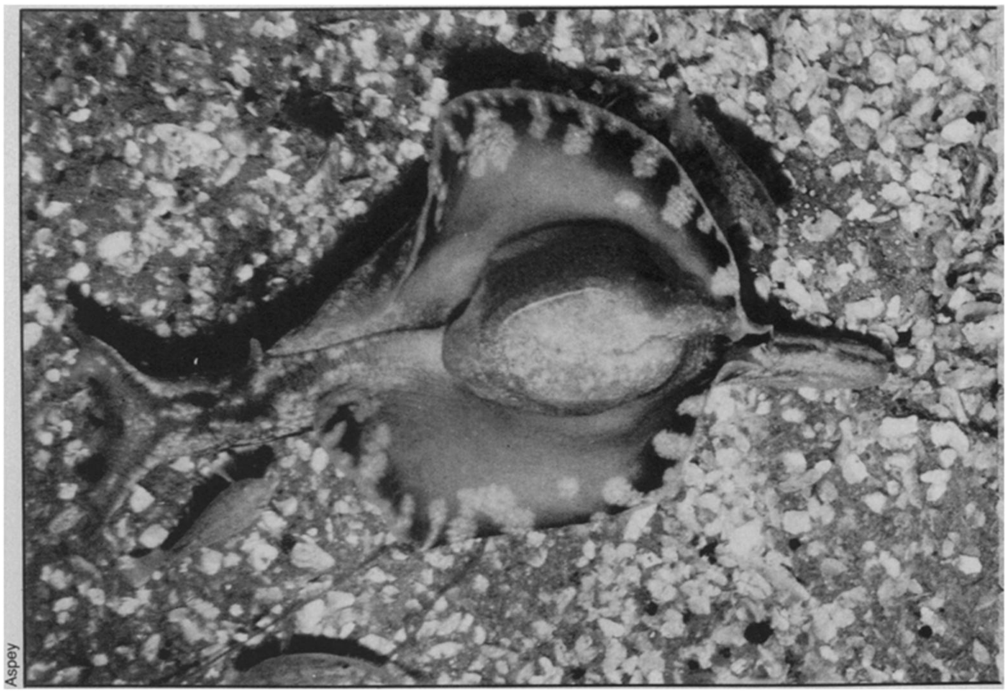
SEX AND THE SEA HARE



Swimming is one behavior of the sea hare, Aplysia brasilina.

Although its complex social behavior is limited primarily to sex, large nerve cells make **Aplysia** a favorite organism for neurobiologists

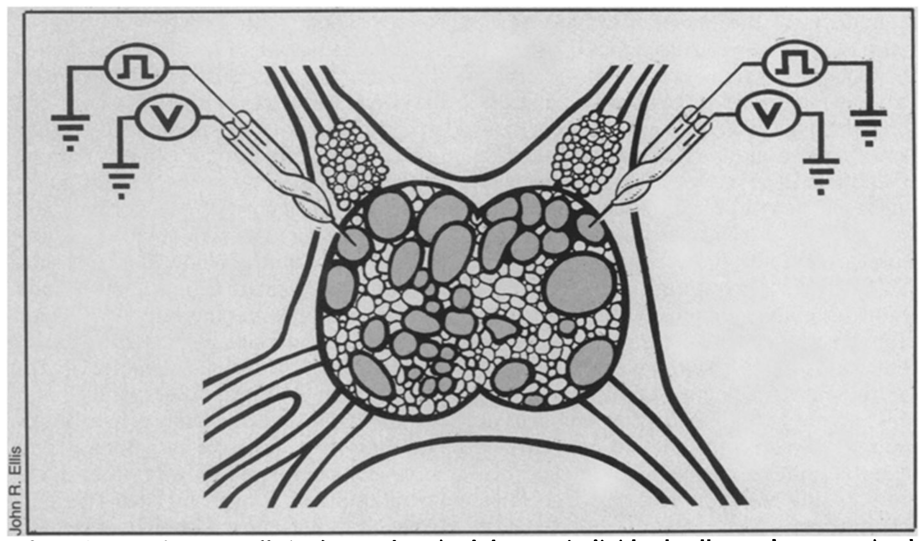
BY JULIE ANN MILLER

What a pity it is that *Aplysia,* the sea hare, doesn't press levers or run mazes or play the piano. If it did it might be the most useful of research animals. This large marine snail with tentacles resembling rabbit ears is already a favorite of neurobiologists because of its convenient nerve cells. What behaviors do those nerve cells direct? The *Aplysia* lay eggs and spews purple ink, it crawls and flaps away from predators and it grazes great quantities of seaweed. Its only complex social behavior seems to be sex — but sex in *Aplysia* is no humdrum affair, as was pointed out at the recent symposium in Syracuse on Chemical Signals in Vertebrates and Aquatic Animals.

Aplysia inched their way into the mainstream of biology by virtue of their convenient nerve cells rather than by any behavioral sophistication. As early as 1893 biologists described the large size of *Aplysia* nerve cell bodies, and in 1942 French biologist Angelique Arvanitaki discovered that certain cells can be identified in any animal of a species.

