Metachronal Limb Movements by Artemia salina: Synchrony of Male and Female during Coupling

Abstract. *Studies of the metachronal rhythms of* Artemia salina *reveal that limb movements of the male and female are synchronous during precopulatory coupling. Synchrony is an adaptation which maintains efficiency in locomotion, respiration, and feeding. The male acts as the pacemaker for the pair and drives the female at a higher rate than she exhibits when alone.*

Synchrony of rhythmic behavior between individuals has been reported for chirping by crickets, flashing by fireflies, and a variety of behaviors by man (*1*). This study demonstrates that the metachronal limb movements of brine shrimp are synchronous when the male and female are joined in precopulatory coupling. The sequence of limb movements begins in the most posterior segment and progresses anteriorly in many arthropods (legs of cockroaches, centipedes, millipedes, and brine shrimp; swimmerets of lobsters and crayfish; wings of locusts; and book gills of *Limulus*) (*2*). This patterning of limb movements coordinates a number of limbs and often aids in hydro- and aerodynamic efficiency. The cyclical movements of the 11 pairs of phyllopodous limbs of the brine shrimp, *Artemia salina*, propel it through the water, generating currents used in respiration and filter-feeding. Coupling occurs in sexually mature *Artemia* when the male attaches himself to the ovisac of the female with his claspers. The pair swims in tandem for 2 to 3 days until the female molts and they copulate. Synchrony of limb movements coordinates the 22 pairs of limbs of the couple and maintains the efficiency of metachrony.

Metachrony of *Artemia* was investigated by macrocinematography with a Bolex H-16 at 64 frames per second; the electrical fields produced during the the cyclical movements of the phyllopods were recorded with external electrodes. Thirty individual and nine pairs of *Artemia* were each placed in distilled water within a Lucite chamber 8 by 20 mm and 5 mm deep. Two silver electrodes coated with AgCl and placed longitudinally in the bottom of the chamber provided differential input to an a-c preamplifier which fed into either an oscilloscope or oscillograph (Narco, DMP-4A). Muscle movements generate potentials of about 100 μv between the electrodes in distilled water; however, these fields weaken as the conductivity of the medium increases, and in saline no potentials are recorded with this apparatus. This technique worked well with *Artemia*, which tolerates distilled water for many hours with no discernible effects upon behavior or physiology (*3*). All experiments were conducted at a room temperature of 22° to 25°C.

Each cycle begins in the posterior limbs and passes anteriorly at frequencies of 2 to 8 cycle/sec in individual *Artemia*. There is an inverse relationship between frequency and specimen size (Fig. 1). Individuals may vary their frequencies twofold. During metachrony, sinusoidal potentials of 50 to 100 μv and 2 to 8 cycle/sec are recorded which correspond to the cycle of limb movement (Fig. 2A). Assurance that these electrical oscillations are caused by limb movements comes from simultaneous macrocinematography of the specimen in the recording chamber and the oscillograph; there is a 1 : 1 correspondence between each potential wave and one point in each metachronal cycle. This is, therefore, an accurate method of recording metachronal frequencies without impeding the animal.

In couples, each metachronal wave in the female is preceded by a wave in the male. Since the wave passes anteriorly in both individuals, the wave appears to pass continuously along the synchronous couple. This occurs even though the spacing between limbs of the individuals of the pair is much larger than the spacing between the limbs of an individual. The spacing for an individual *Artemia* is about 0.1 mm, and the distance between the

