Microscopic Fluorescence Measurement of Fast Interfacial Complexation by Two-Phase Sheath Flow Method

Takahira Tokimoto, Satoshi Tsukahara, and Hitoshi Watarai*
Department of Chemistry, Graduate School of Science, Osaka University, Toyonaka, Osaka 560-0043

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To measure fast reaction processes of the ms order at the interfacial region in the solvent extraction system, we invented a new method using a micrometer-sized two-phase sheath flow system and a laser-induced fluorometry. The reaction between Zn^{2+} with 5-octyloxymethyl-8-quinolinol (Hocqn) in the 1-butanol/water system was examined. The fluorescence intensity of Zn^{2+}-ocqn complex, which increased along the inner organic phase flow, gave the complexation rate of Zn^{2+} with Hocqn.

The liquid/liquid interfaces are playing important roles in the reaction kinetics of the solvent extraction, the liquid-membrane separation, and the interfacial synthesis. Many studies have been carried out to develop the methods to measure reactions at the liquid/liquid interface, so far. For example, the two-phase stopped flow method has succeeded to measure the interfacial reaction in a time range of 0.1 s–a few seconds,[1] and the centrifugal liquid membrane method and the high-speed stirring method were proved to be useful for the measurements of the processes longer than several seconds.[2,3] However, for fast reactions that finish within several ms, the previous methods cannot be applied.

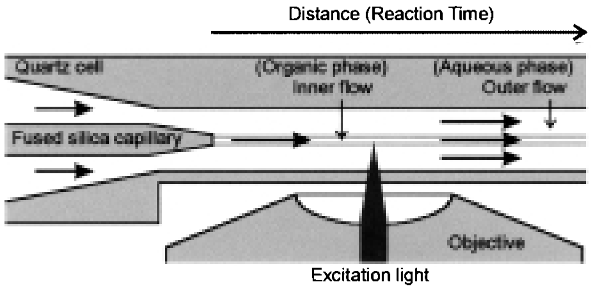

Figure 1. Schematic drawing of the laser-induced fluorescence measurement in the two-phase sheath flow system.

In this study, we made a novel method, a two-phase sheath flow laser-excited fluorometry, to measure fast reactions in the order of ms in the solvent extraction system. Figure 1 is the fluorescence measurement configuration in the two-phase sheath flow system, which consisted of an inner organic solution flow and an outer aqueous solution flow. When the linear velocities of two solutions are matched, a stable two-phase flow was formed without turbulence. Therefore, the interface was regarded as a steady interface except that it was moving from left to right in Figure 1. This means that the distance d along the flows from the capillary outlet can be converted to reaction time t at a constant linear velocity v with the relation $t = d/v$. This unique feature of the sheath flow system allows us to integrate fluorescence at a certain distance for a long time. The cross section of chamber in the quartz cell was 250 μm × 250 μm square, and one side of the cell window was scraped for the observation with an objective.[4] The organic solution was introduced into the cell through a fused silica capillary, whose inner and outer diameters were 50 μm and 150 μm, respectively, and whose outlet was narrowed to 30 μm by pulling under the irradiation with carbon dioxide gas laser.[5] The cell was placed on the stage of an inverted microscope (TE300, Nikon).

In the present study, the 1-butanol/water system has been employed due to its low interfacial tension (1.8 mN m^{-1}), since the inner organic phase with higher interfacial tension (e.g. toluene 36.2 mN m^{-1}) tended to become droplets at the low flow rate adopted in the present study. The extraction system of Zn^{2+} with 5-octyloxymethyl-8-quinolinol (Hocqn) was chosen as a test sample, because the formed complex was highly fluorescent and 8-quinolinol (Hqn) and its derivatives have been widely used for solvent extraction.[6] To detect quite small amount of the complex, a laser-induced fluorescence microscopy was employed.

All reagents were of analytical reagent grade. The organic solution was 1.3×10^{-3} mol dm^{-3} Hocqn in 1-butanol, and the aqueous solution containing 1.1×10^{-2} mol dm^{-3} $ZnCl_2$ and 1.0×10^{-3} mol dm^{-3} 2-morpholinoethanesulfonic acid (MES) as a buffer. The pH of the aqueous solution was kept within 6.0–6.4. Prior to two-phase sheath flow experiments, the aqueous solution was stirred with pure 1-butanol for saturation.

An aliquot (5 cm^3) of Hocqn 1-butanol solution and the same volume of 1.1×10^{-5} mol dm^{-3} Zn^{2+} aqueous solution, including 1.0×10^{-3} mol dm^{-3} MES and 0.01 mol dm^{-3} sodium perchlorate, were shaken for 2 h, and the fluorescence spectra of the organic phase were measured. The wavelengths of excitation and emission maxima of $Zn(ocqn)_2$ were 390 nm and 570 nm, respectively. There were many studies about extracted species in 8-quinolinol extraction systems and the self-adduct form, such as $Zn(qn)_2 \cdot Hqn$.[7] In the present study, the extracted species was assumed to be $Zn(ocqn)_2$, since 1-butanol is a Lewis base solvent.

