

activity was 160 msec, and during alpha, 171 msec. The difference, 11 msec, is so nearly the same as that for the means for switch closure as to force the conclusion that all of the time advantage of the beta condition was prior to the muscle activity.

A 12-msec difference in reaction latency is not a long time. In conditions similar to ours, Lindsley (5) reported a saving of 74 msec by use of a foreperiod of 0.3 to 1 sec compared with the unspecified foreperiod produced by omitting the "ready" signal. Telford (6) found that foreperiods of 1 and 2 sec produced reaction times of 90 msec less than foreperiods of 0.5 sec. Woodrow (6) reported 60-msec savings when he compared foreperiods of 2 sec with foreperiods of 24 sec. Clearly, the alertness indexed by beta waves, if alertness it is, is not the magnitude of alertness caused by optimal forewarning.

But in spite of being small, the difference of 12 msec is real, and we propose two alternative explanations. The first is that there is no fundamental difference between the alpha and beta reaction times, but that we were more successful in hitting the exact peak of excitability in beta than in alpha. Lindsley's failure to find a difference between spontaneous alpha and beta reaction times is irrelevant here because he did not control for the phase of the excitability cycle. But against this possibility of no fundamental difference is the fact that intraindividual variability of beta frequencies is greater than the variability of alpha frequencies. Our experimental error should thus have reduced, rather than produced, the beta advantage.

The second possibility is that the beta advantage comes from an accumulated effect of interneural facilitation produced by heightened activity in the reticular activating system. There is considerable evidence that the reticular system is responsible for beta waves (7) and that it effects interneural facilitation (5, 8). We know of no contradictory evidence, but clarification must await an improvement of technique which will insure hitting the peak excitability of both alpha and beta rhythms.

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#### Action of Acetylcholine on Bivalve Hearts

**Abstract.** The isolated hearts of a comprehensive selection of bivalves were tested to determine the distribution of the excitator and depressor effects of acetylcholine. No broad relationships are obvious although some intrafamily uniformity of response exists. The hearts of most species were both excited and depressed, probably by separate actions of acetylcholine. From these data we can conclude that bivalve heart muscle conforms closely to other molluscan muscle in its pharmacological reactions to acetylcholine.

The depressor effect of acetylcholine on most bivalve hearts and its excitator effect on hearts of *Mytilus* species are well known (1). Pilgrim has also found that some bivalve hearts are depressed at low concentrations and excited at high concentrations of acetylcholine (2). A similar "combination response" has been recorded from hearts of *Spisula solidissima* (3) and *Anodonta cygnea* (4).

We observed the effects of acetylcholine on the isolated ventricles of 39 species from 20 families, in order to determine the distribution of responses within the class Bivalvia. Most hearts were isolated in water-jacketed baths with tension applied between the auricles (5), but the atypical construction of ostreid and anomiid hearts does not allow such an arrangement. In these animals the heart was stretched in the direction of the normal beat. Temperature was controlled throughout each experiment. The perfusion fluid for marine species was either natural or artificial (6) sea water. For freshwater animals a 5-percent artificial sea water solution made with  $10^{-3}M$   $Na_2HPO_4$  (pH 7.6) was used. The isolated hearts of

many species were quiescent and were induced to beat with 5-hydroxytryptamine ( $10^{-7}$  to  $10^{-5}M$ ). Heart beat was recorded by means of an isotonic lever on a smoked drum; tension on the lever was varied between 200 and 1000 mg, depending on the size of the heart.

The responses do not appear to be distributed phylogenetically (Table 1). Thus, depressor, excitator, and combination effects occur both in the Pteriomorphia (7) and Lamellibranchia and in both primitive forms (*Glycymeris*, *Cardita*) and highly evolved forms (*Chlamys*, *Panope*). No depression was observed in the Protobranchiata. Also, no correlation exists between effect and such morphological features as relative size of adductors, symmetry of body and mantle/shell (8), or condition of the gills. Finally, there is no relationship between response and physiological or ecological factors such as activity, burrowing and boring, attachment to substrate, mode of feeding, or exposure during low tides.

Although there are no broad systematic relationships correlated with the acetylcholine responses, there is some uniformity within families. The venerids, which have been widely studied, all have low thresholds ( $10^{-12}$  to  $10^{-8}M$ ) and for the most part show no excitation. Pilgrim (2) obtained similar results from hearts of *Dosinia anus*, *Protothaca crassicosta*, and *Chione stutchburyi*. The Mactridae show only the combination response to acetylcholine. This is also true for the family Amphidesmatidae (Mactracea) (2). Characteristically, hearts from the family Mytilidae have high thresholds ( $5 \times 10^{-8}$  to  $2 \times 10^{-6}M$ ) and excitation predominates. Ostreid hearts have equally high thresholds but no typical excitatory response ever occurs. Frequency always decreases but amplitude changes are variable. Similar effects have been noted for *Ostrea hefferdi* (2), *Ostrea laperousei* (9), and *Crassostrea angulata* (10). The striking uniqueness of the responses of the oysters and mytilids relative to the responses of other Pteriomorphia would tend to support Cox's raising of these two groups to the ordinal level (7).

Two general statements can be made regarding the distribution of these acetylcholine responses. First, there are very few species in which concentrations of acetylcholine lower than those producing excitation did not result in depression. Even in *Mytilus edulis* and *Modiolus modiolus* some depression was observed in most of the hearts tested.

Table 1. Systematic distribution of responses of bivalve hearts to acetylcholine. Symbols: X, excitation; O, depression; OX, combination of excitation and depression; -X, excitation observed, but presence of depression not determined; numerical value, variable response, value of fraction is indicated as percentage of response.

