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ADSORPTION TO CHITIN – A VIABLE AND ORGANISM-PROTECTING METHOD FOR BIOMONITORING METALS PRESENT IN DIFFERENT ENVIRONMENTAL COMPARTMENTS GETTING CONTACTED WITH ARTHROPODS

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ABSTRACT – Among the various biopolymers which cover outer interfaces of organisms, chitin is the most abundant: each year several billion metric tons (possibly even much more) are produced by arthropods and processed in soil and litter, wet sediment (especially in moist soils while otherwise chitin samples can persist virtually unchanged for geological periods of time). Moreover, arthropods, among which Coleoptera are represented by some 400,000 species alone, inhabit almost all ecosystems, way beyond the ecological range of, say, mosses. Given that adsorption of metalliferous analytes (ions, volatile compounds, complexes of whatever net charge) to chitin obtained from arthropods can be demonstrated (and it partly was already), it is feasible to obtain data on environmental element contents in all water, soil and gas phase (atmosphere) by dissolving, analyzing outermost (part of exocuticle) chitin layers. Data on relative uptake contributions/environmental burdens of either compartment can be obtained by both interspecies-comparisons and sampling of different parts of some larger arthropod (abdomen-, outer- and inner wing surfaces of sizable beetles). As just a very thin chitin layer (< 2 µm) is ablated from the animal´s outer surface by dissolution using little toxic components, sampling will not cause harm to them, enabling a) repeated sampling of the same specimen (e.g. for taking t = 0 starting values) and b) use of rare or/and protected species. Applications are with both biomonitoring and a better understanding of metal ion transport in ecosystems, e.g. concerning interfacial M^{n+} binding to dying zooplankton then sinking below the chemokline of euxinic water bodies. An indirect metal levels monitoring of woody plants and underneath soils also appears feasible.

KEYWORDS: CHITIN, ARTHROPODS, ADSORPTION OF METALS AND VOLATILES, INDIRECT MONITORING OF SOIL, AIR AND PLANTS, TRANSPORT OF CHEMICAL ELEMENTS IN ECOSYSTEMS AND ACROSS ECOTONES

INTRODUCTION

When biomonitors are used, certain species are selected for this purpose which, even if they are abundant and distributed over larger areas in the world, yet as a rule keep confined to certain ecosystems or even small parts thereof. If, thus, the monitor organisms are employed near to limits of their ecological valencies, e.g. during relocation for purposes of active biomonitoring, both metabolic activities and responses to environmental chemicals (in terms of either accumulation, metabolic response - like suppression of porphyrine ring biosynthesis by Pb^{2+} - or occurrence of visible damages, to be observed e.g. in "ozone gardens") may alter as the local conditions might produce visible damage or changes of metabolic responses of very much the same kind. Hence there is an obvious advantage in such biomonitoring methods which do not rely on metabolism/internal uptake of "pollutant" chemical compounds/ions but gain data just from outer surface interactions of different organisms with the said analytes; this is tried e.g. in moss monitoring.

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Different multicellular organisms are covered by different kinds of biopolymers (like proteins, keratin, sulfonated and otherwise chemically modified polysaccharides - cellulose, chitin, slime on worms, amphibian or fishes) whereas biofilm outer faces mainly consist of esters (lipids) and long-chained ethers (Archaea biofilms). Among the various covers besides of proteins (skin, fur, feathers) or lipids (bacteria, yeast), cellulose and chitin are most notable as they do cover and protect very different kinds of organisms and accordingly are bioproduced and available in very large amounts also. Hence there should be data from very diverse sites to be got even though interaction of some analytes with either chitin or cellulose should be feeble; for cellulose there is a substantial body of knowledge from paper chromatography of various analytes including metal ions and complexes whereas chitin hitherto was considered an effective sorbent rather in terms of wastewater purification, not so far with biomonitoring. Nevertheless the former use of chitin suggests a high sensitivity given a method to elute the analytes from the chitin cover thereafter.

Organisms covered by chitin are distinguished by very different and uncommon living conditions, from total darkness in caves or deep sea to remote sites of various kinds. Arthropods are distributed over this planet in very many species whereas isolated chitin from appropriate sources (like peeled shrimps) might even be exposed to do "modified biomonitoring" in conditions no arthropod, let alone a green plant, would survive or endure for long. The most diverse group among arthropods (and, in fact, among all animals) are Coleoptera which now are estimated at some 400,000 species (Audisio et al., 2015).

A common feature of such surfaces, whichever their chemical identity may be, is the capability to bind (at least certain) ions and volatile compounds which was used already for long in paper chromatography (Qureshi & Akhtar, 1967) and dyeing of (cotton- or linen-based) textiles where hydroxoaluminates (partly hydrolyzed aluminum sulfate) are applied to link cationic organic dyes to a cellulose surface. This feature – primarily M^{n+} coordination chemistry - is more pronounced in chitin as the latter is "not just" a polysaccharide but comes with additional (metal-)binding sites, namely carboxamide groups. The latter and their anions or dicarbonylimide analogs (e.g. urea, succinimide, pyroglutamate, biuret, hydantoin (Pavlovich & Luthy, 1988; Ishiguro et al., 2004; Merbach, 1982) and N-deprotonated anions thereof) are versatile donors towards metal ions both when acting as (highly to extremely polar [DC range about 35 – 200]) solvents like N-methyl- or dimethyl formamide or ethylene urea or as solid ligands, salts.

