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Biosorption: a solution to pollution?

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Summary To solve the water pollution problem by toxic heavy metal contamination resulting from humans technological activities has for long presented a challenge. Biosorption can be a part of the solution. Some types of biosorbents such as seaweeds, molds, yeasts, bacteria or crab shells are examples of biomass tested for metal biosorption with very encouraging results. The uptake of heavy metals by biomass can in some cases reach up to 50% of the biomass dry weight. New biosorbents can be manipulated for better efficiency and multiple re-use to increase their economic attractiveness.

Key words Pollution · Biosorption · Biosorbents · Heavy metals · Decontamination

Introduction

The knowledge of matter, how it is formed and how it can be modified was a puzzle for people for a long time ever since someone took a piece of clay and shaped it into a pot that could be hardened by fire. That pot, once fired, would retain liquid and resist deformation even when it was set among hot coals. This happened in the Neolithic, which started approximately 9,000 BC. Some 7,000 years later, Greek philosophers speculated that all matter consisted of minute, indivisible particles of the same basic substance. Those early attempts to understand the nature of material things can be taken as the beginning of materials science. It was not until the 19th century that chemistry and physics began to support the empirical efforts of artisans and engineers with the development of applicable theories and novel analytical tools. The key contribution of science was understanding the coupling of external properties of materials to their internal structure. Studies in thermodynamics disclosed the mysteries of how matter responds to environmental factors, particularly to pressure and temperature. People came to comprehend the equilibrium state of matter or of any other system [29].

Modern industry is, to a large degree, responsible for contamination of the environment. Lakes, rivers and oceans are being overwhelmed with bacteria and waste matter. Among toxic substances reaching hazardous levels are heavy metals. Some inland water bodies in Europe and America are closed for fishing. Newspapers are full with reports of frequent ecological disasters in marine environments. In Northern Brazil,

fish from fresh waters are contaminated with mercury as a result of ruthless, illegal, gold extraction.

It was only in the 1990s that a new scientific area developed that could help to recover heavy metals: biosorption. The first reports described how abundant biological materials could be used to remove, at very low cost, even small amounts of toxic heavy metals from industrial effluents. Metal-sequestering properties of non-viable biomass provide a basis for a new approach to remove heavy metals when they occur at low concentrations [49]. Note that metals can be removed from solution only when they are appropriately immobilized, the procedure of metal removal from aqueous solutions often leading to effectively concentrating the metal. That aspect of biosorption makes the eventual recovery of this waste metal easier and economical.

Biomass types

The assessment of the metal-binding capacity of some types of biomass has gained momentum since 1985 [52]. Indeed, some biomass types are very effective in accumulating heavy metals. Availability is a major factor to be taken into account to select biomass for clean-up purposes. The economy of environmental remediation dictates that the biomass must come from nature or even has to be a waste material. Seaweeds, molds, yeasts, bacteria, crabshells, among other kinds of biomass, have been tested for metal biosorption with very encouraging results.

Some biosorbents can bind and collect a wide range of heavy metals with no specific priority, whereas others are specific for certain types of metals [20, 53].

When choosing the biomass for metal biosorption experiments, its origin is a major factor to be taken into account. Biomass can come from (i) industrial wastes which should be obtained free of charge; (ii) organisms easily available in large amounts in nature; and (iii) organisms of quick growth, especially cultivated or propagated for biosorption purposes.

Cost effectiveness is the main attraction of metal biosorption, and it should be kept that way. Not only should microbial biomass be used directly, but biosorbents derived from it in a simple process should be most low-priced for economical metal-removal process applications. If, for any reason, by-products of fermentation processes would not be available, biosorbents could be produced by using relatively unsophisticated and low-cost culture propagation techniques. Nutrients from readily available and inexpensive sources such as carbohydrate-rich industrial wastewaters, which often pose pollution/treatment problems, such as food, dairy and starch industries, might be conveniently used. On the contrary, the costs of biosorbents especially produced could be higher and affect negatively the overall economy of their application [27, 47, 59].

