# Feasibility of $^{19}$F-NMR for Assessing the Molecular Mobility of Flufenamic Acid in Solid Dispersions

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The purpose of the present study was to clarify the feasibility of $^{19}$F-NMR for assessing the molecular mobility of flufenamic acid (FLF) in solid dispersions. Amorphous solid dispersions of FLF containing poly-(vinylpyrrolidone) (PVP) or hydroxypropylmethylcellulose (HPMC) were prepared by melting and rapid cooling. Spin–lattice relaxation times ($T_1$ and $T_{1\rho}$) of FLF fluorine atoms in the solid dispersions were determined at various temperatures ($-20$ to $150\,°C$). Correlation time ($\tau_c$), which is a measure of rotational molecular mobility, was calculated from the observed $T_1$ or $T_{1\rho}$ value and that of the $T_1$ or $T_{1\rho}$ minimum, assuming that the relaxation mechanism of spin–lattice relaxation of FLF fluorine atoms does not change with temperature. The $\tau_c$ value for solid dispersions containing 20% PVP was 2—3 times longer than that for solid dispersions containing 20% HPMC at 50 °C, indicating that the molecular mobility of FLF in solid dispersions containing 20% PVP was lower than that in solid dispersions containing 20% HPMC. The amount of amorphous FLF remaining in the solid dispersions stored at 60 °C was successfully estimated by analyzing the solid echo signals of FLF fluorine atoms, and it was possible to follow the overall crystallization of amorphous FLF in the solid dispersions. The solid dispersion containing 20% PVP was more stable than that containing 20% HPMC. The difference in stability between solid dispersions containing PVP and HPMC is considered due to the difference in molecular mobility as determined by $\tau_c$. The molecular mobility determined by $^{19}$F-NMR seems to be a useful measure for assessing the stability of drugs containing fluorine atoms in amorphous solid dispersions.

Key words    $^{19}$F-NMR; molecular mobility; stability; crystallization; solid dispersion

Amorphous solid dispersions are used for improving the dissolution rate and solubility of poorly soluble drugs. However, drugs in amorphous form are generally less stable than crystalline drugs because of their higher energy state and higher molecular mobility. It is well known that polymeric excipients can reduce the crystallization rate of many amorphous drugs.[1—12] This stabilization by poly(vinylpyrrolidone) (PVP) is partly attributable to its ability to decrease molecular mobility, as indicated by increases in the glass transition temperature ($T_g$).[9] Therefore, it is of great interest to estimate the molecular mobility of drugs in solid dispersions. Although $^{13}$C-NMR relaxation measurements are useful for assessing the molecular mobility of drugs in solid dispersions,[13] the low sensitivity of $^{13}$C because of its low natural abundance is a drawback of $^{13}$C-NMR. In contrast to $^{13}$C, $^{19}$F has very favorable sensitivity in NMR experiments, since it is present in 100% natural abundance, is second only to the proton in its resonance frequency (except $^3$H) and has a spin quantum number of 1/2. The receptivity for $^{19}$F is 83% of that for $^1$H and 4700 times of that for $^{13}$C.[14] Many drugs containing fluorine atoms are listed in The Japanese Pharmacopoeia. In contrast, almost all pharmaceutical excipients do not contain fluorine atoms. $^{19}$F-NMR may therefore have an advantage over $^{13}$C-NMR or $^1$H-NMR for selectivity and sensitivity when assessing the molecular mobility of drugs containing fluorine atoms in pharmaceutical dosage forms such as solid dispersions.

The orientations and molecular mobility of flufenamic acid (FLF)[15] and $^{19}$F-labeled $\alpha$-tocopherol[16] in a lipid bilayer were studied using $^{19}$F-NMR. Structures and molecular mobility of $^{19}$F-labeled peptides and proteins in biological membranes were also investigated.[17—20] To the authors' knowledge, application of $^{19}$F-NMR to studies of drug molecular mobility in solid dispersions has not been reported.

