

SOME ONGOING RESEARCH IN ARCTIC MARINE BIOLOGY



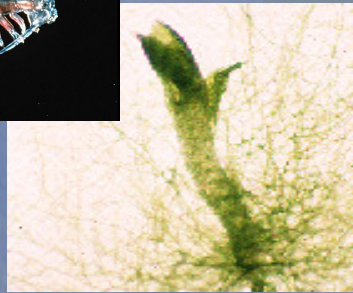
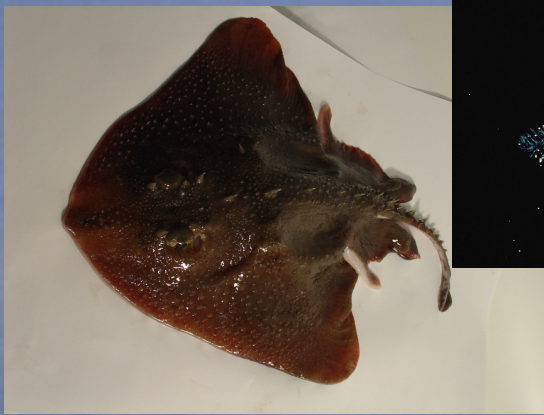
Understanding the present genomic, molecular and phenotypic characters of polar fauna requires consideration of the shaping and engineering of these characters through geological history and their translation into climate-dependent evolution. The recognition of the important role of the Arctic and Antarctic habitats in global climate change has recently awakened great interest in the evolutionary biology of the organisms living in those regions. Recent evidence has indicated that global change is already affecting the physiology and ecology of some species. Uncertainties remain in predicting the ecological responses of increased climatic variability in the Arctic. To assess the biological response, a profound knowledge of the current situation is needed, including understanding of species composition, life history adaptations, physiological capacities and distribution patterns as well as food web interactions and interactions between pelagic and benthic ecosystem.

Much knowledge of the environment effect on vertebrate physiology/evolution comes from fishes, which can be used as sentinels of environmental challenges. Genes and proteins of polar fish offer opportunities to understand thermal adaptation in vertebrates. During cold adaptation, evolution has led to unique specialisations, including modification of the hematological characteristics. Addressing the evolutionary adaptation of a vital process such as respiration, we are investigating the oxygen-transport system of Arctic fish, in comparison with the knowledge gathered on Antarctic species. The remarkable evolutionary adaptations of hemoglobin (Hb) to polar environments can provide new insights into temperate organisms and shed light on convergent processes evolved in response to thermal adaptations in the polar regions.



Most studies of protein thermostability are based on the structural/mutational approach and on sequence comparisons of homologous high- and low-temperature-adapted proteins. In bacteria nowadays, the possibility to sequence whole genomes may provide the necessary amount of data, allowing to reject or accept some of the classical hypotheses currently invoked in protein thermal adaptation. Genome sequences of polar bacteria offer the opportunity to characterise their hemoproteins. The comparison between phylogenetically related bacteria living in freezing and non-freezing habitats will provide additional insights into globin evolution.

Our aims, in collaboration with national and international teams, are manifold. Hbs are studied in terms of multiplicity, expression and regulation of globin genes, structure and function, with the aim to understand the effect of temperature on oxygen transport. The structure-function relationship will be analysed in detail also through molecular modelling and site-directed mutagenesis. The amino-acid sequences will also be used in a molecular phylogeny approach to the study of evolution. Each topic will be investigated in the framework of adaptive evolution and keeping the comparative implications into due account. Such studies are currently under way on the zoarcid spotted wolffish (*Anarhichas minor*), polar cod (*Boreogadus saida*), Atlantic cod (*Gadus morhua*), Arctic cod (*Arctogadus glacialis*), Greenland snailfish (*Liparis tunicatus*), Arctic eelpout *Lycodes reticulatus* and Arctic skate *Raja hyperborea*. Most species display higher Hb multiplicity (three components) than most Antarctic species. The data will be analysed in close connection with those from more cold-adapted Antarctic fish, both phyletically distant and phyletically related (Zoarcidae and Liparidae are also found in Antarctic waters). Our globin-gene studies suggest that the Hb multiplicity and structure/function is correlated with the distinct life style of Arctic vs Antarctic fish: Arctic and Antarctic species follow distinct pathways of evolution. Environment stability allows the phylogenetic signal to be maintained in the Antarctic globin sequences under selective pressure, whereas environmental variations tend to erase this signal in the Arctic sequences



Specific activities carried out to reach the general goals above indicated are:

A) *Isolation, purification and characterisation of Hbs from Arctic fish.* Arctic fish, being exposed to seasonal temperature variations and able to thrive in a wider range of latitudes, exhibit higher physiological plasticity with respect to Antarctic fish. The comparison of structure and function of proteins affords an additional powerful tool to understand cold-adaptation strategies.

B) *Molecular phylogeny of polar fish.* In an attempt to link polar environmental conditions with protein evolution and molecular adaptation, we are also analysing the molecular phylogeny of polar Hbs. The evolution of Antarctic and non-Antarctic globins is compared to that of Hbs of Arctic fish species. We aim at identifying the putative residues involved in cold adaptation.

C) *Purification and characterisation of the recombinant bacterial globins from cold-adapted marine bacteria and their role in biotechnological processes.* The genome sequences of Arctic, Antarctic and temperate bacteria offer the opportunity to study the biochemical and physiological properties of monomeric ("two on two") Hbs and flavoHbs gained and, conversely lost, in response to cold.

Use of the new Marine Biology Laboratory in Ny-Ålesund (run by a Consortium that includes CNR) is planned. Collection of tissues and preliminary processing has been and will be performed during the TUNU expeditions along the coasts of Greenland on board the R/V Jan Mayen, in collaboration with Norwegian colleagues, from 2002. Most of the research is then being carried out at IBP/CNR.

