Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Glucose Directs Amyloid-beta Into Membrane-Active Oligomers

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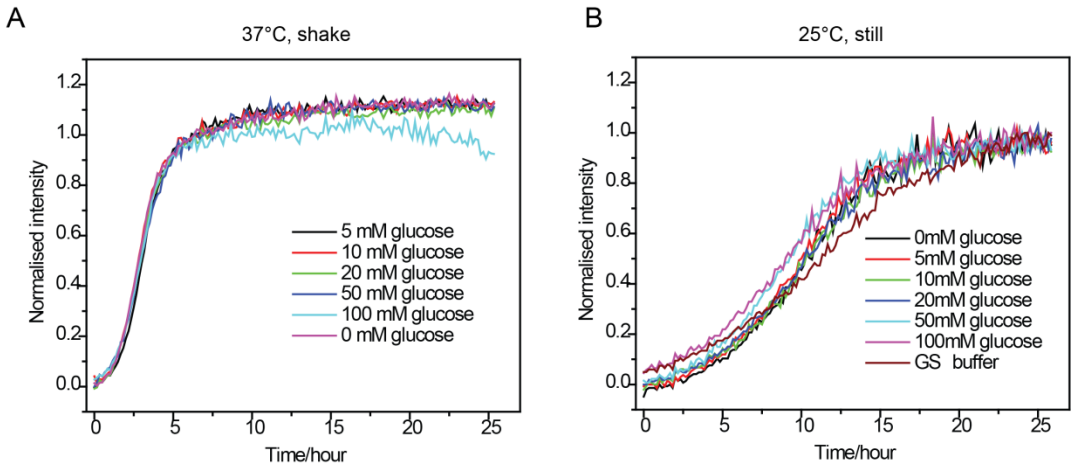
Supplementary figures S1-S7

