These antisera showed high NA against HTLV-III$_B$ and HX10, but NA against HXB2 and HXB3 was substantially lower in every case (Table 2). Neutralizing activity of antiserum to Sub2 against HTLV-III$_B$ and HX10 was seen at dilutions of ≥1:2048 to ≥1:4096, but NA against comparable inocula of HXB2 and HXB3 was lost at dilutions of ≥1:23 to ≥1:281. A murine monoclonal antibody to HTLV-III$_B$–derived gp120 (96–16) displayed similar specificity. In contrast, goat antisera against recombinant gp160 derived from the biologically inactive BH8 molecular clone (IMM-6) showed NA against HXB2 that was 64 times as high as NA against HTLV-III$_B$. The amino acid sequence of HXB2 differs from that of HX10 in the Sub2 region at only two positions (289 and 305), and those of BH8 and HXB3 differ at these same locations, including the presence of arginine or lysine instead of serine (representing a single nucleotide change) within a possible cysteine loop structure (*20*) that lies in the amino-terminal portion of Sub2 (Fig. 2).

In a series of competition experiments, we used synthetic oligopeptides spanning the RP135 or RP136 regions containing residues at the 289 and 305 positions corresponding to those of HX10, HXB2, or HXB3 (Fig. 2 and Table 3). Sera G2935 and 987 completely inhibited infection by HTLV-III$_B$ in the absence of RP136 but showed no inhibition from control values when RP136 was added. In contrast, inhibition by a patient serum (HS-25) was not blocked by RP136. Peptides 51.4 and 80-1 (corresponding to HX10 at position 305) were significantly more effective than 51.20, 51.4.20, 80-2, or 80-3 (peptides containing either threonine or glutamine at position 289 but with arginine or lysine instead of serine at 305 as in HXB2, HXB3, and BH8) in blocking the neutralizing effects of G2935 and 987 on HX10 infection, supporting the conclusion that a change at the 305 position is sufficient to account for the observed restriction of NA in heterologous sera.

Although it is unlikely that differences in areas other than the *env* region are responsible for this restriction of NA, conceivably changes in other genes might alter gp120 processing, presentation, or stability (*21*). Also, differences at distant sites in gp120 or gp41 could induce conformational changes in the RP135 region. Finally, while there was no difference in the average titers of the panel of human sera against these viruses, suggesting HXB2 and HXB3 are not intrinsically more resistant to neutralization, such might not be the case for the specific goat antisera studied.

Our data suggest that a single amino acid change in the virus envelope may result in profound changes in recognition by neutralizing antisera, and extend the previous concept of type-specificity of neutralization (*20*) to differing variants of a single isolate. This suggests that the group-specific NA of some human sera could be directed against a large number of type-specific determinants, as well as conserved epitopes. This could have important implications for vaccine development. Inability to demonstrate protection by candidate vaccines after challenge with homologous virus isolates could represent the selection of minor clonal variants not well recognized by the immunized host (*22, 23*). Only the use of cloned HIV-1 variants will permit direct evaluation of the hypothesis that neutralizing antibodies are capable of protecting against HIV-1 infection.

REFERENCES AND NOTES

1. M. Robert-Guroff, M. Brown, R. C. Gallo, *Nature* **316**, 72 (1985).
2. R. A. Weiss *et al.*, *ibid.*, p. 69.
3. D. D. Ho, T. R. Rota, M. S. Hirsch, *N. Engl. J. Med.* **312**, 649 (1985).
4. S. Rasheed *et al.*, *Virology* **150**, 1 (1986).
5. S. Harada, Y. Koyanagi, N. Yamamoto, *Science* **229**, 563 (1985).
6. S. D. Putney *et al.*, *ibid.* **234**, 1392 (1986).
7. L. A. Lasky *et al.*, *ibid.* **233**, 209 (1986).
8. K. S. Steimer *et al.*, in *Vaccines '88* (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, in press).
9. T. C. Chanh *et al.*, *EMBO J.* **5**, 3065 (1986).
10. T. J. Matthews *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **83**, 9709 (1986).
11. S. Matsushita *et al.*, *J. Virol.* **62**, 2107 (1988).
12. R. A. Weiss *et al.*, *Nature* **324**, 572 (1986).
13. B. H. Hahn *et al.*, *Science* **232**, 1548 (1986); M. Saag *et al.*, *Nature*, in press; A. G. Fisher *et al.*, *ibid.*, in press.
14. G. M. Shaw *et al.*, *Science* **226**, 1165 (1984).
15. B. Hahn *et al.*, *Nature* **312**, 166 (1984).
16. A. G. Fisher *et al.*, *ibid.* **316**, 262 (1985).
17. L. Ratner *et al.*, *AIDS Res. Hum. Retrovir.* **3**, 57 (1987).
18. R. Crowl *et al.*, *Cell* **41**, 979 (1985).
19. W. G. Robey *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **83**, 7023 (1986).
20. J. R. Rusche *et al.*, *ibid.* **85**, 3198 (1988).
21. A. G. Fisher *et al.*, *Science* **237**, 888 (1987).
22. P. L. Nara *et al.*, *J. Virol.* **61**, 3173 (1987).
23. S.-L. Hu *et al.*, *Nature* **328**, 721 (1987).
24. G. Meyers *et al.*, Eds., *Human Retroviruses and AIDS, 1987*, (Los Alamos National Laboratory, Los Alamos, NM, 1987), vol. 2, p. 38; L. Ratner *et al.*, *Nature* **313**, 277 (1984).