Whereas the use of synthetic ion exchangers can be considered a mature technology, biosorption is in its developmental stages and further improvement in both performance and costs can be expected [50]. Biosorbents are prepared from the naturally abundant or waste biomass of mainly algae, fungi or bacteria that have been killed by washing biomass with acids or bases, or even both, before final drying and granulation [6, 26]. Figure 1 schematically summarizes alternative process pathways to produce biosorbent materials which are effective and durable in repeated long-term applications aimed mainly at removing metals from large quantities of toxic industrial metal-bearing effluents. Whereas the preparation of biomass is an extremely important aspect, this type of process development is based on trial-and-error routines but obviously challenging because of the many biomass raw materials.

Abundant natural materials, particularly of cellulosic nature, have been suggested as potential biosorbents for heavy metals. Considering the number of candidate biomass types and the number of metals of interest, all multiplied by the number of experimental or process parameters, the task of prospecting for new and potentially feasible metal biosorbents has a very wide scope [52].

It is mainly the bacterial cell wall that contains chemical compounds with sites capable of passively sequestering metals

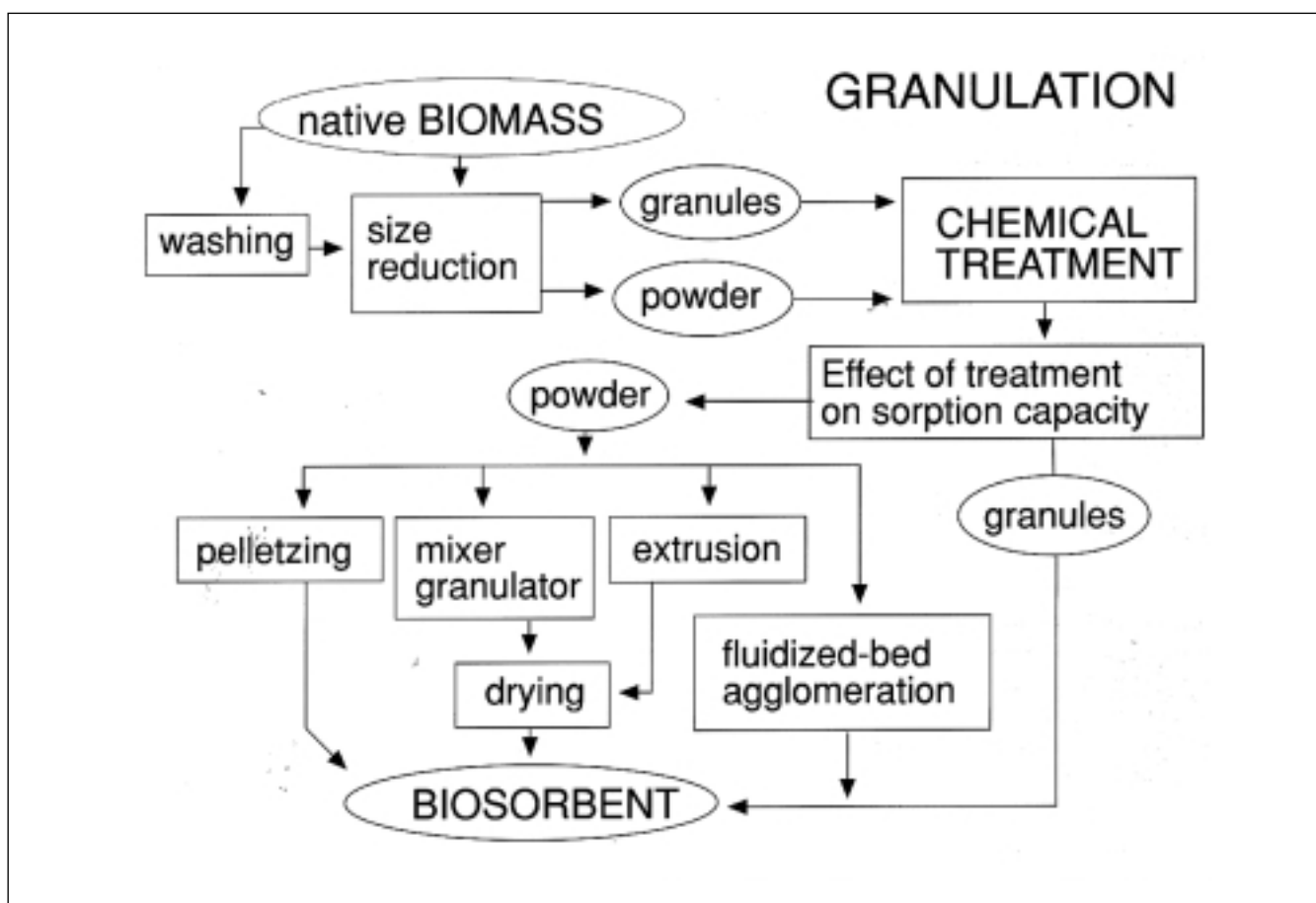


Fig. 1 Schematic diagram of processing different types of microbial biomass into usable biosorption materials

[37]. Figure 2 shows the general structure of the main types of microbial cell walls. Numerous chemical groups have been suggested to contribute to biosorption metal binding by either whole organisms such as algae [10, 17] and bacteria [5, 31] or by molecules such as biopolymers [22, 30]. These groups comprise hydroxyl, carbonyl, carboxyl, sulfhydryl, thioether, sulfonate, amine, imine, amide, imidazole, phosphonate, and phosphodiester groups. The importance of any given group for biosorption of a certain metal by a certain biomass depends on factors such as: the number of sites in the biosorbent material, the accessibility of the sites, the chemical state of the site (i.e. availability), and affinity between site and metal (i.e. binding strength).

