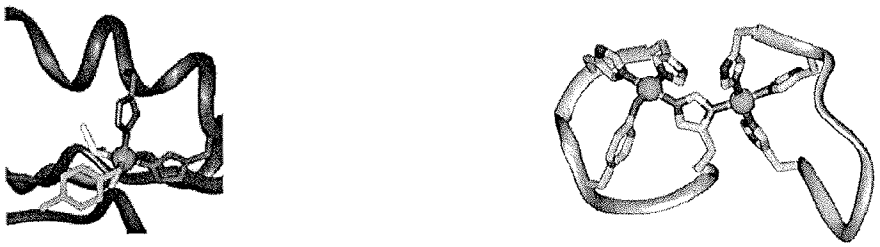
Exploring the Role of Copper in Alzheimer's Disease

Charity Flener Inorganic Literature Seminar September 21, 2004

Alzheimer's disease is a neurodegenerative disease that afflicts over 4.5 million Americans each year.[1] Because no conclusive diagnostic test is available for live patients, Alzheimer's diagnosis is confirmed postmortem by the appearance of amyloid plaques and oxidative damage to brain tissue.[2] Amyloid plaques are generated by aggregation of beta-amyloid peptide(Aβ), a 39-42 amino acid peptide.[2] Beta-amyloid peptide is cleaved from the transmembrane portion of amyloid precursor protein (APP), a 700 amino acid protein found in healthy human brains.[2] The function of APP is unknown but is believed to involve copper transport between neurons.[2,3] Normal copper concentrations in cerebrospinal fluid are much lower (10 μg/g) than those found in the beta-amyloid plaques (19 μg/g) and cerebrospinal fluid (25 μg/g) in Alzheimer's patients[1b] Since copper binds to both APP and Aβ, the disruption of copper homeostasis could play a role in the development of Alzheimer's.

In 2003, using NMR spectra, Multhaup, *et al.* determined the structure of the 65 amino acid residue copper binding domain of APP located in the hydrophilic N-terminus of APP.[4] In these NMR models, copper is bound by one tyrosine, one methionine, and two histidines, providing a possible explanation for the high affinity that APP has for copper (Figure 1a). The tetrahedral structure favors a Cu^{I} center; therefore, coordination of Cu^{II} by APP probably leads to reduction of Cu^{II} to Cu^{I} *in vivo*.[5]



A) B)

Figure 1. A) Copper Binding Domain in APP[4]. B) Imidazole Dimer suspected in Aβ[6].

Upon cleavage of APP and release from the cell membrane, Aβ peptide can bind Cu with attomolar affinity. NMR studies of this copper binding site reveal that copper is bound to three histidines and one tyrosine.[6] Interestingly, EPR and potentiometric studies have suggested that one of the histidines can serve as an imidazole bridge between two copper centers in an Aβ dimer (Figure 1b).[6] An imidazole bridge has been seen in only one other protein, superoxide dismutase, an observation suggesting that the copper chemistry of the two neuro-redox proteins may be similar.[2,7] Free beta-amyloid peptides

are found in healthy human brains, so the neurotoxicity seen in Alzheimer's can not be attributed to free Aβ but to aggregates of Aβ.[2]

In vitro studies show that Aβ aggregates become most neurotoxic in the presence of Cu^{II} ions, most likely due to the favorable Cu^{II}/Cu^{I} reduction potential in Aβ (+0.50 V vs. Ag/AgCl.)[2,8] Once Cu^{I} is formed, reaction with O_2 could initiate catalytic reduction of peroxy radicals through Fenton-type chemistry, leading to oxidative destruction of surrounding amino acids and brain tissue.[3,8] Aβ supports Cu^{II}/Cu^{I} reduction by oxidation of the thioether on methione-35 to a sulfoxide.[8,9] *In vitro* EPR studies have shown that Aβ peptides containing an oxidized methionine or a cysteine inhibit the reduction of Cu^{II} to Cu^{I}.[5] Although oxidation of methionine by Cu^{II} is thermodynamically unfavorable, endergonic reaction processes such as deprotonation of the methionine radical could shift the equilibrium to the right, allowing methionine oxidation to proceed.[8]

Using metal chelators to prevent Cu^{II} binding and subsequent reduction has recently become a focus of research into Alzheimer's treatments. In 1999, Beyreuther, *et al.* demonstrated that injecting metal chelating agents into the brains of rodents genetically modified to express APP significantly reduces brain deterioration.[9a] Possible metal chelators to be used as therapeutic agents for Alzheimer's disease must exhibit low toxicity, be able to cross the blood-brain barrier, display high specificity for copper.[9,10] Clioquinol, like other quinones, shows high binding affinity for copper and low human toxicity.[9,10] Phase II clinical trials study of the metal chelator clioquinol show decreases in the rate of memory loss in 26 patients showing promise that using metal chelating agents can reduce and possibly reverse the rate of memory loss in Alzheimer's patients.[11]

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