Evaluating the Creaming of an Emulsion via Mass Spectrometry and UV–Vis Spectrophotometry

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Supporting Information

ABSTRACT: The creaming behavior of a turbid oil-in-water emulsion was observed via the processes of multiphoton ionization time-of-flight mass spectrometry (MPI-TOFMS) and ultraviolet–visible spectrophotometry (UV–vis), and the results were compared. The transmittance measurement by UV–vis showed that the turbidity of the toluene emulsion was decreased with time. However, non-negligible errors are common in the measurement of a sample with high turbidity. The online measurement by MPI-TOFMS detected many spikes in the time profile, which revealed the existence of toluene droplets in the emulsion. A smooth time profile suggested that the signal intensity had initially increased, and then decreased with time; the initial concentration of toluene was 3 g/L, which had decreased by half after 60 min. The signal behavior obtained using MPI-TOFMS differed only slightly from that obtained using UV–vis. Since a change in turbidity is not the same as a change in the local concentration of an oil component, MPI-TOFMS is useful for the analysis of a turbid emulsion and offers additional information concerning the creaming phenomenon of an emulsion.

INTRODUCTION

An emulsion is a system in which one liquid is dispersed with another and both are immiscible. Emulsions are applied in a wide variety of fields; many types of emulsion products are commercialized such as inks, paints, foods, cosmetics, and pesticide spraying agents.

An emulsion has several specific characteristics that differentiate it from a so-called normal solution. For example, differences in the preparation conditions affect the properties even if the concentrations of the constituents are the same. Moreover, several collapse processes exist in an emulsion such as creaming, aggregation, coalescence, and Ostwald ripening. These processes are sensitive to the constituents and their concentrations, as well as to the preparation conditions. Among the collapse processes, creaming is a phenomenon wherein the emulsion droplets are moved to the upper or lower portions according to the differences in density between the dispersed phase and the continuous phase. The rate of creaming is affected by the droplet size, the viscosity of the dispersed phase, and so on. In any case, the condition of an emulsion changes with time. Also, collapse processes change the qualities of an emulsion product.

Recent analytical studies of emulsions include evaluations of collapse processes, analysis of the interface of droplets, and analysis of emulsion products. Turbidity measured by ultraviolet–visible spectrophotometry (UV–vis) has normally been used for the evaluation of emulsion stability. For instance, the time profile of turbidity was studied to evaluate the coalescence and/or solubilization kinetics of oil in microemulsion droplets and for determining the phase-inversion temperature. However, when an incident light insufficiently passes through an emulsion with rather high turbidity, it is difficult to obtain reliable data. In the case of an oil-in-water (O/W) emulsion, a higher concentration of an oil constituent generally causes higher turbidity, although several other factors can also affect the turbidity. In a normal solution with higher absorbance due to a higher concentration, the solution should be normally diluted when measuring transmittance and absorbance. However, in the case of an emulsion, the size of droplets and the emulsion properties are changed by dilution. As a result, the original behavior of a collapse process cannot be evaluated.

Multiphoton ionization time-of-flight mass spectrometry (MPI-TOFMS) has several notable and practical characteristics. The ionization method uses ultraviolet laser pulses with high optical selectivity and less contamination, although soot is often produced by electron ionization sources. Moreover, TOFMS can detect all of the induced ions in principle, and is a robust and reasonably easy way to accomplish design and handling. MPI-TOFMS is normally applied to atomic/molecular spectroscopy and to the trace analysis of compounds in real samples such as in an environmental setting. Although MPI is generally an ionization method for analytes in a gas phase, several vaporization methods for liquid samples have also been studied.

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Recently, we applied MPI-TOFMS to the online analysis of an emulsion.31–37 We developed a sample introduction technique for an emulsion, and the online mass analysis of an oil phase in an O/W emulsion without pretreatment was achieved.31 We reported the time profile of a peak area of an analyte, which was constructed by plotting the peak areas of the corresponding ions in a series of mass spectra. Spikes often appear in time profiles, particularly when measuring an emulsion with a white turbidity. Such spikes derive from oil droplets, and, in the case of a toluene O/W emulsion, we calculated the minimum diameter of a toluene droplet that could be detected as a spike.35,36 We also reported an online analysis of multiple components in an O/W emulsion33 and that of a multiple emulsion.33 We recently used a styrene O/W emulsion to achieve a quantitative analysis of the oil component in an O/W emulsion, and obtained a linear calibration curve of styrene in a concentration of as much as 5 g/L.37

Thus far, the possibility of using MPI-TOFMS to evaluate the creaming of an emulsion has only been suggested.31 In the present study, the time profiles of an O/W emulsion exhibiting a creaming phenomenon were measured by UV–vis and MPI-TOFMS, and the monitoring of an emulsion in a cuvette via these two types of measurement was aligned. Comparable results established the utility of MPI-TOFMS for the evaluation of creaming.