Accordingly chitin can be expected to withhold metal ions from water or moist soil which form complexes with such carboxamide or carboxamidate moieties which latter fact was used for applying chitin to remove "heavy metal" ions from wastewater flows (Pinto et al., 2011; Plisko et al., 1977), including radionuclide mixtures like those from dissolving "spent" nuclear fuel rods (Muzzarelli, 1970; Muzzarelli, 1973; Moattar & Hayeripour, 2004). On the other hand (much like with cellulose), the network of OH groups in either polysaccharide enables binding of anions which tend to form H bonds with OH groups, therefore readily dissolving in water or alcohols even if gegenions are anything but hydrophilic (e.g. (TBA)- or PPh_4 salts of cyanides, fluorides, fluorometallates or cyanometallates) (Fig.1).

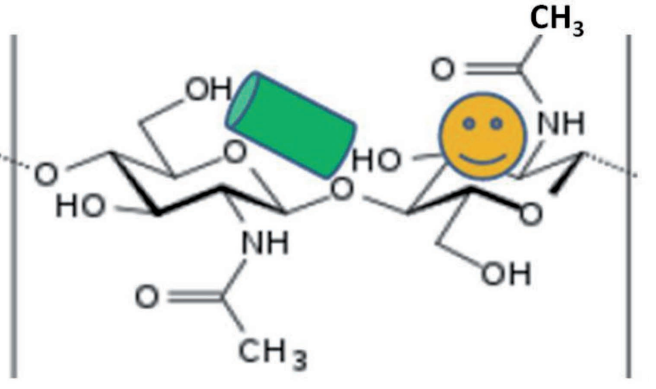


Fig. 1. Binding positions of H-bond-prone anions (the green cylinder represents a cyanide ion ıC≡Nr) or intercalated H-bond-accepting organics (say, sulfoxides, phosphites, amino acid esters) and of metal cations/-complex fragments (orange sphere) on chitin. Saturation of metal ion binding to dissolved chitin (using LiCl or $LiClO_4$ in DMF) corresponds to one metal ion (regardless of identity and being di- or trivalent).

The orange "ball" denotes a metal cation (may also be a complex fragment produced e.g. by thermal replacement of chloro or carbonyl (CO) ligands from chlorometallates or metal carbonyls (M = Fe, Mo, W) which readily form in landfills and are vented with the gas) while the green "stick" represents a cyanide ion.

Bearing this in mind, we started an investigation whether these kinds of retention could be employed for biomonitoring purposes, and to study element cycles in the environment making use of the fact that organisms covered by chitin are almost ubiquitous and can exist in a wide range of surroundings where e.g. moss monitoring would be outright impossible (e.g. caves, ocean, deep sea). Their having an outer cover of chitin means analyte-sorbent interaction takes place without the analytes previously passing the metabolism of the test creature during which event some of the said analytes would possibly (or rather likely) undergo fractionation and chemical modification.

Among the many different organisms which can produce chitin arthropods are finally almost completely covered by one or several (double layered wing arrangements of beetles,

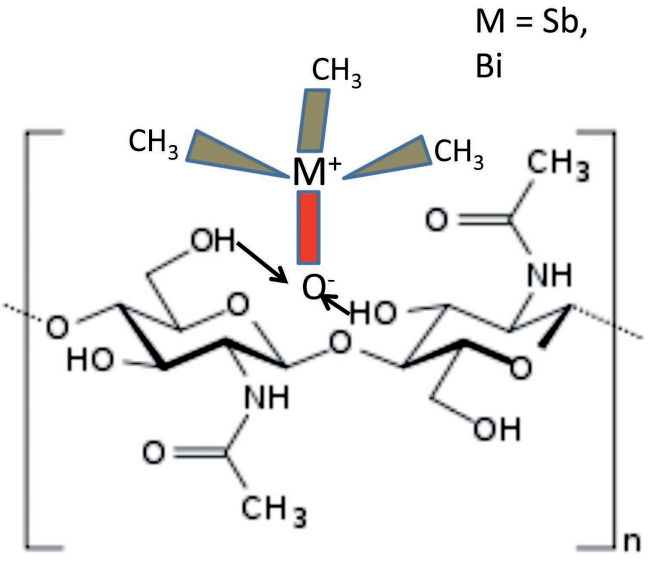


Fig.2. Oxidation of some organoelement (main group semimetal, here $Sb(CH_3)_3$ or $Bi(CH_3)_3$) compound produces an oxide R_3EO with a strongly polarized E=O bond which hence is a good acceptor for H bonds (cp. water miscibility of acetone or dimethyl sulfoxide). Accordingly such an oxidation might increase retention of analyte to chitin if the primary analytes undergo oxidation by air (or other compounds, ions such as nitrate) there before getting into contact with arthropod chitin covers when $Sb(CH_3)_3$ or $Bi(CH_3)_3$ or similar compounds or homoleptic metal carbonyls like $[Fe(CO)_5]$ or $[Mo(CO)_6]$ make their way to the surface (and to arthropods which are most usually aerobic).

locusts) layers of chitin which are permanently or temporally (transparent, thin inner wings covered by cladding structures in beetles) exposed to analytes in either air, soil, or water. While fungi and some fishes (blennies *Paralipophrys trigloides*, Wagner et al., 1993), lancelets and ascidians do also produce chitin, as a rule it is not exposed to the outer surface there, rather it is "buried" in the sporophore interior membranes or fin fortifications, respectively, that is, chitin does not get into direct contact with the environment in those living beings. With some recent arthropods (certain beetles, locusts, lobsters, crabs, hornets, dragonflies and butterflies) being quite sizable (while our sampling method requires about 0.5 cm^2 of chitin surface right now and is going to operate on 1 – 2 mm^2 of sampled active interface soon), many kinds of crabs, beetles, spiders, centipedes/scolopenders, isopods or dragonflies are well-suited for this kind of sampling.

