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Studies of Binding of Metal Ions with Dyes , Detergents and Egg Protein

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ABSTRACT: Chelation is a type of bonding of ions and molecules to metal ions. It involves the formation or presence of two or more separate coordinate bonds between a polydentate (multiple bonded) ligand and a single central metal atom.^{[1][2]} These ligands are called chelants, chelators, chelating agents, or sequestering agents. They are usually organic compounds, but this is not a necessity, as in the case of zinc and its use as a maintenance therapy to prevent the absorption of copper in people with Wilson's disease.^[3]

The word chelation is derived from Greek χηλή, chēlē, meaning "claw"; the ligands lie around the central atom like the claws of a crab. The term chelate was first applied in 1920 by Sir Gilbert T. Morgan and H. D. K. Drew, who stated: "The adjective chelate, derived from the great claw or chele (Greek) of the crab or other crustaceans, is suggested for the caliperlike groups which function as two associating units and fasten to the central atom so as to produce heterocyclic rings."^[4]

Chelation is useful in applications such as providing nutritional supplements, in chelation therapy to remove toxic metals from the body, as contrast agents in MRI scanning, in manufacturing using homogeneous catalysts, in chemical water treatment to assist in the removal of metals, and in fertilizers.

KEYWORDS: chelation, metal ions, dyes, detergents, egg protein, binding

I.INTRODUCTION

Numerous biomolecules exhibit the ability to dissolve certain metal cations. Thus, proteins, polysaccharides, and polynucleic acids are excellent polydentate ligands for many metal ions. Organic compounds such as the amino acids glutamic acid and histidine, organic diacids such as malate, and polypeptides such as phytochelatin are also typical chelators. In addition to these adventitious chelators, several biomolecules are specifically produced to bind certain metals (see next section).^{[7][8][9][10]}

Virtually all metalloenzymes feature metals that are chelated, usually to peptides or cofactors and prosthetic groups.^[10] Such chelating agents include the porphyrin rings in hemoglobin and chlorophyll. Many microbial species produce water-soluble pigments that serve as chelating agents, termed siderophores. For example, species of *Pseudomonas* are known to secrete pyochelin and pyoverdine that bind iron. Enterobactin, produced by *E. coli*, is the strongest chelating agent known. The marine mussels use metal chelation esp. Fe^{3+} chelation with the Dopa residues in mussel foot protein-1 to improve the strength of the threads that they use to secure themselves to surfaces.^{[11][12][13]}

In earth science, chemical weathering is attributed to organic chelating agents (e.g., peptides and sugars) that extract metal ions from minerals and rocks.^[14] Most metal complexes in the environment and in nature are bound in some form of chelate ring (e.g., with a humic acid or a protein). Thus, metal chelates are relevant to the mobilization of metals in the soil, the uptake and the accumulation of metals into plants and microorganisms. Selective chelation of heavy metals is relevant to bioremediation (e.g., removal of ^{137}Cs from radioactive waste).^[15]

Applications

In the 1960s, scientists developed the concept of chelating a metal ion prior to feeding the element to the animal. They believed that this would create a neutral compound, protecting the mineral from being complexed with insoluble salts within the stomach, which would render the metal unavailable for absorption. Amino acids, being effective metal



binders, were chosen as the prospective ligands, and research was conducted on the metal–amino acid combinations. The research supported that the metal–amino acid chelates were able to enhance mineral absorption. During this period, synthetic chelates such as ethylenediaminetetraacetic acid (EDTA) were being developed. These applied the same concept of chelation and did create chelated compounds; but these synthetics were too stable and not nutritionally viable. If the mineral was taken from the EDTA ligand, the ligand could not be used by the body and would be expelled. During the expulsion process the EDTA ligand randomly chelated and stripped another mineral from the body.^[16] According to the Association of American Feed Control Officials (AAFCO), a metal–amino acid chelate is defined as the product resulting from the reaction of metal ions from a soluble metal salt with amino acids, with a mole ratio in the range of 1–3 (preferably 2) moles of amino acids for one mole of metal. The average weight of the hydrolyzed amino acids must be approximately 150 and the resulting molecular weight of the chelate must not exceed 800 Da. Since the early development of these compounds, much more research has been conducted, and has been applied to human nutrition products in a similar manner to the animal nutrition experiments that pioneered the technology. Ferrous bis-glycinate is an example of one of these compounds that has been developed for human nutrition.^[17]