This paper describes the feasibility of $^{19}$F-NMR for assessing the molecular mobility of FLF in PVP or hydroxypropyl-methylcellulose (HPMC) solid dispersions, and discusses the effect of polymer excipients on the crystallization tendency of FLF in solid dispersions in terms of differences in molecular mobility.

## Experimental

**Materials**    FLF (Fig. 1) was purchased from Wako Pure Chemical Industry (Osaka), and PVP and HPMC were from Sigma (St. Louis, MO, U.S.A.). FLF solid dispersions with PVP or HPMC were prepared by melting and cooling of mixtures of FLF with PVP or HPMC. The solid dispersions obtained were confirmed to be amorphous from microscopic observation under polarized light.

**Nuclear Magnetic Relaxation Measurements**    $^{19}$F-NMR measurements were carried out using a model JNM-MU25 pulsed NMR spectrometer (JEOL DATUM, Tokyo) operating at a resonance frequency of 25 MHz. Time profiles of spin–spin relaxation of the $^{19}$F atoms of FLF were measured using the "solid echo" pulse sequence to overcome the dead time of the instrument. Spin–lattice relaxation time in the laboratory frame ($T_1$) was measured using the inversion recovery pulse sequence. Spin–lattice relaxation time in the rotating flame ($T_{1\rho}$) was measured at spin locking intensity of 10 G.

**DSC Measurements**    $T_g$ of FLF-PVP and FLF-HPMC solid dispersions was measured by DSC using a model 2920 differential scanning calorimeter and a refrigerator cooling system (TA Instruments, Newcastle, DE, U.S.A.). Approximately 5 mg of each solid dispersions was put into an aluminum sample pan and then sealed hermetically. $T_g$ was measured at a heating rate of 20 °C/min. Temperature calibration of the instrument was carried out using indium.
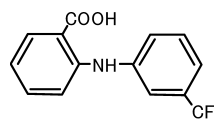


Fig. 1.    Structure of FLF

## Results and Discussion

**Molecular Mobility of FLF as Measured by ^{19}F-NMR Spin–Lattice Relaxation Time** $T_1$ and $T_{1\rho}$ of fluorine atoms of FLF in PVP and HPMC solid dispersions were measured using a pulsed NMR spectrometer in the temperature range from −20 to 150 °C. $T_1$ is sensitive to the molecular motion on the time scale of the resonance frequency (MHz order). On the other hand, $T_{1\rho}$ is sensitive to the molecular motion with a frequency equivalent to the intensity of spin locking field (typically mid kHz order).[21] The temperature dependence of $T_1$ and $T_{1\rho}$ exhibits minimum at a specific temperature at which the molecules of interest have molecular motion with MHz time scale or mid kHz time scale predominantly. The resonance frequency of 25 MHz, lower than that of a conventional high resolution NMR spectrometer, was used to observe $T_1$ minimum in the temperature rage studied. Figure 2 shows the temperature dependence of $T_1$ and $T_{1\rho}$ of FLF fluorine atoms in PVP and HPMC solid dispersions. For FLF–PVP solid dispersions (7:3), the minimum of $T_1$ or $T_{1\rho}$ was observed at about 90 °C and 60 °C, respectively (Fig. 2A). When the PVP content decreased to 20% (w/w), $T_1$ and $T_{1\rho}$ of FLF at temperatures above 70 °C could not be determined due to rapid crystallization. Similar temperature dependence of $T_1$ or $T_{1\rho}$ was observed for the FLF–HPMC solid dispersions (Fig. 2B). The temperature difference between $T_1$ and $T_{1\rho}$ minimum is considered to be due to the difference in the time scale of molecular motion reflected on $T_1$ (MHz order) and $T_{1\rho}$ (mid kHz order). Since the molecular motion on MHz time scale becomes predominant at higher temperature than molecular motion on mid kHz time scale, $T_1$ minimum is observed at higher temperature than $T_{1\rho}$ minimum.

