rat uterus caused by mescaline is due to the release of endogenous mediators, such as serotonin or acetylcholine, that stimulate smooth muscle. Histamine can be ruled out a priori, since it is well known that the rat organs are highly resistant to this amine; this is supported by the lack of effect of tripelennamine. The fact that those drugs causing the most potent inhibition of mescaline-induced contractions were only rarely as active against contractions elicited by serotonin, would seem to eliminate this amine as a potential mediator of the effect of mescaline. Acetylcholine likewise cannot be involved in view of the inactivity of parasympatholytic drugs.

It is difficult to interpret the findings presented above in terms of the possible therapeutic mechanisms of action of the drugs examined, and it is somewhat surprizing that mescaline causes contractions of a smooth-muscle organ such as the uterus. We also have no straightforward explanation for the fact that our findings are somewhat at variance with those of COSTA[3], who did not observe a direct effect of mescaline on the same organ, but only a potentiation of serotonin-induced contractions. On the other hand, it is perhaps relevant that some of the psychoactive drugs examined, e.g. chlorpromazine and amitryptiline, are potent inhibitors of mescaline at peripheral receptors, especially if one assumes that a similar antagonism might take place at mescaline receptors in the central nervous system. Moreover, the unexpected finding that some β-adrenergic blocking agents, notably oxprenolol, antagonize the action of mescaline, could have some bearing on the clinical observations that such drugs in high dosages exert beneficial effects in mania and agitated psychoses[4] or display general psychotropic activities[5]. The capacity of sympathomimetic substances to antagonize mescaline seems to be best explained as being one facet of their non-specific spasmolytic action. In this regard, it may suffice to mention the fact that adrenaline is a highly potent antagonist of contractions induced by trypsin in the rat uterus[2].

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Effect of Acetylcholine on Melanophores of *Rana tigrina*

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Summary. Acetylcholine produced melanin aggregation and blanching of skin colour in *Rana tigrina*, the common Indian frog. The effects were more prolonged in frogs pretreated with an anticholinesterase agent. Acetylcholine effects were not antagonized by either *m*-cholinolytic (atropine) or *n*-cholinolytic (pentolinium) agents, but were markedly inhibited by procaine. The results have been discussed in the light of the well-known membrane-stabilizing effect of procaine.

No phenomenon of nature has probably attracted more attention or has been investigated from more diverse angles than that of colour changes in animals. In amphibians the integumentary colour changes are produced by variations in the skin melanophores, which form a very delicate and responsive system, constantly undergoing changes in response to alterations in the external environment and internal homeostasis[1]. The skin colour of *Rana tigrina*, the common Indian frog, varies from a dirty dark brown to a light yellowish-green colour. These variations in skin colour result from intracellular movement of melanin granules within the melanophores. Dispersion of melanin granules from a perinuclear position out into the melanophore processes, results in darkening of skin colour, while aggregation of melanin granules from the melanophore processes to a perinuclear position causes lightening of skin colour[2].

No reports are available on the effect of acetylcholine, the cholinergic transmitter, on melanophores of *Rana tigrina*. The present investigation reports the effect and possible mode of action of acetylcholine on skin colour and melanophores in this species.

Material and method. Studies were conducted on adult frogs, of either sex, weighing between 150 and 300 g. Frogs were anaesthetized with pentobarbitone sodium (50 mg/kg in ventral lymph sac). Drugs, dissolved in 0.6% saline in a fixed volume, were administered through cannulated left branch of thoracic aorta. The animals were kept submerged throughout the experiment in 0.6% saline for adequate cutaneous respiration. The dorsal skin colour was observed by naked eye and the most lateral web of the left hind limb was observed under low power ($\times 60$) microscope. Individual melanophores were measured with a Leitz micrometer eyepiece by noting the maximum vertical and horizontal diameters. 5 such melanophores were measured and the mean 'melanophore size index' was recorded as the:

$$\frac{\text{maximum vertical} \times \text{horizontal diameter (in } \mu\text{m)}}{100}.$$

Since the cell outline of the melanophores was only distinct when they were in the contracted state, care was taken to measure only the span of the pigment and not the cell boundary. In effect, therefore, the variable recorded is the linear disposition of the pigment in melanophore processes. The melanophore size index was noted before and at different time periods after drug administration and the difference in pre and post drug peak effect was taken into consideration for statistical analysis by Student's *t*-test.

