The effect of catecholamines and β-anti-adrenergic drugs on isolated short-circuited skin of the frog

SIR,—Adrenaline (Koefoed-Johnsen, Levi & Ussing, 1952) and isoprenaline (Santi, Ferrari & others, 1967) increase the short-circuit current in frog isolated skin.

The activity of catecholamines on this preparation was compared by determining their dose-effect curves. The effect of these drugs was evident at surprisingly low concentrations (Fig. 1). Isoprenaline action started at $0{\cdot}5 \times 10^{-9}$M, while noradrenaline and adrenaline activity began from 10^{-8} to 10^{-7}M. The order of affinity* was then the following: isoprenaline $\geqslant$ noradrenaline $>$ adrenaline. The intrinsic activity* was the same for the three catecholamines. Auto-inhibition, developing for the higher doses of catecholamines, follows an opposite order of intensity, being the greatest for adrenaline and least for isoprenaline (Fig. 1). The skin potential variations were similar to those of the short-circuit current.

The antiadrenergic drugs Kö 592 [(3-methylphenoxy)-β-hydroxy-α-isopropylaminopropanol] (Engelhardt, 1965) and D(−)-INPEA (*N*-isopropyl *p*-nitrophenylethanolamine) (Somani & Lum, 1965) antagonized competitively the effect of

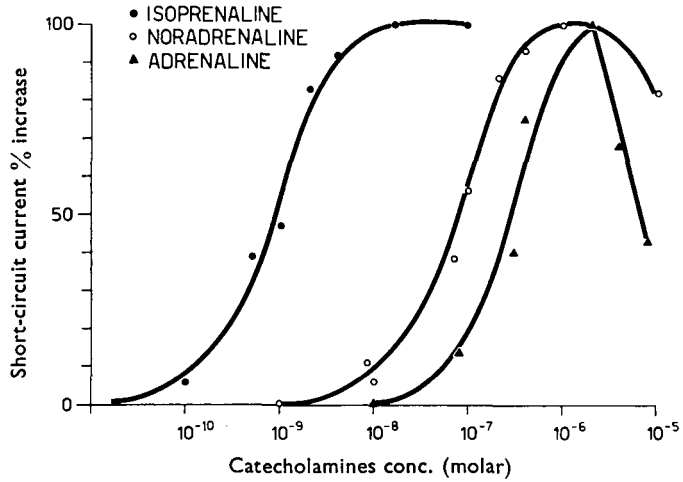

FIG. 1. Log concentration-response curves for the increase of the short-circuit current induced by catecholamines on frog isolated skin. *Abscissa:* molar conc. of isoprenaline, noradrenaline and adrenaline in the solution exposed to the inside of the skin. *Ordinate:* μA % relative increase from control (skin equilibrated for 2 to 3 hr after the dissection and mounting). The short-circuit current maximum increase, induced by isoprenaline 10^{-8}M, by noradrenaline 10^{-6}M and by adrenaline 2×10^{-6}M (263 ± 37 μA) was taken as 100% of the effect. The maximum increase was reached in 10 to 30 min. Each point represents the mean of 4 to 6 experiments. Abdominal skin of *Rana esculenta* (surface 7·06 cm²) was clamped between two lucite chambers containing frog Ringer solution. Short-circuit current and potential difference across the skin were measured according to the technique of Ussing & Zerahn (1951). Drugs were added directly to the solution contained in the chamber bathing the internal side of the skin. After each dose of catecholamine, the skin was repeatedly washed by changing the Ringer solution in the two chambers, and re-equilibrated (2–3 hr) before adding the next dose. Experiments were carried out in July, August and September, at room temperature.

* According to Ariëns (1954) nomenclature.

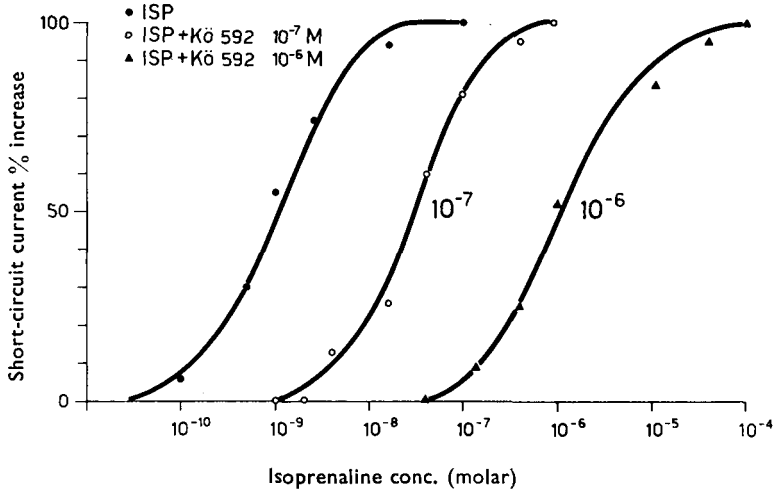

FIG. 2. Log concentration-response curves for the antagonism between isoprenaline and Kö 592 on isolated short-circuited skin of the frog. *Abscissa:* molar conc. of isoprenaline (ISP) in the solution facing the inside of the skin. *Ordinate:* μA % relative increase from control (skin equilibrated for 2 to 3 hr in the presence of frog Ringer solution, with or without addition of the stated conc. of antagonist). Experimental conditions as in Fig. 1. Isoprenaline was added cumulatively in the chamber opposite to the internal side of the skin.

isoprenaline on short-circuit current. Fig. 2 demonstrates the dose-effect curves for the antagonism between isoprenaline and Kö 592. The presence of Kö 592 alone, did not alter, at the lower concentrations (10^{-8} and 10^{-7}M), the skin potential or the resting short-circuit current; at 10^{-6}M it decreased these values.