For the excitation of the complex in the sheath flow system, the second harmonic light (pulse width, 150 fs; wavelength, 400 nm; power, 70 mW; frequency, 82 MHz) of Ti:sapphire laser (Tsunami, Spectra-Physics) was used. The excitation light was reflected by a dichroic mirror (DM455, Nikon) toward the cell and focused on the pillar-shaped organic phase flow by a 20× objective (SuperFluor, Nikon). An intensified cooled CCD camera (PentaMax, Princeton Instruments) detected the fluorescence emitted from the complex formed in the organic phase flow. To measure its fluorescence spectra, a streakscope (C4334, Hamamatsu Photonics) was used in addition. Undesirable light caused by the scattering of excitation light and the Raman scattering of the solvent was removed by the dichroic mirror and a barrier filter (535AF45, Omega Optical; passing 550–580 nm) placed in front of the CCD cam-

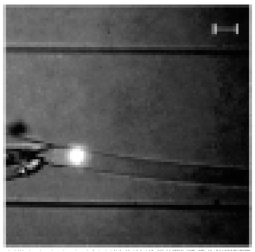

Figure 2. An example of CCD camera image of the two-phase sheath flow system with illumination light. The scale bar represents 30 μm. The linear velocities of two phases are 0.2 m s^{-1}. The white spot on the organic phase flow displays the fluorescence emitted from the complex.

era. By moving the microscope stage, the excitation point was shifted along the inner flow and the fluorescence intensities of the excited spot were measured.

Figure 2 is an example of CCD camera image of this system with illumination light of microscope. We can see that the inner 1-butanol solution, which included $Zn(ocqn)_2$, formed a cylindrical pillar in the sheath flow of the aqueous solution. The linear velocities of the both phases were matched at 0.2 m s^{-1} by controlling the flow rate. The fluorescence was shown as the white spot in Figure 2. This image was used for the determination of the distance between the spot and the capillary outlet. Then, the illumination light was turned off and the fluorescence intensity was measured. The diameter of the inner flow in the Figure 2 was 30 μm and the specific interfacial area, defined as the interfacial area per unit volume of the organic phase, was calculated as 1.3×10^3 cm^{-1}.

Figure 3 shows the results of the kinetic measurement. The fluorescence intensity was converted to the complex concentration by the use of the fluorescence intensities of $Zn(ocqn)_2$ in 1-butanol of a known concentration (1.1×10^{-5} mol dm^{-3}) that was used as an inner phase. The distance between the capillary outlet and the excited spot was converted to the reaction time. The spectra of fluorescence emitted from the excited spots measured by the streakscope were in agreement with that of $Zn(ocqn)_2$ extracted into 1-butanol phase, proving that the detected light was only the fluorescence of the complex. In the system shown in Figure 3, the observed fluorescence intensity was increased with the reaction time, and the slope of 5.4×10^{-4} mol dm^{-3} s^{-1} was thought to be the complexation rate of Zn^{2+} with Hocqn.

In the short contact time less than 5 ms as in the present case, it is difficult to assume the distribution equilibrium of Hocqn between the bulk phases. Simple calculation using 5×10^{-5} cm^2 s^{-1} as a diffusion constant of the solute predicted about 2 μm as its diffusion depth after 5 ms. Therefore, the complexation was postulated to take place only in the interfacial region.

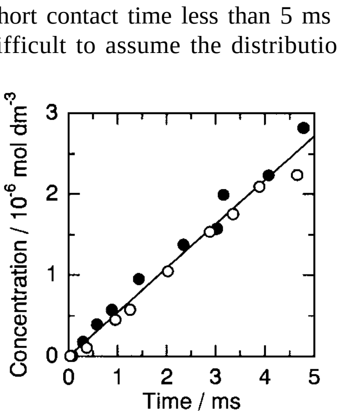

Figure 3. Formation of the fluorescent Zn-ocqn complex in the 1-butanol/water two-phase sheath flow system. The linear flow velocities are 0.26 m s^{-1} (●), and 0.20 m s^{-1} (○). Organic phase was 1.3×10^{-3} mol dm^{-3} Hocqn in 1-butanol. Aqueous phase included 1.1×10^{-2} mol dm^{-3} Zn^{2+} and 1.0×10^{-3} mol dm^{-3} MES, and pH was 6.3.

The rate-determining step of this complexation reaction can be assumed to be the formation of 1:1 complex of Zn^{2+} and Hocqn. When Hocqn is distributed in the aqueous phase close to the interface and reacts with Zn^{2+} there, the extraction rate of $Zn(ocqn)_2$ in the interfacial region may be given by:

$$\frac{d[Zn(ocqn)_2]_{oi}}{dt} = k[Zn^{2+}]_a[Hocqn]_{ai} = \frac{k[Zn^{2+}]_a[Hocqn]_o}{K_D}$$

where $[Zn(ocqn)_2]$, $[Zn^{2+}]$, and [Hocqn] represent the concentrations of the $Zn(ocqn)_2$, Zn^{2+}, and Hocqn, respectively. The subscripts o, oi, a, and ai refer to the organic phase, organic interfacial region, aqueous phase, and aqueous interfacial region, respectively. K_D is the distribution constant of Hocqn. K_D was reported as $10^{5.1}$ in the chloroform/water system.[6] The distribution constant of Hqn in the chloroform/water system and in 1-butanol/water system was reported as $10^{2.37}$ and $10^{1.66}$, respectively.[8,9] With these values, the distribution constant of Hocqn in the interfacial region of 1-butanol/water system was estimated to be about $10^{4.5}$, and hence the rate constant k was calculated as 1×10^6 mol^{-1} dm^3 s^{-1}. The magnitude of this value is close to the values reported in the Eigen mechanism.[6,10] It can be thought that the complexation between Zn^{2+} and Hocqn proceeds in the aqueous phase of interfacial region, where the distribution equilibrium of Hocqn is achieved.

In conclusion, the measurement of the fast (within 5 ms) extraction process of Zn^{2+} with Hocqn was demonstrated by the two-phase sheath flow method with laser-induced fluorometry. The interface in the flow system was stable, and the specific interfacial area was 1.3×10^3 cm^{-1}. The value of the specific interfacial area is much larger than those obtained by the previous methods. Since the contribution of interfacial reaction is extremely enlarged under the high specific interfacial area, the two-phase sheath flow method is promising for the measurement of liquid/liquid interfacial reactions in various extraction systems.

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