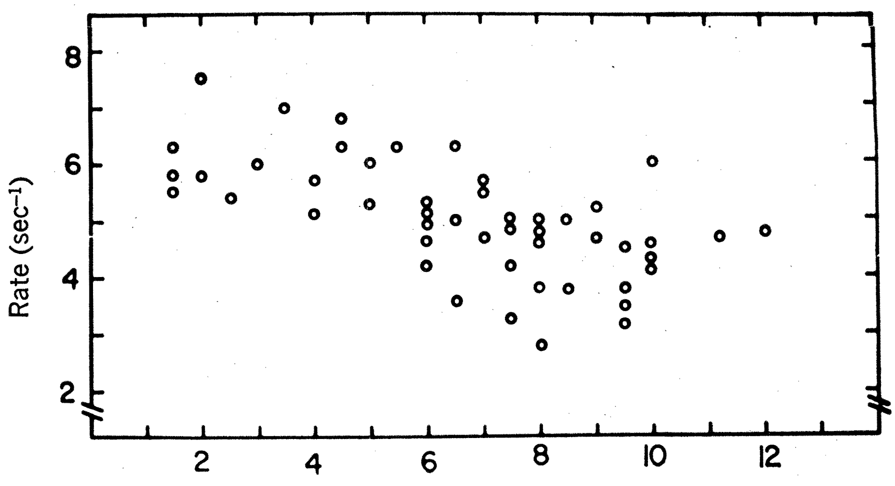

Fig. 1. Frequency of limb movement as a function of overall body length for individual *Artemia*. Observations from both sexes and juveniles. Sex had no relationship to rate; however, the very largest individuals are females.

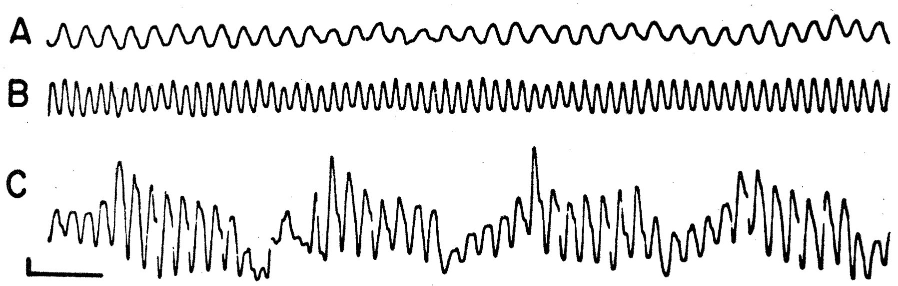

Fig. 2. Oscillograms of potentials generated by *Artemia* and recorded with external electrodes in distilled water. The calibration mark represents 100 μv on the vertical scale and 1 second on the horizontal scale. (A) A record from a single female *Artemia* (9.5 mm long; average rate, 4.0 sec^{-1}; range, 3.2 to 5.5). (B) A record from a synchronous couple (male 7.0 mm, female 9.5 mm, same female as in A; average rate, 5.6 sec^{-1}; range, 4.7 to 6.5). (C) Oscillogram from same couple as in B during a period of asynchrony. The "beat" is the difference in frequency between the two individuals. Beat frequency is 0.25 to 0.35 second. The average frequency of the *Artemia* in this record is 5 sec^{-1}.

posterior phyllopods of the female and the anterior phyllopods of the male is 1 to 3 mm. Electrical recordings of synchronous couples yield sinusoidal potentials of 75 to 125 μv and 3 to 6 cycle/sec (Fig. 2B).

It is noteworthy that couples are not always synchronous. When potentials are recorded from asynchronous couples, the metachronal oscillations are superimposed on a much slower and larger oscillation representing the beat frequency of the unequal frequencies of male and female limb movements (Fig. 2C). When asynchrony occurs, it results from the female's momentarily running at a higher frequency than the male. This happens rarely and the frequency of the female soon returns to the lower rate of the male. I have not observed a female with a lower frequency than that of the male with which she is joined.

When a synchronous couple is separated, the male usually continues at a frequency similar to that of the couple and the female reduces her frequency to a lower value (Fig. 2, A and B). The males of most couples are smaller than the females (6.5 to 7.5 mm, males; 7.5 to 11 mm, females), and the frequencies of the separated individuals correspond to the relation of size and rate (Fig. 1). This observation together with the fact that each wave in the male precedes the wave in the female indicates that the male is the pacemaker for the couple and drives the female at a higher frequency than she exhibits alone.

The remote-recording method is also useful for measuring other behaviors of unrestrained *Artemia*. Shadow-induced escape responses can be measured by simultaneous recording of electrical fields and of the light level at the bottom of the chamber with a photocell. *Artemia* respond to shadows by contracting all limbs, nearly simultaneously, and producing potential changes well over 100 μv. Response times range between 100 and 240 msec. Shadow responses are observed in couples with each member simultaneously beginning vigorous swimming movements.