**EXPERIMENTAL SECTION**

**Materials and Sample Preparation.** Toluene and sodium dodecyl sulfate (SDS), as an oil phase and an emulsifier, respectively, were purchased from WAKO Pure Chemical Industries (Osaka, Japan) and were used without further purification. Purified water was prepared in our laboratory.

For the sample preparation, first, water (20 mL) and SDS were added to a vial that was shaken manually until the solid SDS was dissolved. Then, toluene was added and stirred with a homogenizer (AHG-160D, AS ONE, Osaka) at a speed of 5000 rpm for 10 min. The concentrations of both toluene and SDS were 3 g/L with respect to the water. After stirring, 3.5 mL of the prepared sample was transferred to a 5 mL cuvette.

**UV–Vis.** To establish the degree of turbidity, UV–vis transmittance was measured with a spectrophotometer (Shimadzu, UV-160, Japan). To suppress the vaporization of toluene, a cuvette with a lid was placed on the holder of the spectrophotometer. As will be described later, a local region of the sample in a cuvette was monitored by MPI-TOFMS. Therefore, when measuring turbidity, a horizontal slit (2 mm in width) was placed in the front of the cuvette, as shown in Figure 1a, in a region where the transmittance measurement was limited in terms of height. As a result, the incident light passed through the region 1.8–2.0 cm from the bottom of the cuvette.

The wavelengths of the incident light were set to 600 and 266 nm. While each constituent in an emulsion had no absorption in terms of its former wavelength, the absorption for toluene was in the latter wavelength, which approximated the wavelength of the laser pulse for MPI. In the present study, the turbidity was expressed by $-\log I/I_0$, where $I$ and $I_0$ were the incident light intensity and the transmitted light intensity, respectively.

**MPI-TOFMS.** MPI-TOFMS used in the present study is described in detail elsewhere,31 and only briefly discussed here. The sample introduction port was composed of a pair of concentric capillary columns. An emulsion was placed in a cuvette with a lid that had a small hole (1 mm in diameter) to suppress the vaporization of toluene as much as possible. An inner capillary column was inserted through the hole to introduce an emulsion. The tip of the capillary column was set at the same height as the region of the transmittance measurement described previously, i.e., ca. 2 cm from the bottom of the cuvette. Ambient air flowed through an outer capillary column, and the flow rate was adjusted to 2 mL/min via a flow meter (RK-1250, Kofloc, Tokyo, Japan). The pressure of the vacuum chamber was ca. $1 \times 10^{-2}$ Pa.

The linear-type TOFMS used in the present study was developed at Kyushu University, Japan. The fourth-harmonic of a Nd:YAG laser (GAIA II, 266 nm, 4 ns, 10 Hz, Rayture Systems, Tokyo, Japan) was used for ionization. The laser pulse, which was adjusted to ca. 20 μJ, was focused with a plano-convex lens ($f = 200$ mm). The ion signals were acquired without averaging every 0.1 s via a 1 GHz digitizer (AP240, 1 GS/s, Acqiris/Agilent Technologies, Tokyo, Japan). The mass resolution was typically 220 at $m/z$ 92 when measuring a toluene O/W emulsion with a white turbidity (see Figure S1 in the Supporting Information). The time profile for toluene shown in Figure 4a was constructed by fitting the sum of the peak areas of $m/z$ 92 (a molecular ion) and $m/z$ 91 (a fragment ion) on a series of mass spectra. Figure 4b shows the time profile for the average of every 1200 plots, which corresponded to every 120 s, applied in the three experiments including that in Figure 4a. It should be noted that the flow of ambient air did not influence the time profile of the peak area.
of toluene, as shown in Figure 4, but was used simply to stably introduce an emulsion into TOFMS.