Dentin adhesives were first designed and produced in the 1950s based on a co-monomer chelate with calcium on the surface of the tooth and generated very weak water-resistant chemical bonding (2–3 MPa).^[18]

Chelation therapy is an antidote for poisoning by mercury, arsenic, and lead. Chelating agents convert these metal ions into a chemically and biochemically inert form that can be excreted. Chelation using calcium disodium EDTA has been approved by the U.S. Food and Drug Administration (FDA) for serious cases of lead poisoning. It is not approved for treating "heavy metal toxicity".^[19] Although beneficial in cases of serious lead poisoning, use of disodium EDTA (edetate disodium) instead of calcium disodium EDTA has resulted in fatalities due to hypocalcemia.^[20] Disodium EDTA is not approved by the FDA for any use,^[19] and all FDA-approved chelation therapy products require a prescription.^[21]

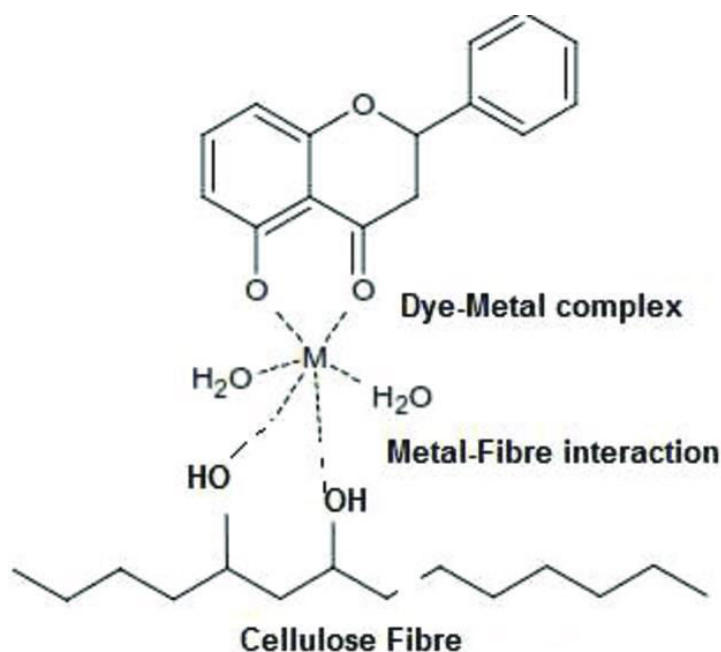
Chelate complexes of gadolinium are often used as contrast agents in MRI scans, although iron particle and manganese chelate complexes have also been explored.^{[22][23]} Bifunctional chelate complexes of zirconium, gallium, fluorine, copper, yttrium, bromine, or iodine are often used for conjugation to monoclonal antibodies for use in antibody-based PET imaging.^[24] These chelate complexes often employ the usage of hexadentate ligands such as desferrioxamine B (DFO), according to Meijs et al.,^[25] and the gadolinium complexes often employ the usage of octadentate ligands such as DTPA, according to Desreux et al.^[26] Auranofin, a chelate complex of gold, is used in the treatment of rheumatoid arthritis, and penicillamine, which forms chelate complexes of copper, is used in the treatment of Wilson's disease and cystinuria, as well as refractory rheumatoid arthritis.^{[27][28]}

Chelation in the intestinal tract is a cause of numerous interactions between drugs and metal ions (also known as "minerals" in nutrition). As examples, antibiotic drugs of the tetracycline and quinolone families are chelators of Fe^{2+} , Ca^{2+} , and Mg^{2+} ions.^{[29][30]}

EDTA, which binds to calcium, is used to alleviate the hypercalcemia that often results from band keratopathy. The calcium may then be removed from the cornea, allowing for some increase in clarity of vision for the patient.