Results and discussion. Results are summarized in the Table. Acetylcholine (0.1 mg/kg) produced centripetal movement of melanin granules, leading to aggregation and resulting in blanching of skin colour. The effect was transient and passed off in 5–10 min. However, in frogs

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Effect of acetylcholine on melanophores of *Rana tigrina*

Drugs (mg/kg)	*n*	Melanophore size index (mean ± SEM)		
		Before	After (peak effect)	Difference
Acetylcholine (0.1)	20	55.0 ± 1.68	17.45 ± 1.06 $P_1 < 0.001$	37.55 ± 1.42
Atropine (5) + Acetylcholine (0.1)	10	52.7 ± 2.51	20.3 ± 1.58	32.4 ± 2.71 $P_2 > 0.05$
Pentolinium (5) + Acetylcholine (0.1)	10	53.6 ± 2.28	17.1 ± 1.38	36.5 ± 1.77 $P_2 > 0.05$
Procaine (2) + Acetylcholine (0.1)	10	56.1 ± 2.19	49.9 ± 1.93	6.3 ± 0.78 $P_2 < 0.001$

P_1, Statistical significance between pre and peak post drug response.
P_2, Statistical significance between differences in blocker pretreated and control acetylcholine groups.

pretreated with a cholinesterase inhibitor (physostigmine, 0.1 mg/kg), the effect of acetylcholine appeared in 3–5 min, reached a peak effect by 20–30 min and lasted for 45–60 min. This dose of physostigmine did not have any significant effect of its own. Pretreatment with either atropine (5 mg/kg) or pentolinium (5 mg/kg) failed to inhibit significantly the effect of acetylcholine. Procaine (2 mg/kg) markedly inhibited the skin colour blanching and melanin aggregating effects of acetylcholine. All blocking agents were administered 15 min prior to acetylcholine and none of them had any significant effect per se on the test parameters.

The effect of acetylcholine on melanophores of *R. tigrina* was probably not mediated through cholinergic receptors, since atropine (*m*-cholinolytic) and pentolinium (*n*-cholinolytic) both failed to antagonize its effects. The marked and highly significant antagonism produced by procaine can be explained on the basis of the membrane stabilizing effect[3] of the local anaesthetic, which would prevent acetylcholine induced ion influx into the melanophores. It has been suggested[4] that sodium is involved in the melanin aggregating and dispersing effects of acetylcholine and melanocyte stimulating hormone (MSH), respectively. Acetylcholine induced inhibition of the melanin dispersal effect of MSH[5,6] has been suggested to be due to a mechanism involving membrane polarity and ion transport across the melanophore membrane[4].

It is interesting to note that, embryologically, melanophores are derived from the neural crest[7,8]. Procaine, which blocks nerve transmission, including cholinergic neurotransmission, has been shown to inhibit cholinergic melanophore response as well, the underlying mechanism probably being similar in either case.

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Action of Stannous and Stannic Chlorides on Bacteria

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Summary. Stannous or stannic chlorides reduced the growth rate of *K. aerogenes*, *Ps. reptilovora* and an unidentified bacterium in a minimal liquid medium and on agar plates. The greatest effect was observed with *K. aerogenes* and was accompanied by a decreased viability, but 100% survival occurred with the other strains. The metal was loosely bound to the cells and there was no direct correlation between the amount adsorbed and the biological response.

This work forms part of a general study of the actions of metal ions on bacteria and their concentration by the organisms during growth. Initial experiments showed that stannous or stannic chloride at concentrations between 0.25 and 2.0 m*M* reduced the growth rate of organisms in liquid culture and accordingly the investigation reported here was undertaken. Other workers have commented on the innocuousness of soluble inorganic tin compounds towards bacteria[2] and fungi[3].

Materials and methods. Klebsiella aerogenes NCIB 418, *Pseudomonas reptilovora* NRRL B 334 and strain X, an organism, as yet unidentified, which was isolated by exposing medium containing stannous chloride to the atmosphere, were used. They were grown aerobically at 28°C in the minimal medium described by CARTER and DEAN[4] supplemented with KCl to increase the K^+ concentration from the normal level of 9 m*M* to 64 m*M*. This is a glucose-inorganic salts medium (pH 7.1) in which exhaustion of the glucose limits growth and the added KCl, besides increasing the stability of the tin chlorides, enabled strain X to grow at its maximum rate. Its growth rate in the presence of 9 m*M* KCl was 66% of that ob-