

Table 1. Antibody activity in the acid eluates. Indirect hemagglutinins (IHA) are expressed as the reciprocal of the titer of type 12 specific hemagglutinins. Bacterial agglutinins (BA): C, complete; 0, no agglutination of type 12 (T12) and type 6 (T6) streptococci. Indirect fluorescent antibody (IFA) assay: degree of fluorescence is expressed as none (0), definite but dull (1+), and maximum brilliance (4+).

Eluate	Protein (mg/100 ml)	IHA T12	BA		IFA		
			T12	T6	T12	T6	Kidney
A-12	112	<4	0	0	1+	0	0
A-12N	35	32	C	0	4+	0	0

the serums of animals in the two groups. Significant deposits of fixed  $\gamma$ -globulin were present in the glomeruli of 20 out of the 26 rats exposed to A12N strain, but no deposits were present in the 15 rats exposed to A12 strain. The tissues from the rats exposed to the two strains were pooled separately. The renal cortices were separated from the medullas, weighed, and ground in a Ten Broeck tissue homogenizer. Dissociation of the fixed  $\gamma$ -globulins was attempted in alkaline (pH 9.0) and acid (pH 3.2) borate buffers (3).

The presence and specificity of antibody in the eluates was assessed with indirect hemagglutination (IHA), bacterial agglutination (BA), and indirect fluorescent antibody (IFA) techniques. In the IHA test we used red cells sensitized with purified M protein and a soluble inhibitor to block cross-reactive antibodies that were not type-specific (4). Bacterial agglutinins were

assayed as follows. A culture of the test strain was grown overnight in tryptic soy broth. The viable bacterial sediment was washed and resuspended in saline to the original volume. The viable suspension (0.5 ml) was incubated with the eluate (1 ml) at 37°C in a water bath for 1 hour. Wet mounts were prepared and examined with a darkfield microscope for complete, partial, or no agglutination. The IFA test consisted of fixation of the washed test strain of bacteria or of 4.0- $\mu$  sections of rat cortex to glass slides and was performed with fluorescein-labeled rabbit antiserum to rat  $\gamma$ -globulin (Antibodies, Inc.) (2).

Immunoglobulins were recovered only in the acid-eluted fraction and were specific for streptococcal M protein. Although the final protein concentration of the nonnephritogenic eluate was at least three times greater than that of the nephritogenic eluate, only the latter contained appreciable amounts of antibody (Table 1). The specificity of this antibody for type 12 M protein was demonstrated by its reactivity with purified type 12 M protein (IHA test) and with group A, type 12 bacteria (BA and IFA test) and by its failure to react with a strain of group A, type 6 (BA and IFA tests). In addition, this antibody did not react with renal antigens as assessed by the indirect fluorescent method.

These antibodies were characterized further by immunoelectrophoresis (5) and sedimentation on a sucrose gradient (6). A single protein band was found on immunoelectrophoresis of the eluate and corresponded to the IgG band of normal rat serum (Fig. 1). Separation of the eluate on a sucrose gradient revealed that the immunoglobulins sedimented in the same range as 7S human immunoglobulins (Fig. 2).

Immunoglobulins eluted from the kidneys of rats with experimental streptococcal glomerulonephritis are of the IgG class and react with type 12 streptococcal M protein but not with other streptococcal antigens or with renal antigens. This observation directly implicates type 12 M protein and type-specific antibody as the immunologic reactants in the production of this disease. Because of the similarity of this experimental model to the disease observed in man, it seems possible that these same reactants may also mediate the production of the disease in man. Still undetermined is the difference between nephritogenic and nonnephritogenic strains which leads to the forma-

tion and fixation of such complexes only in the kidneys of animals and humans exposed to the nephritogenic strains.

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7. Supported by PHS grants T1-AI-185 and AI-06964. L.H. is a National Science Foundation science faculty fellow. We thank M. Klopp and N. Steingold for technical assistance.

19 June 1969; revised 18 August 1969

## Diversity and Composition of Abyssal Benthos

Sanders and Hessler (1) present some important data obtained by use of their anchor dredge and epibenthic sled, and by application of their rarefaction procedure to assess diversity independent of sample size. However, some of their conclusions are not unequivocal and others have been proposed previously.

The theory that "the deep sea harbors a qualitatively restricted fauna" has been adequately disproved (2, 3, and others). Sanders and Hessler say that they did not expect to find so many deposit-feeding species. Research on deep-sea mollusks (3) has already shown that deposit feeders are dominant in the deep sea.

The statement, "... the implication, at least for bivalves is that faunal composition is far more sensitive to change in depth than to the effects of distance" is insufficiently documented. The accompanying discussion also omits any reference to pertinent previous work

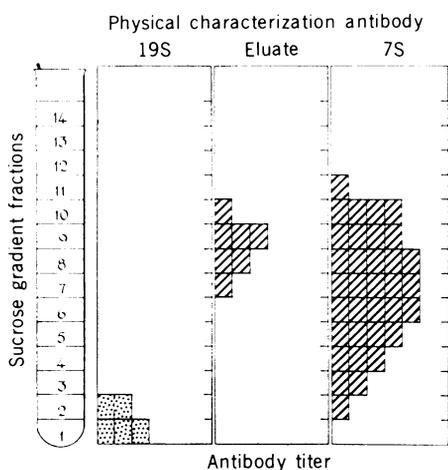


Fig. 2. A comparison of the sedimentation rate of the type 12 specific hemagglutinins in the acid eluate of the kidney with 7S and 19S human hemagglutinins for the lipopolysaccharide of *Escherichia coli* O4. The stippled and cross-hatched blocks represent twofold dilutions of the gradient fractions starting at a dilution of 1:2.

