Membrane and ion transport

001(1aB01)

Analysis of fatty acids and sterols of plasma membrane and tonoplast isolated from salt-stress adaptation of Tobacco cultured cells in suspension

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In order to obtain knowledge of relation between membrane lipids and adaptation mechanisms for salt-stress, we analyzed fatty acid and sterol composition of plasma membrane and tonoplst isolated from Tobacco cells cultured in suspension under conditions of 100 mM and 200 mM saline (adapted cells) and unadapted cells.

There was no significant difference in fatty acid compositions of plasma membrane from cells grown under various salinity. However, there was a significant change in sterol compositions, that is, sitosterol contents decreased with increase in salinity. Moreover, the ratio of more planner/less planner reducted with higher salinity.

Both fatty acid and sterol compositions in tonoplast were not affected the salinity of growth medium.

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ASYMMETRIC STRUCTURE OF MUNG BEAN PLASMA MEMBRANE

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Asymmetric distribution of membrane-lipids and -proteins is essential for membrane functions to play important roles in many cellular events.

Last time, we assayed topography of phospholipids across rightside-out plasma membrane (PM) vesicles obtained by aqueous two-phase partitioning from mung bean (*Vigna radiata* L.) hypocotyles utilizing porcine phospholipase A_2. And it was discovered around 40% of PM phospholipids were inaccessible to the phospholipase A_2 (63rd annual meeting of the Bot. Soc. of Japan, AKITA).

Thereafter, we found that most part of this inaccessible pool was to be located in the inner leaflet by ^{31}P-NMR experiments using a shift reagent, $PrCl_3$.

It is considered the accessibility of the phospholipase A_2 to the substrates might be largely limited by solid structure of peripheral polypeptides of membrane-proteins. Therefore, quantitative asymmetry of peripheral polypeptides could exist in mung bean PM. To elucidate the possibility, the PM vesicles were subjected to trypsin treatment. Consequently, membrane-proteins of sealed rightside-out PM vesicles were hardly hydrolyzed by trypsin, whereas further addition of 0.025% Triton X100 caused the hydrolysis of 30-40% of the proteins. It was suggested there are only small amount of peripheral polypeptides on exofacial leaflet of PM compared with on cytofacial leaflet.

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SOLUBILITIES OF $CaCO_3$ AND $CaHPO_4$ IN PURE DEIONIZED WATER AND 100 mM KCl SOLUTION

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A main chemical compound of calcium bands on Chara cell walls has been reported to be $CaCO_3$. However, $CaHPO_4$ has been identified to coexist in calcium bands by us. When isolated Chara cell walls are immersed in pure deionized water or 100 mM KCl solution, Ca^{2+} releases into them and makes their pH neutral or weakly alkaline.

In the present study, solubilities of $CaCO_3$ and $CaHPO_4$ in pure deionized water and 100 mM KCl solution have been measured using a Ca^{2+}-electrode. Their pH values have also been measured.

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THE DOUBLE-WATER-FILM ELECTRODE MEASURES THE ELECTRICAL PROPERTIES ACROSS THE INTERNODE/NODE -INTERFACE OF *CHARA* AS A FUNCTION OF TIME :

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By scanning the double-water-film electrode, described in detail elsewhere(Ogata, 1998ab1999ab) along the length of two Characean internodes(A&B) joined by a node- complex(N), the specific parallel resistance(Rm) of 30±5 $X10^{-3}\Omega m^2$corresponds to the resistive component of plasmodesmata, the specific parallel capacitance(Cm)of $1.5\pm0.25X10^{-1}Fm^{-2}$ (at 30Hz) corresponds to the capacitive component of plasmalemma and the specific series resistance(Rs) corresponds to the total resistive components including the cell-sap, tonoplast and cytoplasm in series, of $9 \pm 0.1X10^{-3}\Omega m^2$ were estimated at 20 ℃, across the (A or B)/(N) -interface of *Chara*. And these parameters at the vacuole were $8 X10^{-3}\Omega m^2$. With a changing in temperature or turgor-pressure; Rm decreased and Cm increased with a raising temperature or a lowering turgor-pressure. Rm at the vacuole, however, showed an inphasic response with the temperature change. Assuming that the effective size of plasmodesma was a $6x10^{-8}m$ in diameter and a $2x10^{-6}m$ in length, the effective number of plasmodesma per interface (A/N or N/B) calculated was 10^4. This number confirmed, in order, the observations with electron-micrographs demonstrated by Spanswick & Costerton (1967).