For covalent metal binding even an already occupied site is theoretically available. The extent to which the site can be used by a given metal depends on its binding strength and concentration as compared to the metal already occupying the site. For electrostatic metal binding, a site is only available if the metal is ionized.

Seaweed

Seaweeds offer advantages for biosorption because their macroscopic structures offer a convenient basis for the production of biosorbent particles suitable for sorption process applications. Some seaweeds collected from the ocean have indicated impressive biosorption of metals [14, 52, 57, 58]. Brown marine algae tend

particularly to sequester heavy metals [44, 52]. Aderhold et al. [1] studied the efficiency of three species of seaweed *Ecklonia maxima*, *Lessonia flavicans* and *Durvillea potatorum* at sorbing copper, nickel, zinc, lead and cadmium. They found that all three species sequestered metal ions from solution. *L. flavicans* was the poorest at removing lead ions; *D. potatorum* provided the lowest residual metal concentrations in most cases; *E. maxima* released less alginates during experimentation and showed relatively high metal-ion-removal ability. A consistent trend for all three kinds of biomass, in terms of metal-ion uptake, was that cadmium was the most effectively sequestered, followed by copper and nickel. Metal affinity sequences can even be manipulated somewhat, depending on the pretreatment of the biomass to saturate the active sorption sites with different light metals [14].

Volesky et al. [57] worked extensively with one of the best metal-sorbing biomass types, ubiquitous *Sargassum* seaweed (Fig. 3). They compared three different species of non living *Sargassum* biomass for their equilibrium Cd and Cu uptakes from aqueous solutions by using experimental sorption isotherms. Uptakes of Cd at the optimum pH 4.5 were $q_{max} = 87$ mg Cd/g for *Sargassum vulgare*, 80 mg Cd/g for *S. fluitans*, and 74 mg Cd/g for *S. filipendula*. Uptakes of Cu at pH 4.5 were $q_{max} = 59$ mg Cu/g for *S. vulgare*, 56 mg Cu/g for *S. filipendula*, and 51 mg Cu/g for *S. fluitans*.