Organism	Response
<i>Subclass I—Protobranchiata</i>	
NUCULANIDAE	
<i>Yoldia thraciaeformis</i>	-X
<i>Subclass II—Pteriomorpha</i>	
Order Taxodontida	
GLYCYMERIDAE	
<i>Glycymeris subobsoleta</i>	OX
Order Anisomyaria	
MYTILIDAE	
<i>Mytilus californianus</i>	X
<i>M. edulis</i>	OX/X, 63
<i>Modiolus modiolus</i>	OX/X, 86
PECTINIDAE	
<i>Pecten caurinus</i>	OX
<i>Aequipecten irradians</i>	OX*
<i>Chlamys hericulus</i>	OX
<i>Hinnites giganteus</i>	OX
ANOMIIDAE	
<i>Pododesmus macroschisma</i>	O
OSTREIDAE	
<i>Crassostrea virginica</i>	O
<i>C. gigas</i>	O
<i>Subclass III—Lamellibranchia</i>	
Order Heterodontida	
CARDITIDAE	
<i>Cardita floridana</i>	OX/O, 67
VENERIDAE	
<i>Mercenaria mercenaria</i>	O
<i>Protothaca tenerrima</i>	O
<i>P. staminea</i>	O
<i>Tapes philippinarum</i>	O
<i>Compsomyx subdiaphana</i>	O
<i>Humilaria kennerleyi</i>	OX/O, 80
<i>Saxidomus giganteus</i>	O
CARDIIDAE	
<i>Clinocardium nuttallii</i>	OX/O, 20*
<i>Serripes groenlandicus</i>	OX/O, 25
MACTRIDAE	
<i>Schizothaerus nuttallii</i>	OX
<i>S. capax</i>	OX
<i>Spisula solidissima</i>	OX
LUCINIDAE	
<i>Phacoides annulata</i>	OX
ASAPHIDAE	
<i>Gari californicus</i>	OX
SEMELIDAE	
<i>Semele rubripicta</i>	OX
TELLINIDAE	
<i>Macoma secta</i>	OX
Order Schizodontida	
UNIONIDAE	
<i>Anodonta cataraacta</i>	OX/X, 28
<i>A. grandis</i>	OX/X, 78
<i>Lampsilis siliquoidea</i>	OX
Order Adepedontia	
SOLENIIDAE	
<i>Solen sicarius</i>	OX
MYIDAE	
<i>Mya arenaria</i>	O
HIATELLIDAE	
<i>Panomya ampla</i>	OX
<i>Panope generosa</i>	OX
PHOLADIDAE	
<i>Zirfaea pilsbryi</i>	O
<i>Barnea costata</i>	OX*
Order Anomalodesmacea	
LYONSIIDAE	
<i>Entodesma saxicola</i>	O

\* Excitation observed in the presence of benzoquinonium chloride ( $10^{-5}$  g/ml).

Pilgrim (2) observed diastolic arrest in *Mytilus canaliculus*, and we have observed it in *M. edulis*. Thus the hypothesis remains intact that acetylcholine is a transmitter substance of inhibitory cardiac nerves in bivalves. Second, excitation is also a nearly universal response. Excepting ostreids, every family from which more than one species has been tested includes at least one species which has provided some heart preparations which are excitable by high doses of acetylcholine (Table 1). This fact, and particularly such special cases as *Humilaria kennerleyi*, suggests that, when excitation cannot be demonstrated, the effect is masked in some way.

Benzoquinonium chloride (Mytolon) ( $10^{-5}$  g/ml) blocks both the depressor and excitor effects of acetylcholine. However, threshold and effectiveness of blockade are different for the two responses, and the relationship varies from species to species. For example, the depressor response of *Schizothaerus capax* heart is more effectively blocked than excitation. The opposite is true for the heart of another mactrid, *Spisula solidissima*.

The two effects of acetylcholine are clearly separable by their dissimilarity, by the lack of appearance of one or the other in some species (for example, *Saxidomus giganteus*, *Mytilus californianus*), and especially by the differential action of benzoquinonium chloride. A reasonable model of acetylcholine action on bivalve hearts should involve either two separate sites of action or two modes of attachment to the same site at high and low concentrations.

The general excitation of bivalve hearts by acetylcholine brings bivalve heart muscle pharmacologically into line with other well-known molluscan smooth muscle preparations such as the *Mytilus* anterior byssus retractor muscle (11) and the gastropod radula retractor (12). This is especially notable in quiescent heart preparations which in some cases are contracted by acetylcholine and in others are relaxed and induced to beat by 5-hydroxytryptamine. Furthermore, gastropod heart muscle has also been shown to be excitable by acetylcholine (13). Whether these excitatory effects are functionally homologous remains to be seen (14).

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## Effect of Rotation on Flowering Response of *Xanthium pennsylvanicum*

**Abstract.** The flowering response of *Xanthium pennsylvanicum* Wallr. is attenuated when the plant is rotated around a horizontal axis at the rate of 0.25 rev/min. Rotation prior to an inductive dark period has the strongest effect.

*Xanthium pennsylvanicum* is one of the most sensitive short-day plants. It can be maintained indefinitely in a vegetative state if grown under long-day conditions. However, it will initiate floral primordia if exposed to a single dark period in excess of 8.75 hours (1).

When young *Xanthium* plants are rotated around a horizontal axis, the leaves and the petioles of the rotated plants display strong epinastic responses within 24 hours, while stationary control plants show normal diurnal leaf movements (2).

The turntable of the plant rotator used for the experiment has a diameter of 4 feet. The turntable rotates in the same manner as a record player, but the rotating axis is horizontal rather than vertical. Plants were placed in holes on the turntable by hooks and rubber bands. The 24 holes were distributed evenly in a zone 10 to 20 inch-