We made following assumptions in order to estimate the molecular mobility of FLF from $T_1$ and $T_{1\rho}$ of FLF fluorine atoms: first, we assumed that FLF fluorine atoms in the solid dispersions relaxes mainly *via* dipolar interaction, and that the contribution of the spin–rotation interaction mechanism[21] is negligible. While relaxation *via* the spin–rotation interaction mechanism has been reported for liquid sample,[22—24] complete domination of dipolar interactions has been reported for fluorine atoms for polycrystalline van der Waals molecular solid.[25] We also made an assumption that the contribution of the cross-relaxation between fluorine and proton atoms can be considered small. It is known that relaxation is not intrinsically single-exponential when cross-relaxation between fluorine and proton atoms takes place.[14] However, we assumed small contribution of the cross-relaxation, because the relaxation of FLF fluorine atoms in the solid dispersions was exponential within experimental uncertainty. In studies of molecular motions, a large number of models describing molecular motions have been proposed for calculation of the spectrum density function.[26] We used a simple model that the molecular motion reflected on $T_1$ or $T_{1\rho}$ is represented by single correlation time for the purpose of comparing the mobility of FLF in the PVP and HPMC solid dispersions. According to the above assumptions, $T_1$ and $T_{1\rho}$ are described by Eqs. 1 and 2.[21]

$$\frac{1}{T_1} = \frac{6}{20}\frac{\gamma^4\hbar^2}{r^6}\left\{\frac{\tau_c}{1+\omega_0^2\tau_c^2}+\frac{4\tau_c}{1+4\omega_0^2\tau_c^2}\right\} \tag{1}$$

$$\frac{1}{T_{1\rho}} = \frac{3}{20}\frac{\gamma^4\hbar^2}{r^6}\left\{\frac{3\tau_c}{1+4\omega_1^2\tau_c^2}+\frac{5\tau_c}{1+\omega_0^2\tau_c^2}+\frac{2\tau_c}{1+4\omega_0^2\tau_c^2}\right\} \tag{2}$$

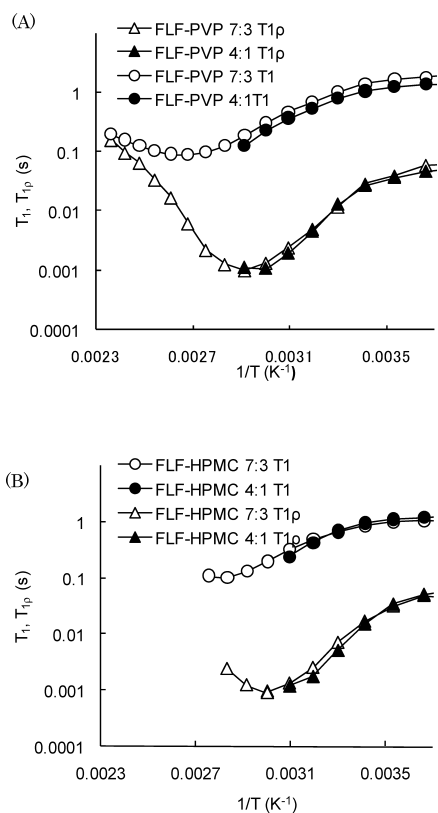where $\tau_c$ is the correlation time that characterizes molecular reorientations, and $\omega_0$ and $\omega_1$ are the resonance frequencies of fluorine atoms in the static magnetic field and spin locking field, respectively. $\gamma$, $r$ and $\hbar$ are the gyromagnetic ratio of fluorine, the distance of neighboring fluorine atoms, and the Plank constant divided by $2\pi$, respectively. Equations 1 and 2 infer that $T_1$ and $T_{1\rho}$ become minimal when $\omega_0\tau_c$ is approximately $0.62$[27] and $\omega_1\tau_c$ is approximately $0.52$,[21] respectively. When the minimum of $T_1$ or $T_{1\rho}$ is observed, we can calculate the unknown value, $r$, in Eqs. 1 and 2. If $r$ is known, the $\tau_c$ value can be calculated from the observed $T_1$ or $T_{1\rho}$ value, assuming that $r$ does not change with temperature.