Materials and Methods

All reagents were reagent-grade, p.a. or higher purity and checked for relevant metal contents prior to use (includes chitin, nylon garment, nitric acid for back-exchange and ion exchanger resin granules). Chitin dissolution and analysis was done as described elsewhere (Bauer, 2014, Fränzle, 2015, and Fig. 3) making use of the fact that certain lithium salts (chloride, perchlorate, nitrate, or thiocyanate) cause dissolution of chitin in carboxamide solvents (here: N,N-dimethyl formamide - DMF) given a sizable ionization (thus electrical conductivity) in the liquid. Chitin is soluble in conc. HCOOH or $CHCl_2COOH$, or in carboxamides such as DMF (Plisko et al., 1977) given the latter does contain considerable Li^+ ions, that is, Li salts which undergo ionization to rather large extents when dissolved in the said medium, like LiCl or $LiClO_4$ (metal perchlorates tend to be readily soluble in DMF, Kolthoff et al., 1970) are added. Moreover, chitin is even reported to dissolve in water saturated with LiSCN. Saturation levels of chitin in all these solvents are similar, some 2 – 3 % by weight of solvent. A 1.5-molar solution of $LiClO_4$ in DMF is put to the surface (spot of 1 cm diameter) for 30 seconds, then an ion exchanger resin is brought into contact with the chitin-loaded solution. Aromatic sulfonic acids of which the Amberlite-IR 120 resin is a polymeric representative, are strong acids in DMF, like in water, even though acidities of neutral acids, unlike ammonium-, alkylated ammonium- or pyridinium ions, in DMF are considerably weaker than in water (Kolthoff et al., 1970). Ion exchange by H-forms of cation exchangers (H^+ vs. Mn^+ ions) should thus occur smoothly in either solvent. Nevertheless, cations bigger and less strongly bound (not in a partly covalent mode) than H^+ should be removed and replaced more readily. As a variation increasing efficiency of ion uptake by the exchanger resin from this chitin/DMF/Li^+ solution according to preliminary experiments, the above sulfonic acid form was converted into a tributyl ammonium form of exchanger resin by stirring it with tributyl amine (0.40 ml [311 mg] tris-n-butyl amine per g of exchanger resin, according to the specified exchange capacity of 1.8 mval/g of moist (45 – 50% water content) resin) in methanol which causes some heating. The resin is completely stable in all water, methanol and DMF. CH_3OH is removed by evaporation and the resin put to an Eppendorf vial covered by nylon garment. First results indicate the extent of metal retention (here: Co^{2+}) by the Bu_3NH form of ion exchanger to be much larger (by some factor of ten) than when using the H form.

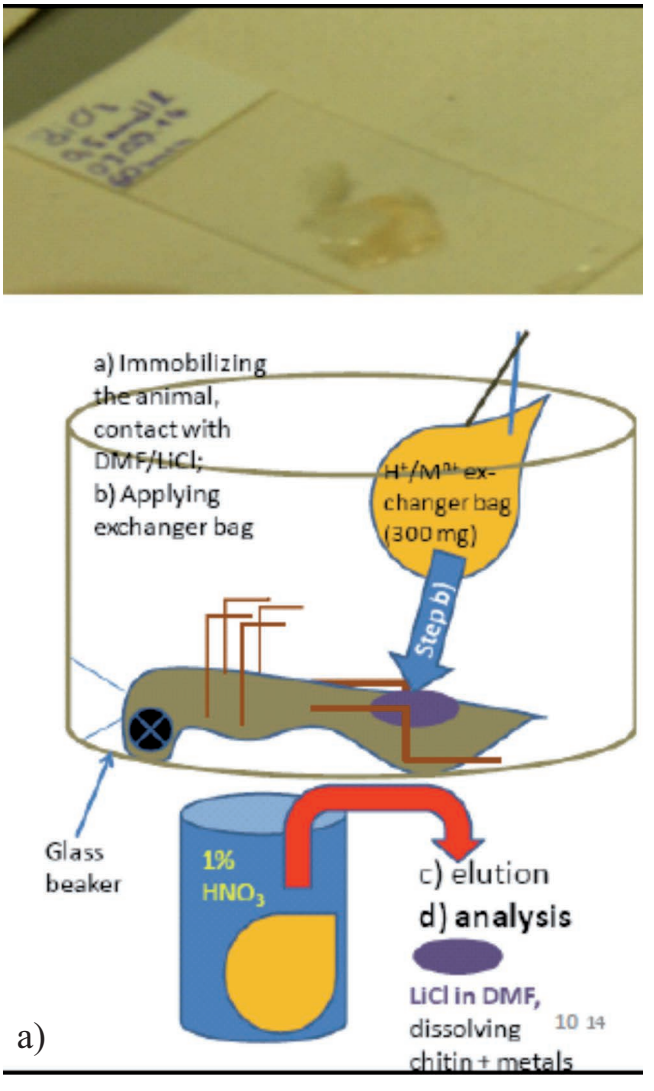
Experiments were done on both isolated chitin (either flakes from chitin scrap obtained by peeling marine crabs or exuvia of dragonfly *Aeshna cyanea*) and living arthropods (cricket *Gryllus assimilis*) in order to determine the extent of metal ion- or other analyte transfers from different environmental compartments. Both living insects and isolated chitin grafted on glass were sampled in the same manner which permits for multiple sampling of one item or specimen without causing significant harm to it: the animals are going to survive the

procedure meaning that:

- a time-zero dataset prior to active biomonitoring can be obtained;
- rare and/or protected species can also be employed in this kind of biomonitoring; generally speaking the method of removing the analytes from the arthropod chitin covers can be considered non-invasive as the animal is neither killed nor permanently harmed, just dissolving a very thin film of chitin from its outer surface. Hence the procedure can be repeated in the same specimen for defining a starting value prior to as well after active biomonitoring.

The acidic cation exchanger both adsorbs the dissolved chitin (≈ 1 mg corresponding to a surface < 2 µm thick) and the bound analytes. The latter (if cationic) pass over to the ion exchanger polymer (mixed polystyrene-/poly-1,4-divinylbenzene sulfonic acid resin) from which they can be eluted by 1% aq. nitric acid, permitting direct injection into ICP-MS (fig. 3b). Molecular ions possibly derived from metal ions still coordinated to DMF after this procedure, like MO^+, $M\text{-}CO^+$, MN^+ or $M(NC/CN)^+$ were not specifically looked for in ICP-MS as the objective simply was quantitative determination of metal(-oid) contents in the solution.

For reasons of experimental convenience and workbench safety, we replaced gaseous volatiles with solutions of volatile or less-volatile (p_{vapor} ≈ 1µPa – 5 kPa at 20°C) possible speciation forms of a number of metals (Al, Fe, Mo, W, Sb, Bi) with solutions in toluene: This enabled easy preparation of dilution series while a simple comparison of the concentrations of a) saturated vapor and b) a saturated solution in toluene provided information on solvation energies. In addition, reactions of metal carbonyls with oxidants like iodine (I_2) had been studied in either hexane or toluene solutions before (Dobson et al., 1973). Solutions of 10 µM L^{-1}, 0.5 and 25 mM L^{-1} were used. The latter level was close to saturation for several of the analytes, including iron pentacarbonyl even though this liquid compound (fp = – 20°C; bp = 103°C) is reported to be completely miscible with benzene and should behave similar in toluene. If adsorbed amounts in the latter two or all three



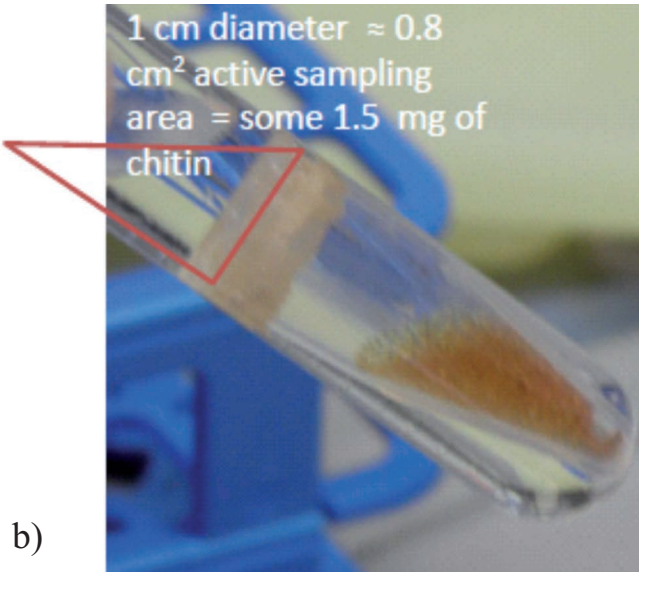


Fig. 3. a) A "mock animal" (chitin flakes fixed on glass) and the protocol of removing a thin chitin surface layer from epicuticle of an immobilized animal and putting analytes to ICP-MS analysis. The yellow drop denotes the Eppendorf vial filled with 300 mg ion exchanger resin and covered by a nylon garment; b) for ion back-exchange the Eppendorf vial containing the cation exchanger is soaked in dilute (1 % = 0.16 M) nitric acid (Suprapur).

concentrations were identical, it was concluded that this was at or beyond the saturation level of the chitin surface. Solution levels in toluene were >> vapor saturation density of all the volatiles, indicating substantial solvation of $[M(CO)_x]$ by toluene.

