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Rangia cuneata on the East Coast: Thousand Mile Range Extension, or Resurgence?

Abstract. Rangia cuneata, *a valuable clam of the estuarine zone where fluctuating salinities (from 0 to 15 parts per thousand) exclude most animals, is now developing large populations in many estuaries from Florida to Maryland. Before 1955 it was thought to be extinct on the East Coast since the Pleistocene and to be living only in Gulf Coast estuaries.*

The brackish-water clam *Rangia cuneata* (Gray in Sowerby, 1831) is a member of the marine family Mactridae but is included in some freshwater handbooks (*1*). As a living species, its range was until recently always designated as Gulf Coast estauaries of the United States and Mexico (*1, 2*). The Pleistocene range is said to have been much broader, extending north as far as New Jersey (*3*). On the Atlantic Coast from Florida to New Jersey, shells of *R. cuneata* are common in many places but have always been considered very old, mostly Pleistocene. No living individuals were reported from the East Coast until a few years ago. Now the species is extremely abundant in many estuaries from Florida to northern Maryland, and is apparently still increasing.

Wells (*4*) listed *Rangia cuneata* among the species living "in the substrate between oysters or under them" in Newport River, North Carolina, in 1955–1956. This seems to be the first recorded finding of living *R. cuneata* on the Atlantic Coast. Godwin (*5*) described dense populations of this species in the Altamaha River delta, Georgia, and quoted J. P. E. Morrison as saying that specimens had been collected there "at least as early as 1958." In Virginia (*6*) living *R. cuneata* were found in 1960 by W. G. Hewatt in Back Bay near the North Carolina line, where it was abundant by 1962, and Jon Shidler found the species living in the James River in 1963. Pfitzenmeyer and Drobeck (*7*) found *R. cuneata* abundant at several places in the Potomac River, Maryland, and gave reasons for believing that it had not been there before 1960. Chanley (*8*) reared the swimming larvae from James River *Rangia*, and quoted Morrison as believing that the "scattered populations" in Maryland and Virginia had been established longer than the 5 years suggested previously (*7*). Wolfe (*9*) measured ^{137}Cs in large populations of *R. cuneata* in the Neuse River and its tributary the Trent in North Carolina. Wolfe and Petteway (*10*) measured growth of clams in the dense Trent River population. Also in North Carolina, Tenore *et al.* (*11*) studied the effect of substrate on growth of *R. cuneata* from the Pamlico River population, which they described as abundant in a 64-km stretch, from fresh water to water of 18 parts per thousand (ppt) salinity at the river mouth.

In 1969 *R. cuneata* is even more abundant and widespread on the East Coast. It now occurs in the upper end of Chesapeake Bay and in the Sassafras River in northern Maryland (*12*), probably in other Maryland rivers, and certainly in several Virginia rivers. It seems well on the way to reoccupying all the range occupied in Pleistocene or warmer Recent times. *Rangia* populations now "pave the bottom" in many places where frequent sampling revealed none a few years ago. Shellfishery biologists familiar with the phenomenon have two theories: (i) that *R. cuneata* is a recent invader from the Gulf Coast, or (ii) that some unknown ecological change sparked resurgence of a small undiscovered population surviving since the Pleistocene in East Coast rivers. Either explanation is hard to believe, but it is undeniable that we are now witnessing a population explosion.

The distribution of *Rangia* in an estuary overlaps that of *Crassostrea virginica,* but *R. cuneata* becomes much more abundant farther up the estuary where the salinity, usually 0 to 10 ppt, is too low for oysters and for almost all other estuarine competitors or influents. A population of *R. cuneata* 40 to 50 km above the mouth of the Neches River in Texas lives in fresh water (salinity below 0.3 ppt) for at least 7 months of the year, and in salinity up to 13 ppt during low river periods, without apparent mortality. This population averages approximately 250 4-year-old clams (45 mm long) per square meter. It was estimated to produce annually 12,400 pounds of shell and 2,560 pounds of meat (wet weight) per acre (13,900 kg of shell and 2,900 kg of meat per hectare). At current prices paid to producers, the shell would be worth $25 and the meat more than $750 for a total value of at least $775 per acre ($1914 per hectare) per year (*13*). *Rangia cuneata* is of enough economic value and food value to justify expenditures to keep rivers free from pollution. It is also an ecological asset because it converts detritus into meat feeding many fishes and crustaceans (*11, 14*). It is an especially desirable addition to the estuarine fauna because it populates the zone of salinity tension where few other invertebrates can live.

On the Gulf Coast *R. cuneata* tolerates water temperatures as low as 3°C for at least a few hours, and as high as 32°C for months, without conspicuous mortality. Some mortality does occur even under "normal" conditions, however, and there are occasional die-offs without apparent cause. The Maryland populations either must withstand water temperatures lower, for longer periods, than Gulf populations ever encounter, or they will be killed by severe winters. Much of the basic information needed is still lacking, and *R. cuneata* remains perhaps the most mysterious of our common mollusks (*15*).

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15. J. P. E. Morrison, refereeing this paper, calls attention to M. Smith, *East Coast Marine Shells*, 1937, p. 65 (listing Jacksonville and Georgia as collection localities for *R. cuneata* without saying that the clams were found alive), and K. Woodburn, *Florida State Board of Conservation, Marine Lab.*, 1 Aug. 1962, p. 10 (mentioning specimens from Florida East Coast without giving collection dates), and quotes "the Game Warden of the Back Bay Region," Virginia, as saying *R. cuneata* was living in that area in his boyhood, about 1907.
16. Contribution 334 from the Virginia Institute of Marine Science, Gloucester Point, Virginia.