Homogeneous catalysts are often chelated complexes. A representative example is the use of BINAP (a bidentate phosphine) in Noyori asymmetric hydrogenation and asymmetric isomerization. The latter has the practical use of manufacture of synthetic (–)-menthol.

Citric acid is used to soften water in soaps and laundry detergents. A common synthetic chelator is EDTA. Phosphonates are also well-known chelating agents. Chelators are used in water treatment programs and specifically in steam engineering. Although the treatment is often referred to as "softening," chelation has little effect on the water's mineral content, other than to make it soluble and lower the water's pH level.



I. SCHEMATIC REPRESENTATION OF DYE-METAL-CELLULOSE FIBER INTERACTION

Metal chelate compounds are common components of fertilizers to provide micronutrients. These micronutrients (manganese, iron, zinc, copper) are required for the health of the plants. Most fertilizers contain phosphate salts that, in the absence of chelating agents, typically convert these metal ions into insoluble solids that are of no nutritional value to the plants. EDTA is the typical chelating agent that keeps these metal ions in a soluble form.^[31]

Metal-complex dyes are a family of dyes that contain metals coordinated to the organic portion. Many azo dyes, especially those derived from naphthols, form metal complexes by complexation of one of the azo nitrogen centers. The insertion of the metal into the organic ligand often involves redox reactions, e.g. pre-reaction of sodium dichromate with glucose. Phthalocyanine (Pc) complexes, such as CuPc, are another important family of metal complex dyes.^[1]

II. DISCUSSION

Dishwasher detergent is a detergent made for washing dishes in a dishwasher. Dishwasher detergent is different from dishwashing liquid made to wash dishes by hand.

A. Uses

When using a dishwasher, the user must select a special detergent for its use.^[1] All detergents are designed for use after the user scrapes leftover food from the dishes before washing.^[2] To function, the user places dishes in the dishwasher in such fashion that the surface of all dishes is open to the flow of water.^[2]

Most dishwasher detergents are incompatible for use with silver, brass, cast iron, bronze, aluminum, pewter, and goldleaf.^[2] They can also harm disposable plastic, anything wood, knives with hollow handles, and fine glassware.^[2]

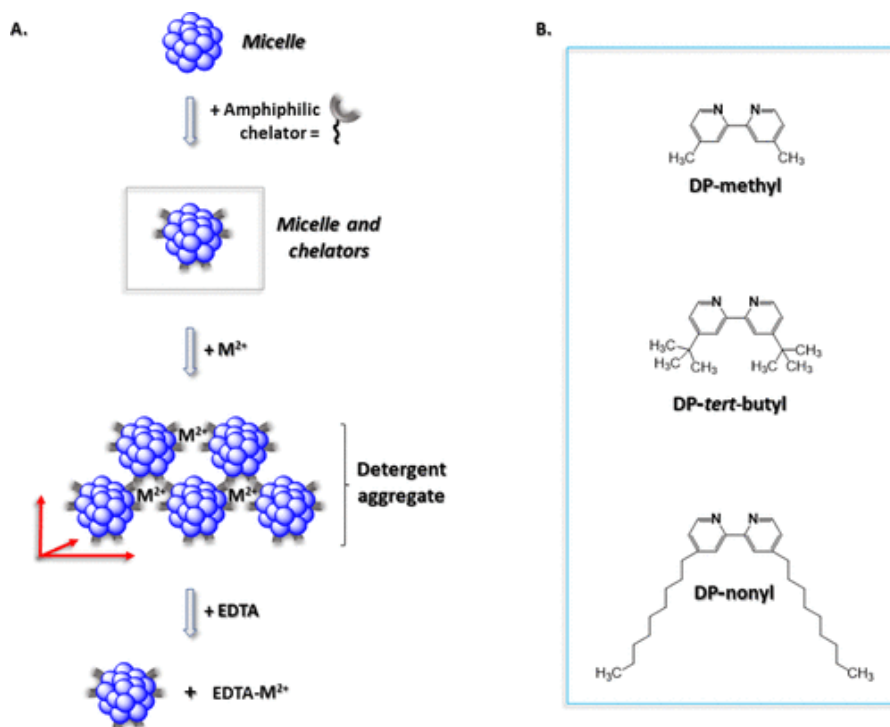
There are specific examples of chemical reactions we use in our everyday lives. For example, a dishwasher detergent uses sodium hypochlorite and sodium carbonate (simple bleach) in a chemical reaction to clean the dishes. Dishes washed in cold water are less clean than dishes washed in hot water.^[30]



