found evidence in rabbits that drugs acting directly upon the central nervous system exert a modifying influence on serum cholesterol and on development of experimental atherosclerosis. Raab(15) recently reviewed literature showing relationship of neurohumoral agents to atherogenesis, while Vedeneeva(16) obtained histologic and electrocardiographic evidence of development of myocardial infarctions several days after administration of a large dose of adrenalin. These observations tending to implicate the neuroendocrine system in pathological deposition of lipid material in vascular walls may be related to older findings of Hausberger(17) more recently confirmed by Sidman and Fawcett(18) that lipid mobilization from, and deposition in, fat depots depends upon intact innervation to these areas.

The influence of the nervous system upon development of experimental atherosclerosis has not been studied as extensively as the possible importance of endocrine and dietary factors. The effect of chronic cardiovascular pathology upon integrative functions of the neuroendocrine system is virtually unexplored.

Our results indicate that, at least in the dog, such pathology—produced by prolonged period on an atherogenic diet—is associated with an altered hypothalamic-pituitary-adrenal response to stress. Further work is needed to clarify the site and mechanism of this altered reactivity.

Summary. Dogs maintained on an atherogenic diet to 2 years exhibited a greater response to electric shock or cold stress than did normal control animals.

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Antimicrobial Effect of Abalone Juice. (25580)

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Meat of the abalone (*Haliotis refescens*) (1) has been a common food item in China for many generations. A familiarity with the characteristics of this sea animal, which has access to a wide variety of organic material of biological origin led the author to consider it as a potentially valuable source of material in which to search for an antimicrobial agent. The investigation reported here indicates that there is, indeed, an effect of abalone upon growth of *Staphylococcus aureus* and to some extent also upon the course of experimental poliomyelitis in mice although the mechanisms involved are obscure.

TABLE I. Effect of Abalone on Experimental Poliomyelitis in Mice in 7 Trials.

Virus type	MID_{50}/mouse	Days treatment started before infection	Treated		Control	
			Paralysis or death*	%	Paralysis or death*	%
III	32	3	12/20	60	18/19	95
"	32	3	14/19	74	19/20	95
"	3	3	7/20	35	12/20	60
"	3	7	17/50	34	32/50	64
"	3	7	11/50	22	24/50	48
II	5	2	7/12	58	12/12	100
I	1	7	18/50	36	29/50	58

* No. paralyzed or dead/No. inoculated.

Materials and methods: *Assay of abalone with experimental poliomyelitis.* Abalone purchased as the canned product* was used. Each can contained about 200 ml of juice and about 250 g of meat. The juice used for experiments was removed aseptically, pooled and stored at 4°C. After a few days storage the juice separated into 2 layers; the upper two-thirds was brownish yellow in color and fairly clear, and the lower one-third was grayish white in color and quite cloudy. Sometimes the upper layer became a gelatinous semi-solid. The whole juice was warmed and the 2 layers well mixed before use. Mouse and tissue culture adapted type I (Mahoney) type II (Lansing) and type III (Leon) polioviruses all maintained in our laboratory and grown in monkey kidney tissue culture were used. General purpose Swiss mice (obtained from Animal Production Section, DBS, NIH), weighing 12-15 g were inoculated with one to 32 mouse infective doses in 0.02 ml of type I or III virus by intraspinal (IS) route and observed for 2 weeks. For type II virus, mice weighing 17-22 g were inoculated with 5 mouse infective doses in 0.03 ml by intracerebral (IC) route and observed for 3 weeks. Treated mice were fed with abalone juice. Approximately 1 ml of the juice was given to each mouse/day placed on whole wheat bread or mixed with commercial mouse diet. The control mice were fed with regular mouse diet. *Assay of Abalone with Staphylococcus aureus.* At beginning of present study, the canned abalone juice was tested for inhibiting effect on *Staphylococcus aureus* by absorbent paper disc method and results were inconsistent. Toward end of the work, frozen fresh abalone† was secured and some brownish juice was obtained upon thawing. This brownish quice was centrifuged and heated to 70°C for 30 minutes before use. A penicillin sensitive strain of *Staphylococcus aureus*, FDA 209, was used as test organism. The test was carried out as follows.§ Ten-fold dilutions of a 24 hour culture of *Staphylococcus aureus* in buffered saline were made and a loopful of each dilution was inoculated into a Wassermann tube containing 1.8 ml of nutrient broth with 0.2 ml of the heated abalone juice. The same dilutions were also inoculated into control tubes without abalone. After incubating at 35°C for about 18 hours, 0.1 ml from each tube was spread over surface of nutrient agar plate. The plates were then incubated overnight and colonies on each plate counted.

