application News No. 04-AD-0283-EN.
2. Metode Analysis Uji Cemaran Etilen Glikol (EG) dan Dietilen Glikol, Badan Pengawas Obat dan Makanan, 24 Oct 2022.

04-AD-0288-EN First Edition: Jan. 2023

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