Supporting Information

Nutritional aspects of *β*-carotene and resveratrol antioxidant synergism in giant unilamellar vesicles

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Extraction and separation of chlorophyll *a* (Chl*a*)

Acetone, dichloromethane (DCM), *n*-hexane, sodium chloride and anhydrous magnesium sulfate (all AR) were purchased from Beijing Chemical Works (Beijing, China). Quartz sand was purchased from Tianjin Fuchen Chemical Reagent Factory (Tianjin, China). Silica gel (200–300 mesh) were purchased from Qingdao Haiyang Chemical Branch Works (Qingdao, China). HPLC grade methanol was purchased from J&K Scientific Ltd. (Beijing, China). All operations were carried out under reduced light in order to minimize photodegradation.

Fresh spinach leaves (~500 g) were pulverized and extracted with 200 mL of acetone and then transferred to a separatory funnel. Excess water and acetone were removed by addition of saturated NaCl solution. DCM was then added to concentrate chlorophylls for 3 times until the acetone phase was nearly colorless. Anhydrous $MgSO_4$ was added to the DCM solution of chlorophylls to remove trace water. The crude extraction was then filtered and rotary-evaporated.

The chromatographic column (2.5 cm inner diameter, 30 cm length) was mounted using wet method. Crude chlorophylls re-dissolved with DCM were added to the top of the column. The eluent was initially pure DCM and the polarity was gradually increased by increasing the volume ratio of acetone. Chl*a* solutions were collected and rotary-evaporated.

HPLC separation of Chl*a* was based on ref (S1) using HP1100 HPLC System (Agilent Technologies, Inc., Santa Clara, CA, USA). Pure HPLC grade methanol was used as the eluent and the flow rate was 1 mL/min, and Chl*a* was detected at 450 nm. The HPLC spectrum was shown in Figure S1, and the Chl*a* component was >95% determined by peak integral.

Figure S1. HPLC spectrum for Chlorophylls separation

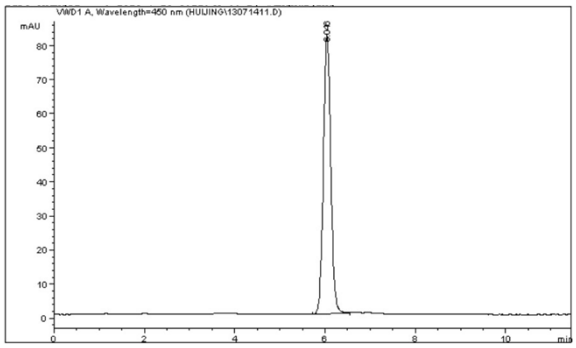


Figure S2. Inverted fluorescence microscopy experimental setup

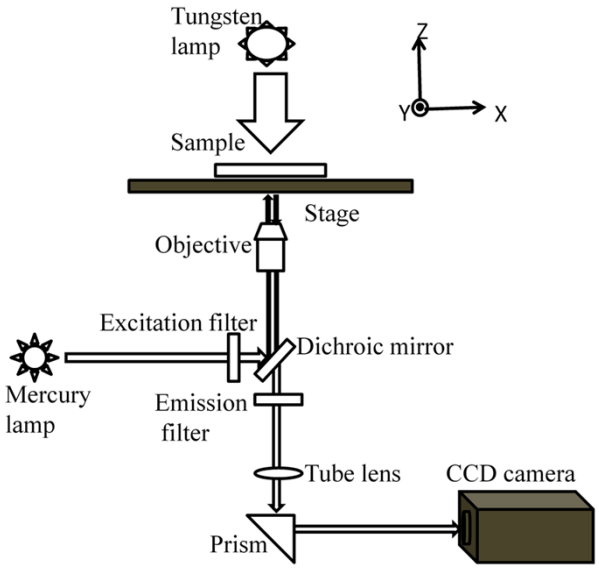


Figure S3. Images of blank GUV sample exposed to light (400−440 nm, 66 mW/mm^2) for up to 60 min.

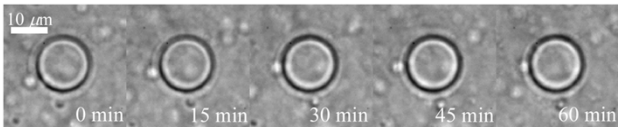
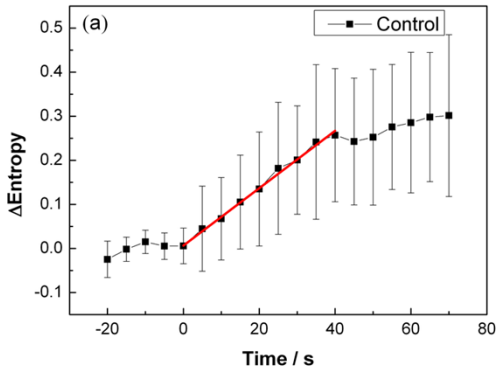
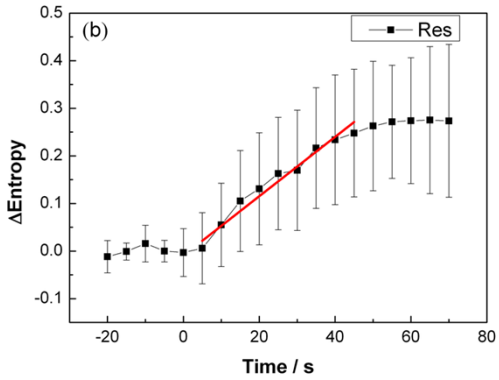
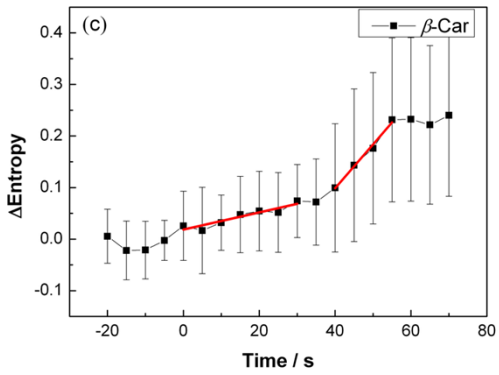
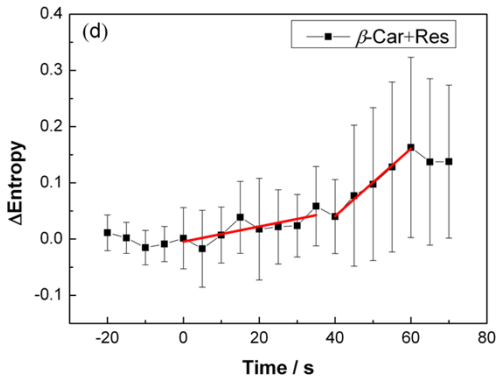


Figure S4. Δ*E* as a dimensionless scalar measurement of heterogeneity for GUVs fluorescently labeled by Chl*a* for up to 80 s. Statistics: mean±SD, *n*=15.









Reference

(S1) Y. Shioi, R. Fukae and T. Sasa, Chlorophyll analysis by high performance liquid chromatography, *Biochimica et Biophysica Acta – Bioenergetics*, 1983, **722**, 72–79.