Results. The results on experimental poliomyelitis in mice are summarized in Table I. In treated mice the rates of paralysis and death were lower, average incubation period was longer and paralysis was less severe than in control mice. All experiments showed a similar curve of accumulative percentage of paralysis or death. Examples are shown in Figs. 1 and 2.

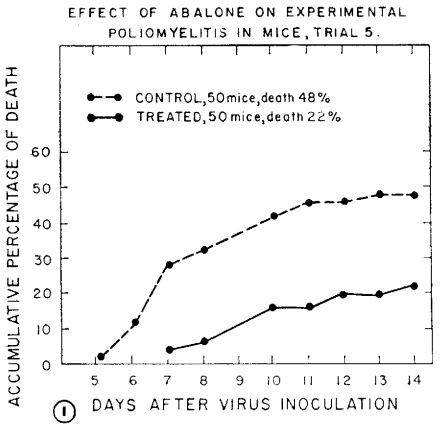
The effect of abalone on growth of *Staphylococcus aureus* is illustrated by 2 sample experiments shown in Table II. Both experiments were carried out with the penicillin sensitive strain, FDA 209. They were repeated

* Calmex Brand of Marine Products Co., San Diego, Calif. or Brand of Chong Kee Co., N. Y.

† Frozen fresh abalone was kindly supplied by Dr. H. G. Orcutt, Bureau of Marine Fisheries, Calif.

‡ This culture was kindly supplied by Dr. A. L. Schade of National Institute of Allergy and Infectious Diseases.

§ This method of testing was suggested by Dr. C. W. Hiatt of this laboratory.



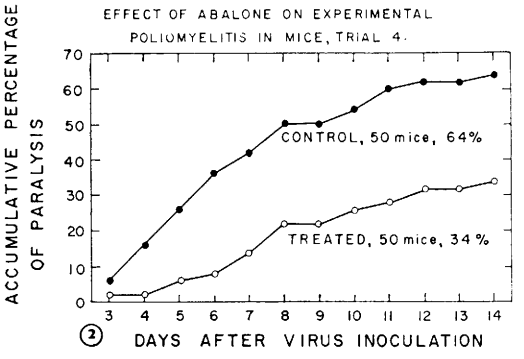


FIG. 1 and 2.

several times and consistent and similar results were obtained. In some experiments the crude juice from frozen fresh abalone was heated to 95°C for 45 minutes or dialyzed against water overnight and its effect on *Staphylococcus aureus* was not impaired.

Discussion. In reviewing the literature for antiviral(2-4) and bacterial agents, it was noticed that efforts have been largely concentrated in the field of microbial products. It is now felt necessary to shift toward some other material. Because of abundance and variety of organic materials in sea water, and the known ability of gastropods to filter and concentrate suspended solids of their aquatic milieu, certain sea animals such as abalone, may be regarded as a potential source in the search for antimicrobial agents. Our experiments demonstrated clearly antimicrobial action of crude fresh abalone juice against *Staphylococcus aureus,* although the effect of canned abalone juice against experimental poliomyelitis in mice was only moderate. Thermostability and nondialyzability of the agent suggested that it is not an ordinary antibiotic. One of the chief advantages of its possible use for therapeutic purpose is that it is edible as a food, and therefore non-toxic. Further work on purification and concentration is in progress.

TABLE II. Effect of Abalone Juice on Growth of *Staphylococcus aureus*, FDA 209 Strain.

Trial	Abalone in broth	Colonial growth from 0.1 ml of 18 hr broth cultures inoculated with one loopful of serial dilutions 10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	Control*
1	+	36†	8	0	0	0	0
	—	Con.‡	Con.	Con.	Con.	Con.	0
2	+	84	4	0	0	0	0
	—	Con.	Con.	Con.	Con.	TN§	0

* Control: not inoculated.
† Besides 36 colonies there were 2 small patches of growth.
‡ Confluent growth.
§ Colonies too numerous to be counted but not confluent.

Summary. Commercial canned abalone juice given to Swiss mice orally possessed sparing effect against intraspinal infection of type I and III poliovirus and intracerebral infection of type II poliovirus. Crude fresh abalone juice also possessed marked inhibitory effect against *Staphylococcus aureus.*

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‖ Dialysis was kindly carried out by Dr. A. K. Saz of Nat. Inst. of Allergy and Infectious Diseases.

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