25. We thank S. Petteway, L. Ivanoff, D. Waselefsky (Dupont), and J. Derge (Frederick Cancer Research Facility) for synthesis of oligopeptides, F. Dorner (IMMUNO) for goat antisera to gp160, A. Bodner (Biotech Research) for monoclonal antibody 96-16, and L. Arthur and G. Robey for goat antisera to gp120. We also thank A. Rose, S. Kensek, V. Kao, L. Simek, A. Bodner, and J. Lemp for technical support and assistance.

12 January 1988; accepted 6 May 1988

Acoustical Detection of High-Density Krill Demersal Layers in the Submarine Canyons off Georges Bank

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High-density demersal layers of krill have been detected in the submarine canyons off Georges Bank by means of a high-frequency, dual-beam bioacoustical technique. Krill densities in these demersal layers were observed to be two to three orders of magnitude greater than the highest densities observed in water-column scattering layers. Such abundances may help explain the unusually high squid and demersal fish production estimates attributed to the Georges Bank ecosystem.

EUPHAUSIIDS TYPICALLY PLAY A MAJOR role in the economy of pelagic marine ecosystems. In the productive waters of the North Atlantic, the species *Meganyctiphanes norvegica* provides an important link in the food chain between lower trophic level plankton and higher trophic level consumers, including decapod crustaceans, squids, fishes, marine mammals, and birds (*1, 2*). Early Norwegian whalers referred to *M. norvegica* as krill, a term that has now been expanded to encompass all species of euphausiids (*3*). A feature of krill ecology that makes them particularly vulnerable to successful exploitation by higher trophic level consumers is their tendency to form highly aggregated distributions (*1, 2, 4*). Krill aggregations have been categorized into four basic types: patches, shoals, swarms, and schools, with the last two types corresponding to high-density aggregations of more than 1000 animals per cubic meter (*2*). Such high-density aggregations of *M. norvegica* have been reported, but those reports have been of surface swarms often associated

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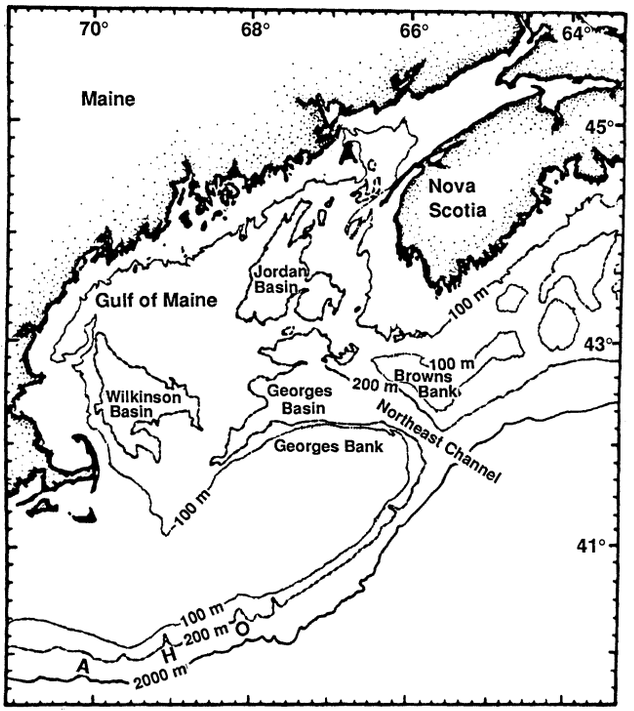

Fig. 1. Locations of submarine canyons south of Georges Bank where submersible dives were conducted: Atlantis Canyon, A; Hydrographer Canyon, H; and Oceanographer Canyon, O.

