SUPPORTING INFORMATION

Peptide-Phospholipid complex formation at liquid-liquid interfaces

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The calculated charge values for AngIII and LeuEnk as a function of pH are presented. Details concerning the BESI source as well as the mass spectrum of DPPC analyzed with the commercial ESI source and the BESI and the mass spectra of DPPC-Lys and DPPC-FF are shown with some comments

Dual channel microchip

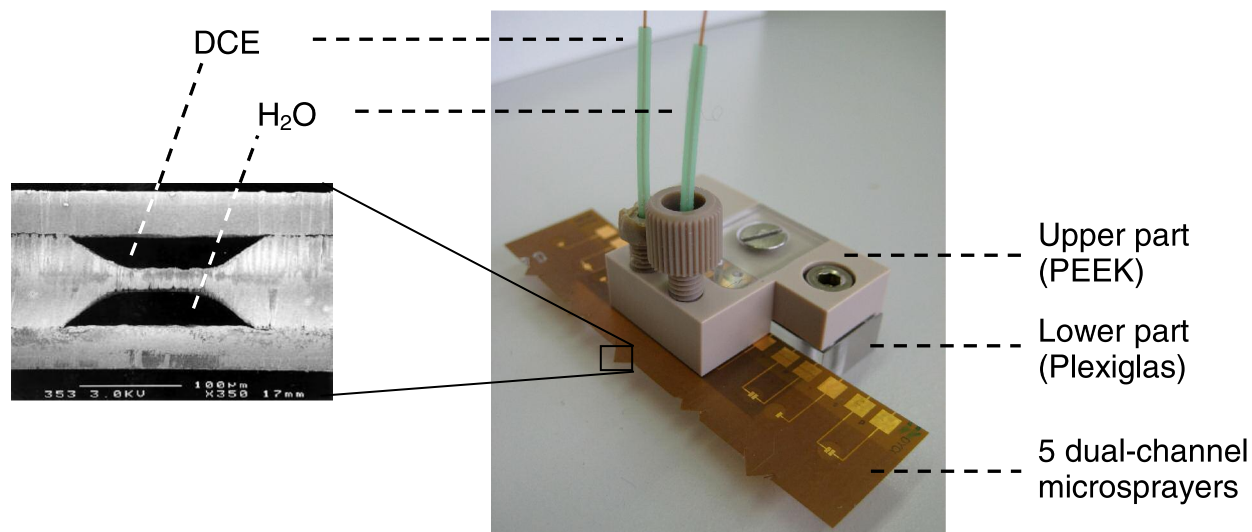

Figure S-1. Right: bar of 5 dual-channel microsprayers mounted inside a holder. Left: scanning electron microscopy picture of a cross section of a microchip tip (larger microchannels are used as example).