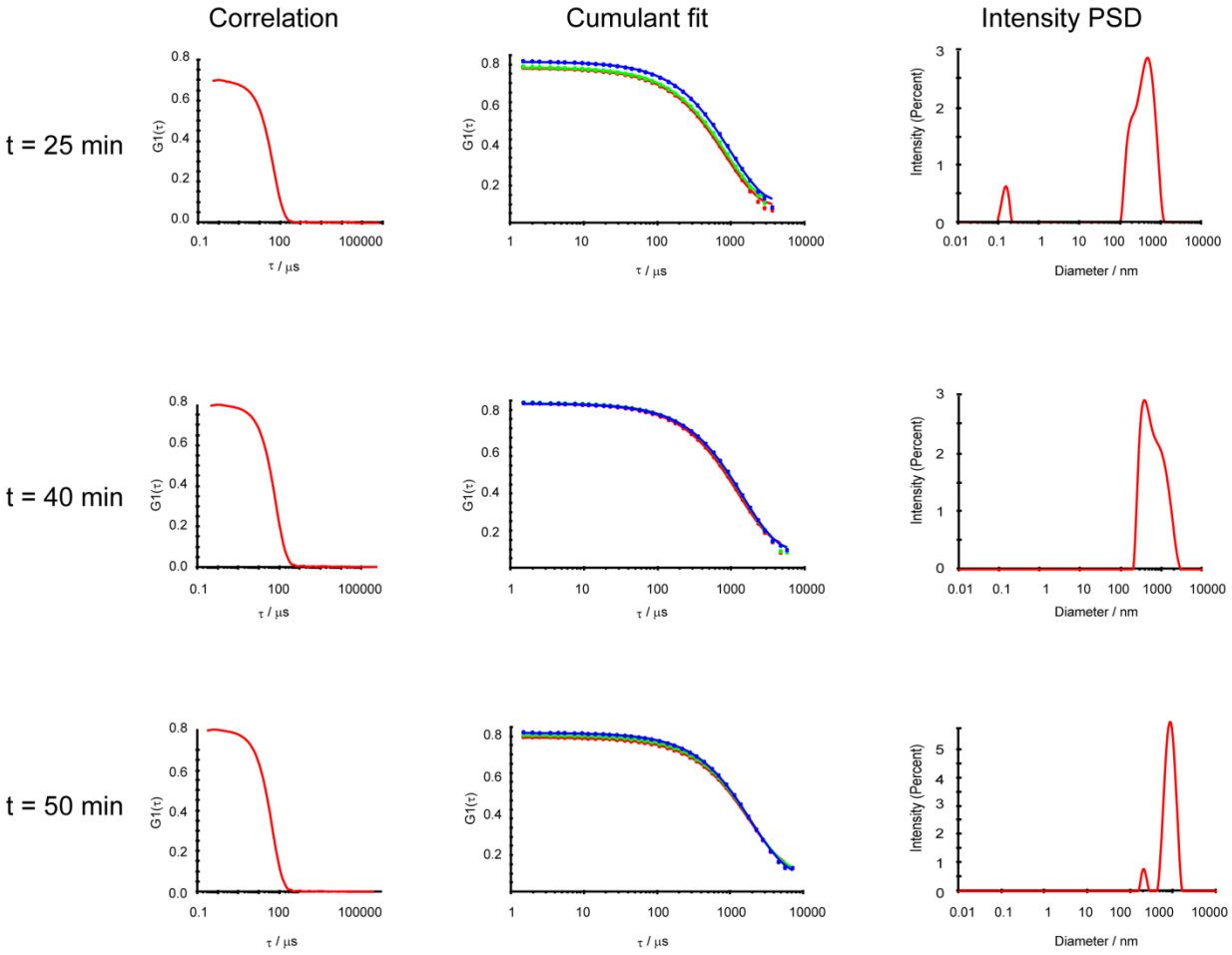
Supplementary Figure S1



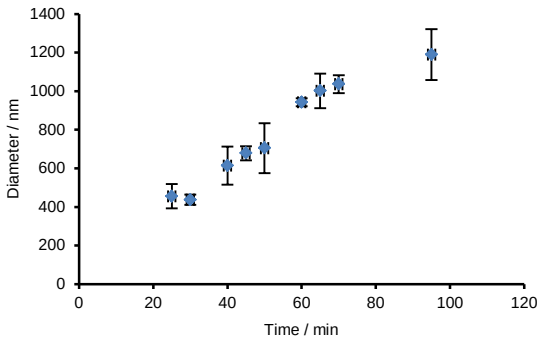
(A) Aggregation kinetics of 5 µM freshly filtered Aβ-42 in PBSN buffer containing different concentrations of glucose (0 to 100 mM) at 37°C temperature. Intermittent agitation (5s every 10 min) was used for this assay. (B) Aggregation kinetics of 5 µM freshly filtered Aβ-42 in PBSN buffer containing different concentration of glucose (0 to 100 mM) or GS-buffer at 25°C temperature. Samples were measured without agitation. Both kinetics were monitored by measuring fluorescence intensity of the ThT dye (20 µM).

Supplementary Figure S2

A



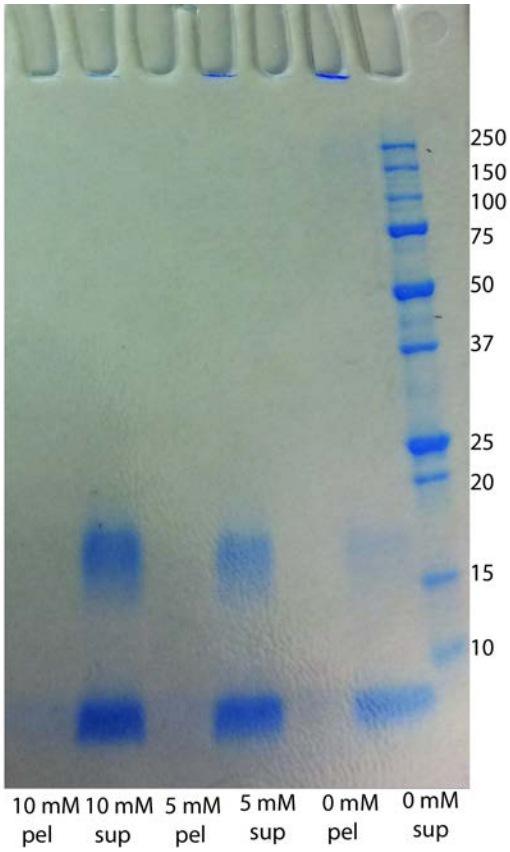
B



A) Dynamic light scattering autocorrelation curves, cumulant analysis, intensity particle size distributiondiameter distribution of Aβ-42 incubated in PBSN containing 10 mM glucose buffer for 25 min, 40 min and 50 min. B) Average particle diameters from intensity distribution of Aβ-42 oligomers