Results

All metal ions (with the selectivity depicted in the following graph, Fig.4), inorganic volatiles and organometal compounds were demonstrated to bind to chitin exposed to dilute solutions in either water or toluene within a few minutes. When solutions of Al, Sb and Bi trichlorides (all anhydrous) at 25 mM L^{-1} got into contact with chitin flakes, immediate and permanent clouding of the solution (formation of white turbidity) was observed, suggesting partial hydrolysis of the analytes and chlorination of chitin, possibly also acetal cleavage to separate the saccharide rings from each other. In column experiments it turned out that Al and Be are capable of increasing the amount of other ions (commonly being rather constant at 25 – 40 µM g^{-1} DW chitin, regardless of tri- or divalent ions) binding to chitin from aqueous solution. This is significant given the

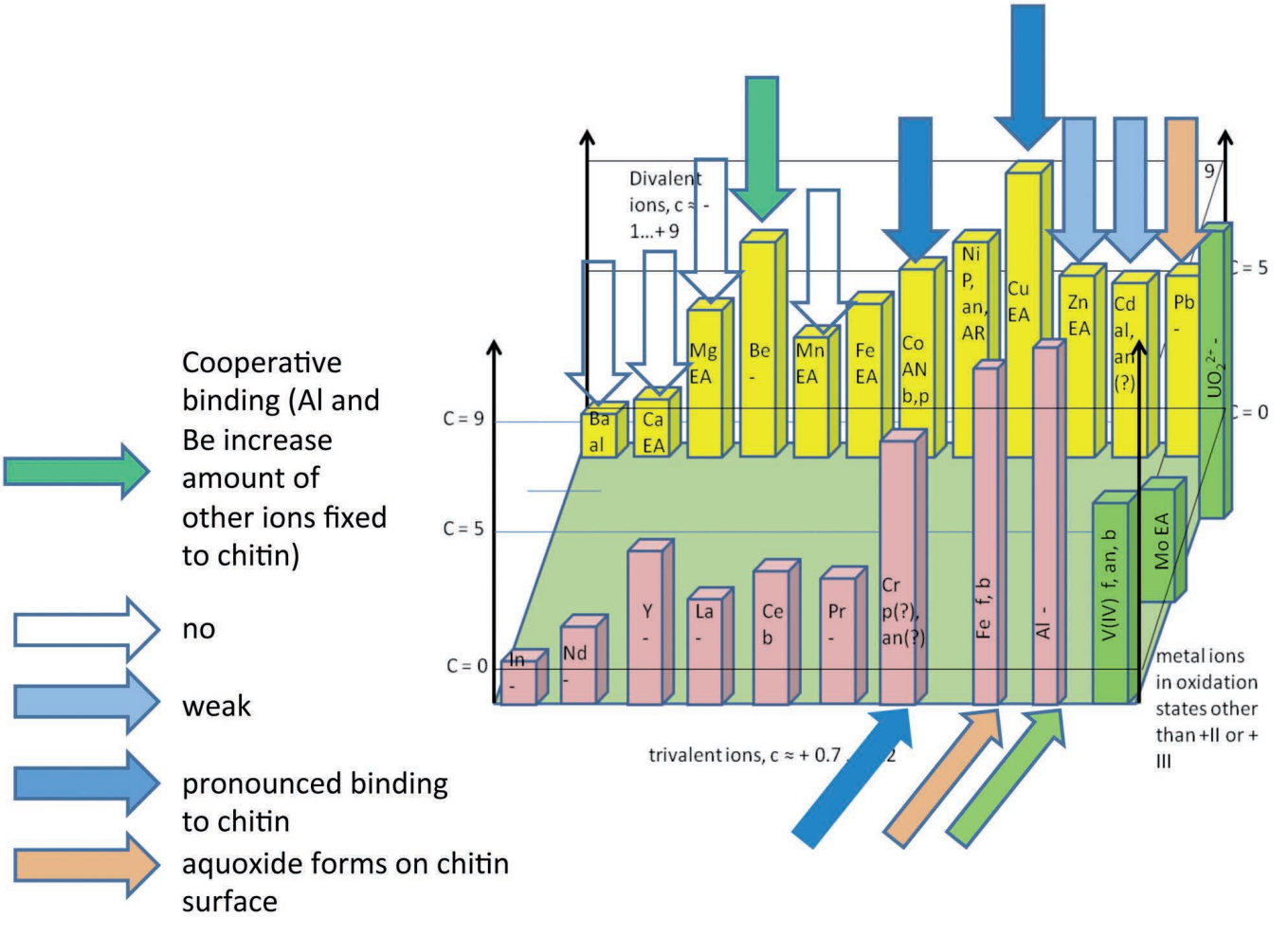


Fig. 4. Relationship between retention of various metal ions by chitin (colorless arrow: no; light blue: little/only from more concentrated solutions; dark blue: strong; green: cooperative [Al^{3+} and Be^{2+} increase binding capacities of other ions except of Mn^{2+} beyond the amount to be bound in their absence]; brown: formation of aquoxide precipitates on the chitin surface) and the two-parameter equation introduced by this author (e.g. Fränzle et al. 2007; Fränzle 2010; Fränzle 2013) to predict ion complexation in both aqueous and organic media: $-\log k_{diss} = x^{*}E_L(L) + c$
Where x is the slope of a regression equation, $E_L(L)$ the electrochemical ligand parameter of a ligand divided by its denticity (according to Lever 1990) and c the axis intercept of the regression equation for a set of complexes of the same metal ion with ligands such as salicylate, oxalate, glycinate, ethylene diamine, malonate or oxamidate. For carboxamide donors like DMF solvent or chitin $E_L(L) \approx 0$ (Lever 1990), implying that binding can be directly predicted using just c (tables in Fränzle 2010; Fränzle 2013) as shown above.

