Data article

X-ray diffraction and NMR data for the study of the location of idebenone and idebenol in model membranes

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**Abstract**

Here we present some of our data about the interaction of idebenone and idebenol with dipalmitoyl-phosphatidylcholine (DPPC). In particular, we include data of small angle X-ray diffraction (SAXD) and wide angle X-ray diffraction experiments, obtention of electronic profiles of the membranes, 2H-NMR and 31P-NMR, as part of the research article: “Both idebenone and idebenol are localized near the lipid-water interface of the membrane and increase its fluidity” (Gomez-Murcia et al., 2016) [1]. These data were obtained from model membranes that included different proportions of idebenone and idebenol, at temperatures both above and below of the gel to fluid phase. The X-ray experiments were carried out by using a modified Kratky compact camera (MBraun-Graz-Optical Systems, Graz Austria), incorporating two coupled linear position sensitive detectors. The NMR data were collected from a a Bruker Avance 600 instrument.

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### Specifications Table

| Subject area More specific subject area | Biology | Biophysical Chemistry of membranes |
|----------------------------------------|---------|-----------------------------------|
| Type of data                           | Table, figure | X-ray diffraction; NMR |
| How data was acquired                  | Raw and analyzed | Samples prepared as described in Gómez-Murcia et al. [1]. |
| Data format                            | SAXRD form DPPC-idebenol samples; d-spacings of samples DPPC/idebenone and DPPC/idebenol; electronic profiles of the membranes obtained from the diffractograms; $^{31}$P-NMR and $^2$H-NMR of d62-DPPC/idebenone and d62-DPPC/idebenol |
| Experimental features                  | Murcia, Spain |
| Data source location                   | Data is with this article |

### Value of the data

- These diffractograms, electronic profiles and spectra may be useful to establish comparisons with the data obtained by other workers exploring these biomolecules or other similar.
- Providing these details to the scientific community may be of help when trying to interpret experiments carried out by using the same techniques.
- The supplied repeat distances obtained from X-ray diffraction may be of great help to study in detail these systems.

### 1. Data

These data were obtained as described in detail in [1]. SAXD diffractograms, SAXD and WAXD spacings, $^{31}$P NMR and $^2$H NMR are included obtained from idebenone-DPPC and idebenol/DPPC samples.

### 2. Experimental design, materials and methods

**Materials.** 1-Palmitoyl-2-oleoyl-sn- glycerol-3-phosphocholine (POPC); 1, 2-dipalmitoyl-sn-phosphatidylcholine (DPPC) and 1, 2-d$_{62}$-sn- dipalmitoyl-sn-phosphatidylcholine (DPPC-d$_{62}$) were obtained from Avanti Polar Lipids (Birmingham, Alabama, USA). Idebenone was from TCI Europe N.W. (Zwijndrecht, Belgium) and all other chemicals were highly pure from Sigma Chemical Co. (Madrid, Spain).

### 3. Methods

#### 3.1. Reduction of idebenone to idebenol

Idebenol was prepared from idebenone by reduction, using sodium borohydride, as described for the reduction of ubiquinone to ubiquinol [2].
Fig. 1. Small angle (A, C and E) and wide angle (B, D and F) diffraction profiles of DPPC/idebenol mixtures. Molar ratios and temperatures are shown next to the traces.
3.2. X-ray diffraction

Samples for X-ray diffraction were prepared by dissolving 15 mg of DPPC/DPPCd_{62} (2:1 M ratio) and idebenone or idebenol to give the desired ratio, in chloroform/methanol (2:1), evaporating the organic solvent under a stream of nitrogen and leaving the samples under vacuum for 3 hours and the appropriate amount of idebenone or idebenol in chloroform/methanol (2:1). Multilamellar vesicles were formed by hydrating the samples in 0.5 mL of 100 mM NaCl, 25 mM Hepes pH 7.4 buffer.