The uranium uptake by *Sargassum* was higher than expected, the metal constituting more than half of the biomass dry weight (> 500 mg U/g) [58]. That exceeded the stoichiometric ion-

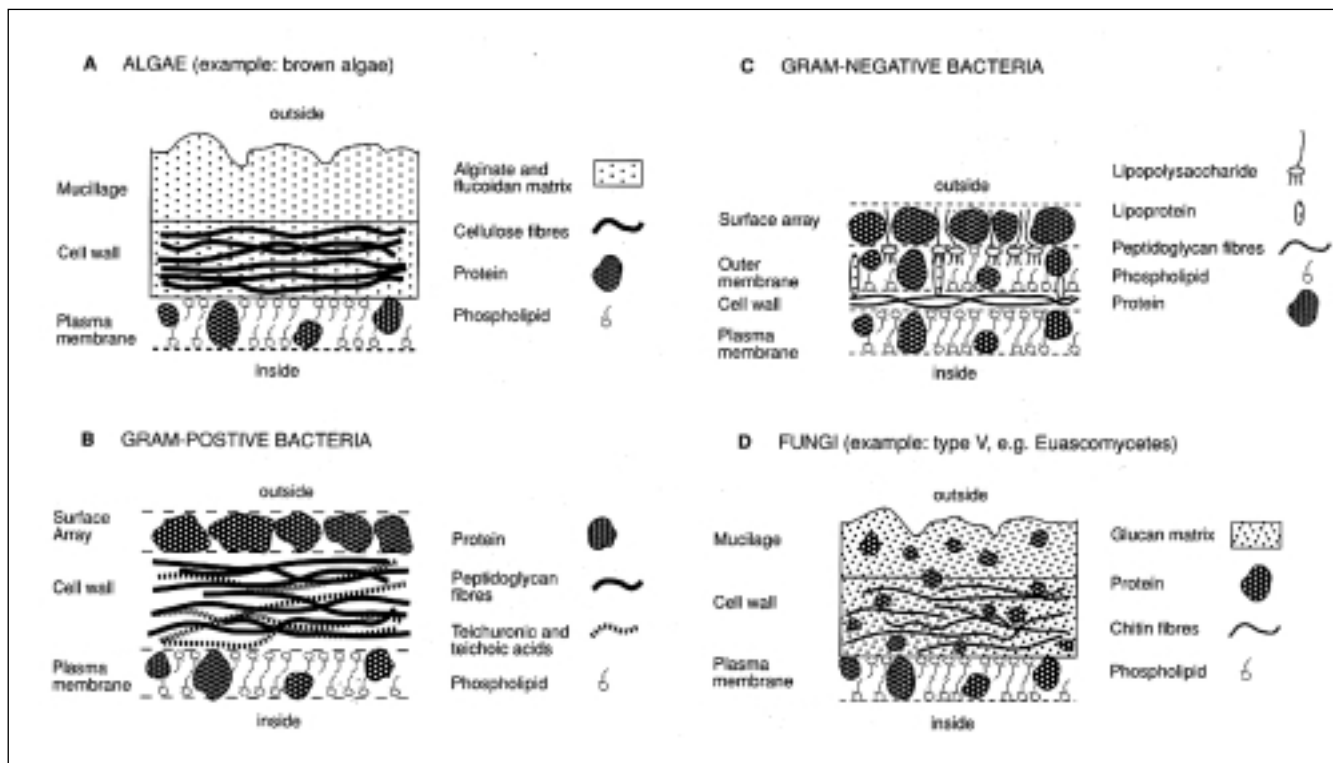


Fig. 2 Cell wall structure in A: algae (example: brown algae), B: Gram-positive bacteria (in part after Beveridge [2] and Remacle [37]), C: Gram-negative bacteria (in part after Beveridge [2] and Remacle [37]), D: fungi (example: type V, e.g. Euscomycetes) (in part after Moore [32])

