The Natural Antioxidant Quercetin Induces Changes in Membrane Electrical Parameters and Fluidity

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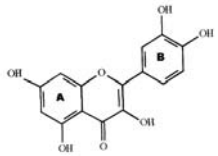


Fig. 1 Quercetin is a flavonol with remarcable properties in its capacity to behave as an antioxidant, inactivating the free radicals of oxygen. At the level of cell membrane it has the capacity to prevent the lipid peroxidation induced by the free radicals.

Abstract. The effects of the flavonoid quercetin, well known for its strongly antioxidant properties, on lipid artificial and natural bilayers are studied by means of electrophysiological and spectrofluorimetrical methods. Thus, we are measuring the variations of the electrical parameters (conductance, capacitance) of the artificial membranes in the presence or absence of some lyotropic anions, in the presence of quercetin inserted in the lipid bilayers, by using the BLM (Black Lipid Membrane) method. Our results show that quercetin inserts itself into lipid membranes, modifying their properties. On adding lyotropic anions the conductance of artificial lipid membranes increases considerably, depending on the type of the anion and on its concentration.

BLM measurements

- for small quercetin concentrations the conductance of the bilayer increases drastically for kosmotropic acetate and chaotropic nitrate (fig. 3 a, b), suggesting the formation of pores which results in the permeabilization of the membrane; a similar effect was noticed for cations (Jorgensen, 1991, 1993)
- the capacitance of the bilayer increases with quercetin concentration and remains constant in the presence of lyotropic anions and quercetin suggesting that quercetin and liotropic anions have compensatory effects

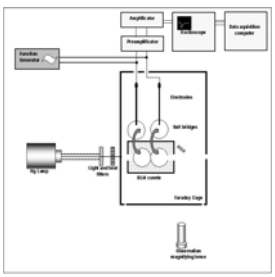


Fig. 2 The BLM set-up.

Fluorescence anisotropy measurements

Fluorophores:

DPH (1,6-diphenyl-1,3,5-hexatriene)

TMA-DPH (1-[(4-trimethyl-amino)phenyl]-6-phenyl-1,3,5-hexatriene)

Steady-state fluorescence anisotropy :

$$r = \frac{I_{VV} - GI_{VH}}{I_{VV} + 2GI_{VH}}$$

where

$$G = \frac{I_{HV}}{I_{HH}}$$

I_{VV}, I_{VH}, I_{HV} and I_{HH} are the fluorescence intensit components; *VV, VH, HV, HH* stand for vertical/horizontal positions of the excitation / emission polarizers respectively

Lipid order parameter

$$S = \sqrt{\frac{r_\infty}{r_0}}$$

where the limiting initial values of the fluorescence anisotropy:

$r_0^{DPH} = 0.362$ $\quad r_0^{TMA\text{-}DPH} = 0.362$

and the limiting long-time values of the fluorescence anisotropy:

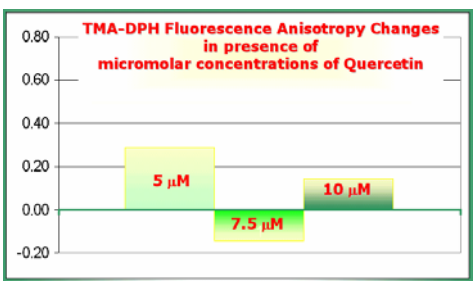
$r_\infty = 1.270\, r - 0.076$ for $0.13 < r < 0.28$

$r_\infty = 1.100\, r - 0.032$ for $0.28 < r < 0.34$

(formula based on experimental data published by Blitterswijk, Van Hoeven, Van der Meer, 1981)

Membrane fluidity

in the bilayer polar headgroup region	in the core of the membrane lipid bilayer
$\text{Fluidity} = \frac{1}{S^{TMA\text{-}DPH}}$	$\text{Fluidity} = \frac{1}{S^{DPH}}$



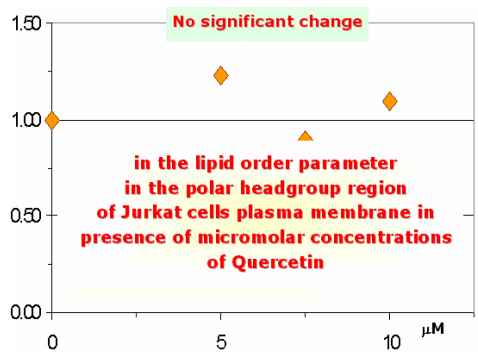
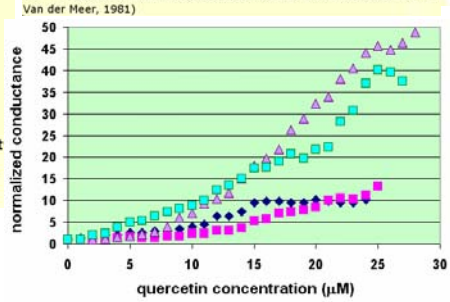
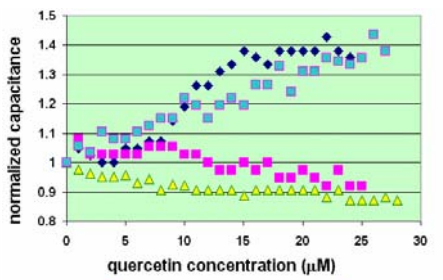
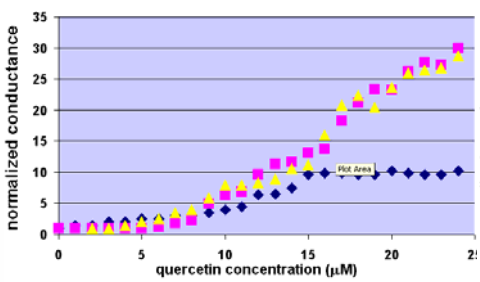


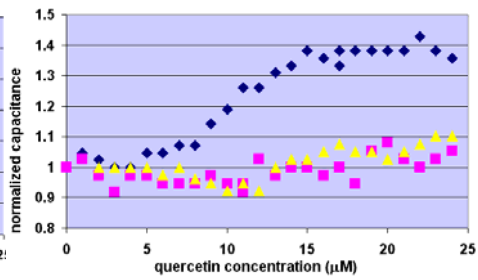
Fig. 5 Membrane fluidity in the presence of quercetin





◆0 mM Na nitrate ■16 mM nitrate △52 mM nitrate ■192 mM nitrate ◆0 mM nitrate ■16 mM nitrate △52 mM nitrate ■192 mM nitrate





◆0 mM Na acetate ■16 mM Na acetate △52 mM Na acetate ◆0 mM Na acetate ■16 mM Na acetate △52 mM Na acetate

Fig. 3 Normalized conductance of the bilayer versus quercetin concentration in the presence of various a) Na nitrate and b) Na acetate concentration added on both sides of the artificial membrane

Fig. 4 Normalized capacitance of the bilayer versus quercetin concentration in the presence of various a) Na nitrate and b) Na acetate concentration added on both sides of the artificial membrane

References

- Jorgensen et *al.*, **BBA**, 1062, 227-238 (1991)
- Jorgensen et *al.*, **Chem. Phys. Lipids**, 65, 205-216 (1993)
- Movileanu et *al.*, **Int. J. Pharm.**, 205, 135-146 (2000)
- Shirai et *al.*, **J. Agric. Food Chem.**, 49, 5602-8 (2001)

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