The values of $r$ calculated from the $T_1$ and $T_{1\rho}$ minimum observed for the FLF–PVP solid dispersion (7:3) were 2.3 and 2.4 Å, respectively, and similar $r$ values were obtained for the FLF–HPMC solid dispersion (7:3). These values are comparable to the reported value (2.174 Å) for 3-(trifluoromethyl)phenanthrene,[25] indicating that dipole interaction between neighboring fluorine atoms can be considered the predominant relaxation mechanism of FLF fluorine atoms in the solid dispersions. The difference between the $r$ values obtained in this work and the reported value suggests that the possibility of the spin–rotation interaction mechanism and/or dipole interaction between fluorine and proton atoms cannot be excluded as a relaxation mechanism of FLF fluorine atoms.

Figure 3 shows the temperature dependence of $\tau_c$ calculated from $T_1$ and $T_{1\rho}$ for FLF fluorine atoms in the solid dis-



(A)

- △ FLF-PVP 7:3 T1ρ
- ▲ FLF-PVP 4:1 T1ρ
- ○ FLF-PVP 7:3 T1
- ● FLF-PVP 4:1T1



(B)

- ○ FLF-HPMC 7:3 T1
- ● FLF-HPMC 4:1 T1
- △ FLF-HPMC 7:3 T1ρ
- ▲ FLF-HPMC 4:1 T1ρ

Fig. 2. Temperature Dependence of $T_1$ and $T_{1\rho}$ of FLF Fluorine Atoms in PVP (A) and HPMC (B) Solid Dispersions

persions. The $\tau_c$ of FLF fluorine atoms in PVP solid dispersions calculated from $T_{1\rho}$ was 8.2 $\mu$s at 50 °C, which was about 3 times larger than that in HPMC solid dispersions (2.6 $\mu$s), indicating that the molecular mobility of FLF was lowered more strongly by PVP than by HPMC.

The $\tau_c$ values calculated using $T_1$ values differ from those calculated from $T_{1\rho}$ values. The slope of temperature dependence of $\tau_c$ changed around $T_g$. These findings suggest that the assumption that the molecular motion reflected on $T_1$ and $T_{1\rho}$ is represented by a single $\tau_c$ may be too simple to describe the molecular motion of FLF in the solid dispersions at temperatures studied, and that two or more molecular motions, such as rotation of trifluoromethyl group and motions with larger scales than rotation of trifluoromethyl group, may be reflected on $T_1$ and $T_{1\rho}$. Further studies including $^1$H-NMR relaxation measurement and dielectric relaxation measurements will be needed to identify the detailed molecular motion of FLF in the solid dispersions.

**Correlation between Crystallization Tendency and Molecular Mobility of FLF in Solid Dispersions** Crystallization proceeds *via* formation of crystal nuclei and crystal growth. As a measure of the crystallization tendency of amorphous FLF in solid dispersions, the overall crystallization rate of amorphous FLF in the solid dispersions was estimated from the time profiles amorphous FLF remaining in the solid dispersions instead of measuring the nucleation rate and growth rate. Amorphous FLF remaining in the solid dispersions was estimated by analyzing solid echo signals of FLF fluorine atoms. Figure 4 shows the solid echo signal of
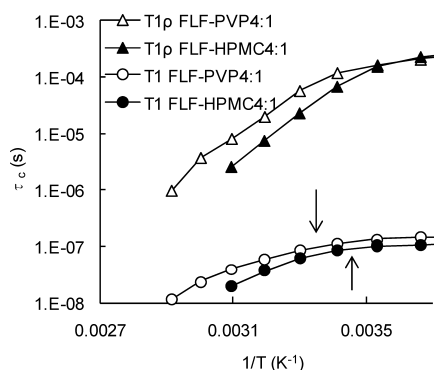
fluorine atoms of FLF in solid dispersions containing 20% (w/w) PVP and that of fluorine atoms of crystalline FLF. The signal for the solid dispersions was describable by the Lorentzian relaxation equation (Eq. 3), and its relaxation time ($T_{2L}$) was approximately 140 $\mu$s. Crystalline FLF exhibited Gaussian relaxation signals (Eq. 4), and its relaxation time ($T_{2G}$) was approximately 30 $\mu$s. These results indicate that amorphous FLF in solid dispersions is considered to exhibit Lorentzian relaxation signals.