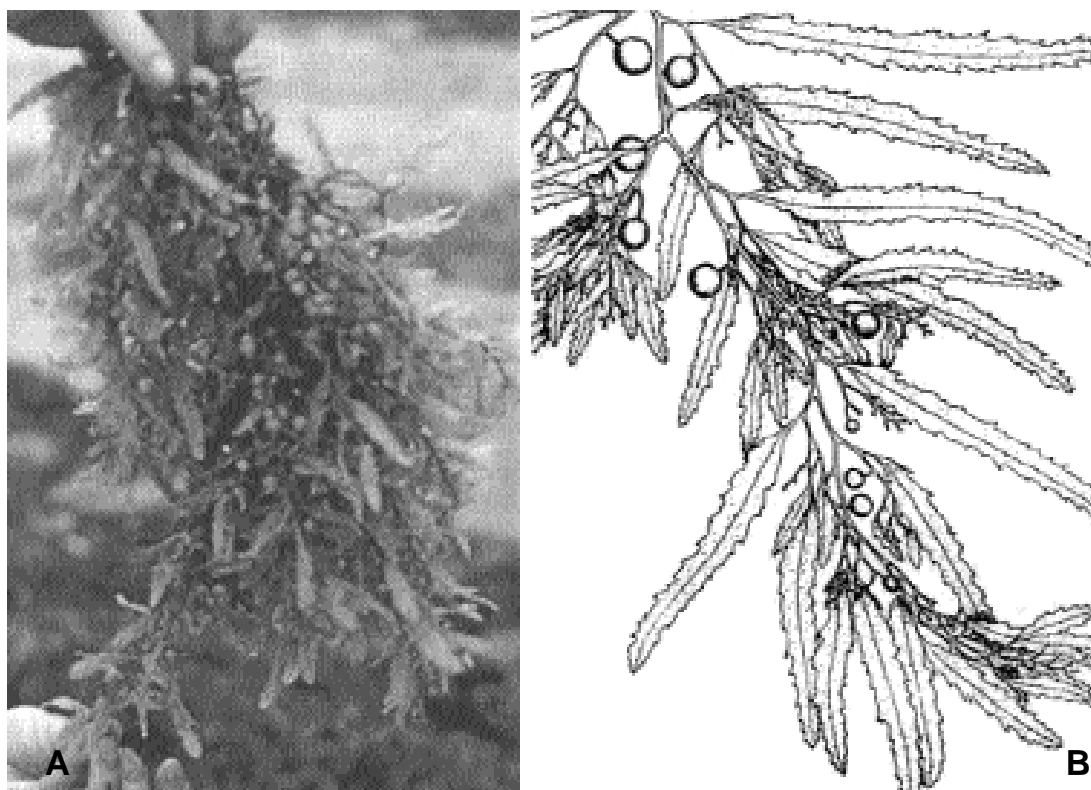
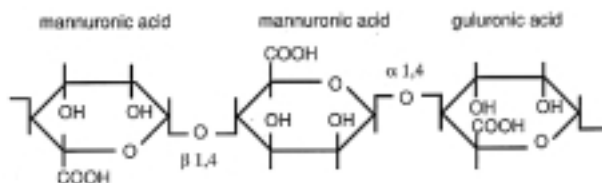
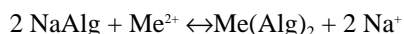


Fig. 3 *Sargassum* brown marine alga (seaweed): A: a medium size plant (Caribbean Sea). B: Drawing of *Sargassum fluitans*

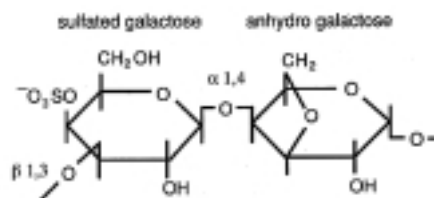
exchange predictions, highlighting thus the importance of considering carefully the solution chemistry of sequestered metals. At the same time, the weight-based expression of the metal uptake naturally favors the heavier elements, molar-based uptake reflects more the chemists point of view. In brown alga *Sargassum* biomass, alginate in the cell wall is the main component responsible for the metal sorption [15]. It is present in a gel form in the cell wall which appears very porous and easily permeable to small ionic species [11, 36]. This molecular-level aspect also sets biosorption apart from physical sorption based on the surface area concept. Apart from its abundance and metal sequestering capacity, *Sargassum* biomass is especially suitable for biosorption because its polysaccharide content is not as easily leached out as are other brown seaweeds used commercially as a source of alginate. The ion exchange properties of certain natural polysaccharides have been studied in detail and it is well established that bivalent metal ions exchange with counter ions of polysaccharides such as alginic acid (in brown algae):



as it is shown in the following reaction [18, 35]:



Ion exchange has been confirmed to be highly involved to a large degree in the metal sequestering by algal biomass [19, 44]. Although other algal polysaccharides such as abundant carageenan have potential binding sites:



red marine algae containing carageenan do not have outstanding metal-sorbing properties.