abundance of Al in clay suspensions and the likely increase of detection sensitivity caused by this cooperative effect.
It was shown that bound amounts of some analytes and co-reagents, like $SbCl_3$ and the oxygen transfer reagent [$Co^{II}Cl$(lauroyl sarcosinate)] (possibly dimeric) by far exceeds one monolayer. If saturation was demonstrated (approximately identical levels of metals on chitin samples which had been exposed to two or more different levels of metal compound), the above solvation data can be used to deduce when saturation from vapor phase can be expected, the corresponding vapor concentration (e.g., 10 μM L^{-1} at solvation-induced increase by factor 10^5 (solvation energy some 28.5 kJ mol^{-1} in toluene) → 100 pM L^{-1} in "genuine" air vapor ≈ 2.4 ppb(v) in air (close to the level of [$Mo(CO)_6$] in landfill gas vents; Feldmann & Cullen, 1997).
The model compounds selected among the range of compounds produced by biomethylation and (biochemically not yet understood) metal carbonyl formation in e.g. landfills do indeed undergo an oxidation to increase the amount of analyte binding to chitin from toluene solutions yet do so only with very high analyte levels (Fränzle, 2015). Except for this unrealistic case it is safe to state that subsequent oxidation of such analytes (which is almost inevitable after they get to aerated regions where arthropods can live) does not substantially alter uptake of elements, that is, speciation does not considerably change readings of analytes on element level after chitin adsorption.

DISCUSSION

Chitin as a widely-distributed, highly abundant biogenic sorbent (> 10^{13} kg/a are produced - and mainly decomposed again within some months to years in moist soils/sediments while included, fortifying protein fibers are even way less stable (Stankiewicz et al., 1998)) can be used to retain a broad range of analytes from all environmental compartments for purposes of analysis. As this includes features of speciation, it is significant whether, or to which extent, speciation does modify the aptitude to bind chemical elements to a chitin interface, the more as e.g. biomethylation which can rather readily convert electrophilic M^{3+} or $M(OH)^{2+}$ ions (M = Sb, Bi) into ligands ($M(CH_3)_3$) which rather bind to ("soft") metal ions. Both studies on separation of metal ions/complexes on cellulose and the unlike behavior of different speciation forms in dimethyl formamide, ethylene urea and similar solvents (cp. Plisko et al., 1977; Pinto et al., 2011) suggest polysaccharides to respond to speciation of metal ions interacting with their surface. Work is underway to determine whether this does include (reversible or, more likely, irreversible) redox reactions of the biopolymer at potentials between those of $Fe^{2+/3+}$ or $Cu^{+/2+}$ at near neutral and $MnO_4^{-/2-}$ at alkalinic pH, using different vanadium salts. Cr(III) is readily adsorbed to chitin whereas chromate – unlike permanganate – will neither bind to nor oxidize the interface molecules.
Secondary oxidations of volatile speciation forms by either dioxygen/exocellular enzyme model or molecular iodine are negligible in terms of analyte equilibrium retention (even though very large amounts of cobalt amino acid complexes get bound also) for all the realistic abundance range of the said volatile analytes; at levels close to saturation enhancement actually occurs. These values by far exceed those measured for trimethyl pnictogenes and oxidation products thereof; accordingly speciation does matter at least with Sb and Bi (and similarly in case of Fe introduced as either anhydrous $FeCl_3$ or [$Fe(CO)_5$]), indicating occurrence of metal-organic compounds when total element levels in the environment are known.
Many insects inhabit dead wood, develop there or parasitize at or below bark of trees or live within cavities of tree stems. Generally speaking, the wooden parts of trees connect their

Fig. 5. Example of a beetle (*Lucanus cervus*), larvae of which develop in decaying wood. When adult, the skinny wings used for thrust in flight are only exposed to the environment when the animal is actually flying, thus sample air and volatiles existing there only whereas the rugged brownish covering wings and the ventral abdomen are permanently exposed and going to take up metal ions and other matter from litter, wood residues or soil as well. Courtesy of picture: Wikipedia.

photosynthetic organs to the roots. Thus, the xylem, although as a rule (except for some *Carya* and other tree species like hickory) wood has substantially lower contents of "heavy metals" than the average plant or its leaves or needles, it connects and transports elements many of which, e.g. Fe, Pb, Zn or Cu, can be intercepted and monitored by chitin interfaces when there is contact somewhere when going from roots/soil to leaves or needles or fruits. These bark- inhabiting beetle-or woodwasp imagoes and larvae will do, provided (which is most likely given the additional carboxamide moiety of chitin with respect to structures of both cellulose and lignocelluloses) that chitin absorbs metal ions like those mentioned above directly from living or dead or decaying wood, enabling an indirect analysis of soil chemistry without either taking and digesting (requiring hydrogen fluoride!) soil samples or removing (drilling, sawing or cutting out) wood from stem or large twigs. Experiments on birch-, beech-, spruce-, ash-, Scot´s pine, oak-, willow-, and Douglas fir wood samples for transfer are under way, modeling partition of metals between the respective kinds of wood and (isolated and grafted) chitin.

Of course, for suitable sites the method should be compared and benchmarked to results of moss monitoring; a mesocosm was designed for this purpose specifically (fig.7).