$$I = I_0 \exp(-t/T_{2L}) \tag{3}$$

$$I = I_0 \exp\{-t^2/(2T_{2G}^2)\} \tag{4}$$

where $I_0$ and $I$ represent the signal intensities at time 0 and $t$, respectively. Figure 5 shows solid echo signals for the fluorine atoms of FLF in the solid dispersions stored at 60 °C. Samples stored at 60 °C exhibited biphasic decay signals, and signals were describable by summation of the Gaussian (solid line) and Lorentzian (dashed line) equations (Eq. 5).

$$I = I_0\{P_L \exp(-t/T_{2L}) + P_G \exp(-t^2/2T_{2G}^2)\} \tag{5}$$

where $P_L$ and $P_G$ are the ratio of fluorine atoms exhibiting Lorentzian and Gaussian relaxation process, respectively, and $P_L + P_G = 1$. Assuming that the $T_{2L}$ and $T_{2G}$ values are 140 and 30 $\mu$s, respectively, $P_L$ values of FLF in the solid dispersions were estimated by curve fitting. $P_L$ values of the solid dispersions decreased with increasing storage time, indicating that crystallization of amorphous FLF in solid dispersions takes place during storage at 60 °C. To certify the reliability of the $P_L$ values obtained by $^{19}$F-NMR measurements, change in the heat capacity at $T_g$ ($\Delta C_p(T_g)$) was determined for the solid dispersions stored at 60 °C for various periods as a measure of amorphous FLF remaining, and was compared with the value of $P_L$. As shown in Fig. 6, the $P_L$ value was proportional to the $\Delta C_p(T_g)$ value, and was considered to be a useful measure of amorphous FLF remaining in the solid dispersions.

Figure 7 shows the time profiles of the $P_L$ values for FLF solid dispersions containing 20% (w/w) PVP or HPMC at 60 °C. The decrease in the ratio of Lorentzian fluorine atoms was faster for HPMC solid dispersions than for PVP solid dispersions, indicating that the overall crystallization rate of FLF in HPMC solid dispersions is larger than that in PVP solid dispersions. The overall crystallization rate depends on both molecular mobility (the rate of diffusion across the interface between crystalline and amorphous phase) and ther-

Fig. 3. Temperature Dependence of $\tau_c$ of FLF Fluorine Atoms in PVP and HPMC Solid Dispersions
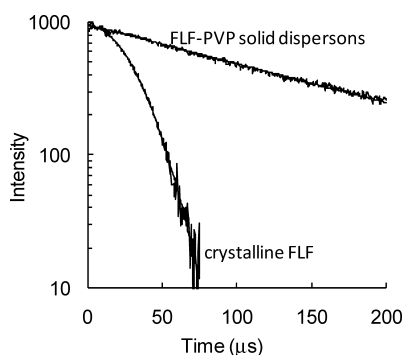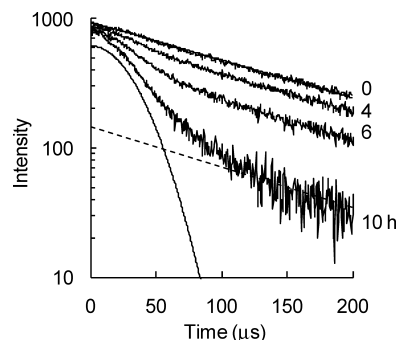
Arrows in the figure represent $T_g$.