Table 1
Repeat spacings (d, in nm) in the low-angle and wide angle regions of pure DPPC and DPPC/idebenone mixtures, at the molar ratios indicated, obtained after X-ray diffraction.

| Mixture                  | Temperature (°C) | Low-angle |        | Wide angle |        |
|--------------------------|------------------|-----------|--------|------------|--------|
|                          |                  | h, k      | d (nm) |            | d (nm) |
| DPPC                     | 52               | 1.0       | 67.29  | 3.0        | 33.66  |
|                          |                  | 2.0       | 74.90  | 4.0        | 39.13  |
|                          | 38               | 1.0       | 65.62  | 4.20–4.10  | 32.70  |
|                          |                  | 2.0       | 39.13  |            | 21.73  |
|                          | 28               | 1.0       | 45.38  |            | 16.34  |
|                          |                  | 2.0       | 32.70  |            | 13.15  |
|                          | 3.0               | 4.0       | 16.29  |            | 8.10   |
|                          | 5.0               |           | 21.55  |            | 8.10   |
|                          | 18               | 1.0       | 76.34  | 4.20–4.10  | 42.70  |
|                          |                  | 2.0       | 33.56  |            | 24.09  |
|                          |                  | 3.0       | 21.55  |            | 15.92  |
|                          |                  | 4.0       | 16.29  |            | 10.89  |
|                          |                  | 5.0       | 13.10  |            | 8.10   |
| DPPC/Idebenone 15:1)     | 52               | 1.0       | 75.62–68.45 | 3.0  | 34.36 |
|                          |                  | 2.0       | 75.62  | 4.18       | 37.83  |
|                          | 38               | 1.0       | 75.62  | 4.18       | 37.83  |
|                          |                  | 2.0       | 37.83  |            | 21.73  |
|                          | 28               | 1.0       | 76.36  | 4.15       | 42.70  |
|                          |                  | 2.0       | 32.70  |            | 24.09  |
|                          |                  | 3.0       | 21.55  |            | 16.34  |
|                          | 18               | 1.0       | 76.34  | 4.14       | 42.93  |
|                          |                  | 2.0       | 33.56  |            | 24.09  |
|                          |                  | 3.0       | 21.55  |            | 16.34  |
| DPPC/Idebenone (10:1)    | 52               | 1.0       | 75.62–67.86 | 3.0  | 34.36 |
|                          |                  | 2.0       | 75.62  | 4.19       | 37.83  |
|                          | 38               | 1.0       | 75.62  | 4.18       | 37.83  |
|                          |                  | 2.0       | 37.83  |            | 21.73  |
|                          | 28               | 1.0       | 76.34  | 4.14       | 42.70  |
|                          |                  | 2.0       | 42.70  |            | 24.24  |
|                          |                  | 3.0       | 24.24  |            | 16.34  |
|                          | 18               | 1.0       | 76.34  | 4.14       | 42.25  |
|                          |                  | 2.0       | 42.25  |            | 24.24  |
|                          |                  | 3.0       | 23.95  |            | 16.34  |
| DPPC/Idebenone (5:1)     | 52               | 1.0       | 75.62–69.65 | 3.0  | 34.36 |
|                          |                  | 2.0       | 75.62  | 4.19       | 37.83  |
|                          | 38               | 1.0       | 74.90  | 4.15–3.92  | 37.83  |
|                          |                  | 2.0       | 37.83  |            | 21.73  |
|                          | 28               | 1.0       | 80.20  | 4.15–3.92  | 45.38  |
|                          |                  | 2.0       | 45.38  |            | 24.17  |
|                          |                  | 3.0       | 24.17  |            | 16.34  |
|                          | 18               | 1.0       | 85.37  | 4.15       | 46.72  |
|                          |                  | 2.0       | 46.72  |            | 24.24  |
|                          |                  | 3.0       | 24.24  |            | 16.34  |
by extensive vortexing, followed by centrifugation at 13,000g; the pellets were collected. Simultaneous small (SAXD) and wide (WAXD) angle X-ray diffraction measurements were carried out using a modified Kratky compact camera (MBraun-Graz-Optical Systems, Graz Austria), incorporating two coupled linear position sensitive detectors (PSD, MBraun, Garching, Germany) to monitor the s-ranges \[ s = 2 \sin \theta / \lambda, \quad 2\theta = \text{scattering angle}, \quad \lambda = 1.54 \text{Å} \] between 0.0075-0.07 and 0.20–0.29 Å⁻¹, respectively. Nickel-filtered Cu KR X-rays were generated by a Philips PW3830 X-ray generator operating at 50 kV and 30 mA. The detector position was calibrated using Ag-stearate (small-angle region, D-spacing at 48.8 Å) and lupolen (wide angle region, D-spacing at 4.12 Å) as reference materials. Sample pellets were placed in a steel holder with cellophane windows, which provided good thermal contact with the Peltier heating unit. X-ray diffraction profiles were obtained for 10 min exposure times after 10 min of temperature equilibration. (Fig. 1; Tables 1 and 2) Background corrected SAXS data were analyzed using the program GAP (global analysis program) written by Georg Pabst and obtained from the author [3,4]. This program allowed to retrieve the membrane thickness, from a full q-range analysis of the SAXS patterns [5]. The parameters \( z_H \) and \( \sigma_H \) are the position and width, respectively, of the Gaussian used to describe the electron-dense headgroup regions within the electron density model (Fig. 2).

