

### The effects of saponins on the transporting epithelium of isolated frog skin

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The stimulus for this work was the observation that saponins (0.05–1.0 mg/ml) applied to the outer surface of frog skin (*Rana temporaria*) caused a precipitous fall in trans-epithelial potential (PD) with only a minor effect on sodium transport as measured by short circuit current (SCC). The actions of saponins differ from those of antidiuretic hormone, which cause both SCC and PD to increase, and from those of theophylline, which cause SCC to increase and PD to fall. In the presence of saponins and in spite of low PD values antidiuretic hormone was still able to activate the transport mechanism and increase SCC and PD.

The effects of saponins were associated with a fall in DC resistance, both in the absence (sodium replaced by choline) and presence of sodium. Sodium and chloride diffusion SCCs in skins which were not actively transporting were increased by saponins. Experiments with  $^{22}\text{Na}$  showed that saponins increased both the inward and outward sodium fluxes, although on one occasion the PD fell while the SCC and forward flux remained constant.

When skins were exposed on the outside to  $^{14}\text{C}$ -inulin the penetrable space was only  $1.3 \pm 0.2\%$  (mean  $\pm$  S.E.M.; three experiments), whereas if saponins were also present on the outside the penetrable space increased to  $12.2 \pm 2.3\%$  (three experiments). These values were significantly different ( $P < 0.01$ ). When inulin space was determined in the conventional manner it was  $36.7 \pm 1.8\%$  (six experiments) and in the presence of saponins it increased to  $44.1 \pm 1.7\%$  (six experiments). These values were also significantly different ( $P < 0.05$ ). The increase in inulin space caused by saponins is probably an underestimate since some inulin remained in the saponin-treated skins, probably by resealing of the cells when they were leached in distilled water. In two experiments saponins caused an increase of 26.7% in the inulin space when allowance was made for the inulin left in the tissue.

These findings show that inulin, and hence ions, can penetrate into and perhaps between the cells when saponins are added to the outer bathing fluid. This effect is not surprising considering the well known lytic actions of saponins (Dourmashkin, Dougherty & Harris, 1962). Thus the sodium-selective diffusion barrier at the outer skin surface is short-circuited and the PD falls. The classical model of a sodium transporting epithelium is an outer rate-limiting diffusion barrier and an inward facing unsaturated ion pump (Koefoed-Johnson & Ussing, 1958) predicting that the rate of sodium transport is controlled by the permeability of the outer surface to sodium. Yet the increase in permeability induced by saponin never caused an increase in SCC, implying that ions were not entering the transporting pool.

#### REFERENCES

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### Platelet aggregation by very low intensity acoustical energy and its inhibition by drugs

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A new technique was developed in which the aggregation of blood platelets in