Since then Eric Kandel of Columbia University and others have used criteria such as size, position, color, firing pattern and response to neurotransmitter chemicals to recognize many more cells in the *Aplysia* abdominal ganglion, a group of about 1,500 nerve cell bodies. The scientists have distinguished 55 individual cells and recognized about 1,000 more cells as members of characteristic-sharing clusters. In contrast, individual nerve cells have not been identified among the large number of small cells in any mammalian brain.

Experiments with *Aplysia* and other invertebrate animals have led to important insights into the nervous system. Such research, for example, is largely responsible for the concept that individual nerve cells can be unique, with characteristic receiving, integrating and signaling properties.



In a cluster of nerve cells in the sea hare's abdomen, individual cells can be recognized. Identified cells are shaded in the diagram above. With electrophysiological techniques, scientists can record the activity of several cells simultaneously.

Aplysia are also helping to bridge the gap between studies of single cells and of behavior. Scientists can use the well-mapped ganglia to determine interconnections between nerve cells. The researchers insert electrodes simultaneously into several of the *Aplysia* nerve cell bodies. By stimulating one cell and looking for reactions in the others, they can determine which cells are connected, whether connections are inhibitory or excitatory and how influential each connection is. With that technique Kandel and colleagues have discovered principles of nerve cell connections — for instance, that a signal of a single nerve cell may be interpreted as inhibitory to some of its follower cells and excitatory to others.

Although the behavioral repertoire of *Aplysia* is limited, the scientists have been able to investigate simple examples of learning at the nerve cell level. One productive topic has been the habituation of a reflex. When disturbed, an *Aplysia* protectively withdraws its gill into a respiratory chamber. However, if the animal is disturbed again and again but suffers no dire consequences, it will stop withdrawing its gill after 10 or 15 stimuli.

Kandel and collaborators found that such habituation is the result of fewer packets of neurotransmitter molecules being released by a particular nerve. That release, in turn, is due to a smaller amount of calcium flowing into the nerve cell at the release site. Reduced release of transmitter now seems to be a general mechanism for habituation. That mechanism has been indicated in studies of crayfish and cats. The large size of the *Aplysia* nerve cells gives the investigators hope that they may eventually pursue learning to subcellular, biochemical and membrane mechanisms.

While some researchers are focusing on the cellular descriptions of simple behaviors, others are exploring more thoroughly *Aplysia*'s natural behavioral repertoire — especially its versatile sexual style. *Aplysia* are hermaphroditic and take full advantage of that fact. Wayne P. Aspey of Ohio State University has observed a surprising variety of sexual encounters among *Aplysia brasiliana,* a species found in the Gulf of Mexico. "*Aplysia* have one of the most exciting copulatory repertoires around," he says.

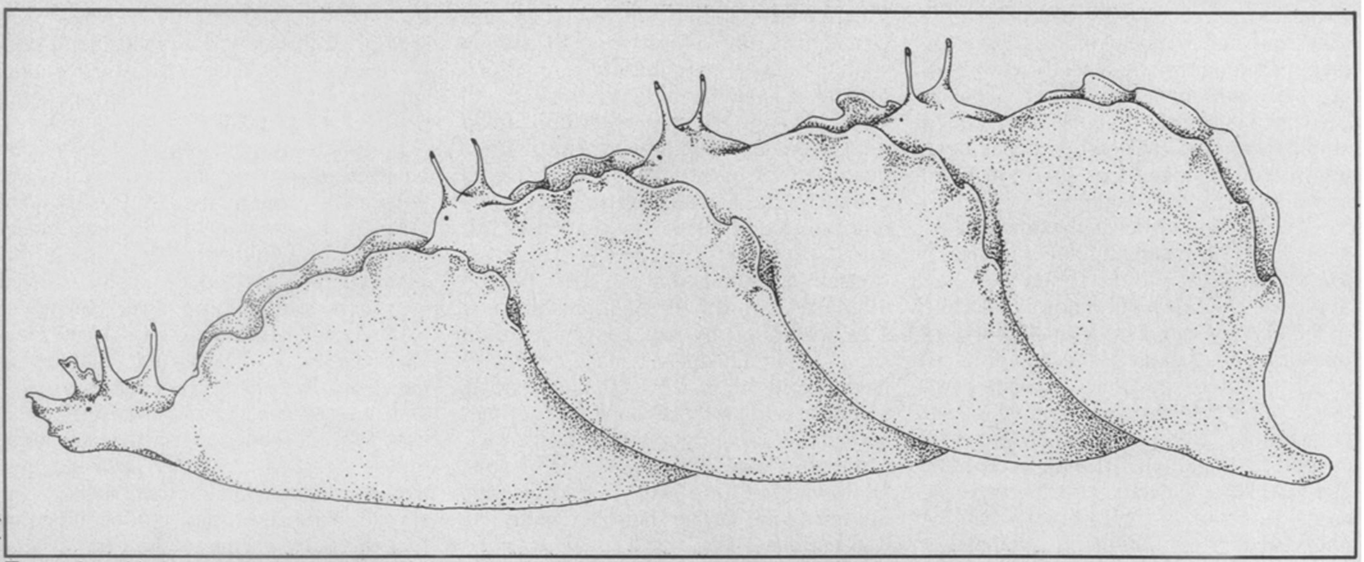
Each *Aplysia* has an extensible penis located just to the right of its mouth and a vagina in the center of its back. When two *Aplysia* mate, Aspey says, one may act as male and the other as female. Or they may both act each role simultaneously in a reciprocal copulation.

