Single molecules in soft matter : a study of biomolecular conformation, heterogeneity and plasmon enhanced fluorescence
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4 Crystallite nucleation in supercooled glycerol near the glass transition

Abstract – Heterogeneity and solid-like structures found near the glass transition provide a key to a better understanding of supercooled liquids and of the glass transition. However, the formation of solid-like structures and its effect on spatial heterogeneity in supercooled liquids is neither well documented nor well understood. In this work, we reveal the crystalline nature of the solid-like structures in supercooled glycerol by means of neutron scattering. The results indicate that inhomogeneous nucleation happens at temperatures near T_g. Nevertheless, the thermal history of the sample is essential for crystallization. This implies such structures in supercooled liquids strongly depend on thermal history. Our work suggests that different thermal histories may lead to different structures and therefore to different length and time scales of heterogeneity near the glass transition.

4.1 Introduction

Supercooled liquids have been intensively studied near their glass transition in the past decades, but a full understanding of their structure and dynamics still remains elusive [54, 55, 143]. Heterogeneity, which has been revealed by different experimental methods [93, 144, 145], is a key feature which must be considered when dealing with the glass transition. Fluorescence microscopy of single molecules confirms the presence of large variations in the local viscosities [57–59, 146, 147]. Zondervan *et al.* observed that these variations can be surprisingly long-lived, even at temperatures well above the glass transition temperature (T_g) [58]. This may indicate the existence of a nearly static solid-like network at temperatures above T_g, which was also revealed by rheological measurements [65]. Besides heterogeneity, aging and structural changes were also observed in supercooled liquids near the glass transition. Xia *et al.* observed micrometer-sized structures in thin films of glycerol near the glass transition by fluorescence microscopy [148]. Later on, Möbius *et al.* found that solidification appeared after glycerol aged at a temperature above its T_g [66]. Besides supercooled glycerol, solid-like structures were also observed in other supercooled liquids such as orthoterphenyl (OTP), triphenyl phosphite (TPP) and n-butanol, and have often been attributed to a different state of the material called glacial phase [60, 62, 149, 150] or to a liquid-liquid transition [61]. Such solid-like structures are a natural source of heterogeneity near the glass transition.

In a glacial phase, clusters are seen to form and grow near the glass transition [151]. The clusters are described as an "apparently amorphous phase" or a "defect-ordered phase" by the frustration-limited domain theory (FLD), a thermodynamic theory of supercooled liquids [152–154]. In the FLD theory, supercooled liquids consist of domains with locally preferred structures. Such domains prefer to grow as the temperature is low enough. But the growth of the locally preferred structures is limited by frustration, which is due to strains over large distances. Thus, solid-like amorphous clusters with a limited size can form and exist in supercooled liquids. However, neutron and X-ray experiments suggest the glacial phase to be a liquid and nanocrystalline mixed phase [150, 155–157].

Although theories of supercooled liquids such as the the mode coupling theory (MCT) [67], the random first-order transition theory (RFOT) [158] and the frustration-based theory [154] have been proposed, none of them includes the competition between the supercooled liquid state and the crystalline state. Recently, Stevenson and Wolynes extended the RFOT theory of glasses to account for the existence of a crystalline state [74]. The RFOT theory describes

the existence of a length scale of cooperative reconfiguration of molecules that grows with deeper supercooling in liquids [158]. Just below melting, the classical nucleus size is much larger than that of a reconfiguration region, so that none of local structural reconfigurations toward a crystalline phase can be stable due to their entropic disadvantage. However, the critical size of the classical crystallization nucleus shrinks upon deeper supercooling. Thus, it is possible that the length scale of reconfiguration becomes comparable with the classical nucleus size at deep enough supercooling. When this happens, fluctuations or driving forces can be sufficient for local crystalline configuration (nanocrystallites) to cross the critical size and to initially form and grow [74]. It is worth to note that the crystal morphology evolves with temperature and time in this scenario. The nanocrystallites are expected to be finite in size and have a fractal shape at early stage. Then they can develop into a ramified network as time elapses [74]. This newly developed theory suggests the solid-like structure can be crystalline. Hence, the structural origin of the solid-like structure developed in supercooled glycerol near T_g becomes significantly important for a better understanding on previous experimental observations [58, 65, 66, 148]. To get more insight into the structural organization of the solid-like network and on the solid-like fraction of the sample, we performed neutron diffraction and inelastic neutron scattering.