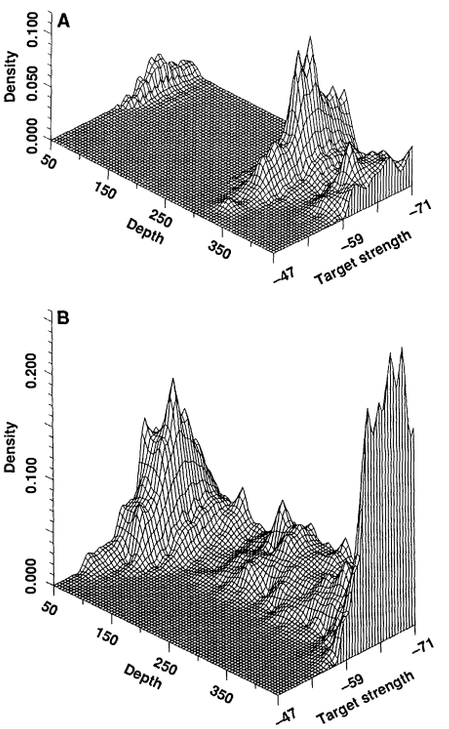

Fig. 2. (**A**) Daytime vertical profile of animal densities from the water column above Hydrographer Canyon. Densities (in animals per cubic meter) for each depth interval (in meters) were apportioned to different target strength classes (in decibels) (*6*). (**B**) Nighttime vertical profile from water column above Hydrographer Canyon.

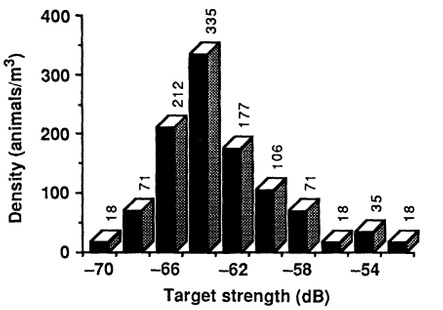

Fig. 3. Density estimates of different acoustical size classes of animals in the 50-m-thick demersal layer near the bottom of Oceanographer Canyon. Krill size range corresponds to target strength classes between −70 and −62 dB. Functional regressions for converting target strengths to lengths and wet weights are in (*6*).

with breeding behavior (*1, 2, 5*). Subsurface swarms and schools of krill are much more difficult to detect and survey with conventional sampling gear. In this report, we present acoustical evidence for the occurrence of high-density demersal layers of *M. norvegica* in the submarine canyons off Georges Bank.

The high-density layers of krill were detected during our September 1987 cruise to field test a high-frequency, dual-beam bioacoustical technique for studying zooplankton and micronekton distributions. This technique differs from previous acoustical techniques in that it enables investigators to analyze the echoes returning from individual animals, thereby providing direct estimates of the size distribution of the animal assemblage (*6*). We conducted acoustical profiling surveys of the water column from approximately 30 m off the bottom to the sea surface in three of the submarine canyons east of southern New England and south of Georges Bank (Fig. 1). All surveys were conducted on dives aboard the Johnson Sea Link submersible (*7*).

Typical profiles of the daytime and nighttime water-column distributions of acoustical targets in these canyons are presented in Fig. 2. The most abundant acoustical size classes, between approximately −62 and −71 dB, correspond to animals the size of mature krill. *M. norvegica* dominated the krill biomass at all dive sites, although several species of *Thysanoessa* were also common. During the day, krill densities were low in the upper 300 to 350 m of the water column and higher at greater depths. In contrast, during the night, krill were more uniformly distributed throughout the water column but often formed a strong near-surface scattering layer. A simple day-night comparison of the distributional patterns reveals two important features of krill ecology. First, changes in animal density in the upper 300 m of the water column suggest that krill exhibit vertical migrations of at least 300 to 400 m each way per day; second, an apparent increase in animals integrated over the water column during the night relative to the day suggests that more krill reside by day in high-density layers on or near the bottom (*8*).