Yeasts and other fungi

Other kinds of high metal-sorbing biomass such as yeast can also be considered [16]. However, the most common yeast biomass (*Saccharomyces cerevisiae*) is not usually a waste, but a commercial commodity (feed-lot uses). Some chemical compounds of yeast cells can also act as ion exchangers with rapid reversible binding of cations. Volesky et al. [54] working on cadmium biosorption by *Saccharomyces cerevisiae* demonstrated that this yeast is a reasonably potent biosorbent material for cadmium. Their findings agree with those of

Strandberg et al. [45], who earlier demonstrated the rapid uptake of uranium and postulated that polyphosphate groups and carboxyl groups in *S. cerevisiae* cell walls are active in metal complexation. Phosphoryl groups form stable complexes with uranium whereas carboxyl groups become involved only when the phosphoryl groups are saturated. However, *S. cerevisiae* only exhibits a moderate level of metal biosorption activity [55].

Note that fungal biomass, for example of *Penicillium chrysogenum*, can extract gold from a cyanide solution [34]. However, the biosorption capacity was not encouraging. The main mechanism of gold biosorption involved anionic AuCN_2^- species adsorption onto N^- , P^- or O^- containing functional groups on biomass through ion pairing ($\text{H}^+ - \text{AuCN}_2^-$) [34]. These and other observations [24, 25, 56] confirm that biosorption can also sequester anionic species whose behavior is obviously quite different from the usual heavy metals occurring as cations.

Some mucoralean fungi have shown intriguing metal biosorbent properties, particularly high for uranium and thorium [47], whereby different metal deposition patterns could be clearly distinguished (Fig. 4A, B). Note also that a similar and conveniently available biomass of *Aspergillus* species is not very active in biosorption of metals [23, 33, 38, 46]. Niu and Volesky [34] examined selected bacteria, algae and the fungus

Penicillium chrysogenum and found that gold biosorption from cyanide solution is higher at lower pH values, indicating that, in the uptake of anions, biosorbents may act as weak-acid ion exchangers. At pH 2, the gold uptake by *Bacillus* biomass was $8.0 \mu\text{mol g}^{-1}$, by *Penicillium* $7.2 \mu\text{mol g}^{-1}$ and by the seaweed *Sargassum* $3.2 \mu\text{mol g}^{-1}$. The relatively low uptake of the anionic gold complex by *Sargassum* in this work contrasts with excellent uptakes of cationic gold form observed earlier [28, 53]. The results confirmed that waste microbial biomaterials do have some potential for removing and concentrating gold from solutions where it occurs as an anionic goldcyanide complex.

Bacteria

The evaluation of bacterial metal-sorbing properties has aroused some controversy. Most of the experiments done with metals and bacteria have really concerned metabolically mediated bioaccumulation, while the basic principle of biosorption is the use of dead biomass. According to Volesky and Holan [52], who presented an extensive review of biosorption results, the strong biosorbent behavior of certain types of microbial biomass toward metallic ions is a function of the chemical makeup of