4.2 Experimental

Inelastic neutron scattering (INS) was performed on the backscattering spectrometer IN10 (ILL, Grenoble, France) in the so-called fixed-window scan mode, in which the elastic scattering intensity is monitored as a function of the sample temperature or time. The instrumental resolution determines the probed dynamical range: any motions with characteristic timescale faster than 550 ps will lead to quasielastic broadening, therefore a decrease in elastic intensity. The measurements are performed in the momentum transfer range 0.1 - 2 Å^{-1}, close to the maximum of the structure factor.

Neutron diffraction was performed in the Q-range of 0.02-1.70 Å^{-1} on the small momentum transfer diffractometer D16 (ILL, Grenoble, France). The sample was placed into a cryostat with quartz windows and thermalized within 0.1 K. Deuterated samples were used in order to decrease the incoherent background arising from hydrogen atoms.

Deuterated glycerol (CD$_2$OD-CDOD-CD$_2$OD, Cambridge Isotope Laboratories) and protonated glycerol (CH$_2$OH-CHOH-CH$_2$OH, Sigma-Aldrich) were used without further purification, but handled under an inert and dry
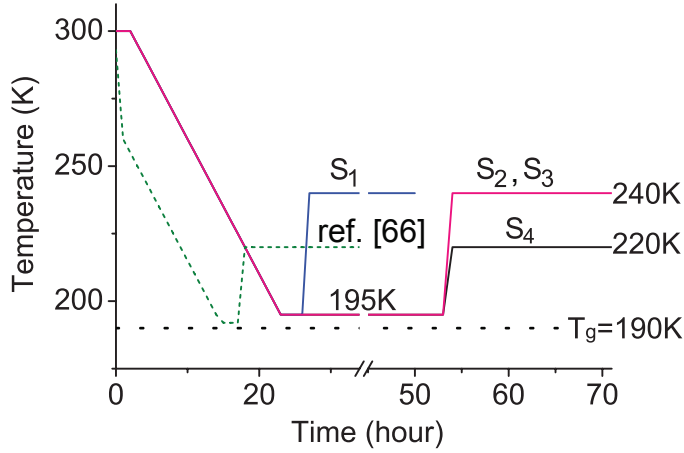
Figure 4.1: Thermal histories of the samples. S_1, S_2, S_3 and S_4 correspond to four samples which underwent three different thermal histories. S_1, S_3, and S_4 are deuterated samples. S_2 is a protonated one. The dashed line shows the thermal history to start solidification in protonated glycerol [66].

atmosphere. Protonated glycerol was filled into an aluminium cell with a sample thickness of 0.25 mm. Deuterated glycerol was filled into a fused silica cell (Hellma, 110QS) with an optical path of 2 mm. All materials were only quenched a single time to the low temperature for measurement, without being rewarmed. The samples were submitted to the same quenching process but different aging histories. All samples were cooled from 300 K (9 K above the melting temperature) to 195 K (5 K above T_g) at a cooling rate of 5 K/h, which was reported to be crucial for creating the solid-like state [66]. After quenching, they were kept at 195 K for 3 or 30 hours before being transferred to the instruments. During the transfer, the samples were immersed in liquid nitrogen (77 K) for less than 10 minutes. The temperature of the experimental cryostat was stabilized at 195 K for an hour after a sample was inserted, then increased to 240 K or 220 K. The thermal profiles are shown in figure 4.1.

4.3 Results

As described in the experimental section, we monitored the elastic intensity arising from the sample as a function of time in INS measurements. Although the high-frequency fast relaxation that contributes to the intensity is much faster than alpha-relaxation, it has a similar temperature dependence with
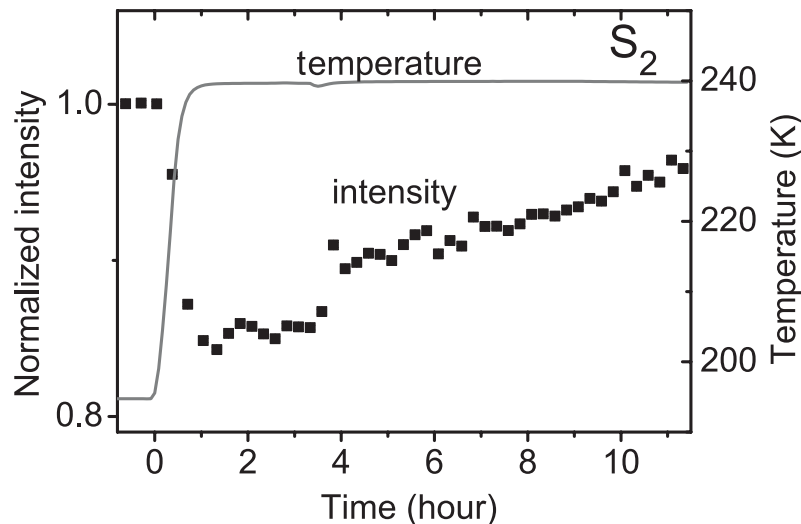
Figure 4.2: The scattering intensities on IN10 from sample S_2 versus time at $240\,K$. The solid curve shows the temperature profile during measurements. The squares shows the normalized scattering intensities.