### 3.3. ²H-NMR measurements

Samples were prepared as described above for X-ray diffraction. Sample pellets were dispersed in 300 μl of buffer in deuterium-depleted water and transferred to NMR glass tubes. ²H-NMR

| Mixture                  | Temperature (°C) | Low-angle | Wide angle |
|--------------------------|------------------|-----------|------------|
|                          |                  | h, k      | d (nm)     | d (nm)     |
| DPPC/idebenol (15:1)     | 52               | 1.0       | 76.36-67.86|            |
|                          |                  | 2.0       | 34.10      |            |
|                          | 38               | 1.0       | 76.34      | 4.18       |
|                          |                  | 2.0       | 39.56      |            |
|                          | 28               | 1.0       | 76.34      | 4.15       |
|                          |                  | 2.0       | 41.80      |            |
|                          |                  | 3.0       | 23.32      |            |
|                          | 18               | 1.0       | 76.34      | 4.16       |
|                          |                  | 2.0       | 41.80      |            |
|                          |                  | 3.0       | 23.32      |            |
| DPPC/idebenol (10:1)     | 52               | 1.0       | 67.29      |            |
|                          |                  | 2.0       | 33.71      |            |
|                          | 38               | 1.0       | 74.20      | 4.19       |
|                          |                  | 2.0       | 42.47      |            |
|                          |                  | 3.0       | 23.27      |            |
|                          |                  | 4.0       | 15.73      |            |
|                          | 28               | 1.0       | 76.34      | 4.15       |
|                          |                  | 2.0       | 23.27      |            |
|                          |                  | 3.0       | 15.73      |            |
|                          | 18               | 1.0       | 77.08      | 4.15       |
|                          |                  | 2.0       | 23.27      |            |
|                          |                  | 3.0       | 15.73      |            |
| DPPC/idebenol 5:1        | 52               | 1.0       | 73.52      |            |
|                          |                  | 2.0       | 36.61      |            |
|                          | 38               | 1.0       | 77.08      |            |
|                          |                  | 2.0       | 38.56      |            |
|                          | 28               | 1.0       | 82.70      | 4.15       |
|                          |                  | 2.0       | 40.94      |            |
|                          | 18               | 1.0       | 83.57      | 4.15       |
|                          |                  | 2.0       | 43.88      |            |
experiments were carried out on a Bruker Avance 600 instrument (Bruker, Etlingen, Germany) at 92.123 MHz using the standard quadrupole echo sequence [6]. The spectral width was 150 KHz, with a 10 µs 90° pulse, 40 µs pulse spacing, 3.35 µs dwell time, 1 s recycling time and 50 Hz line broadening, with an accumulation of 15000 transients. Spectra were acquired at temperatures ranging from 18 °C to 52 °C, raising the temperature in 2 °C steps (Fig. 3).

Spectra were dePaked by numerical deconvolution with the software supplied by Bruker, Amix-tools versión 3.5.5, using the Thikonov regularization method. The dePaked spectra correspond to the spectra that would be obtained from a planar membrane with its bilayer normal aligned parallel to the applied static magnetic field, thus enhancing resolution and facilitating analysis of individual spectral peaks [6–9]. These spectra were compared with the original ones to ensure that the relevant features were maintained during the dePaking process.

3.4. 31P-NMR measurements

The same samples and the same spectrometer operating at 242.9 MHz were also used to collect static 31P-NMR spectra. All spectra were obtained in the presence of a gated broad band proton decoupling (5 W input power during acquisition time), and accumulated free inductive decays were
obtained from up to 8000 scans. A spectral width of 48,536 Hz, a memory of 48,536 data points, a 2 s interpulse time, and a 90° radio frequency pulse (11 μs) were used with inverse gated decoupling $^1$H. Prior to Fourier transformation, an exponential multiplication was applied, resulting in a 100 Hz line broadening (Fig. 4).

**Fig. 3.** $^2$H-NMR spectra of pure DPPC and DPPC/idebenone and DPPC/idebenol mixtures. Molar ratios and temperatures are indicated. In all the cases equimolar mixtures DPPC and DPPC-d$_{62}$ are used.
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Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at http://dx.doi.org/10.1016/j.dib.2016.03.064.
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