Previous studies of *M. norvegica* with conventional sampling gear have yielded qualitative results consistent with our findings of extensive vertical migrations and high-density demersal layers (*1, 2, 9*). An unexpected finding was the magnitude of the densities observed in these demersal layers. During one dive in Oceanographer Canyon (Fig. 1), we conducted an acoustical survey of the demersal layer from 50 m above the sea floor to the bottom. Krill densities in this layer exceeded 1000 animals per cubic meter in many places and averaged 813 animals per cubic meter over the full 50-m layer (Fig. 3). These densities are two to three orders of magnitude greater than the highest densities that we estimated in the upper part of the water column. Such densities are unusually high for *M. norvegica* and other North Atlantic krill (*1, 2*), corresponding to an average biomass concentration of 614 g wet weight per cubic meter and a layer-integrated biomass of 30.7 kg wet weight per square meter.

We hypothesize that these high-density demersal layers are formed as the result of the interaction of depth and topography with the behavioral ecology of krill. Earlier studies have shown that underwater light levels regulate the diel vertical migrations of krill, which remain in coherent scattering layers as they migrate (*10*). In shallow oce-

anic regions, these scattering layers may coalesce in a high-density layer at the bottom when the krill migrate downward in the morning, thereby greatly concentrating the animals present (*11*). In addition, the topography of the submarine canyons may cause a funneling effect that would further concentrate the animals at the canyon bottom (*12*).

Better documentation of the spatial extent and temporal persistence of high-density demersal layers of krill will be essential for assessing the importance of these layers in high-latitude marine ecosystems (*13*). The Georges Bank ecosystem provides a good example. Georges Bank is one of the world's most productive fishing grounds (*14–17*), and krill are an important but variable dietary component of the Bank's commercially important squid and demersal fish stocks, including long- and short-finned squid, cod, flounder, haddock, hake, pollock, and redfish (*1, 18–20*). Recent analyses of secondary production on Georges Bank indicate that the levels of zooplankton and benthic production there are insufficient to support the high levels of squid and demersal fish production (*16, 21*). Hypotheses suggesting that there are high trophic efficiencies within the ecosystem do not seem to explain this discrepancy (*16*). A more plausible hypothesis is that squid and demersal fish stocks on Georges Bank are subsidized by exploiting krill production in the canyons and deep waters surrounding the Bank (*22*). Although krill rarely intrude on the shallower parts of Georges Bank, many squid and demersal fishes seasonally inhabit the deep, surrounding waters (*23*). Closer examination of the spatial and temporal coupling between predator and prey populations will determine the viability of this hypothesis. If it proves correct, then the excess squid and demersal fish production associated with Georges Bank may be attributed to the existence of high-density demersal layers of krill in the deep, surrounding waters.