Whether the action of catecholamines on frog skin should be regarded as one affecting permeability, sodium pump, or other functions of this organ cannot be stated, very little information being available. According to Koefoed-Johnsen & others (1952), adrenaline induces in the frog skin a transitory increase of the short-circuit current by causing an unusual active transport of Cl^- outward, probably as a result of the action on the mucous glands.

However, the action of catecholamines on isolated short-circuited frog skin shows features similar to those characterizing a typical β-tropic function, if the wide difference in the affinities between isoprenaline and noradrenaline and the specific interaction between isoprenaline and β-lytic agents are considered. On the other hand, the unusual affinity sequence isoprenaline > noradrenaline > adrenaline is a typical feature of the adrenergic lipid mobilization *in vitro* (Wenke, Mühlbachová & others, 1964; Rudman, Garcia & others, 1964; Barrett, 1965; Fain, 1967).

Isolated short-circuited frog skin could, thus, represent a useful test for adrenergic and anti-adrenergic drugs.

Institute of Pharmacology,
University of Padua,
Largo E. Meneghetti 2,
I 35100, Padua, Italy.

G. FASSINA
F. CARPENEDO
G. FIANDINI

November 15, 1967

References

Ariëns, E. J. (1954). *Archs int. Pharmacodyn. Thér.*, **99**, 32–49.
Barrett, A. M. (1965). *Br. J. Pharmac. Chemother.*, **25**, 545–556.
Engelhardt, A. (1965). *Arch. exp. Path. Pharmak.*, **250**, 245–246.
Fain, J. N. (1967). *Ann. N.Y. Acad. Sci.*, **139**, 879–890.
Koefoed-Johnsen, V., Levi, H. & Ussing, H. H. (1952). *Acta physiol. scand.*, **25**, 150–163.
Rudman, D., Garcia, L. A., Brown, S. J., Malkin, M. F. & Perl, W. (1964). *J. Lipid Res.*, **5**, 28–37.
Santi, R., Ferrari, M., Tóth, C. E., Contessa, A. R., Fassina, G., Bruni, A. & Luciani, S. (1967). *J. Pharm. Pharmac.*, **19**, 45–51.
Somani, P. & Lum, B. K. B. (1965). *J. Pharmac. exp. Ther.*, **147**, 194–204.
Ussing, H. H. & Zerahn, K. (1951). *Acta physiol. scand.*, **23**, 110–127.
Wenke, M., Mühlbachová, E., Elisová, K., Schusterová, D. & Hynie, S. (1964). *Int. J. Neuropharmac.*, **3**, 283–292.

The penetration temperature of aqueous sodium dodecyl sulphate solutions into solid long-chain alcohols

SIR,—When a surfactant, amphiphile and water are mixed, a spontaneous formation of ternary liquid crystalline phase occurs in one of two ways depending on how the components are mixed (Lawrence, 1959, 1961a, b). If a piece of surfactant-amphiphile mixture is flooded with water, myelinic tubular forms appear. These are strongly birefringent close to the original mixture but the birefringence decreases as the myelins become more elongated and finally the outer part dissolves to form an isotropic solution.

If, however, the solid amphiphile is immersed in surfactant solution, spontaneous formation of ternary mesophase again occurs, but with two differences. Firstly, true myelins are not formed but only tubular myelin-like protuberances. These are deformed spherulites which, unlike true myelins, do not possess a central core of isotropic solution (Lawrence, 1958). Secondly, there is a sharply defined temperature, T_{pen}, below which an isotropic solution occurs very slowly, while at and above T_{pen} penetration of surfactant solution into the amphiphile occurs by formation of ternary liquid crystal phase. The first step is the formation of a film of mesophase around the amphiphile, followed by myelinic-like protuberances, formed as the surfactant solution penetrates into the amphiphile. These extrude into the aqueous solution and then break up into liquid crystal spherulites as they become fluid enough for surface tension to act.

T_{pen} varies with the nature of the hydrophobic and hydrophilic groups of the surfactant and amphiphile. It has been stated that T_{pen} does not vary with concentration of surfactant over wide limits (Lawrence, 1958; Lawrence, Bingham & others, 1964) or that it varies only slightly with concentration (Lawrence, 1961a, b). We have investigated this for aqueous sodium dodecyl sulphate solutions in the range 0·5% to 20·0% w/w using the normal C_{14}, C_{16} and C_{18} alcohols. The purity of the alkyl sulphate and 1-hexadecanol was as given by Barry & Shotton (1967), 1-octadecanol was Fluka purrissimum grade, and the 1-tetradecanol was Fluka purum grade which was purified by preparative gas chromatography. Dry and wet melting points were respectively: for 1-tetradecanol 39·5, 40·0; 1-hexadecanol 49·5, 52·0; 1-octadecanol 58·5, 61·5.

A few mg of an alcohol were melted on a cavity slide and agitated with a needle whilst cooling so as to yield a thin solid layer when cold. The cavity was filled with one of the sodium dodecyl sulphate solutions, a cover slip added and the slide was placed on a Kofler micro hot stage fitted to a polarizing