Mechanisms of Hard Tissue Destruction

REIDAR F. SOGNNAES

Mechanisms of Hard Tissue Destruction

A Symposium Presented at the Philadelphia Meeting of The American Association for the Advancement of Science December 29 and 30, 1962

Edited by

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Preface

Mechanisms of Hard Tissue Destruction is based on a four-session symposium organized by the Section on Dentistry of the American Association for the Advancement of Science, held during the 129th annual meeting of the AAAS in Philadelphia, Pennsylvania, on December 29 and 30, 1962.

The symposium was cosponsored by the AAAS Sections on Dentistry (Nd), Medicine (N), and Zoology (F) and by the International Association for Dental Research, North American Division, the American College of Dentists, and the American Dental Association.

A multidisciplinary approach was chosen with a view to covering a theme which could serve as a logical sequence—the other side of the coin, as it were—to a previous AAAS symposium which dealt with the formative aspects of hard tissue biology (*Calcification in Biological Systems,* AAAS Publication No. 64, Washington, D. C., 1960).

In the present volume the oral presentations have been supplemented by three additional manuscripts (chapters 5, 10, and 23), making a total of twenty-six chapters by forty-eight authors and coauthors, including fourteen from institutions outside the United States.

The international participation in this symposium was made possible in part by a conference grant from the National Institute of Dental Research, negotiated together with Dean Lester W. Burket and Dr. Ned B. Williams, University of Pennsylvania School of Dentistry, the latter serving as 1962 AAAS Vice President and Chairman of AAAS Section on Dentistry (Nd).

Serving with me as co-chairmen of the individual symposium sessions were Drs. Seymour J. Kreshover, National Institute of Dental

Research (now Secretary, AAAS Section Nd); Franklin C. McLean, University of Chicago; and George Nichols, Jr., Harvard University.

The cost of the color plate (facing page 218) was covered by a grant from the Miami Valley Laboratories of the Procter & Gamble Company, Cincinnati, Ohio.

From the initial planning until the final preparation of subject and author indexes, I have enjoyed assistance beyond the hours of duty from Mrs. Dorothy Good, Administrative Assistant, and Mrs. Phyllis Lessin, Secretary, School of Dentistry of the University of California, Los Angeles.

I am grateful for the excellent cooperation of all the authors here and abroad, and for other expert help in editing this volume.

Each of the individual chapters that follow is concluded with a summary, and the brief tabulation below may serve to orient the

SUMMARY OF CONTENTS

CHAPTER NUMBERS	DESTRUCTIVE PROCESSES	STRUCTURES INVOLVED	BIOLOGICAL INFLUENCES
1 to 4	Boring canals	Calcareous rocks Coral reefs Shells Tusks and teeth	Mollusks Sponges Gastropods Postmortem fungi (?)
4 and 5	Attrition	Teeth	Mastication, bruxism
	Abrasion	Teeth	
	Erosion	Enamel, dentin	Saliva, bacteria (?)
	"Erosion"	Enamel	Postmortem algae (?)
6 to 10	Caries	Teeth	Bacteria
10 to 24	Resorption	Antler Bone Cementum Dentin Enamel	Multinucleated giant cells
	"Osteolysis"	Bone	Osteocytes
25	Chelation	Shells, bones, teeth (?)	Sequestering agents
26	Proteolysis	Collagen	Collagenase

reader regarding the extent to which different reports have shed light on related mechanisms.

The primary purpose of the symposium was to examine the conditions in which mineralized structures—including rocks, corals, shells, antlers, bone, ivory, cementum, dentin, and enamel—are subject to destruction by various marine and subterranean organisms such as boring sponges, mollusks, snails, octopuses, worms, algae, and fungi, as well as by the action of the giant cells typical of lacunar resorption and the oral bacteria responsible for tooth decay. Beyond the morphological and cellular levels of observation, the symposium also served to delineate present knowledge and various areas of ignorance regarding specific chemical agents which lead to the disruption and dissolution of the inorganic salts and organic matrices of mineralized structures; e.g., glandular secretions and various extracellular and intracellular metabolites, acids, chelators, enzymes, and combinations of chemical and physical factors.