When more than two animals participate, the possibilities multiply. Two "males" may thrust their penises into one "females'" vagina or the animals may line up with the *Aplysia* at one end acting as a male, the intermediate animals serving both female and male functions and the final animal acting as a female. Four or more copulating *Aplysia* can even swing the chain into a loop, so that the end animals also couple.

Aspey noticed that *Aplysia* in a laboratory tank generally copulate in large groups. If he puts 25 *Aplysia* in the center of the tank, they disperse randomly to the edges. But within several hours, they concentrate in copulating aggregates. Aspey wanted to find out how the animals locate the action.

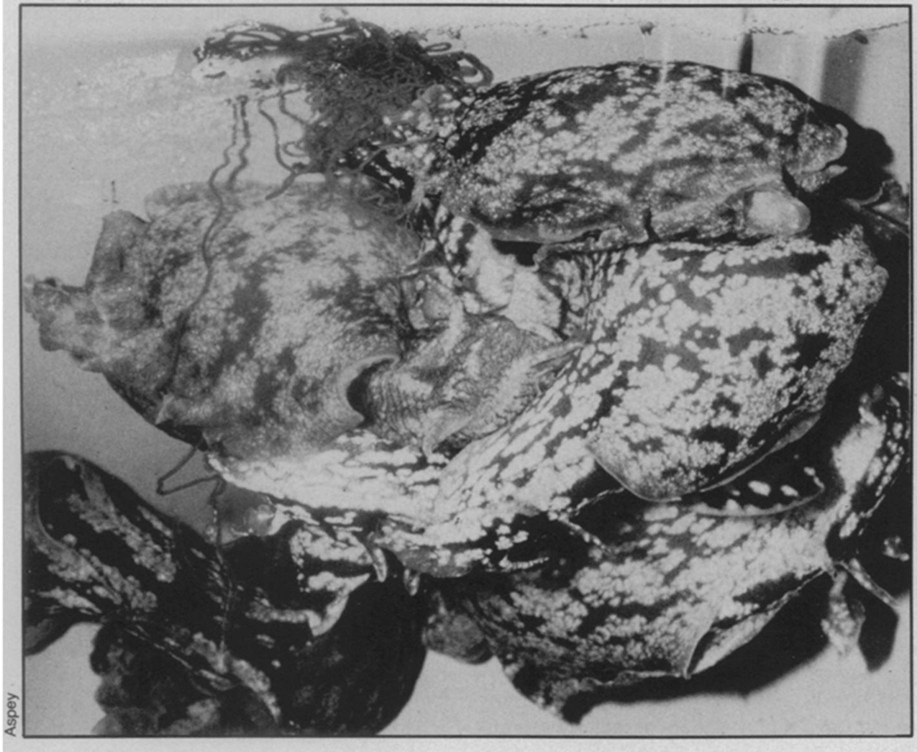
He first suspected that eggs release a chemical signal, because at the base of each aggregate usually one animal was laying eggs copiously. When an egg-laying animal is placed in the tank, other *Aplysia* orient toward it and soon swim over. That aggregation requires water flow.

Experiments with eggs and with egg extracts, however, showed that egg components can't substitute for an egg-laying animal. Aspey then began to examine extracts of various glands. He found that material from a gland called the accessory genital mass does attract *Aplysia.* Aspey suspects the aggregation signal is a product of that gland during egg laying.



Aspey

Two styles of Aplysia *"orgy": In the "Daisy chain," animals act simultaneously as male and as female. In copulating aggregates, one animal usually lays thread-like eggs copiously.*



Aspey

Although many *Aplysia* act as female and male simultaneously, some behave exclusively as one gender or the other. The animals most prolific as egg layers usually play the female role. Others, whose ovitestes contain few or no eggs, act as "exclusive" males.

Aspey and his colleagues discovered that the same sex hormones that regulate human development may govern *Aplysia* gender. In preliminary experiments, they find a surplus of the female hormone 17-beta estradiol in circulating fluid of the behavioral females and excess testosterone in the behavioral males.

In the book *Behavioral Biology of Aplysia,* Kandel predicts that analysis of mating in cellular terms will be possible in the near future. That analysis should include more biochemistry of gender and mapping of the neural circuits involved in chemical signals between animals. Even if the innate behavior of the sea hares is a far cry from human reasoning, *Aplysia* promises to be a valuable source for discovering the basic elements of nervous system function. □