Calculated charge Vs. pH curves for AngIII and LeuEnk

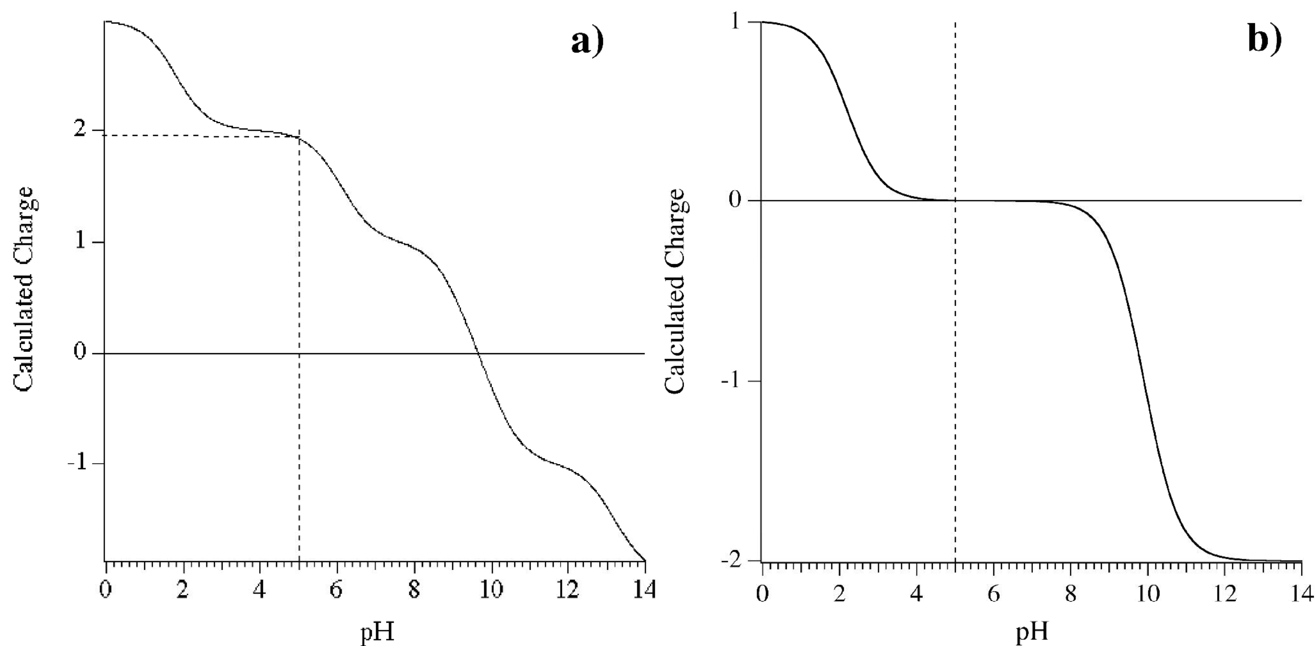

Figure S-2. Calculated charge values as a function of pH for a) AngIII and b) LeuEnk, respectively. The dashed line demarks the pH value of 5.

Influence of DPPC concentration

The influence of the DPPC concentration was tested with the commercial ESI source and the BESI source (see Figure S-3). The empty markers stand for the experiments carried out with the commercial ESI source and the solid markers for those carried out with the dual microsprayer, where the aqueous channel was filled with acidified water. In the first case, the abundance obtained for 2DPPC (square markers) increases until reaching a constant value at concentrations beyond 50 µM. The recorded abundance of the monomeric form (triangle markers) is weaker and does not really increase over the whole concentration interval tested. In the second case, the abundance of 2DPPC always increases and that of DPPC increases until 100 µM and then it goes down. In addition, the dimer becomes more intense than the monomer at 100 µM. At higher concentrations, the formation of oligomers is favored. In the case of the commercial ESI source where the dimer is the most intense peak, this effect can be explained by the presence of only one phase and the absence of acid (pure DCE), which could enhance the formation of non-covalent complexes.

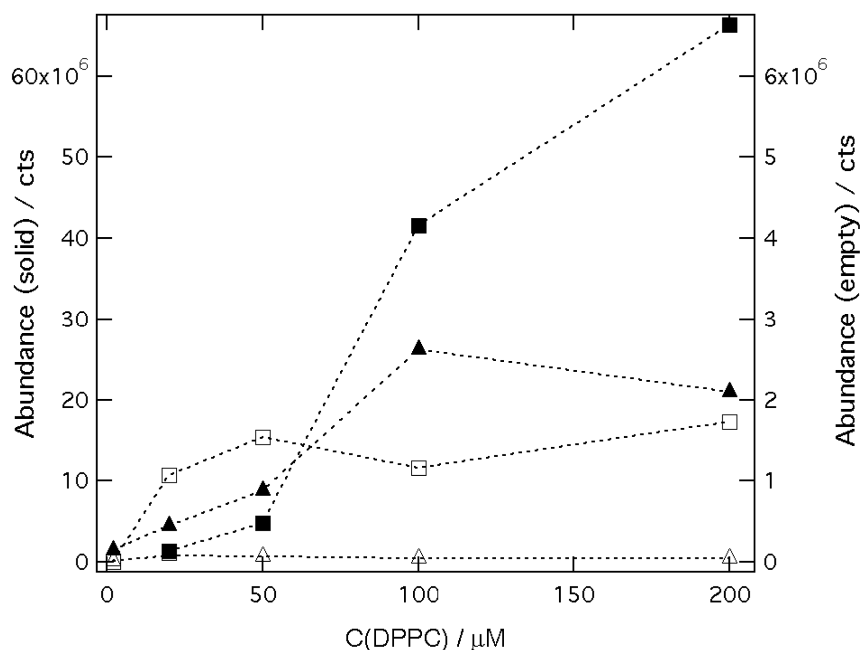

Figure S-3. Relative abundances of DPPC and 2DPPC as a function of the phospholipid concentration. Empty and black markers correspond to the experiments carried out with the commercial ESI source in pure DCE, and with a dual-channel microsprayer where one line was filled with AcOH 1%. All the spectra were obtained using the ion trap mass spectrometer. Triangle and square markers stand for DPPC and 2DPPC, respectively. Mass spectra were optimized at the phospholipid dimer *m/z* value.