■ RESULTS AND DISCUSSION

Features of an Emulsion. The features of the emulsion prepared in a cuvette with time are shown in Figure 2. Just after the preparation, the emulsion developed a white turbidity, the lower portion of which turned slightly transparent with time, while the turbidity in the middle portion, monitored using UV−vis and MPI-TOFMS in the present study, seemed unchanged.

Time Profile of Turbidity by UV−Vis. Figure 3 shows the results of transmittance measurement obtained by UV−vis spectroscopy where the wavelength of the incident light was 600 nm. This indicates the turbidity of an emulsion because the three constituents of the present emulsion, i.e., toluene, water, and SDS, each had no absorption. In this study, the turbidity of the first 5 min was seldom unchanged or was slightly decreased, and was then clearly decreased with time at the height of the monitoring position, although the change in turbidity could not be confirmed by the naked eye (Figure 2). In this manner, the UV−vis time profile measurement of turbidity was useful for evaluating the creaming in an emulsion. However, as shown in Figure 3, the value of turbidity (−log I/I₀) was more than 1.0 during the first 35 min. This indicates the turbidity of an emulsion because the three constituents of the present emulsion, i.e., toluene, water, and SDS, each had no absorption. In this study, the turbidity of the first 5 min was seldom unchanged or was slightly decreased, and was then clearly decreased with time at the height of the monitoring position, although the change in turbidity could not be confirmed by the naked eye (Figure 2). In this manner, the UV−vis time profile measurement of turbidity was useful for evaluating the creaming in an emulsion. However, as shown in Figure 3, the value of turbidity (−log I/I₀) was more than 1.0 during the first 35 min. That is, the transmittance was less than 10%, which means that the incident light was seldom transmitted through the emulsion. In conventional UV−vis spectroscopy, such an experimental condition often results in an increase in the incidence of errors. That is, non-negligible errors easily occur in the measurement of a sample with turbidity such as the one in this study.

Incidentally, the value of −log I/I₀ obtained at 266 nm was saturated at 2.1 throughout the measurement time because, in addition to the presence of turbidity, toluene has absorption at this wavelength. As a result, the creaming of the present emulsion could not be evaluated using the transmittance at this wavelength. Therefore, it is quite difficult to use UV−vis to explain the relationship between the local concentration of a constituent of a dispersed phase and the creaming phenomenon. The proper absorbance might have been obtained by adding water to dilute toluene and also to reduce the turbidity, but the properties of the resultant diluted emulsion, including any creaming behavior, would surely be changed. A shortening of the light path is one possible experimental condition that could enhance transmittance, which could allow an estimation of the concentration. However, the use of a thinner cuvette would be impractical and would likely influence the creaming phenomenon of the emulsion because situations such as convection differ between a thin cuvette and the rather large storage containers that are used in practical situations.

Time Profile of the Concentration of the Oil Phase by MPI-TOFMS. A time profile of the peak area of toluene obtained by online MPI-TOFMS is shown in Figure 4. Figure 4a shows the results of a time profile without averaging. In this measurement, the recording began at the same time that the capillary column was inserted into an emulsion. In Figure 4a, although the signal for toluene was very small, it was surely found after ca. 100−113 s in three measurements and was then saturated at 2.1 throughout the measurement time because, in addition to the presence of turbidity, toluene has absorption at this wavelength. As a result, the creaming of the present emulsion could not be evaluated using the transmittance at this wavelength. Therefore, it is quite difficult to use UV−vis to explain the relationship between the local concentration of a constituent of a dispersed phase and the creaming phenomenon. The proper absorbance might have been obtained by adding water to dilute toluene and also to reduce the turbidity, but the properties of the resultant diluted emulsion, including any creaming behavior, would surely be changed. A shortening of the light path is one possible experimental condition that could enhance transmittance, which could allow an estimation of the concentration. However, the use of a thinner cuvette would be impractical and would likely influence the creaming phenomenon of the emulsion because situations such as convection differ between a thin cuvette and the rather large storage containers that are used in practical situations.

Figure 2. Photograph of an O/W emulsion with time.

Figure 3. Time profile of the turbidity of an O/W emulsion measured by UV−vis at 600 nm. The error indicates the standard deviation (n = 3).

