The passive ascorbate transport across DPPC membranes

Hanspeter Strittmatter, Helmut Sapper, Hans-Helmut Paul, Wolfgang Lohmann
Institut für Biophysik, Strahlenzentrum der Justus Liebig Universität, Leihgesterner Weg 217, 6300 Gießen

Differences in vitamin C activity of several analogues of ascorbic acid have often been ascribed to differences in their uptake mechanism. As the membrane permeation has been discussed in this context as an essential point for their efficiacy, nuclear magnetic resonance (NMR)spectroscopy has been applied for determining the passive transport of ascorbate and isoascorbate through lipid bilayers.

The pH-dependence as well as local differences in the magnetic susceptibility are used to differentiate between distinct NMR signals of the ascorbate under investigation. The substance is partially entrapped in DPPC vesicles (DPPC: L-α-dipalmitoyl-phosphatidylcholine) caused by ultrasonification and ultra-filtration. Lowering the outside pH and concentration causes an efflux of ascorbate resulting in an increase in NMR intensities of external ascorbate.

The balancing of a pH gradient induced across the membranes demonstrates that the vitamin C molecules permeate as mono-anions. This is also confirmed by the fact that their transport is considerably delayed if the outside pH-value is below the pK-value of ascorbate (pK=4.1). The temperature dependence observed in the range from 20 to 50^{o}C indicates the fluidity of the membranes as an other decisive quantity. The permeation rate is most enhanced in the range between the pre- and main phase transition temperature known for DPPC membranes. Configurational factors seem to be also important since iso-ascorbate is less permeable (by about a factor 3 at 30^{o}C) than ascorbate. This fact may explain the lower vitamin C activity of this stereoisomer as well as of other ascorbic acid analogues with respect to their physiological meaning.