B. Composition

Different kinds of dishwashing detergent contain different combinations of ingredients. Common ingredients include:

- Phosphates: Bind calcium and magnesium ions to prevent 'hard-water' type limescale deposits. They can cause ecological damage, and have been partially banned or phased out.
- Oxygen-based bleaching agents (older-style powders and liquids contain chlorine-based bleaching agents): Break up and bleach organic deposits.
- Non-ionic surfactants: Lower the surface tension of the water, emulsifies oil, lipid and fat food deposits, prevents droplet spotting on drying.
- Alkaline salts: These are a primary component in older and original-style dishwasher detergent powders: Highly alkaline salts attack and dissolve grease, but are extremely corrosive (fatal) if swallowed. Salts used may include metasilicates, alkali metal hydroxides, sodium carbonate etc.^[3]
- Enzymes: Break up protein-based food deposits, and possibly oil, lipid and fat deposits.^[4] The enzymes used are similar to the ones used in laundry.^[5]
- Anti-corrosion agent(s): Often sodium silicate, this prevents corrosion of dishwasher components.^[25,26,27]



(A) Cartoon of micelle conjugation via amphiphilic metal:chelator complexes generated at the micelle/water interface. Upon addition of EDTA, a strong, water-soluble chelator, the micelle aggregates disassociate. (B) Chemical structures of amphiphilic dipicolinate (DP) chelators.

Dishwashing detergent may also contain:

- Anti-foaming agents: Foam interferes with the washing action. Foam may affect operation of the machine's water-level sensors and will leak past the door seals.
- Additives to slow down the removal of glaze & patterns from glazed ceramics
- Perfumes



- Anti-caking agents (in granular detergent)
- Starches (in tablet based detergents)[20,21,22]
- Gelling agents (in liquid/gel based detergents)

Dishwasher detergents are generally strongly alkaline (basic).^[6]

Inexpensive powders may contain sand. Such detergents may harm the dishes and the dishwasher. Powdered detergents are more likely to cause fading on china patterns.^[7]

Besides older style detergents for dishwashers, biodegradable detergents also exist for dishwashers. These detergents may be more environmentally friendly than conventional detergents.

Hand-washing dish detergent (washing up liquid) creates a large foam of bubbles which will leak from the dishwasher.^[8]

1) Rinse aid

Rinse aid (sometimes called rinse agent) contains surfactants and uses Marangoni stress to prevent droplet formation, so that water drains from the surfaces in thin sheets, rather than forming droplets.[18,19,20]

Rinse aid prevents "spotting" on glassware (caused by droplets of water drying and leaving behind dissolved limescale minerals), and can also improve drying performance as there is less water remaining to be dried. A thinner sheet of water also has a much larger surface area than a droplet of the same volume, which increases the likelihood of water molecules evaporating[30,31]