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L-Asparaginase-Induced Immunosuppression: Effects on Antibody-Forming Cells and Serum Titers

Abstract. *Treatment of mice with* L-*asparaginase from* Escherichia coli *resulted in a marked suppression of the immune response, as assessed both cellularly and humorally. Suppression occurred only when the enzyme was injected together with the sheep erythrocytes used as antigen. There was little or no effect when the enzyme was injected before the antigen. Simultaneous injection of asparagine prevented suppression, an indication that the effect of the enzyme was due to depletion of an amino acid probably essential for normal lymphoid cell function during antibody production.*

L-Asparaginase, purified from *Escherichia coli,* can suppress the development of leukemia, lymphoma, or sarcoma in experimental animals (*1*). The efficacy of this enzyme in suppressing leukemic cells may reflect a unique difference between normal and tumor cells in the requirement for asparagine as an essential amino acid (*1, 2*). Maintenance of a relatively high concentration of the enzyme in the blood apparently prevents sufficient quantities of asparagine reaching the tumor cells, resulting in their "starvation." Thus, the enzyme does not seem to affect leukemic cells directly, but merely decreases the extracellular concentration of an essential nutrient (*1, 2*).

Most chemotherapeutic drugs used for treatment of leukemia and other neoplastic diseases also depress the immune response (*3*). Such agents are usually metabolic "poisons" or inhibitors directly affecting rapidly dividing cells. Although it is not known whether asparagine is an essential amino acid for normal leukocytes, it seemed plausible that administration of asparaginase to a normal animal could affect the response to an antigenic stimulus which stimulates rapid proliferation of specific immunocompetent lymphoid cells. Thus we attempted to determine whether injection of this enzyme into mice, at the time of antigen injection, would affect their immune response.

Mice were injected with relatively small doses of L-asparaginase (Worthington Biochemical Corp.) at a concentration known to affect leukemic cells (*1, 2*). These mice, as well as untreated controls, were then challenged with sheep erythrocytes, and the cellular and humoral immune responses were assessed. Individual antibody plaque-forming cells (PFC) appearing in the spleens of these mice were enumerated by the hemolytic immunoplaque assay in agar gel, essentially as described by Jerne *et al.* (*4*). Serum antibody was determined by microtitration (*4*).

Normal mice injected with sheep erythrocytes alone had a rapid appearance of specific PFC's in their spleens, with the peak number appearing on day 4 after immunization (Fig. 1). Mice treated with 10 international units (I.U.) of asparaginase before immunization had essentially the same response. However, when test animals were injected with the enzyme on the day of immunization and on the following 2 days, there was a diminution of the number of antibody-forming cells detected on day 4 and on subsequent days. In general, there was a 70 to 90 percent suppression of the peak number of PFC's in these animals, as compared to controls, either when calculated per whole spleen or per million spleen cells (Fig. 1 and Table 1). Injection of asparaginase during the first 4 days after immunization resulted in almost a complete suppression of the PFC response (Fig. 1).

The effect of enzyme dose and time of injection was also studied. Mice receiving one injection of enzyme either the same day or 1 or 2 days before immunization had only a slight to moderate decrease in the number of PFC's detected 4 days later (Table 1). Two injections on days 0 and 1 resulted in a significant suppression. However, the greatest suppression occurred in mice treated with the enzyme during the first 3 or 4 days after immunization. In addition, a greater degree of suppression occurred with 10 or 50 units of enzyme, as compared to 0.5 or 5 units.

Serum antibodies were most suppressed in mice treated with enzyme on the day of immunization and the following 2 to 4 days (Table 1). One injection of enzyme had a slight to moderate effect on the titers, which were generally parallel to the effects

Table 1. Effect of time and dose of asparaginase administration on antibody response to sheep erythrocytes and spleen weight 4 days after challenge immunization. The PFC response is the average response of five or more mice per group; the differences between animals within a group was never greater than ± 30 percent.

Time	Asparaginase per injection (I.U.)	PFC response		Spleen weight (percent of control)	Serum titer (mean)
		Per spleen	Per 10^6 spleen cells		
		None			
		57,700	237	100	1:220
		Single injection of asparaginase			
Day −2	10	38,640	207	76	1:106
Day −1	10	26,290	155	66	1:64
Day 0	10	22,720	148	71	1:16
Day 0	50	10,950	59	68	1:6
		Multiple injections of asparaginase			
Days 0, +1	10	7,490	47	61	<1:2
Days 0, +1, +2	10	5,000	24	55	<1:2
Days 0, +1, +2, +3	10	2,850	3	50	<1:2
Days −6, −5, −4	10	54,000	231	100	1:200
Multiple injections of asparaginase plus 10 mg of asparagine per day					
Days 0, +1, +2, +3	10	50,650	206	92	1:235
		*Inactivated asparaginase**			
Days 0, +1, +2, +3	10	53,300	238	95	1:158

* Enzyme solution agitated in glass flask or heated at 80°C for 30 minutes; 90 to 95 percent or more of the activity was decreased.