Fig. 4. Typical Solid Echo Signal of Fluorine Atoms of FLF in the Freshly Prepared Solid Dispersion Containing 20% (w/w) PVP and That of Fluorine Atoms of Crystalline FLF

Fig. 5. Typical Solid Echo Signals of Fluorine Atoms of FLF in the Solid Dispersions Containing 20% (w/w) PVP Stored at 60 °C
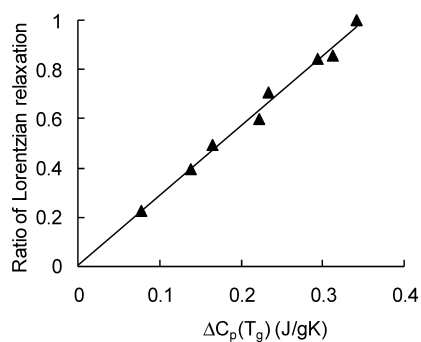
Fig. 6. The Ratio of FLF Fluorine Atoms Exhibiting Lorentzian Relaxation as a Function of Changes in the Heat Capacity at $T_g$
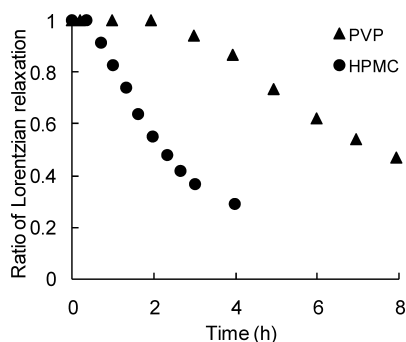


Fig. 7. Time Profiles of the Ratio of FLF Fluorine Atoms Exhibiting Lorentzian Relaxation in PVP and HPMC Solid Dispersions Stored at 60 °C

modynamic factors, such as free energy difference between crystalline and amorphous form.[2,3,10] Differences in the overall crystallization rate of amorphous FLF are consistent with those in the molecular mobility (Fig. 3), suggesting that the molecular mobility as determined by the ^{19}F-NMR spin–lattice relaxation times may be one of the factors determining crystallization rate, and useful as a measure of the physical stability of FLF in solid dispersions. The $T_g$ values of the solid dispersions containing 20% PVP and 20% HPMC were 23 °C and 15 °C, respectively, indicating that molecular mobility reflected on $T_g$ is higher for the solid dispersion containing HPMC than for that containing PVP. The $T_g$ data seem to support the speculation obtained from NMR data. However, the scale of molecular mobility reflected on $T_g$ is considered to be larger than that reflected on $\tau_c$. Further studies should be conducted to elucidate the quantitative correlation between the physical stability of amorphous FLF and the molecular mobility determined by ^{19}F-NMR.

In conclusion, ^{19}F-NMR is useful for elucidating the molecular mobility of drugs containing fluorine atoms in amorphous solid dispersions. $\tau_c$ values of FLF fluorine atoms were calculated from the ^{19}F-NMR spin–lattice relaxation data. The $\tau_c$ value for solid dispersions containing 20% PVP

was 2—3 times longer than that for solid dispersions containing 20% HPMC at 50 °C. Molecular mobility of FLF in the solid dispersions containing 20% PVP was lower than in those containing 20% HPMC, and this was consistent with the fact that the overall crystallization rate of amorphous FLF in the solid dispersion containing PVP was smaller than in that containing HPMC. The molecular mobility determined by ^{19}F-NMR seems to be useful as a measure of the physical stability of an amorphous drug in solid dispersions.

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