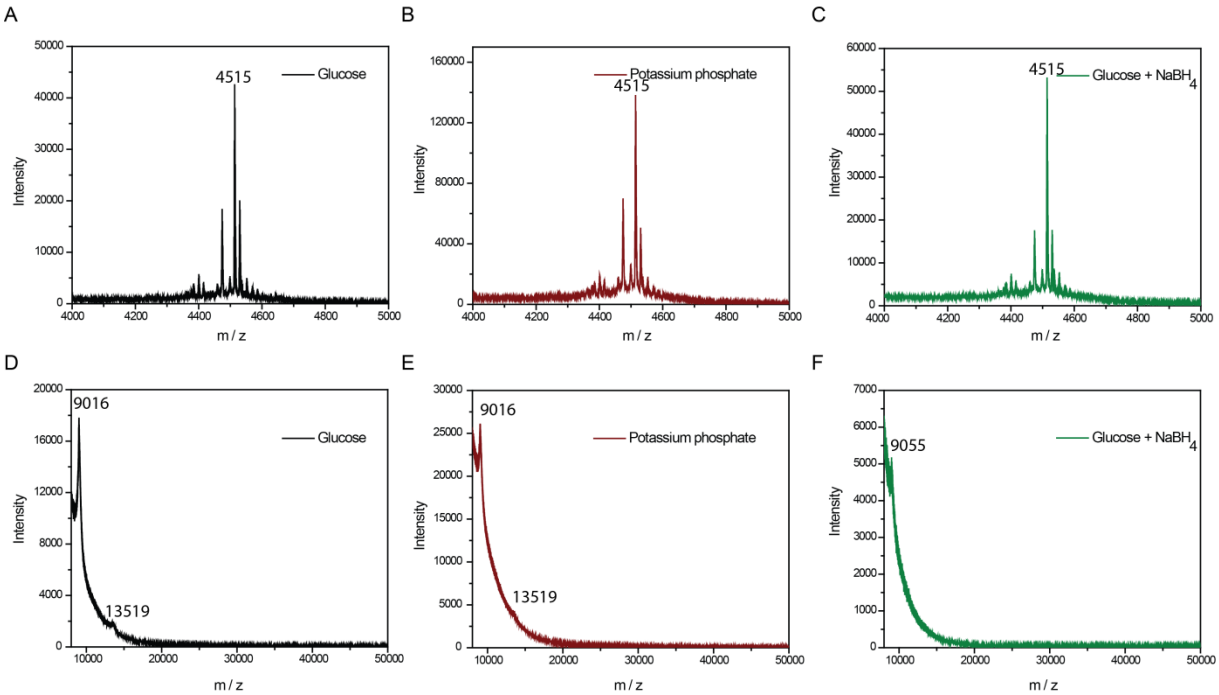
in PBSN containing 10 mM glucose derived from triplicate measurements, mean ± SD. Correlation analysis found fairly homogenous Aβ-42 particles in presence of glucose, whose size increased with incubation time.

Supplementary Figure S5



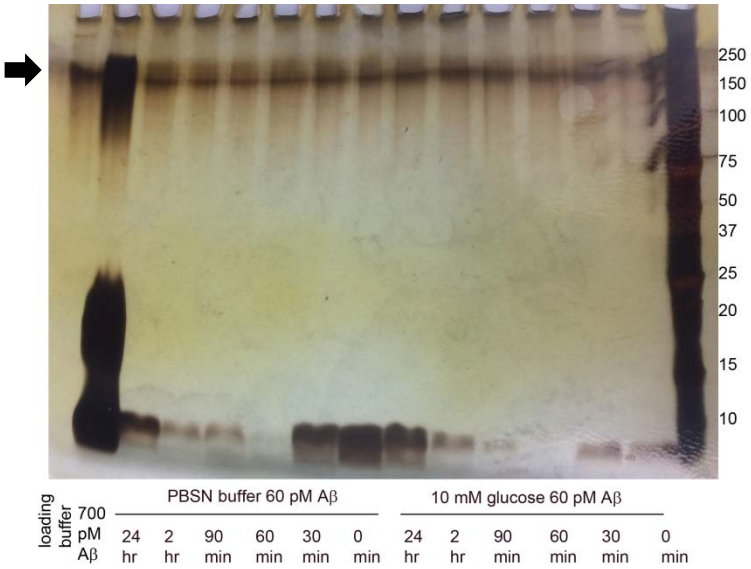
Quantification of soluble Aβ-42 by ultracentrifugation. 60 µM Aβ-42 was incubated in PBSN (K-phosphate 8 mM, pH 6.8; NaCl 150 mM) containing different concentration of glucose (0 mM, 5 mM and 10 mM) for 1 h. Soluble and insoluble forms of the peptides were separated by ultracentrifugation at 70,000 rpm (200,000 x g) for 20 minutes at 4^{o}C. After ultracentrifugation supernatants were collected and pellets were dissolved by 10 min sonication in the same volume of the same buffer as the supernatant. 2% SDS was added and supernatant and pellet fractions were boiled for 10 min. Then, samples were separated by SDS-PAGE and visualized by Coomassie brilliant blue staining. All Aβ-42 was found in the supernatant fractions in the form of monomers and SDS-stable trimers.