When comparing the various mineralized structures that succumb to decalcification in biological systems, one is impressed by the broad spectrum of their chemical and physical properties, as well as by the variety of the biological organisms and biochemical agents involved in their dissolution. Rock-boring organisms can disintegrate not only relatively soft sedimentary rock, but also densely mineralized calcareous products. Boring sponges burrow not only into corals, but also into limestone and shells whether composed of calcite or of aragonite. Gastropods "drill" into the shells of bivalves as well as those of their own fellow snails. Excised gastropod boring organs can act on hard tissues other than shells and will produce etchings when the calcium phosphate crystals of human enamel and dentin are exposed to them *in vitro*. Indigenous oral microorganisms evidently are endowed with a dual capacity to produce agents which can dissolve and digest hard tissues of as contrasting composition as enamel and dentin, so as to cause tooth decay, now definitely established as being of bacterial origin. Subterranean fungi under postmortem conditions produce boring canals by a dissolution of the collagen and calcium phosphate in buried bone, ivory, cementum, and dentin, but leave dental enamel alone. Marine fungi attack the shells of bivalves (calcite) and snails (aragonite) and

have the biochemical capacity to digest the organic conchiolin shell matrix.

Vertebrate hard tissues prone to biological destruction are not entirely uniform with regard to the nature of their organic scaffolding and inorganic building blocks. Presumably there may also be different molecular bonds which bridge the two, i.e., the organic-inorganic linkage which renders the biological whole—be it shell, pearl, ivory, or bone—something far more complex (as well as more beautiful) than a simple summation of the chemical parts. The complexity of analyzing this problem has been illustrated by a number of biological systems described in this volume.

All together at least half a dozen destructive influences appear to be at work: acid demineralization, chelation, enzymatic digestion, proteolysis, molecular bond disruption of the organic-inorganic linkage, cellular ingestion, possibly phagocytosis, physical motion, and mechanical friction. A combination of two or more if not all of these mechanisms may well exert their influence at some stage of destruction within one and the same biological system. The more readily understood physical forces are at work in the case of large multicellular organisms, e.g., the twisting motion of rock-boring bivalves and the drilling action of the snail rasping holes in a shell region partially softened by decalcification. Yet anyone who has observed the cinematographic recordings of the lively process of experimental bone resorption in tissue culture will have a vivid impression that the osteoclast—aside from its complex biochemical apparatus—does in fact move around in a slow-motion "twist," rubbing its pseudopodia along the presumably softened walls of an eroding Howship's lacuna.

Though the discussion is primarily focused on the destructive aspects of hard tissue biology, it is noteworthy that a variety of systems in fact exhibit closely related constructive (biopositive) and reparative phenomena; in other words, that we are dealing with interrelated three-way processes: formation, destruction, and reformation at the cytological level; mineralization, demineralization, and remineralization at the molecular level; in short, cellular and chemical remodeling.

In coral reef remodeling the extensive "erosion" of the dying

coral skeletons by boring sponges is countered by extensive rebuilding through the calcifying powers of the living coral polyps. As rock-boring mussels channel their way into mineralized structures, the dissolved calcareous material is deposited on the walls of the burrows. In vertebrate hard tissues, redeposition of new large inorganic crystals takes place, at least at the ultrastructural level, within superficially altered tooth substance both in erosion and in caries. Such intermittent recrystallization may in part have a "reparative" significance. For example, the large crystals filling the dentinal tubules in dental erosion could possibly explain the failure of oral microorganisms to invade the dentin substance; and, similarly, in dental caries the large crystals noted in partially demineralized areas have been found to contain an exceptionally high amount of fluoride, which presumably would make such tooth substance less soluble. Moreover, when the dental enamel is exposed to a demineralizing solution, the "first order" diffusion-controlled reaction can be inhibited by deposition of protective reaction products in equilibrium with acid solutions (dicalcium phosphate and calcium fluoride on the surfaces of hydroxyapatite and fluorapatite respectively). In brief, dental erosion and caries, destructive as they are, can no longer be looked upon as entirely one-way processes, at least not from the point of view of molecular biology.

In terms of protective mechanisms there is limited knowledge regarding certain organic coatings which appear to modify the destructive processes in teeth as well as in shells, and possibly in bone. Thin salivary films which cover the tooth surfaces appear to have a significant bearing on the relative protection of the thin external layer of enamel in early caries. Furthermore, it has been suggested that dental erosion may in part be due to the absence of the protective action of such a salivary film. The mussels, whose rock-boring capacity is assisted by a calcium-dissolving secretion, have an organic protection against decalcification of the mussels' own shells, these being covered by a thick periostracal horny covering. When snails and octopuses erode the shells of oysters and abalone, it must be presumed also that a preferential shell destruction of their prey can occur only if the gastropod's own radula is protected from both mechanical and chemical action through the presence of

either some organic coating or a different crystal structure of the denticles, or both.

