Heavy Metal Tolerance in Plants: Evolutionary Aspects

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PREFACE

There has been a great deal of interest during the last 15 years in the interactions between living organisms and heavy metals. This is a result of concerns about the environmental impact of heavy metal pollution, as well as from interests in the biochemistry, physiology, ecology, and evolution of plant and animal populations in nature. A large number of review papers and books have appeared, which deal with various aspects of metals and biology. So why one more book?

The excellent selection of books available in the general area of metals and living organisms covers the subject thoroughly from the environmental impact standpoint. Such questions as — What are the sources of metals in ecosystems? How do they circulate through the biosphere? What is their apparent impact on plants and animals? — are critically important environmental issues and have been addressed in previous books. These volumes highlight the need for ongoing research and periodic syntheses of information.

Heavy metal toxicity and tolerance has also provided a model system for studying evolutionary changes in natural populations. The evolution of heavy metal tolerance in plants is now one of the most thoroughly documented examples of evolution in action, comparable in many ways to the evolution of industrial melanism in moths, or more recently, to the evolution of pesticide resistance in insects or herbicide resistance in weeds. It is this aspect of metal tolerance, in providing an exciting and unique opportunity to study the mechanisms involved in the development of plant adaptations, with which the present volume is primarily concerned.

The first part of the book provides descriptions of habitats contaminated by metals; the ecological theaters within which the evolution of tolerance occurs. A brief essay by A. Borovik addresses such topics as what metals are, and how their chemical properties might impose constraints on the evolution of tolerance mechanisms. Also considered here is the imprecise nature of the term, "heavy metal" itself. Subsequent chapters consider metals in ecosystems, and the ecology of sites contaminated by atmospheric deposition and other anthropogenic sources. These chapters set the stage for developing insights into the evolution of tolerance mechanisms in wild plants.

The second section of the book contains a systematic survey of tolerance in plants. The only major taxonomic group not represented here is the angiosperms, since they provide much of the substance for the remaining chapters. A perusal of this section highlights, by their absence, the scarcity of information on tolerance in such plant groups as Pteridophytes and Gymnosperms.

Physiological and molecular aspects of tolerance are considered in the third section, and a chapter on the metallothioneins of animals is included here for comparison with data on plants. In the last section of the book, microevolutionary processes involved in the development of tolerance are considered in detail. Again, a chapter on the evolution of tolerance in animals is included for comparison. To conclude, Bradshaw, Putwain, and McNeilly summarize the essential qualities of metal tolerance which make this such a unique and valuable system for studying evolutionary change.

This volume demonstrates that exciting progress has been made in understanding the evolution of tolerance in natural populations; however, it is at least as exciting to note that much potential still exists for gaining additional insights into evolutionary processes, utilizing heavy metal tolerance as a model system. It is hoped that publication of *Heavy Metal Tolerance in Plants: Evolutionary Aspects*, will provide a stimulus for continuing research.

THE EDITOR

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Dr. Shaw received his B.S. degree in Agriculture from Cornell University, Ithaca, New York, in 1977. He obtained his M.S. degree in Botany from the University of Alberta in 1980, and his Ph.D. degree, also in Botany, from the University of Michigan in 1983. After completing a 1-year post-doctoral fellowship at the National Museum of Natural History, The Smithsonian Institution, he did 3 additional years of post-doctoral research at Duke University. In 1987, he was appointed an Assistant Professor of Biology at Ithaca College.

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TABLE OF CONTENTS

PART 1. ECOLOGY OF METAL-CONTAMINATED HABITATS

PART 2. MACROEVOLUTIONARY ASPECTS OF METAL TOLERANCE

PART 3. PHYSIOLOGICAL AND MOLECULAR ASPECTS OF TOLERANCE IN HIGHER PLANTS

Part 1.
Ecology of Metal-Contaminated Habitats

Chapter 1

CHARACTERIZATIONS OF METAL IONS IN BIOLOGICAL SYSTEMS

A. S. Borovik

In the past 50 years the importance of metal ions in biological processes has become apparent. Upon reflection this is not surprising, since metals constitute more than 75% of all known elements. From their grouping in the periodic table (Figure 1), the metallic elements can be roughly divided into three general classes: (1) the representative metals (groups 1A to 6A); (2) the transition metals (groups 1B to 8B); and (3) the lanthanide and actinide metals. Metallic species share common physical properties which include high electrical conductivity, malleability, ductility, and lustrous surfaces. In biological systems, metals exist in their ionic forms, specifically as cations. While the representative metal ions usually occur only as singly charged species (e.g., K^+ and Mg^{2+}), the transition metal ions can have a variety of different charges or oxidation states. Metal ions can bind a number of different species (referred to as ligands) to form metal complexes. The chemical and physical properties of the metal complexes are governed by the charge of the metal ion and the type of ligands bound to it. Thus, two ions of the same metal that have different oxidation states or ligands can have vastly different properties. The discipline of chemistry that examines the structure and reactivity of metal ion complexes is called corrdination chemistry.