In the other arthropods for which synchronous rhythmic behavior has been reported, the behavior is involved with communication (*1*). In *Artemia* the rhythmic synchrony is primarily locomotor and its adaptive significance is obvious: the efficiency of metachrony in locomotion, feeding, and respiration is maintained during the period of coupling. Asynchrony would interrupt the smooth currents necessary for respiration and feeding and would reduce the efficiency of swimming. Even the escape response of the pair is synchronous; hence, sexually mature brine shrimp are not subject to adverse selection as a result of coupling.

CHARLES M. LENT

Department of Zoology,
Ohio University, Athens 45701

References and Notes

1. J. Buck and E. Buck, *Science* **159**, 1319 (1968).
2. G. M. Hughes, *J. Exp. Biol.* **29**, 267 (1952); S. M. Manton, *J. Linn. Soc. Zool.* **42**, 118 (1952); *ibid.* **44**, 299 (1954); W. J. Davis, *J. Exp. Zool.* **51**, 547 (1968); K. Ikeda and C. A. G. Wiersma, *Comp. Biochem. Physiol.* **12**, 107 (1964); D. M. Wilson, *J. Exp. Biol.* **37**, 471 (1961); C. R. Fourtner, C. D. Drewes, R. A. Pax, *Comp. Biochem. Physiol.* **38**, 751 (1971).
3. P. C. Crogan, *J. Exp. Biol.* **35**, 234 (1958).
4. I thank Andrew Wilson and Douglas Smith for technical assistance. Supported in part by Ohio University Research Committee grant number 340.

29 July 1971

■

Auditory Evoked Potentials during Speech Perception

Abstract. *Neural responses evoked by the same binaural speech signal were recorded from ten right-handed subjects during two auditory identification tasks. One task required analysis of acoustic parameters important for making a linguistic distinction, while the other task required analysis of an acoustic parameter which provides no linguistic information at the phoneme level. In the time interval between stimulus onset and the subjects' identification responses, evoked potentials from the two tasks were significantly different over the left hemisphere but identical over the right hemisphere. These results indicate that different neural events occur in the left hemisphere during analysis of linguistic versus nonlinguistic parameters of the same acoustic signal.*

The relation between an acoustic speech signal and its phonetic message appears to be a complex and highly efficient code, which requires a specialized linguistic "decoder" for its perception (*1*). Dichotic listening experiments with normal (*2, 3*) and brain-damaged subjects (*4*) have further suggested that the specialized neural mechanisms required for the perception of speech are lateralized in one cerebral hemisphere, usually the left. This interpretation is consistent with clinical analyses of language disorders following brain damage (*5*), and may be related to anatomical differences between left and right temporal lobes (*6*).

In a recent review of hemispheric specialization for speech perception, Studdert-Kennedy and Shankweiler (*3*) concluded that ". . . specialization of the dominant hemisphere in speech perception is due to its possession of a linguistic device. . . . [W]hile the general auditory system common to both hemispheres is equipped to extract the auditory parameters of a speech signal, the dominant hemisphere may be specialized for the extraction of linguistic features from those parameters."

Despite the large body of behavioral and clinical evidence for specialization of one hemisphere in speech perception, there is no evidence which clearly distinguishes neural activity specifically related to linguistic processing from that which occurs during the processing of any auditory stimulus (*7, 8*). Empirical evidence for such a distinction requires a direct comparison of neural activity during linguistic and nonlinguistic processing conditions with other sources of variation in neural activity eliminated between conditions. We have therefore compared neural activity evoked by the same consonant-vowel syllable during two auditory identification tasks: one that required analysis of acoustic parameters which provide linguistic information (Stop Consonant task) and one that required analysis of acoustic parameters which provide no linguistic information at the phoneme level (Fundamental Frequency task). For convenience, we shall use the terms "linguistic and nonlinguistic parameters" to refer to those acoustic parameters that do and do not, respectively, provide linguistic information at the phoneme level.

In the Stop Consonant task, subjects were required to indicate which of two possible stimuli had occurred on each trial: /ba/ or /da/. The stimuli were generated by the parallel resonance synthesizer (Haskins Laboratories), and were prepared to be identical in duration (300 msec), initial fundamental frequency ($F_0 = 104$ hz), frequency contour (falling), and intensity contour (falling). Thus, the two sylla-