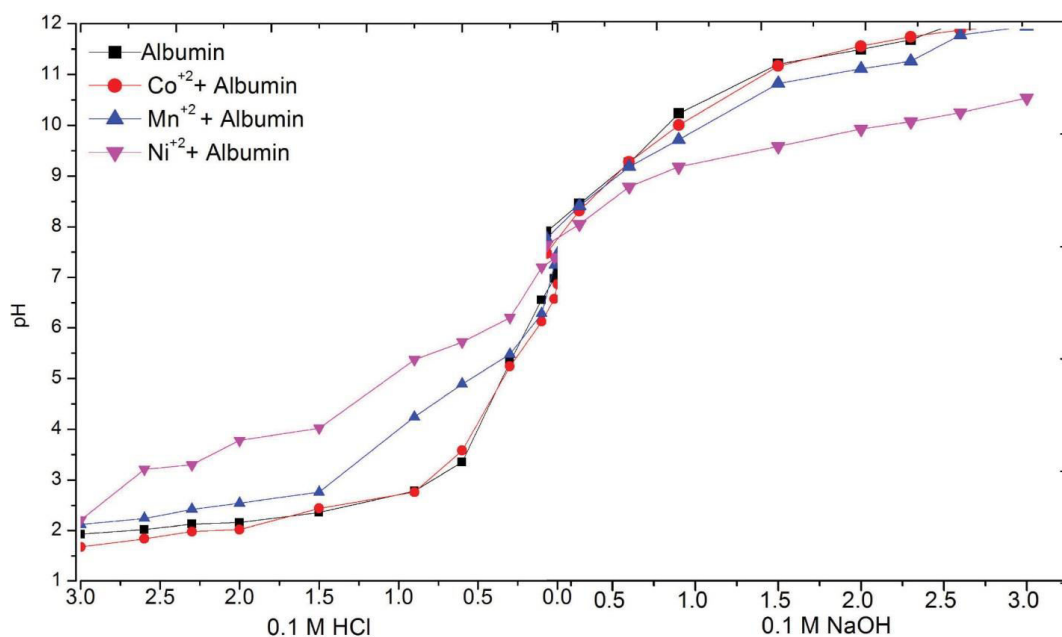
III.RESULTS

In this paper we report the binding of Mn (II), Ni (II) and Co (II) with albumin are reported employing spectrophotometric and pH metric method. In order to distinguish between ionic and colloidal linking, the binding of metal by using pH metric and viscometric methods and the result are discussed in terms of electrovalent and coordinate bonding.[32,33,34]

The binding of Ni⁺², Co⁺² and Mn⁺² ions have been studied with egg protein at different pH values and temperatures by the spectrometric technique.

The binding data were found to be pH and temperature dependent. The intrinsic association constants (k) and the number of binding sites (n) were calculated from Scatchard plots and found to be at the maximum at lower pH and at lower temperatures. Therefore, a lower temperature and lower pH offered more sites in the protein molecule for interaction with these metal ions. Statistical effects seem to be more significant at lower Ni⁺², Co⁺² and Mn⁺² ions concentrations, while at higher concentrations electrostatic effects and heterogeneity of sites are more significant.[11,12,13]

The pH metric as well as viscometric data provided sufficient evidence about the linking of cobalt, nickel and manganese ions with the nitrogen groups of albumin. From the nature and height of curves in the three cases it may be concluded that nickel ions bound strongly while the cobalt ions bound weakly.



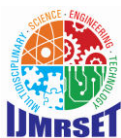
Titration curve of albumin in absence and presence of metal ions.

IV. CONCLUSION

Insoluble expression of intrinsically soluble proteins with native activity is potentially a promising alternative to soluble expression of folded protein or insoluble expression of unfolded protein requiring refolding. The highly soluble halophilic His-rich metal binding protein (HP) as insoluble inclusion bodies with native metal-binding activity using insolubilizing nona-peptide (Ins), GILQINSRW, derived from hen egg white lysozyme (His-InsHP). About 80% of expressed His-InsHP was localized in inclusion bodies in Na-phosphate/NaCl buffer, pH 7.4, while His-HP without Ins peptide was exclusively expressed in soluble supernatant. We report expression, purification and characterization of this insoluble His-InsHP, and its possible application for efficient biosorption and recovery of environmental metal ions, for example, by using whole bacterial cells expressing insoluble His-InsHP as a new cost-effective metal ion-adsorbent. Conalbumin/ovotransferrin is a glycoprotein which has the capacity to bind the bi- and trivalent metal cations into a complex and is more heat sensitive than ovalbumin. At its isoelectric pH (6.5), it can bind two cations and assume a red or yellow color.[35,36,37]

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