The metal ions that are most essential for life are the representive metals ions: Na^+, K^+, Mg^{2+}, and Ca^{2+}; and the transition metals: V, Mn, Fe, Co, Ni, Cu, Zn, and Mo. There are no known actinide and lanthanide metals that are essential for life. The essential metal ions have a variety of functions in biological systems. Their functions range from regulators of biological processes to important structural components in proteins. One area of particular interest is the functional role of metal ions in proteins. In many proteins the metal ion binding site is also the center of activity. Two of the more important functions that metal ions are involved in are electron transfer and oxygen binding and/or activation. For example, plastocyanin, a copper based protein, is found in the electron transport chain in photosynthesis, while hemoglobin is an iron protein whose function is to transport dioxygen in mammals. There are several factors that allow metal ions to have such diverse roles in nature, with two of the most important being their abilities to accept and donate electrons, and bind several donor groups. This is particularly true of the transition metal ions. They can easily act as oxidants or reductants in biochemical processes or can expand their coordination sphere to bind substrates.

Where do heavy metal ions fit into this discussion of metal ions in biology? To begin with, the term heavy metal is unfortunately somewhat vague. Most coordination chemists would define heavy metals as the second and third row transition metal ions (i.e., the metal ions from Y to Cd and La to Hg), and the representative metal ions, tin and lead. These metal ions are "heavier" than most other metal ions and, with the exception of Mo, none are essential for life. Yet there are no specific chemical or physical properties that link these metal ions together. Other chemists have defined heavy metal ions as metal ions that are not essential for life. The problem with this definition is that while several metal ions are essential for life (such as Ni, Cu, and Zn), they also have harmful effects if present in excess. Biologists, on the other hand, usually use the term to include virtually any transition metal ion. Clearly, different scientists follow different conventions in defining heavy metal ions. Probably the best way to avoid this confusion in nomenclature is to eschew the term "heavy metal" and simply state the specific metal ions concerned.[1]

Transition metals

1A	2A	3B	4B	5B	6B	7B	8B	8B	8B	1B	2B	3A	4A	5A	6A	7A	8A
1 H																	2 He
3 *Li*	4 *Be*											5 B	6 C	7 N	8 O	9 F	10 Ne
11 *Na*	12 *Mg*											13 *Al*	14 Si	15 P	16 S	17 Cl	18 Ar
19 *K*	20 *Ca*	21 *Sc*	22 *Ti*	23 *V*	24 *Cr*	25 *Mn*	26 *Fe*	27 *Co*	28 *Ni*	29 *Cu*	30 *Zn*	31 *Ga*	32 Ge	33 As	34 Se	35 Br	36 Kr
37 *Rb*	38 *Sr*	39 *Y*	40 *Zr*	41 *Nb*	42 *Mo*	43 *Tc*	44 *Ru*	45 *Rh*	46 *Pd*	47 *Ag*	48 *Cd*	49 *In*	50 *Sn*	51 Sb	52 Te	53 I	54 Xe
55 *Cs*	56 *Ba*	57 *La* *	72 *Hf*	73 *Ta*	74 *W*	75 *Re*	76 *Os*	77 *Ir*	78 *Pt*	79 *Au*	80 *Hg*	81 *Tl*	82 *Pb*	83 *Bi*	84 *Po*	85 At	86 Rn
87 *Fr*	88 *Ra*	89 *Ac* †	104 *Rf*	105 *Ha*	106 *Unh*	107 *Uns*		109 *Une*									

* Lanthanide Series	58 *Ce*	59 *Pr*	60 *Nd*	61 *Pm*	62 *Sm*	63 *Eu*	64 *Gd*	65 *Tb*	66 *Dy*	67 *Ho*	68 *Er*	69 *Tm*	70 *Yb*	71 *Lu*
† Actinide Series	90 *Th*	91 *Pa*	92 *U*	93 *Np*	94 *Pu*	95 *Am*	96 *Cm*	97 *Bk*	98 *Cf*	99 *Es*	100 *Fm*	101 *Md*	102 *No*	103 *Lr*

FIGURE 1. The periodic table of the elements. The metallic elements are in italics.

The deleterious effects of metal ions can be manifested in many ways. As pointed out by Ochiai,[2] the molecular mechanisms of metal ion toxicity can be divided into five general groups:

1. Displacing essential metal ions from biomolecules and other biologically functionaly units
2. Blocking essential functional groups of biomolecules, including enzymes and polynucleotides
3. Modifying the active conformation of biomolecules, especially enzymes and polynucleotides
4. Disrupting the integrity of biomolecules
5. Modifying some other biologically active agent

The basis for these five mechanisms, especially the first three mechanisms, is the ability of metal ions to bind strongly to oxygen, nitrogen, and sulfur atoms. These atoms are abundant in biological systems and can serve as ligands to all essential metal ions. In many cases, the metal ion at the active site can be displaced by a different metal ion, forming a derivative with altered or no biological activity. In addition, toxic metal ions can coordinate to essential functional groups of proteins which can render the protein inactive. This is especially true of Hg(II), which has a tremendously high affinity for sulfur (the binding constant of Hg[II] to S^- is 1×10^{24}). Mercury(II) is so selective for sulfur that it is commonly used in biochemical analyses to determine the amount of cysteine present in proteins.