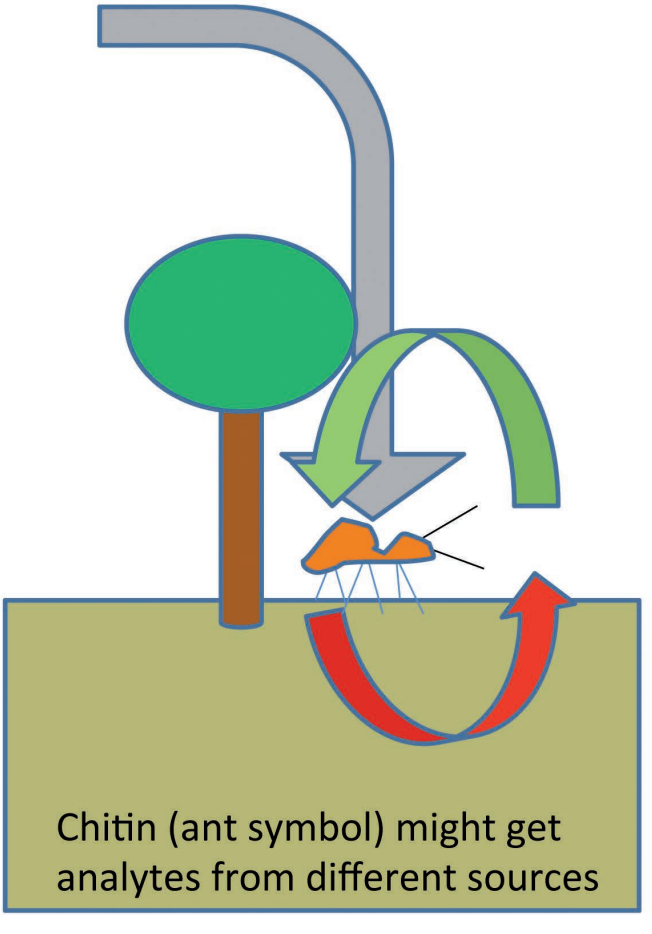


Fig.6. Element cycles involving modification/speciation in reducing (water-logged soil) soil layers (red) as well as inputs from aerosol (grey) and element speciation forms converted into organic acid complexes by vegetation (green).

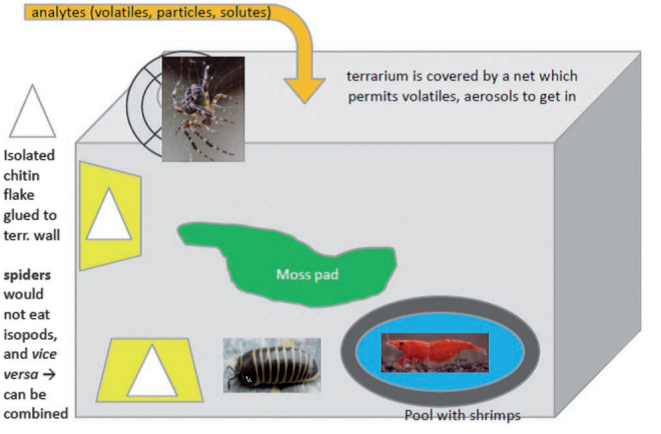


Fig. 7. A setup (mesocosm) for quantitative comparison of airborne metal uptake by various conventional and uncommon recipients during active biomonitoring.

For the moment we do not yet apply these mesocosms but co-expose dried aquatic mosses and grafted chitin in colloquial moss-bags to running waters (creeks, small rivers) expected to bear chemical signatures from upstream (former) mining activities for different metals while releasing additional ones such as Sb, Cd, Zn, W. Besides of applications in biomonitoring and forensic sciences (i.e., obtaining additional data from analyzing the chemicals which adhere to a dead and decaying corpse rather than just estimating the time/date of death by development state and composition of fly (-larval) fauna to be found there) the key objective is to analyze matter flows over concentration- and/or chemical gradients. Such gradients form around ecotones, in caves or old mines/sinkholes where often strongly reducing (sulfides like ZnS, PbS or $CuFeS_2$, arsenides such as NiAs or FeAsS, $FeAs_2$, metalliferous lignite - "salt coal") mineral phases get into contact with air oxygen (Gerth, 2013), often forming secondary minerals (sulfates, arsenates). The same should happen around "black-" or "white smokers" or sulfurous (H_2S-containing) warm and hot springs, or when a metal-rich (more or less acidic) creek or river flows into the ocean or some larger, metal-depleted neutral or alkalinic body of water. Metal ions adhering to chitin covers of zooplankton next to estuaries or in meromictic, euxinic lakes will sink down when zooplankton is dying: quite conceivably there is a contribution in vertical metal transport from this process.

In fact, most of dissolved and particle organic nitrogen in the oceans – from the surface to > 4,000 m depth – is carboxamide rather than amines, amino acids, N heterocycles or anything else (evidence from ^{15}N-NMR spectra; McCarthy et al., 1997) which probably corresponds to chitin trickling downward in this manner.

Conclusions

Chitin, retaining metal ions and volatile compounds from all the environmental compartments as well as due to direct mechanical contact with mineral-/dust grains, does intercept both toxic environmental analytes (Pb, Sb, Co, Cu, organometal[-oid]s, Be, little Cd) and essential trace metals (Fe, Cu, Zn, V, to some extent Mn but not Mg) and certain REEs (which are neither particularly toxic nor essential, except for a Ce-dependent alcohol oxidase enzyme) from their stationary (soil, aqueous oxide suspensions) and mobile forms (salts or complexes) in the environment, enabling to construct models for transport and spreading of metals in ecosystems. By selection of suitable arthropods different pathways and transport over given ecotones, or pollution/emission sites can be pinpointed.

