

to 13.5 kcal per mole unlike the activation energy determined in the presence of vasopressin.

There is a considerable amount of evidence supporting a separation of water and Na^+ transport effects of vasopressin⁶⁻⁹. The observation that 10 mM Ca^{2+} inhibits the vasopressin stimulated water transport but has no effect on vasopressin stimulated sodium transport led PETERSEN and EDELMAN⁹ to suggest that sodium and water transport effects of vasopressin may be due to separate adenylate cyclase systems. However, we have studied the vasopressin stimulation of cyclic AMP levels in the toad bladder and have shown a dose response characteristic of a single activation process¹⁰. The dose response characteristic of respiration increases following vasopressin treatment shows evidence of two activation processes⁴ one of which is entirely insensitive to theophylline, the second is theophylline sensitive. The first of these can be related to the dose response characteristic for vasopressin stimulated Na^+ transport. The second can be related to the effects of vasopressin on the stimulated adenylate cyclase⁶.

We believe that interaction of vasopressin with membrane receptor sites¹¹ leads directly to an increase in the mucosal permeability to Na^+ , to a release of membrane

bound Ca^{2+} ions and activation of the adenylate cyclase. The release of Ca^{2+} from the membrane and its subsequent mobilization is believed to mainly affect water transport. Such a mechanism of action involving 2 types of permeability effects would provide an alternative basis for understanding the separate effects of the hormone on Na^+ and water transport. Although cyclic AMP may help to mobilize tissue Ca^{2+} probably as a result of a biochemical effect on glycolysis¹⁰ and mitochondrial bound calcium, the principle effect of cyclic AMP on glycolysis is possibly to increase the available supply of ATP to the ion pump. Cyclic AMP may affect membrane bound calcium ion and thereby increase the number of 'pores'. This effect is not equivalent to the hormone permeability effect which is believed to have an effect on the size of effective 'pores'.

Zusammenfassung. Der Temperaturkoeffizient für den durch das zyklische AMP und Theophyllin stimulierten Natriumtransport in der isolierten Krötenblase wurde analysiert. Die Enzypumpe wurde stärker beeinflusst als die mukosale Permeabilität und die Aktivität von Na^+/K^+ ATPase korrespondiert mit der Energie der Pumpenaktivität.

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Activation energy EA of the Na^+ transport across the isolated toad bladder stimulated by various agents including results of ref.³ and present work

Treatment	EA (kcal/mole)
Control	13.6
Cyclic AMP ($10^{-3}M$)	13.8
Theophylline ($10^{-2}M$)	13.4
Aldosterone ($10^{-7}M$)	9.4
Vasopressin ($10^{-6}M$)	9.0
Amphotericin B ($10^{-7}M$)	9.0
Na^+/K^+ ATPase	8.9

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Morphological Evidence for Electrical Synapse of 'Gap' Junction Type in Another Vertebrate Receptor

'Gap' junctions¹ – synapses presumed to be electrically mediated² – are such cases of close apposition of synaptic membranes as show a 20–30 Å gap³, traversed by a hexagonal array of subunits⁴, in between. The over-all thickness of the synaptic junction is 135–180 Å^{1,5} and, for a given synapse, is not affected by differences in methods of treatment^{1,6}. This last feature, therefore, serves as a good criterion for locating and identifying a 'gap' junction⁶.

Using the above criterion, it is possible to demonstrate the presence of 'gap' junction between the sensory cells and afferent nerve endings in large tuberous organ^{7,8} of *Sternarchus albifrons*, a gymnotid weakly electric fish. For electron microscopy, the material was fixed in osmium tetroxide, acetate veronal buffered, and sections were double stained with uranyl acetate and lead citrate. Each sensory cell receives at its basal end a single large bouton terminal of an afferent nerve fibre (Figure 1). The latter carries its myelin sheath very close to the base of the bouton⁸. The cell membranes of the sensory cell and the nerve termination are separated by an interspace of 300–450 Å, except where they enter into formation of synapses. Two types of synapses are found at this interface. One type (Figure 1) is of what has come to be known as

chemical synapses⁹ which are readily recognized by the presynaptic structures, viz., electron dense body surrounded by vesicles. The synaptic membranes in this case are separated by a uniform cleft of 200 Å, and vesicles and mitochondria are found close to the synapse in the nerve ending.

The other type of synapses, at once conspicuous by the closeness of the 2 junctional membranes, appears in section as a single dark line in low magnification electron micrographs (Figure 1), as if a fusion has occurred here

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between the cell membranes of the sensory cell and the bouton. But at higher magnification 2 separate dark lines can still be seen in the region of the synapse (Figure 2), representing the inner leaflets of the 2 cell membranes as the outer leaflets are not stained by the method employed^{6,9}. In sections perpendicular to the synapse, the over-all thickness of the junction is about 150 Å, measured between the cytoplasmic faces of the apposing

membranes¹. The light line between the two dark lines is about 70 Å wide and represents the total distance between the external faces of the inner leaflets of the adjacent cell membranes including the 20 Å gap of the junction. Neither vesicles nor mitochondria are seen in the bouton near the synapse (Figures 1 and 2). Sections partly tangential to the synapse reveal at high magnification dark striations criss-crossing the interspace between the 2 junctional membranes, presenting a pattern what may be obtained if a honeycomb is cut obliquely (Figure 3). Undoubtedly these synapses are different from chemical synapses. These above-mentioned features of the synapse provide strong evidence to suggest that these are 'gap' junction type of electrical synapses.