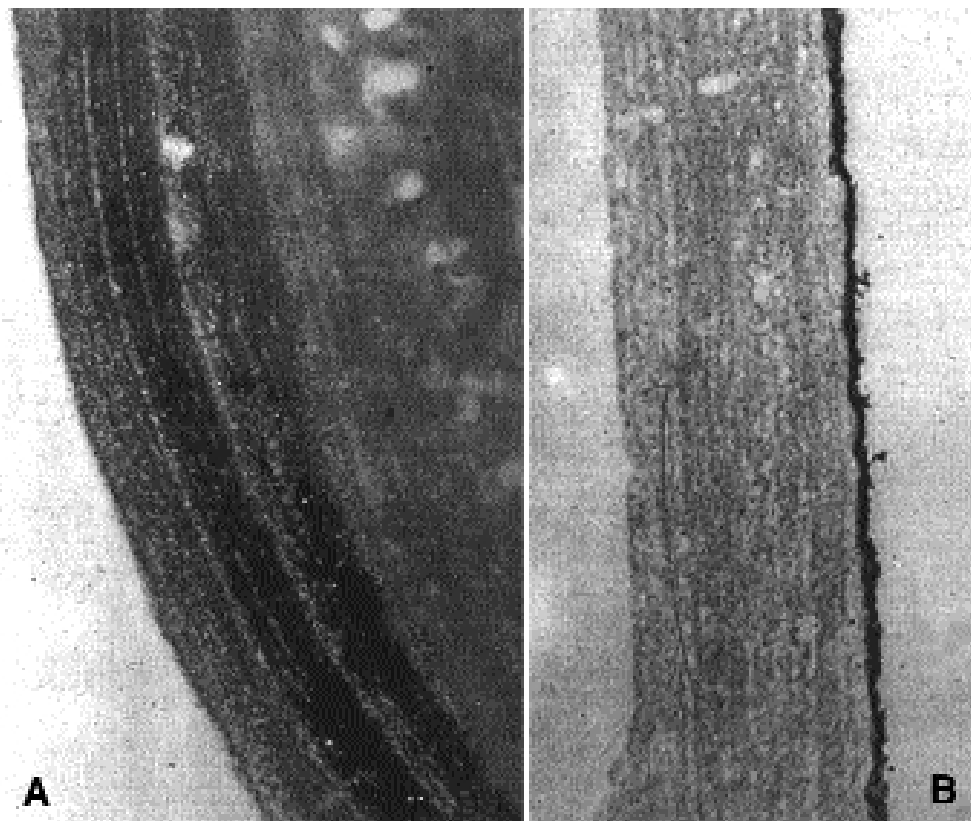


Fig. 4 Transmission electronmicrographs of metal deposition in and on the cell wall of the fungus *Rhizopus arrhizus*. A: Uranium sequestered in the layers of the cell wall material (magnification approx. 100,000 \times). B: Thorium sequestered on the surface of the cell wall (magnification approx. 80,000 \times)

microbial cells. In fact, the biomass is dead and all cells are metabolically inactive. There is also much confusion in the methodology used and in the evaluation and expression of experimental data which shows an inadequate understanding of sorption principles [25, 51, 56].

The most relevant work on true bacterial biosorption has been done by the Brierleys [4–6], who took the metal biosorption concept all the way to the commercial stage [7]. Fein et al. [12] used *Bacillus subtilis* to examine further the bacterium interaction with Cd, Cu, Pb and Al. Their results quantified not only the deprotonation constants for the important organic functional groups on the bacterial cell wall, but also the stability constants for adsorption of environmentally important metal species onto the individual sites. Bacterial cell walls are negatively charged under acidic pH conditions and the cell wall chemically functional groups display a high affinity for metal ions in solution [9].

According to Beveridge [3], bacteria make excellent biosorbents because of their high surface-to-volume ratios and a high content of potentially active chemisorption sites such as on teichoic acid in their cell walls. Churchill et al. [8] used two Gram-negative strains *Escherichia coli* K-12 and *Pseudomonas aeruginosa* and a Gram-positive strain *Micrococcus luteus* to demonstrate biosorption of Cu^{2+} , Cr^{3+} , Co^{2+} and Ni^{2+} . Their sorption binding constants suggested that *E. coli* cells were the most efficient at binding copper, chromium and nickel and *M. luteus* sorbed cobalt most efficiently.

Hu et al. [21] worked with *Pseudomonas aeruginosa* strain CSU, a genetically not altered bacterial strain known to bind dissolved hexavalent uranium. *P. aeruginosa* CSU biomass was sorbing significantly more uranium than certain novel, patented biosorbents derived from algal or fungal biomass sources. A good basis for ongoing work in the field has been provided by Mann in one of the most relevant reviews of bacterial metal biosorption [31], which remains very attractive and exciting for study and contribution.