that of alpha-relaxation [159]. Due to the temperature dependence of the Debye-Waller factor and that of the fast high-frequency relaxation, the elastic scattering intensity decreases when the temperature increases. We can clearly see such behavior on protonated sample S_2, as shown in figure 4.2. At the lowest temperature ($195\,K$), the sample gives the maximal intensity. The elastic scattering intensity decreases while the temperature increases from $195\,K$ to $240\,K$. At a fixed temperature, any increase of the scattering intensity indicates the growth of solid-like structures. Such intensity increase can be clearly identified in figure 4.2. We did not observe any increase of intensity on deuterated sample S_1 (data not shown).

Neutron diffraction patterns of deuterated samples were measured at two fixed temperatures ($240\,K$ and $220\,K$) at different aging times. Each diffraction pattern was accumulated for one hour. As we can see in figure 4.3 (a), after three hours at $240\,K$, a few Bragg peaks appeared on the diffraction patterns and their intensities grew with time. The positions of the peaks ($1.089\,Å^{-1}$, $1.260\,Å^{-1}$, $1.337\,Å^{-1}$, $1.480\,Å^{-1}$, $1.545\,Å^{-1}$ and $1.607\,Å^{-1}$) correspond to the crystal indexes (1 1 0), (0 2 0), (1 0 1), (1 1 1), (1 2 0) and (0 2 1) respec-
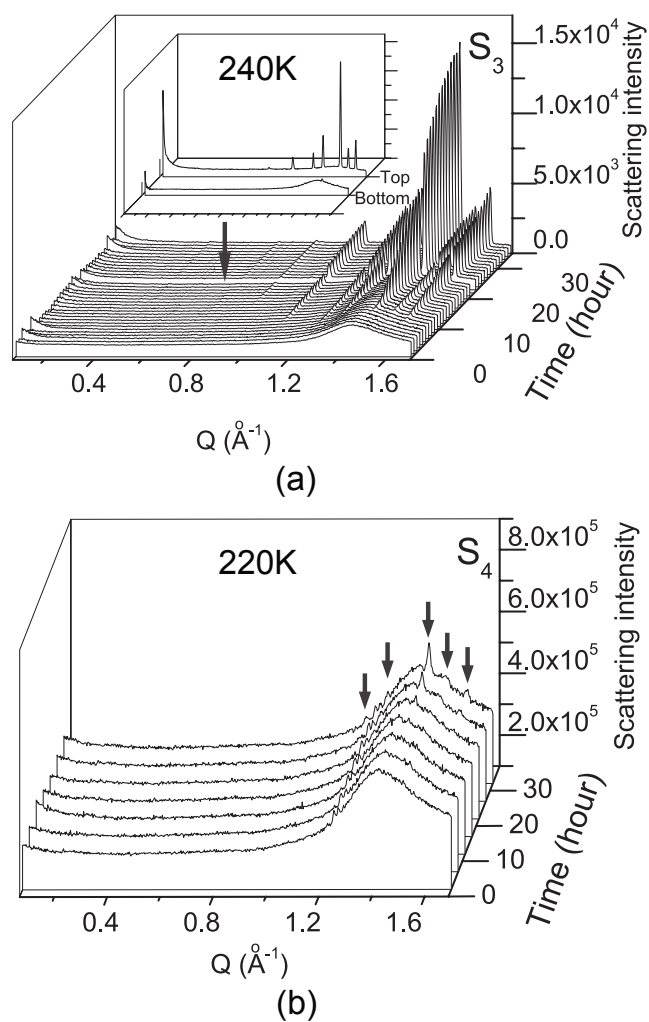
Figure 4.3: (a) shows the diffraction patterns of sample S_3 recorded with time at 240 K. The arrow indicates a point in time when we measured diffraction patterns of the top and bottom halves of the sample separately (shown in insert). (b) shows the diffraction patterns of sample S_4 recorded with time at 220 K. The arrows point out the appearance of five diffraction peaks.