Amino-acids/dipeptide-phospholipids interactions

The interactions between phospholipids and amino acids or a dipeptide have been tested with two polar amino acids, one basic (Lys) and one acidic (Glu) and a hydrophobic dipeptide (FF). These experiments were carried out with the dual microsprayers, with a temperature of the heated capillary fixed at 200°C. Thus, the reactions were conducted at the liquid-liquid interface formed between the aqueous solution containing the amino acid and the DPPC organic solution. In general, from the obtained mass spectra it is always possible to observe the peaks of both reactants (except for Glu). However, the complexes between DPPC and the amino acids or the dipeptide are hardly observed, unless the heated capillary temperature be decreased to 100°C, as shown in Figure S-4. It should be mentioned that the spray current keeps constant without changing any parameter during these variations of temperature, as also reported by Page et al.[55]

As expected, the mass spectrum obtained for Glu in the aqueous phase shows only the presence of the phospholipids (data not shown). In the case of Lys (Figure S-4a), only a non-covalent complex (DPPC-Lys) is formed at *m/z* = 880.2 Th and no other complexes, such as 2DPPC-Lys, were observed. The relative abundance was around 13% and decreased drastically when increasing the temperature (see square markers in Figure 7a). Moreover, it was difficult to acquire tandem mass spectra according to the binding weakness of the complex. Thus, the selection of the ion was only possible at low collision energies (less than 20%) and the signal was lost beyond 25%. For the dipeptide (FF), the formation of a

non-covalent complex (DPPC-FF) at *m/z* = 1046.0 Th was obtained. As before, this complex could not be observed when increasing the temperature beyond 140°C (diamond markers in Figure 7a). Although the tandem mass spectra exhibits a weak peak at *m/z* = 313 Th (FF), no relevant information can be extracted from it due to the poor quality, like in the case of DPPC-Lys. In addition, the less intense peak at *m/z* = 1780.1 Th also decreases with the temperature. This behavior fits well to that observed for the noncovalent complexes so far. Thus, it can be attributed to the existence of the doubly charged complex $[4DPPC\text{-}2FF + 2H]^{2+}$.

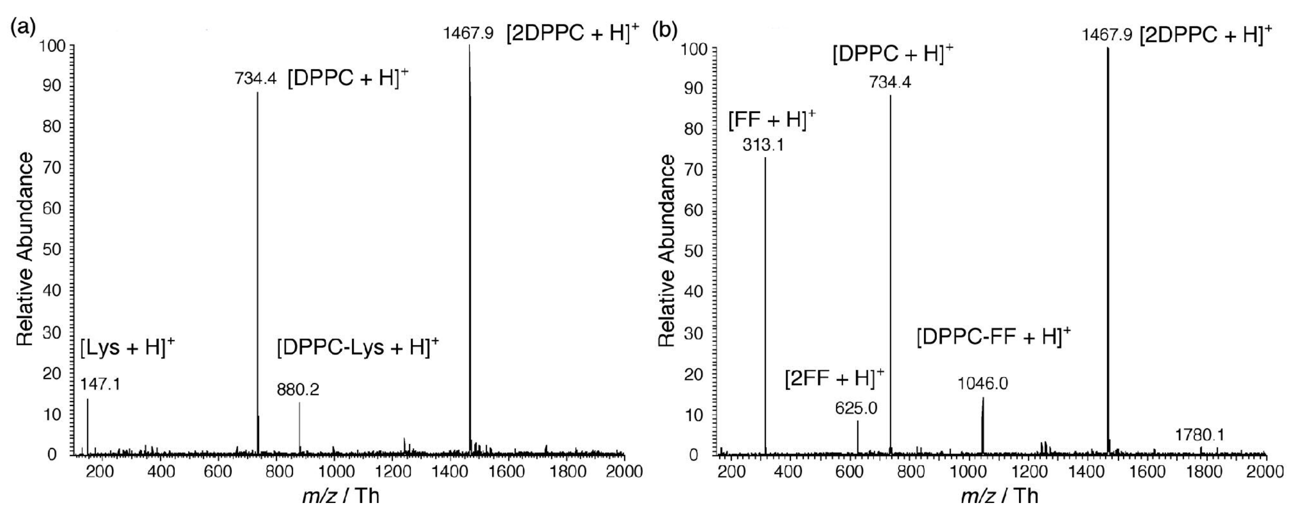

Figure S-4. Mass spectra of the reaction between DPPC 200 µM in DCE and (a) Lys and (b) FF at 200 µM in acidified water (1% acetic acid) obtained with a BESI source coupled to the ion trap mass spectrometer. The temperature of the heated capillary was fixed at 100°C.