

Cardiac Stimulant Action of Extracts of Coelenterates on Rat Atria

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Abstract □ Crude aqueous ethanol extracts of nine out of 12 species of anthozoans and the one scyphozoan examined showed varying degrees of positive inotropic and chronotropic effects on isolated rat atria. This is the first report of what may be rather widely occurring heart stimulants of possible practical utility among the Coelenterata.

Keyphrases □ Coelenterates—cardiac stimulant action of extracts, rat atria □ Anthozoans—ethanol extracts, cardiac stimulant activity, rat atria □ Scyphozoan—ethanol extract, cardiac stimulant activity, rat atria □ Cardiac stimulant activity—ethanol extracts of coelenterates, rat atria

While examining extracts of coelenterates for anti-tumor activity (1, 2) and central stimulant action (3), the toxicity of the extracts aroused interest in examining their effect on cardiac muscle. Extracts of nine of 12 species of anthozoans and the one scyphozoan examined showed varying degrees of positive inotropic effect.

About 70 of some 9000 known species of coelenterates have been reported capable of causing intoxication in humans, either from contact with the nematocysts (stinging organelles) or by ingestion of uncooked tissues (4). Many workers have examined the chemical nature and pharmacology of these toxins, and several have examined their cardiac action (4). Huang and Mir (5) noted that an extract of the tentacles of *Calliactis polyopus* (a sea anemone) caused a brief lowering of coronary outflow, heart rate, and amplitude of cardiac contractions in isolated rabbit heart, while higher doses produced irregular cardiac contractions, usually resulting in cardiac arrest.

This is the first report of what may be rather widely occurring heart stimulants of possible practical utility among the Coelenterata.

EXPERIMENTAL

The sea animals were preserved in ethanol immediately after collection. They were homogenized with 50 ml 30% ethanol/100 g animal wet weight and allowed to stand for 24 hr. The solid residue was removed by centrifugation, and the combined supernate and preserving alcohol were flash evaporated at $\leq 40^\circ$ to a volume equal in milliliters to the wet weight of the animal in grams. The aqueous solution was filtered or centrifuged and the clear solution was used for testing. This crude extract was lyophilized for dry weight determination.

Bioassay of the solutions was performed on isolated atria of rat hearts. The atrium was separated from the rest of the heart and suspended in an isolated organ bath (20, 25, or 50 ml) containing Krebs-Ringer bicarbonate medium (pH 7.4) of the following composition in distilled deionized water (in mmoles): Na^+ , 145; K^+ , 6.02; Ca^{+2} , 1.22; Mg^{+2} , 1.33; Cl^- , 126; HCO_3^- , 25.3; PO_4^{-3} , 1.2; SO_4^{-2} , 1.33; and glucose, 5.5. The temperature of the organ bath was maintained at 30° , and the Krebs-Ringer medium was continuously aerated with 95% O_2 -5% CO_2 .

The spontaneously beating atrial preparation was connected by

a thin silk thread to a force-displacement transducer¹, and the contractile movements were recorded on a six-channel polygraph². The preparation was allowed to equilibrate under 750 mg tension for 60 min prior to beginning an experiment. After this equilibration, during which the preparations were washed out every 30 min, the spontaneous beat rate of the atria remained constant, the change during a 10-min observation being less than 5 beats/min. The changes in contractile force and rate produced by the test agents are expressed as a percentage increase or decrease in tension and rhythm, with the period immediately preceding addition of test solution to the tissue bath as the baseline for comparison. These data are preliminary, one solution being tested on preparations from three different animals at most.

RESULTS AND DISCUSSION

Table I summarizes the cardiac stimulant action (inotropic and chronotropic) of crude extracts from 13 species of coelenterates. Figure 1 shows the tracings for the crude extracts of *Anthopleura xanthogrammica* and *A. elegantissima*, where the effects were observed for 70 and 30 min, respectively.

Pretreatment of the preparations with propranolol and metolol³ (both 10^{-8} M, both β -adrenergic receptor blocking agents) and with phentolamine (10^{-6} M, an α -adrenergic receptor blocking agent) had no effect on the inotropic response of the atria to the stimulatory test agents. Atria from either 6-hydroxydopamine-treated rats (50 mg/kg iv, 24 hr prior to sacrifice) or reserpine-treated rats (4 mg/kg im, 24 hr prior to sacrifice) showed responses identical to those of normal animals. Therefore, it appears that the action of the coelenterate stimulant substance(s) is

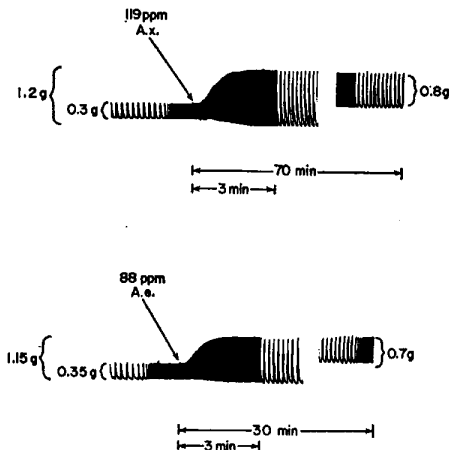


Figure 1—Tracings for the crude extracts of *A. xanthogrammica* (top) and *A. elegantissima* (bottom) (rat atria, 1 g rest tension, chart speed of 10 mm/min and 5 mm/sec).

¹ Grass model FT.03.

² Grass model 7.

³ MJ-1998, Mead Johnson.

Table I—Effect of Extracts of Various Species of Coelenterates on Rat Atria

Species	Source	Concentration, ppm	Percent Increase, Inotropic ^a	Percent Increase, Chronotropic ^a
<i>Boloceroides mcMurrichi</i> (Kwiatniewski)	Oahu, Hawaii	— ^b	20–51	0
<i>Palythoa psammophila</i> Walsh and Bowers	Oahu, Hawaii	— ^b	0	0
<i>Zoanthus pacificus</i> Walsh and Bowers	Oahu, Hawaii	— ^b	0	0
<i>Macranthea cookei</i> Verrill	Oahu, Hawaii	— ^b	0	0
<i>Tealia coriacea</i> (Cuvier)	Bodega Bay, Calif.	500	>100	0
<i>Tealia lofotensis</i> (Danielssen)	Bodega Bay, Calif.	350	10–20	0
<i>Metridium senile</i> (L.)	Bodega Bay, Calif.	50	125–150	67
<i>Anthopleura xanthogrammica</i> (Brandt)	Bodega Bay, Calif.	119	300 ^c	30
<i>Anthopleura elegantissima</i> (Brandt)	Bodega Bay, Calif.	83	230 ^d	14
<i>Tealiopsis nigrescens</i> Verrill	Oahu, Hawaii	100	230	85
<i>Stoichactis kenti</i> Haddon and Shackleton ^e	Tahiti	10	50–85	0–30
<i>Tubastrea aurea</i> Quoy and Gaimard	Oahu, Hawaii	100	210	43
<i>Cassiopeia mertensi</i> Brandt	Oahu, Hawaii	100	0	0

^a Increases were noted for at least 5 min. ^b A 0.1-ml crude extract diluted 200:1. ^c Increase observed for 70 min, dropping to 170% at 70 min. See Fig. 1. ^d Increase observed for 30 min, dropping to 100% at 30 min. See Fig. 1. ^e Purified fraction. Procedure described in Turlapaty *et al.* (6).

not related to any adrenergic mechanism and that the compound(s) probably acts directly on the heart muscle.

The isolation, characterization, mode of action, dose-response, and other pharmacological studies of the active principle(s) are being pursued.

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