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References

Audisio P., Alonso-Zarazaga M.A., Slipinski A., Nilsson A., Jelinek J., Vigna Taglianti A., Turco F., Otero C., Canepari C., Kral D., Liberti G., Sama G., Nardi G.L., Loebl I., Horak J., Kolibac J., Hava J., Sapiejewski M., Jäch M., Bologna M.A., Biondi M., Nikitsky N.B., Mazzoldi P., Zahradnik P., Wegrzynowicz P., Constantin R., Gerstmeier R., Zhantiev R., Fattorini S., Tomaszewska W., Rücker W.H., Vazquez-Albalate X., Cassola F., Angelini F., Johnson C., Schawaller W., Regalin R., Baviera C., Rocchi S., Cianferoni F., Beenen R., Schmitt M., Sassi D., Kipennberg H., Zampetti M.F., Trizzino M., Chiari S., Carpaneto G.M., Sabatelli S., De Yong Y., 2015. Fauna Europaea: Coleoptera 2 (excl. series *Elateriformia, Scarabaeiformia, Staphyliniformia* and superfamily *Curculionoidea*). Biodiversity Data Journal 3, e4750. DOI: 10.3897/BDJ.3.e4750.

Bauer A., 2014. Orientierende Untersuchungen zur Bindung von Metallionen an Chitin und zur davon abhängigen Eignung von Arthropoden zur Bestimmung von Metallionenkonzentrationen in der Umwelt. Master thesis IHI Zittau (Dresden Tech), submitted and accepted in January, 2014.

Mc Carthy M., Pratum T., Hedges J., Benner R., 1997. Chemical composition of dissolved organic nitrogen in the ocean. Nature 390, 150-154.

Dobson G.R., Jernigan R.T., Chang P.T., 1973. The kinetics and mechanism of the thermal reaction between iron pentacarbonyl and iodine in n-hexane. Journal of Organometallic Chemistry 54, C33-C36.

Fränzle S., Markert B., Wünschmann S., 2007. Dynamics of trace metals in organisms and ecosystems: prediction of metal bioconcentration in different organisms and estimation of exposition risks. Environmental Pollution 150, 22-33.

Fränzle S., 2010. Chemical Elements in Plants and Soil. Springer, Berlin.

Fränzle S., 2013. Estimating and predicting chemical potentials, distributions, speciation modes and mobilities of radiometals in soil, water and biomass. Journal of Environmental Radioactivity and Ecological Radiochemistry 48, 102-115.

Fränzle S., 2015. Adsorption on chitin covers of arthropods and least-invasive retrieval of analytes: a novel item/strategy in biomonitoring – results of model studies. Ecological Indicators, submitted for publication.

Gerth A., 2013. Reise in die Erdgeschichte der Oberlausitz, des Elbsandsteingebirges und Nordböhmens. Oberlausitzer Verlag, Spitzkunnersdorf.

Ishiguro S.I., Umebayashi Y., Ryuzuki R., 2004. Characterization of metal ions in coordinating solvents by means of Raman spectroscopy. Analytical Sciences 20, 415-21.

James D.W., Mayes R.E., 1984. Ion-ion-solvent interactions in solutions. 8. Spectroscopic studies of the lithium perchlorate/N,N-dimethylformamide system. Journal of Physical Chemistry 88, 637-642.

Kolthoff I.M., Chantooni M.K., Smagowski H., 1970. Acid-base strengths in N,N dimethylformamide. Analytical

Chemistry 42, 1622-1626.

Lever A.B.P., 1990. Electrochemical parametrization of metal complex redox potentials, using the ruthenium(III)/ruthenium(II) couple to generate a ligand electrochemical series. Inorganic Chemistry 29, 1271-85.

Merbach A.E., 1982. Solvent exchange rates in metal complexes. Pure and Applied Chemistry 54, 1479-98.

Moattar F., Hayeripour S., 2004. Application of chitin and zeolite absorbents for treatment of low level radioactive wastes. International Journal of Environmental Science and Technology 1, 45-50.

Muzzarelli R.A.A., 1970. Uptake of nitrosyl 106-Ruthenium on chitin and chitosan from waste solutions and polluted seawater. Water Research 4, 451-55.

Muzzarelli R.A.A., 1973. Natural Chelating Polymers. Pergamon, New York.

Pavlovich G.Z., Luthy R.G., 1988. Complexation of metals with hydantoins. Water Research 22, 327-36.

Pinto P.X., Al-Abed S.R., Reisman D.J., 2011. Biosorption of heavy metals from mining-influenced water onto chitin products. Chemical Engineering Journal 166, 1002-1009.

Plisko E.A., Nudga L.A., Danilov S.N., 1977. Chitin and its chemical transformations. Russian Chemical Reviews 46, 1470-87.

Qureshi M., Akhtar I., 1967. Paper chromatography of metal ions in solvents containing methylamine. Fresenius´ Zeitschrift für analytische Chemie 227, 89-95

Stankiewicz B.A., Mastalerz M., Hof C.H.J., Bierstedt A., Flannery M.B., Briggs D.E.G., Evershed R.P., 1998. Biodegradation of the chitin-protein complex in crustacean cuticle. Organic Geochemistry 28, 67-76.

Wagner G.P., Lo J., Laine R., Almeder M., 1993. Chitin in the epidermal cuticle of a vertebrate (*Paralipophrys trigloides*, Blenniidae, Teleostei). Experientia 49, 317-19.