Supplementary Figure S4



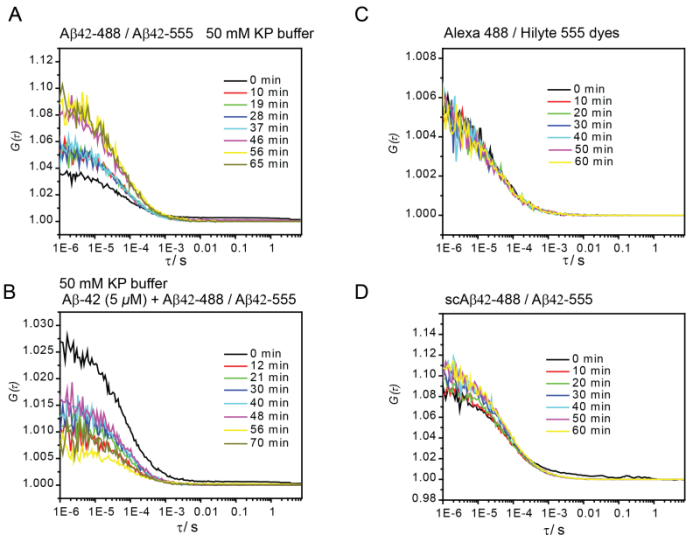
MALDI mass spectrometry analysis of 5 µM Aβ-42 incubated for 60 minutes with PBSN buffer containing 10 mM glucose (A, D), PBSN buffer (B,E) or PBSN buffer containing 10 mM glucose in which the sample was reduced by $NaBH_4$ (2 mM) added after incubation to stabilize potential Schiff base adducts (C,F). Plots A-C analyze intensities at m/z from 4000 to 5000, D-F from 8000 to 50000. In all cases m/z of main peaks represent unmodified Aβ42 monomer (4515), dimer (9016), and trimer (13,519). No peaks were found that corresponded to a Schiff base adduct of glucose (+ 162 m/z).

Supplementary Figure S5



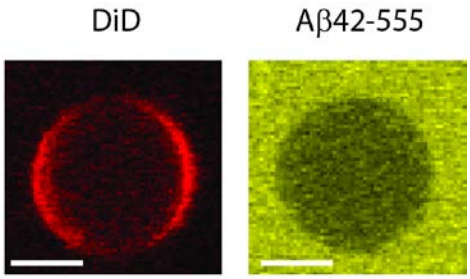
5 µM Aβ-42 (60 pmol) incubated with PBSN buffer or PBSN buffer containing 10 mM glucose for 0-24h was run through SDS PAGE gel and visualized by silver staining. Monomeric Aβ-42 (100 µM, 700 pmol) in NaOH and loading buffer were run as control samples. We found no evidence of covalent crosslinking of Aβ-42 monomers via Maillard reaction. The arrow indicates an unspecific staining band also present in the buffer control sample. Note that monomeric Aβ-42 (700 pmol) was strongly overstained, revealing trace amounts of oligomers.

Supplementary Figure S6



Fluorescence cross-correlation curve collected at different time points for (A) Aβ42-Hilyte 488 (50 nM) and Aβ42-Hylite 555 (40 nM) in 50 mM KP buffer, pH 6.8; (B) 5 μ Aβ-42 with Aβ42-488 (50 nM) and Aβ42-555 (40 nM) in 50 mM KP buffer, pH 6.8; (C) Free Hilyte 555 dye and Alexa 488 dye in GS buffer; (D) scrambled Aβ42-488 (16 nM) and Aβ42-555 (10 nM) in GS buffer.

Supplementary Figure S7



Confocal microscopy images of DiD labeled DOPC GUV and of a GUV in the presence of Aβ42-Hilyte 555 (50 nM) + Aβ-42 (5 µM).