Although 'gap' junctions are long known to occur between neurons in the central nervous system^{6,9}, their incidence in receptors is rare. The only case so far known is that of the calyceal synapses of the mammalian vestibular organs¹⁰. Thus, the large tuberous organs of *Sternarchus* become the first such case among non-mammalian vertebrate receptors. Electrophysiological data for these receptors are not available, but ultrastructural and electrophysiological studies on tuberous organs of other weakly electric gymnotids do not show the presence of electrical synapses^{11,12}. It is likely that such synapses are specific of *Sternarchus* alone, a weakly electric fish with very high frequency¹³ and unique electric organs of neural origin¹⁴ and reported not to possess usual electroplates with synapses¹⁵. A rapid pathway system¹⁶ has been postulated to account for the striking coordination that exists between electroreception and electric organ discharge frequency. The present morphological evidence of electrical synapses of presumably 'gap' junction type between the receptor cells and their afferent nerve endings, affecting negligible synaptic delay at receptor level, fits well with the theory of rapid pathway system, as far as *Sternarchus* is concerned.

Résumé. Une étude des organes tubéreux de grande taille chez un poisson électrique à faible décharge, *Sternarchus albifrons* a été effectuée en microscopie électronique. Elle a permis d'observer la présence, en plus de la synapse chimique, d'une jonction en «gap» de la synapse électrique.

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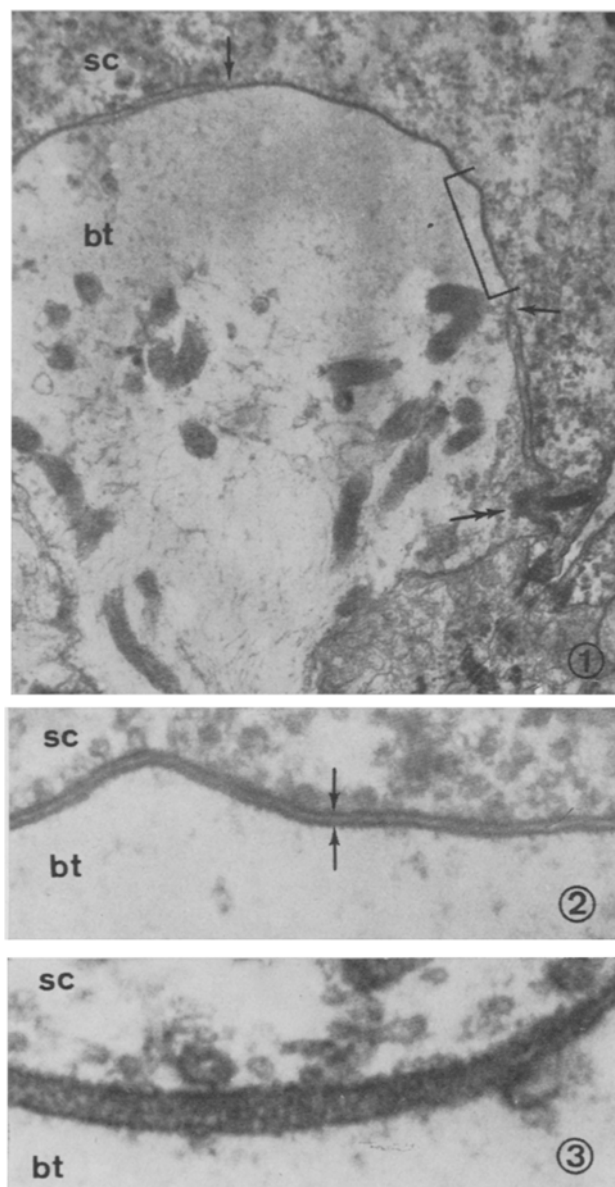


Fig. 1. Large tuberous organ of *Sternarchus albifrons*, in section. A bouton terminal (bt), synapsing with a receptor cell base (sc). Chemical synapse on the right (double-headed arrow). Electrical synapse appearing as single dark line, between the arrows, beyond which the 2 cell membranes are widely separated. Vesicles and mitochondria not seen near the electrical synapse, unlike the chemical synapse. $\times 20,800$.

Fig. 2. Magnified view of the area marked with a bracket in Figure 1. The single dark line resolved into 2 dark lines separated by a light line. The cytoplasm of the receptor cell (sc), and the nerve terminal (bt) separated by the 150 Å thick electrical synapse, presumably a 'gap' junction, between the arrows. $\times 104,000$.

Fig. 3. An electrical synapse in partly tangential section at receptor cell (sc) nerve terminal (bt) interface. See text. $\times 104,000$.

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