The above mechanisms of metal ion toxicity are directly related to the modes of metal ion binding in biological systems. In order for a biomolecule to bind a metal ion it must posess a number of chemical characteristics, the most important being: (1) a region that has a high concentration of oxygen, nitrogen, or sulfur atoms; (2) the proper number of donor atoms to stabilize the metal ion (depending on the metal ion this can be from two to eight donors); and (3) sufficient space in the metal ion site that allows an appriopriate three-dimensional geometry about the metal ion. In proteins, the sources of the oxygen, nitogen, and sulfur atom donors usually come from the amino-acid residues. Common amino acids that serve this function are tyrosine, aspartic acid, glutamic acid, histidine, cysteine, and methionine. Metallothioniens are excellent examples of biomolecules that contain these characteristics. These proteins have regions that are rich in cysteine which bind several different metal ions (see Chapter 14). In addition, the metal ion-binding polypeptides found in plants also have a high concentration of cysteine (see Chapter 13). There have been numerous investigatons into the structure of the metal ion binding sites in proteins.[2,3] One of the major reasons for interest in these sites is that, in most cases, the activity of the protein occurs at the metal ion center. In various organisms it has been found that the amino-acid sequences of the metal ion binding site(s) of the proteins are highly conserved. This suggests that the chemistry of metal ions is an important factor in the evolution of metal-containing proteins. Thus, the chemical properties of metal ions limit the possibilities of the structure of the metal ion binding site in biological systems.

We are just beginning to understand the effects that metal ions have in biological systems. This area of science has so many aspects that it easily lends itself to a multidisciplinary approach. Chemists are exploring the molecular aspects of metal ion binding, molecular biologists are investigating how metal ions are involved in gene regulation, while other biologists are studying population and species differences in metal ion tolerance. It is only through the interactions of scientists from different backgrounds that we will be able to completely determine the roles of metal ions in biology.

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Chapter 2

THE MOVEMENT OF METALS THROUGH SOILS AND ECOSYSTEMS

Andrew J. Friedland

TABLE OF CONTENTS

I. INTRODUCTION

The cycling of heavy metals in soils and ecosystems has received a great deal of attention in recent decades due to the increased release of metals that occurs during human-related activities such as smelting and the combustion of fossil fuels. Anyone studying evolutionary aspects of heavy metals and plants today must appreciate the relatively recent changes that have occurred in the amounts of heavy metals in terrestrial ecosystems throughout most of the world.

The processes and global patterns described in this chapter will pertain to heavy metals in general. However, when specific examples are given, seven elements will be used whenever possible: lead, copper, zinc, nickel, cadmium, aluminum, and mercury. These elements were chosen because they cover a range from toxicity to necessity in plant metabolism and health. Many of the processes and global patterns described in this chapter will pertain to natural forested ecosystems. The principles should be consistent throughout a number of ecosystems and descriptions can be applied to other systems with necessary modifications.

II. THE BEHAVIOR OF HEAVY METALS IN ECOSYSTEMS

When considering the behavior of a particular metal in a specific ecosystem, it is important to consider the function of that metal as well as its chemical properties. For example, under most conditions, copper and nickel have similar degrees of mobility and complexation with organic matter.[1] However, because copper is a plant-essential trace metal, it will usually become incorporated into and cycled by plants more readily than nickel.[2] Even a generalization such as this must be applied with caution; there are indications that nickel may be an essential micronutrient and thus may be cycled by some plants, such as legumes.[3]

Of the seventeen plant-essential elements, eight (iron, manganese, zinc, copper, boron, molybdenum, cobalt, and chlorine) are required in small quantities and are thus called trace elements.[4] Of these eight, iron, manganese, zinc, copper, and molybdenum are sometimes referred to as heavy metals. Yet the term "heavy metal" does not distinguish between those metals that are toxic at some level. In fact, many plant-essential trace elements including copper, zinc, iron, and manganese can be toxic to plants at levels encountered in some natural systems.[5] Accordingly, the terms "trace metal", "heavy metal", and descriptions of an element's necessity to plants must be considered carefully when used.

III. SOURCES OF HEAVY METALS IN ECOSYSTEMS

There are three major sources of heavy metals in most terrestrial ecosystems: the underlying parent material, the atmosphere, and the biosphere. Biotic sources of metals are originally obtained from one of the other two sources. In different systems, the relative input from each of these three sources varies.

A. PARENT MATERIAL INPUTS

In a natural, undisturbed ecosystem, the primary source of most heavy metals is the underlying bedrock[6,7] or surface material transported via the atmosphere from another location.[8] Except in areas of ore deposits and other unusually high concentrations of metals, the heavy metal content in most parent material is quite low. The average or range of concentrations for lithosphere material and sedimentary rocks for six heavy metals is shown below (Table 1). Trace metals in soils are usually derived from the underlying parent material and are often similar to parent material concentrations. Organic soil metal concentrations are generally higher than the underlying mineral soil[9,10] (Table 1).

Bedrock is not always the parent material. In certain environments, glacial till or fluvial and other sediments can be transported hundreds and even thousands of kilometers.[11] The subse-