REFERENCES AND NOTES

1. J. Mauchline and L. R. Fisher, *Adv. Mar. Res.* **7**, 1 (1969).
2. J. Mauchline, *ibid.* **18**, 371 (1980).
3. ———, *Euphausiid, Stomatopod and Leptostracan Crustaceans* (Brill-Backhuys, London, 1984).
4. P. F. Brodie, D. D. Sameoto, R. W. Sheldon, *Limnol. Oceanogr.* **23**, 1264 (1978).
5. R. G. B. Brown, S. P. Barker, D. E. Gaskin, *Can. J. Zool.* **57**, 2285 (1979); S. Nicol, *Mar. Biol.* **18**, 241 (1984).
6. The dual-beam technique of in situ target strength estimation provides data on the relative proportions of different acoustical target strength classes [J. J. Traynor and J. E. Ehrenberg, *J. Fish. Res. Board Can.* **36**, 1065 (1979)]. In combination with echo integration estimates of volume backscattering, the two techniques can be used to estimate total density and to apportion that density among the various target strength classes as follows: $sv = N\Sigma p_i \cdot \sigma_i$ where sv is the volume backscattering coefficient, N is the total density of targets, p_i is the proportion of targets in target strength class i, and σ_i is the backscattering cross section of targets in target strength class i. With 420-kHz sound, acoustical target strengths (TS) can be related to animal lengths (L) and wet weights (WW) by the following functional regressions: $TS = -114.2 + 31.1\log(L)$ and $TS = -93.7 + 10.3\log(WW)$ (C. H. Greene, P. H. Wiebe, J. Burczynski, *Limnol. Oceanogr.*, in press).
7. M. J. Youngbluth, *Proceedings of Submarine Technology '83 Symposium*, London (Society of Underwater Technology, London, 1984), p. 335.
8. We interpret the observed nighttime increase in integrated water-column abundance as evidence that a greater fraction of the krill population resides by day in a dense demersal layer not included in our initial surveys. Krill avoidance of the submersible during daylight hours could be an alternative explanation for the observed nighttime increase. However, we believe that this explanation is unlikely because in situ acoustical experiments revealed no evidence of krill being attracted to or repelled by the submersible when its lights were off. Zooplankton patchiness could also have accounted for the apparent increase in nighttime krill abundance, especially because some of the other profiles do not reveal such dramatic day-night differences. Additional replicate comparisons are required to confirm our interpretation of these day-night differences.
9. C. F. Hickling, *J. Mar. Biol. Assoc. U.K.* **13**, 735 (1925); H. B. Bigelow, *Bull. Bur. Fish. Wash.* **40** (no. 2), 1 (1926); R. MacDonald, *J. Mar. Biol. Assoc. U.K.* **14**, 753 (1927); J. Hjort and J. T. Ruud, *Rapp. P.-V. Reun. Cons. Int. Explor. Mer* **56**, 5 (1929).
10. G. H. Tucker, *J. Mar. Res.* **10**, 215 (1951); B. P. Boden and E. M. Kampa, *Symp. Zool. Soc. Lond.* **19**, 15 (1967).
11. J. D. Isaacs and R. A. Schwartzlose, *Science* **150**, 1810 (1965).
12. J. A. Koslow and A. Ota, *Biol. Oceanogr.* **1**, 107 (1981).
13. Quantitative surveys of krill demersal layers are virtually impossible to conduct with conventional sampling techniques that make use of plankton nets or benthic sleds (*1, 2*). The recent advent of remote, high-resolution bioacoustical techniques will allow such quantitative surveys to be feasible in the near future [R. R. Pieper and D. V. Holliday, *J. Cons. Int. Explor. Mer* **41**, 226 (1984); K. E. Richter, *Deep-Sea Res.* **32**, 163 (1985); C. H. Greene and P. H. Wiebe, *Sea Technol.*, in press].
14. D. W. Bourne, in *Georges Bank*, R. H. Backus and D. W. Bourne, Eds. (Massachusetts Institute of Technology Press, Cambridge, 1987), pp. 252–255.
15. B. E. Brown, *ibid.*, pp. 480–493.
16. E. B. Cohen and M. D. Grosslein, *ibid.*, pp. 383–391.
17. M. P. Sissenwine, *ibid.*, pp. 347–350.
18. H. B. Bigelow and W. W. Welsh, *Bull. Bur. Fish. Wash.* **40** (no. 1), 1 (1924).
19. R. E. Bowman and W. L. Michaels, *Natl. Oceanic Atmos. Adm. Tech. Mem. Natl. Mar. Fish. Service/Northeast Center 18* (1984).
20. M. D. Grosslein and T. R. Azarovitz, *N.Y. Bight Atlas Monogr.* **15** (1982).
21. The paradox is in Georges Bank squid and demersal fish production because pelagic fish production is comparable to that in the North Sea and in line with estimates of zooplankton secondary production [E. B. Cohen, D. G. Mountain, S. H. Clark, *J. Cons. Int. Explor. Mer* **50**, 47 (1982); K. Sherman *et al.*, in *Georges Bank*, R. H. Backus and D. W. Bourne, Eds. (Massachusetts Institute of Technology Press, Cambridge, 1987), pp. 268–282; F. W. Steimle, *ibid.*, pp. 310–314].
22. High-density demersal layers of krill have been observed visually in the deep waters of the Gulf of Maine as well [M. J. Youngbluth, T. G. Bailey, P. J. Davoll, *Eos* **66**, 1264 (1985); M. J. Youngbluth *et al.*, *Natl. Oceanic Atmos. Adm. Symp. Ser. Undersea Res.*, in press]; however, acoustical surveys of these layers have not been conducted yet.
23. Deep-water fish assemblages adjacent to Georges Bank include the Gulf of Maine deep, Northeast Peak, intermediate, and slope and canyon assemblages (*15*). Several of the species in these assemblages migrate seasonally on and off Georges Bank (*20*); E. D. Anderson, *Natl. Oceanic Atmos. Adm. Tech. Mem. Natl. Mar. Fish. Service/Northeast Center 29* (1984); A. M. Lange and J. E. Palmer, *Natl. Oceanic Atmos. Tech. Mem. Natl. Mar. Fish Service/Northeast Center 39* (1985)].

24. We thank the captain and crew of the R.V. *Seward Johnson* and sub crew of the *Johnson Sea Link* for contributing to a successful cruise. We thank the director and staff at Friday Harbor Laboratories for providing facilities to conduct our calibration studies. This research was supported by the Office of Naval Research with ship and submersible time provided by the National Oceanic and Atmospheric Administration National Undersea Research Program.

14 March 1988; accepted 7 June 1988