In addition to such potential protection of the inorganic phase against demineralization, other factors appear to control the enzymatic breakdown of the underlying organic framework. Observations on the action of collagenase suggest that the amorphous ground substance may serve as a protective coating which could modify the collagenolytic activity. The pertinence of this concept in dissolution of bones and teeth has not been fully established. The very same substance, presumably an acid mucopolysaccharide, has been thought to be involved in the process of calcification (see *Calcification in Biological Systems*). The two concepts could conceivably be reconciled, however, were one for the moment simply to suggest that the ground substance serves a stabilizing function.

When all is said and done, it will appear that the weakest link in our present fundamental understanding of the mechanisms involved in dissolution of mineralized structures relates to the specific chemical agents located in *immediate juxtaposition* to the dissolving surfaces. Whereas the living culprits of destruction generally can be identified at the "scene of the crime"—be they gastropods, mollusks, sponges, algae, fungi, osteoclasts, or bacteria—the precise microenvironments in which these biological systems operate present great difficulties in research and consequently certain differences in interpretation.

It is hoped that this volume may serve as a springboard for further research on hard tissue biology throughout the animal kingdom, especially at the relatively unexplored level of molecular biology.

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September, 1963

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Contents

1

Rock-Boring Organisms

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BEARING in mind the problems presented by this mode of life, the habit of rock boring is surprisingly widespread among marine organisms. Among plants, it is found in a variety of green, blue-green, and red algae and also in some fungi. Boring animals include certain sponges, a flatworm (Turbellaria), various sipunculid and polychaete worms, certain echinoid echinoderms, a genus of barnacles, and a diversity of gastropod and, above all, bivalve molluscs. In general these organisms are inhabitants of shallow, most often intertidal, waters. Although of wide occurrence where suitable substrates exist in temperate and tropical seas, they are undoubtedly most abundant on tropical coral reefs and within mudstones in temperate waters. Personal experience of boring organisms has been gained during the course of the Great Barrier Reef Expedition of 1928–1929 and, more recently, on the central California coast while working during 1949 and subsequently at the University of California, Berkeley, and at the Hopkins Marine Station of Stanford University at Pacific Grove.

The habit of boring is obviously not primitive. The substrate bored is either relatively soft sedimentary rock or else the calcareous product of animal secretion, notably coral skeletons and the shells of molluscs, especially the larger Bivalvia. Certain organisms, it may be noted, such as serpulid polychaetes which secrete a calcareous tube and the neogastropod Coralliophilidae, e.g. *Magilus,* settle

upon corals and extend their shells to keep pace with the growth of these. The final appearance gives a misleading impression of boring. Organisms bore either by mechanical or, if the rock be largely or in part calcareous, by chemical means. The firmer the substrate (e.g. many bivalve shells), the greater the need for at least some chemical assistance in boring.

Significance and Mode of Boring

The ability to bore invariably confers a high degree of protection, and this certainly represents the biological reason for the prevalence of the habit. Unlike the wood-boring bivalve Teredinidae (shipworms) or the crustacean *Limnoria* (gribble), which obtain much of the energy for boring from the material into which they penetrate, it is the exception for rock borers to obtain energy in this manner. Nevertheless we may conveniently begin by considering cases where boring is either certainly or possibly associated with feeding.

Some Association with Nutrition

Plants. The one certain case where energy is obtained by the borer is that of the fungi which ramify through dead or living bivalve shells, utilizing the energy present in the organic conchiolin matrix of the shell. The best-known instance is provided by the causal agent of Dutch shell disease, which formerly did great damage to European stocks of oysters, often spreading as spores to the living oyster shells from dead shells used as a settling surface or "cultch." Korringa (1952) has described the life history; although oysters die if the shells become heavily infected, this is due to reaction by the molluscan tissues. The fungus itself never penetrates the tissues. Presumably boring is along the areas of conchiolin, although some actual penetration of the calcified regions by either mechanical or, more probably, chemical means may well be necessary.

A variety of filamentous green, blue-green, and also some red algae penetrate into calcareous rock or shells, either deeply or superficially. Blue-green algae are abundant between tidal levels on coral reefs, causing a softening which may be due to the action of CO_2 or other