	3hrs at 195 K	30hrs at 195 K	aging at 220 K	aging at 240 K	deuterated	protonated
S$_1$	Y			Y	-	+ [66]
ref [66]	Y		Y		NA	+ [66]
S$_2$		Y		Y	+	+
S$_4$		Y	Y		+	NA

Table 4.1: Thermal histories and solidification status after aging. "Y" shows the thermal treatment applied to the sample. "-" means solidification did not take place. "+" means solidification happened. "NA" means no experimental result available.

tively [160]. This observation reveals the crystalline nature of the solid-like structures formed in the deuterated sample S$_3$. By comparing the thermal histories used to trigger solidification (see table 4.1), we find that deuterated glycerol needs a significantly longer aging time near T$_g$. The differences in bond energy and in mobility between deuterated and protonated glycerol [161] are sufficient to induce significant differences in the thermal histories causing solidification. Despite these small differences, we believe that the solid-like structure appearing in protonated glycerol is crystalline, as the one found in deuterated glycerol with a similar thermal history.

The full-widths at half-maximum (FWHM) of the Bragg peaks do not vary much with time and are mainly limited by the instrument's resolution. We can only estimate the average crystallite size to be larger than 45 nm according to Scherrer's equation [162]. To check whether the crystallization is homogeneous, we moved the beam slits to measure the diffraction patterns of the bottom and top halves of the sample. The top half of the sample gave strong crystal peaks while there were weak crystal peaks at the bottom half of the sample, as shown in the insert of figure 4.3 (a). This inhomogeneous solidification at the early stage is in good agreement with the observation by Möbius *et al.* [66]. On deuterated sample S$_4$, as shown in figure 4.3 (b), Bragg peaks started to appear and grow after 22 hours at 220 K. Until the beam time was over, five peaks appeared on the diffraction pattern. At a temperature as low as 220 K, crystallization could still happen, but at a rate more than one decade slower. This corresponds well with the slow down of the alpha-relaxation of glycerol at these temperatures [93].

4.4 Discussion

Our experiments reproduced the solid-like structures previously found in supercooled glycerol and revealed a crystallization process in supercooled glycerol near the glass transition. The neutron scattering results indicate the crystalline nature of the solid-like structures. We suppose nuclei form and grow near T_g since the crystallization only occurred after long time aging near T_g. These observations agree with the nanocrystallite theory proposed by Stevenson and Wolynes [74]. In the nanocrystallite theory, nanocrystallites are predicted to form and grow when the length scale of dynamical correlation becomes comparable to or larger than the critical nucleus size at deep enough supercooling. In our case, glycerol near the glass transition could have been sufficiently deeply supercooled to allow nanocrystallites to exceed the critical nucleus size and grow. At a higher temperature, when the scale of dynamical correlation is well separated from the critical nucleus size, for example at 240 K in this experiment, nanocrystallites cannot form anymore. After a raise in temperature, already existing nanocrystallites will disappear if they do not exceed the critical size at this higher temperature. In contrast, if the nanocrystallites have exceeded the critical size by the time of increasing the temperature, they can grow at a faster speed because of accelerated alpha-relaxation. Hence, the onset of solidification strongly depends on thermal history of the sample under investigation. Furthermore, the nanocrystallite theory may also help to understand the different patterns observed in supercooled glycerol with different thermal histories [148]. The nanocrystallite theory describes a developing morphology of the crystalline structures in supercooled liquids. Isolated nanocrystallites are first surrounded by liquid at the early stages, then grow and connect into a solid-like network. In addition, one expects different shapes of the cooperatively rearranging regions in supercooled liquids to be either a compact nearly spherical shape or a extended string-like shape depending on the depth of supercooling [163]. Hence, morphology of the crystalline structure strongly depends on the thermal history of supercooled liquids as well. It is worth to note that different heterogeneity length scales were revealed experimentally in supercooled glycerol. For instance, 1 nm heterogeneity near T_g was found by NMR [164]. This is smaller than at least a few nanometers revealed by single-molecule microscopy [58]. These different experimental results may be due to the different thermal histories applied on the samples. Therefore, heterogeneity should be probed under full control of the thermal history.

To conclude, we reveal the crystalline nature of the solid-like structure indicated in previous studies on supercooled glycerol [58,65,66,148]. We have

evidence that a nucleation process takes place during aging at a temperature near T_g. Nonetheless, crystallization strongly depends on the thermal history that the sample was submitted to. The experimental findings agree with the newly developed nanocrystallite theory. Our results emphasize the importance of controlling thermal history when studying heterogeneity length scales in supercooled liquids.

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