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Supporting Material

Designed Fluorescent Probes Reveal Interactions between Amyloid-β-peptides (1-40) and G_{M1}-Gangliosides in Micelles and Lipid Vesicles

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Supporting Material

Preparation of vesicles. Large unilamellar vesicles were prepared by means of the extrusion technique. For this an extruder manufactured by Lipex Biomembranes Inc. (Vancouver, Canada) was used. Briefly, the dried lipid film, containing bovine sphingomyelin, cholesterol, ganglioside G_{M1} and the labelled G_{M1}, was hydrated to the necessary concentration by using a 20 mM Tris-HCl buffer (pH 7.4) which contained 1 mM disodium salt of EDTA. This mixture was freeze-thawed five times and then passed ten times through double polycarbonate filters (Nucleopore) with a pore size of 100 nm at 50 ^{o}C.

Steady-state and time-resolved fluorescence experiments. The emission and excitation fluorescence spectra were recorded using a Fluorolog®-3 (Jobin Yvon Inc. U.S.A.) spectrometer equipped with Glan-Thompson polarisers. The spectral bandwidth of the excitation and emission monochromators was 2 nm. The fluorescence spectra were corrected. Fluorescence lifetime decays were measured by using the time-correlated single-photon-counting (TCSPC) technique. The instrument was a PRA 3000 (PRA, Canada and the excitation source was NanoLED N-14, 471 nm, (IBH, Scotland) pulsed diode, operated at 800 kHz or PicoQuant laser, 470 nm. The excitation and emission wavelength were selected using a set of filters (Melles Griot, The Netherlands). For the excitation an interference filter centred at 470 nm (FWHM = 9.3 nm) was used, and for the emission interference filters centred at 520 nm (FWHM = 28 nm), 540 nm (FWHM = 28 nm), and long pass filters from 550 nm and 610 nm. The fluorescence decays were collected over 1024 channels with the resolution of 50 ps/ch, with at least 10 000 photons in the peak maximum for the lifetime experiments, which were made with the emission polariser set at the magic angle (54.7^{o}) relative to the excitation polariser. For the time-dependent anisotropy experiments, fluorescence decays were collected with the difference of 50 000 counts in the peak maxima of the fluorescence decays, which were obtained for the standard polarised settings VV and VH.

Absorption spectra were recorded on Cary 5000 (Varian, UK).

Modelling of donor-acceptor electronic energy transfer. A model is described for calculating the average distance from a donor group to an acceptor group, which are localised at the distances R_{D} and R_{Cn} from the centre of a spherical micelle (*cf.* Fig. S1). The average rate of

energy transfer (ω) depends on the distribution of distances $\left|\vec{R}\right| = \left|\vec{R}_{\mathrm{D}} - \vec{R}_{\mathrm{Cn}}\right|$ which, due to the spherical symmetry, is the same for all relative orientations (α, β). The transfer rate also depends on donor and acceptor number densities, $\rho_{\mathrm{D}} \propto 1/R_{\mathrm{D}}^2$ and $\rho_{\mathrm{Cn}} \propto 1/R_{\mathrm{Cn}}^2$. The transfer rate can be written as:

$$\omega \propto \rho_{\mathrm{D}}(R_{\mathrm{D}})\rho_{\mathrm{Cn}}(R_{\mathrm{Cn}}) \int_{R_{\min}}^{R_{\max}} \frac{dR}{R^6} \qquad \text{(S1)}$$

In Eq. S1 the donor-acceptor distance depends on R_{Cn}, R_{D} and the orientation angle β, according to:

$$R = \left(R_{\mathrm{D}}^2 + R_{\mathrm{Cn}}^2 - 2R_{\mathrm{D}}R_{\mathrm{Cn}}\cos\beta\right) \qquad \text{(S2)}$$

In the following we are interested in an expression for the relative average rates of energy transfer between a donor and acceptors located at the distances R_{Cn} and R_{Cm}. This means that the rates between two experiments are compared, in which the average number of donors per micelle is the same in each experiment, as well as the number of acceptors. For each of the two cases the transfer rate is obtained by averaging over the orientations (α, β). This yields that the relative transfer rates are given by the following formula:

$$\frac{\omega_{\mathrm{Cm}}}{\omega_{\mathrm{Cn}}} = \xi(R_{\mathrm{D}}) = \frac{R_{\mathrm{Cn}}^2}{R_{\mathrm{Cm}}^2}\left[\frac{\left(\frac{1}{R_{\mathrm{D}}^2 + R_{\mathrm{Cm}}^2 - 2R_{\mathrm{Cm}}R_{\mathrm{D}}}\right)^{5/2} - \left(\frac{1}{R_{\mathrm{D}}^2 + R_{\mathrm{Cm}}^2 + 2R_{\mathrm{Cm}}R_{\mathrm{D}}}\right)^{5/2}}{\left(\frac{1}{R_{\mathrm{D}}^2 + R_{\mathrm{Cn}}^2 - 2R_{\mathrm{Cn}}R_{\mathrm{D}}}\right)^{5/2} - \left(\frac{1}{R_{\mathrm{D}}^2 + R_{\mathrm{Cn}}^2 + 2R_{\mathrm{Cn}}R_{\mathrm{D}}}\right)^{5/2}}\right] \qquad \text{(S3)}$$

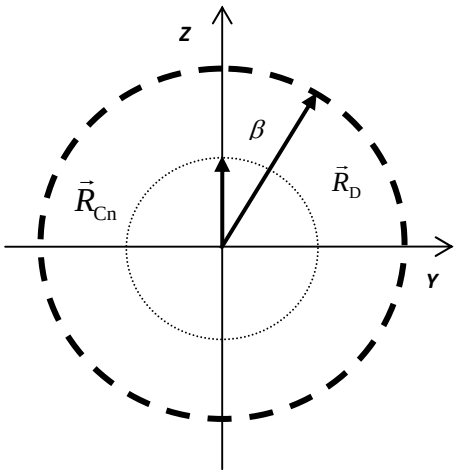

Fig. S1. Schematic of a spherical micelle in which the positions of the donor ($\vec{R}_D$) and acceptor ($\vec{R}_{Cn}$) groups are indicated by vectors. The donor positions are given in spherical polar coordinates $\vec{R}_D = R_D\left(\cos\alpha\sin\beta, \sin\alpha\sin\beta, \cos\beta\right)$.