Figure 4. Time profile of the peak area of toluene in an O/W emulsion measured by MPI-TOFMS. (a) Time profile recorded without averaging (by every 0.1 s). (b) Time profile of an average of every 1200 plots, which corresponds to every 120 s, was applied in three experiments including that in (a). The first plot (corresponding to the average of the first 120 s) was ignored because a time from 0 to ca. 120 s was required to pass the sample through the capillary column (see the text). The intensity of the second plot was set to 100%, and the right-hand scale indicates the concentration of toluene where an intensity of 100% is compatible with 3 g/L of toluene (see the text). The error indicates the standard deviation (n = 3).
adopted as the time required for sample introduction. After that, many spikes appeared on the time profile, which decreased both in number and intensity with time. The appearance of spikes suggested the existence of highly concentrated toluene, i.e., toluene droplets.\(^3\) In the present study, the number and size of the toluene droplets seemed to decrease at the monitoring position in the cuvette with the occurrence of the creaming of an emulsion.

Next, an average of every 1200 plots, which corresponded to every 120 s, was re-plotted. The time profile is shown in Figure 4b. The first plot (corresponding to the average value of the first 120 s) was ignored (masked) because a time that ranged from 0 to ca. 120 s was required for the sample to pass through the capillary column. In addition, the average intensity of the second plot was set to 100% in Figure 4b.

Previously, we reported the quantitative analysis of an O/W emulsion wherein a linear calibration curve of the oil phase of styrene in an emulsion with up to 5 g/L was obtained.\(^3\) Although toluene was used in the present study, the results in Figure 4b suggest a change in the concentration of toluene at the sampling position. The concentration of toluene at the beginning of the measurement was 3 g/L. In this figure, the right-hand scale indicates the concentration of toluene where a signal intensity of 100% is compatible with 3 g/L of toluene.

Interestingly, the concentration increased ca. 25% during the first few minutes, which could have been due to the introduction of a large number of, and/or a large size of, toluene droplets. After that, the concentration of toluene decreased at a rate of ca. 0.2 g/L (min) from 8 to 16 min. Finally, the signal intensity was decreased by half compared with the initial (at 2 min) signal intensity, which amounted to a decrease of ca. 1.5 g/L after 60 min. These toluene concentrations were still rather high and not suitable for the measurement of absorbance by UV−vis at 266 nm, and, of course, the decrease in transmittance that was caused by turbidity should also be taken into account. In this manner, the creaming behavior can be quantitatively evaluated using MPI-TOFMS, which can demonstrate the advantage of the present method.

The changes in the signal intensities obtained by UV−vis (Figure 3) and MPI-TOFMS (Figure 4b) differed slightly. That is, the turbidity was either almost constant or slightly decreased for the first few minutes when using UV−vis, while the concentration was somewhat increased when using MPI-TOFMS. Although further studies are needed, one conceivable reason is as follows. The present emulsion had a white turbidity and was polydispersed, and at the monitoring point in the middle part of the cuvette, relatively large oil droplets, which were moved from a lower position, were first detected by MPI-TOFMS. Therefore, the local concentration in the middle portion was increased first. This concentration change, however, only scarcely contributed to the turbidity. In addition, assuming that the latter part of the results (from 14 min or later) of Figures 3 and 4b were a single exponential decay, the decay time constants were calculated to be 34 and 10 min, respectively. In this manner, the differences in behavior between the turbidity and the concentration could be detected.

MPI-TOFMS directly provides information concerning the concentration of an oil component in an emulsion. This method can also be used to monitor the multiple constituents in an emulsion.\(^3\) Therefore, the influence of creaming on each constituent would be evaluated. We are now studying the formulation for the signal behavior obtained by MPI-TOFMS for a quantitative evaluation of the creaming of an emulsion.

### CONCLUSIONS

In the present study, an O/W emulsion that showed creaming was measured via UV−vis and MPI-TOFMS. Transmittance measurement provided information concerning turbidity, but the measurement of emulsions with higher levels of turbidity is difficult in principle. On the other hand, with online mass analysis using MPI-TOFMS, the change in the concentration of an oil phase accompanied by creaming could be evaluated. The tendency of the change in the signal intensities obtained from both methods was slightly different, which suggested that the two methods provided different information, i.e., the turbidity and the local concentration of an oil phase. The information for a local concentration that was obtained by MPI-TOFMS would be difficult to obtain via conventional UV−vis, which makes MPI-TOFMS a useful tool for the evaluation of the collapse process of an emulsion.

### ASSOCIATED CONTENT

Supporting Information
The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acsomega.8b02283.

Mass spectrum of toluene of a toluene O/W emulsion (Figure S1) (PDF)

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**Notes**

The authors declare no competing financial interest.

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