Comparing biosorption performance

Some confusion persists even in the published literature when it comes to quantitatively expressing and evaluating biosorption performance. The quantitative foundation for comparing any sorption process is in the relatively simple batch equilibrium contact experiment (Fig. 5A) [48, 51]. It allows enough time for establishing equilibrium between the metal immobilized, sequestered in the solid material (sorbent) and the metal still left in the solution. Meaningful comparison of biosorption performance must be done on an equal basis best provided by comparing the sorbent metal uptake (q) at the same equilibrium, final, residual metal concentration (C_f). The equilibrium (metal) uptake and concentration relationship is expressed by the conventional sorption isotherm curve (Fig. 5B). The isotherm plots for two sorbent materials A and B in

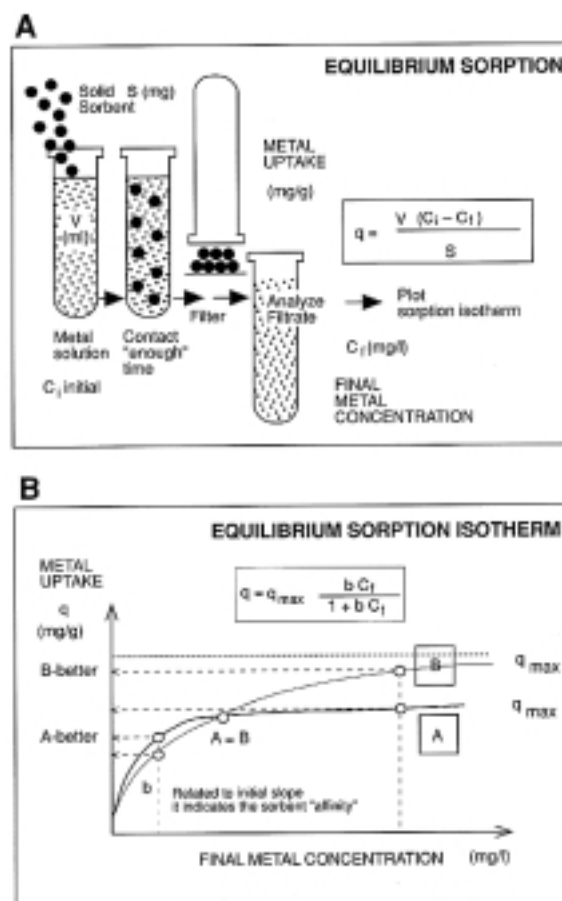


Fig. 5 Evaluation of metal biosorption performance. A: Schematics of the batch equilibrium sorption experimental procedure. B: Resulting sorption isotherm plot of the equilibrium metal uptake (in the solid sorbent) versus the equilibrium metal concentration (left in the liquid phase)

Fig. 5B show that, in the range of high residual concentrations, sorbent B features the highest uptakes at any given residual (final, equilibrium) concentration. The case is opposite in the low equilibrium concentration range whereby sorbent A is better than B.

Note that the conventional isotherm plots must be done at the same equilibrium (final!) pH that should be controlled throughout the experiment. Eventually, a proper evaluation of the biosorption performance could become a fairly complex task as can be seen in the relevant literature [13, 25, 39, 40–44, 56].

Conclusion

Apparently, there are many modes of non-active metal uptake by (microbial) biomass. Any one or a combination of them can be functional in immobilizing metallic species on biosorbents. A number of anionic ligands participate: phosphoryl, carbonyl, sulfhydryl and hydroxyl groups can all be active to various degrees in binding the metal [47]. Due to the accumulated

knowledge and due to the extremely significant economic margin for application in the metal removal/detoxification process, some new biosorbent materials are currently well poised for commercial exploitation. However, there are no limits to expanding the science of biosorption required to provide deeper understanding of the phenomenon and to support effective application attempts.

Many scientific studies are currently under way and contributions to welfare are welcome in this world which grows each second and which needs to be in equilibrium with so much progress. Some pollution seems inevitable, and one can wonder what one should do to minimize it? Human populations need methods and technologies to clean waters and diminish the environmental dangers related to progress. Biosorption can be a solution to clean the environment contaminated by heavy metals. When matter was first tamed, nobody could foresee how many problems humans would have to face in the future.

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