(3) in which a conflicting viewpoint is developed in regard to deep-sea bivalves.

The statements, "Among the bivalves, the transition of faunal change at the shelf-slope break is even more pronounced" and "We believe that this faunal break is related to temperature" (1, p. 1421), are not accompanied by any reference to evidence that this break may be related to the lower limit of the euphotic zone (3, 4) and to the shifts in feeding mechanisms necessitated by reduced concentrations of suspended food below this limit.

Many will disagree with the application of "the principle of competitive exclusion" as related to sparse populations and to extrapolation of results, principally derived from studies of the deep-sea fauna between Woods Hole and Bermuda, to form the basis of generalizations applicable to the world ocean. There are reasons to believe that the deep-sea region near Bermuda is one of unusually high diversity.

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- 25 April 1969; revised 14 July 1969

We have once again read the papers cited by Clarke (1) and find no direct statement to the effect that the benthic fauna of the deep sea is diverse. If such was the implication it is not evident to the reader, nor do the data presented warrant such a conclusion.

We agree with Clarke's statement that deposit feeding plays a major trophic role among abyssal bivalves. However, we are attempting to arrive at some general conclusions about the deep-sea benthos, which are based on a closer approximation of its total fauna rather than limiting ourselves to a restricted element of that fauna. Bivalves contribute less than 10 percent of the species in our initial five epibenthic sled samples, a percentage about typical of what we found in our later samples. The deposit-feeding habit also plays a dominant role for the vast species array of peracarid crustaceans (40 percent of the species in the same five epibenthic samples, a percentage generally applicable for unwinned

samples by this sampling gear), the polychaetes (approximately 22 percent of the species), and such less taxonomically diverse groups as the sipunculids, holothurians, and, to a lesser degree, the gastropods, ophiuroids, and asteroids.

Regarding Clarke's criticism that there is insufficient documentation for our statement "the implication, at least for bivalves is that faunal composition is far more sensitive to change in depth than to the effects of distance," there is adequate documentation. If, for example, we chose 2890 or 3840 meters instead of 1400 meters as reference depths, for a change of 1000 meters vertically (depth) it is necessary in each case to move thousands of kilometers horizontally (distance) at the reference depths to obtain an equivalent faunal change. Clarke (2) documents the fundamental differences between the benthic mollusk fauna of the abyss as compared to shallow water. On the other hand, he claims that the deep-sea benthos present in the different regions of the ocean are derived from the fauna of the adjacent shallow seas. Since diversity in shallow tropical seas is greater than in the boreal zone, he therefore concludes that diversity in the abyss at low latitudes is greater than at high latitudes. We agree with Clarke's first conclusion but not with his second. Because of the fundamental differences between the shallow and abyssal mollusks (equally true for many other groups), the great radiation of deep-sea taxa occurred *in situ*. Many species at bathyal and abyssal depths are found both at low and high latitudes. We are unable to perceive a pattern in our samples suggesting a strikingly greater benthic diversity in low latitudes.

We think that Clarke's next point is not relevant. All faunal samples described in our report, both on the shelf above the zone of transition and at bathyal and abyssal depths, came from soft oozes and, therefore, are depositional environments. In such sediments the deposit feeder is the dominant feeding type. Thus there is no need to confront the issue with "shifts in feeding mechanisms necessitated by reduced concentration of suspended food. . . ." Both the constituent species and the kinds of deposit feeders are decidedly different above and below the zone of transition. This we show in Fig. 3 of our paper and discuss in some detail (3, p. 1421).

If Clarke is correct in his prop-

osition that competition is significantly reduced where animal density is low, there should be a greater number of species per unit number of individuals in low as compared to high density situations. A comparison of the numerically impoverished abyss under the Sargasso Sea with the large density of animals on the continental slope south of New England suggests that such an interpretation is not tenable.

Besides collecting from our transect between Woods Hole and Bermuda, we have collected thousands of deep-sea benthic animals from the Cape, Angola, and Sierra Leone basins off West Africa, and from the Brazil Basin, and have received numerous bivalves from the West European and Canaries basins. All the material examined so far supports our generalizations.

Concerning the remark that "There are reasons to believe that the deep-sea region near Bermuda is one of unusually high diversity," we cannot find the evidence to support it. Certainly it is not to be found in the three published reports of the deep-sea mollusks of that area: at H.M.S. *Challenger* station 56 (4), where a total of 13 mollusk species were collected; the valves of seven species dredged from two stations by the R.V. *Caryn* (5); and the two stations of the R.V. *Theta* (6), which yielded a total of eight species. Considering only bivalves, our Bermuda station 119 provided 18 species from a total of 515 living individuals at time of capture. With corrections for the differing numbers of individuals, the diversity found at this station is in no way higher than the values we find in other regions of the Atlantic Ocean.

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1 August 1969; revised 9 October 1969

# Science

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*Science* **166** (3908), 1033-1034.  
DOI: 10.1126/science.166.3908.1033

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