

TRIAL OF SIMULTANEOUS RECORDING OF MEMBRANE CURRENT AND CONCENTRATION CHANGE OF INTRACELLULAR CALCIUM IN THE OLFACTORY RECEPTOR CELL.

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Although many evidences indicate that the olfactory transduction is performed through the cAMP second messenger system, some reports argued the existence of Ca/IP₃ second messenger system. This argument dictated us to try simultaneous measurements of membrane current and intracellular Ca concentration in the olfactory receptor cell: When cAMP was injected into the isolated newt olfactory receptor cell loaded with Ca sensitive dye, Fura2 by achieving the whole cell recording mode with the patch pipette containing the 5 mM cAMP under the holding potential of -70 mV, current influx and Fura2 fluorescence change indicating the intracellular Ca increase were recorded simultaneously. The differentiated function of the time course of Ca increase at the olfactory knob near cilia exhibited very similar wave form to the current influx. This result strongly suggests that Ca ions influxed through the ion channels gated by cAMP in the cilia. The reported Ca increase induced by odor stimulation may occur as the part of the cAMP pathway.

IP₃-ACTIVATED ION CHANNELS IN FROG OLFACTORY RECEPTOR CELL MEMBRANE

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To clarify the possible role of IP₃ as a second messenger in vertebrate olfactory transduction, I have studied the effect of IP₃ on ion channel activity in inside-out membrane patches excised from the soma membrane of olfactory receptor cells. In a microflow superfusion chamber, patches were perfused with divalent cation-free internal solution or low calcium K-internal solution (Ca²⁺ 1 × 10⁻⁸ M), in which 1 μM cAMP or 5.3 μM IP₃ was dissolved. With divalent cation-free external solution or Na-external solution containing divalent cations, cAMP-activated ion channels which were identified by their response characteristics of I-V relation did not respond to IP₃. With Ba-external solution (Ba²⁺ 88 mM), however, ion channels which exhibited burst-like openings in response to IP₃ were found at both -75 mV and +75 mV. These IP₃-activated ion channels responded to neither cAMP nor voltage change itself. The unit conductance determined from the total amplitude histograms was 26 pS at -75 mV and 44 pS at +75 mV. The present finding of IP₃-activated ion channels in the olfactory receptor cell membrane, which probably belong to a type of second messenger-operated Ca-channels, suggests that the multiple pathways, both an adenylate cyclase cascade and a phosphoinositidase C cascade, may be involved in vertebrate olfactory transduction.

EFFECTS OF ODOR STIMULI TO VOMERONASAL ODOR SAMPLING MOVEMENTS IN MICE

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In rodent vomeronasal organ, odorants are received by pumping action of cavernous vascular tissue situated along the organ. To ascertain whether the vomeronasal organ is specialized sex pheromone receptor, the effects of various kinds of odor to the vasomotor action of the organ was investigated.

When, spontaneous vasomotor action was suppressed by an excessive anesthesia, electrical stimulations delivered to the naso-palatine nerve caused odor sampling movements consisted of relaxation and contraction. Stimulations with adequate frequency yielded the movement which simulated natural sampling actions such as periodical contractions or a long lasting large contraction. Various vapor odors slightly affected the frequency and amplitude of periodical contracting movement with the exception of impinging acetic acid odor which caused large contraction. As aquatic stimulus, direct urine stimulus caused a long lasting large contraction, indicating that the mouse vomeronasal system would appear to be essential to the reception of non-volatile urine pheromone.

VOMITING ABILITY OF AMPHIBIANS

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As an aid to understand the vomiting mechanism of vertebrates, the emetic response of amphibians was studied. Ten species of frogs and two salamanders (from the genera *Xenopus*, *Rana*, *Hyla*, *Rhacophorus*, *Cynops* and *Hynobius*) were tested.

Xenopus laevis and *Rhacophorus schlegelii* were sensitive to apomorphine-HCl, while the other species were less sensitive. Copper sulfate was a potent emetic to both frogs and salamanders. The most notable phenomenon during emesis was contraction of the abdominal wall, suggesting a rise in intra-abdominal pressure. Frogs with their abdominal walls denervated by cutting the spinal nerves could not vomit. Electromyograms of the rectus abdominis muscle confirmed contractions in association with the ejection of gastric contents.

In mammals, the primary force for ejecting the gastric contents is a rise in intra-abdominal pressure produced by the contractions of body wall and respiratory muscles. From a mechanistic standpoint, there seems no difference between amphibians and higher vertebrates in how they generate force to eject foodstuff. Among tetrapods, the ability to vomit characterizes carnivorous bulk-feeders and is not a simple reflection of evolutionary stage.