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Glutathione As Biomarker for Neurodegenerative Diseases

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Mini Review

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Abstract

Brain imaging is one of the main obstacles of modern disease diagnostics since it must allow accurate and quantitative measurements on a living organ that is placed in adults (in neonatal humans' skull penetration is somewhat easier) inside a bone cage. Inside is the brain, the main function controller of our body that must be done in a non- invasive way on the living operation brain. Many spectroscopic methods are trying to overcome the difficult access to the brain: Photoacoustic microscopy, Confocal microscopy, Two-photon microscopy, Optical coherence tomography, Scanning Laser Acoustic Microscopy, Acoustic microscopy, Ultrasonography, positron tomography, fluorescence methods, photo caustic microscopy and multi (two) photon imaging spectroscopy. The situation is complex since the brain is wrapped in the blood brain barrier allowing only selected molecules to pass from the blood stream to and out of the brain. In this chapter, we will survey the current situation of brain diagnostics with the aid of the spectroscopic methods. Brain research is integrated in aging research as a major area of interest. Aging is in most cased coupled with the loss of brain function and dementia. The neurodegenerative diseases, although identified by Dr. Alzheimer and his collaborators more than a century ago, continue as the leading causes of mortality among the elderly. Brain research in trying to give hope to those people but unfortunately our understanding in this area is limited. Oxidative stress contributes to neurodegenerative diseases pathophysiology and progression. The target was to describe central and peripheral metabolites of redox metabolism and to describe correlations between glutathione status, age, and disease severity.

Opening Words

The brain is the organ known to have its own guarding system, a huge blood vessels net that allows the entry of essential nutrients while blocking other substances. Unfortunately, this barrier is so effective in protecting against the passage of foreign substances that it may prevent life-saving drugs from being able to repair the damaged brain [1,2]. A partial list of more than 20 amyloid-related diseases includes Alzheimer's Disease, Parkinson's Disease, Huntington's Disease, prion diseases, familial amyloidosis, type II diabetes, Creutzfeldt–Jakob disease, **Lewy Body Dementia** and more than 20 more uncurbable and therefore fatal diseases. New studies are guiding researchers toward a breakthrough in the cure of non-infectious neurodegenerative diseases are associated with the accumulation of fibrillar proteins. These diseases exhibit features that are reminiscent of those of prionopathies, including phenotypic diversity and the propagation of pathology of Proteopathy (refers to a class of diseases in which certain proteins become structurally abnormal, and thereby disrupt the function of cells, tissues and organs of the body.). Retinopathies are the abnormal accumulation and toxicity of proteins in certain disease states. [3] Also, selective hyper proteolytic diseases have been referred to this category, e.g. critical illness myopathies or tumor cachexia [2]. The retinopathies comprise at least 30 diseases that affect a variety of organs and tissues, including Alzheimer's Disease (AD), Parkinson's Disease, type 2 diabetes, amyloidosis, and a wide range of other disorders [4,5].

In some retinopaths, an abnormal assembly can be designed on an exogenous protein, usually in the folded form of the same protein. In this way the disease state in a susceptible host can be induced by inserting a diseased tissue extract from an infected donor. The best known form of such an infectious (or transmitted) protopathy is a fertility disease, which can be transmitted by exposure of a host organism to a purified fertility protein in a disease-causing structure [6,7]. There

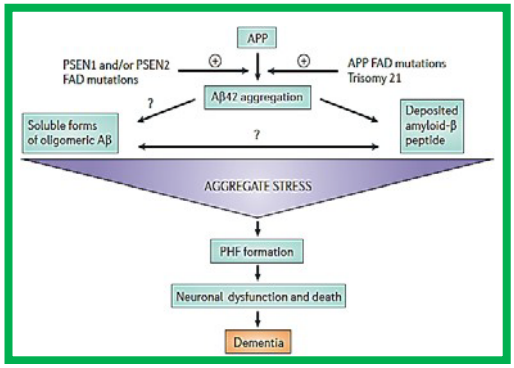
is now evidence that other protopaths can be injected by a similar mechanism, including amyloidosis AA, apolipoprotein AII amyloidosis, and amp amyloidosis [8,9]. In all of these cases, an aberrant form of the protein itself appears to be the pathogenic agent. Already one hundred years ago, Jacob Heinrich Lewy described intracellular eosinophilic inclusions in the brains of patients with "paralysis agitos ", commonly known as Parkinson's Disease [10].

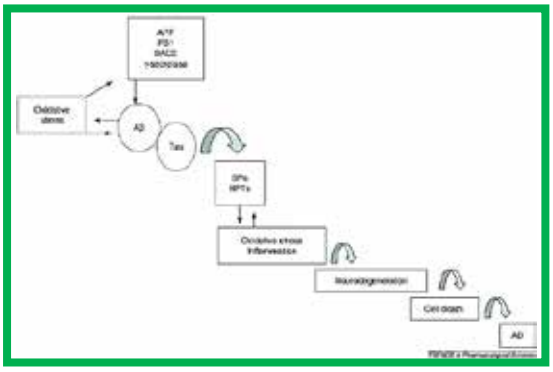
A group of psychiatrists at the Psychiatric Clinic at the University of Munich: A. Alzheimer and Solomon

Fuller sit in the front row; Standing in the back row, from left to right: Bronciini, Bronciini, von Norbert, Ranky, and unidentified.

Mighty progress has been made over the past decades in analyzing the content and formation of Levy bodies and their relationship to degenerative diseases. Multitudes of researchers have been engaged in the last hundred years since the identification of the phenomenon. However, only little has been achieved in the diagnosis of the initial stages of the diseases. Here we try to survey the chemistry aspect of the diagnostic efforts.

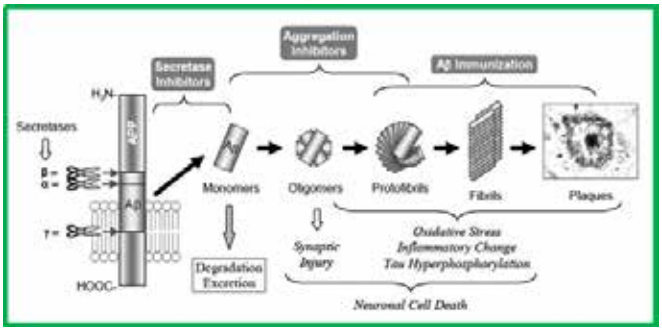
The health authorities in the USA gathered some information on the effort in the chemistry of neurodegenerative diseases. A sample could be found on the Internet [11].





Amyloid Beta Hypothesis Oxidative stress Hypothesis, Glutathione [12-14]

It has been more than 10 years since it was first proposed that the neurodegeneration in Alzheimer's disease (AD) may be caused by deposition of amyloid β-peptide (Aβ) in plaques in brain tissue. According to the amyloid hypothesis, accumulation of Aβ in the brain is the primary influence driving AD pathogenesis. The rest of the disease process, including formation of neurofibrillary tangles containing tau protein, is proposed to result from an imbalance between Aβ production and Aβ clearance.



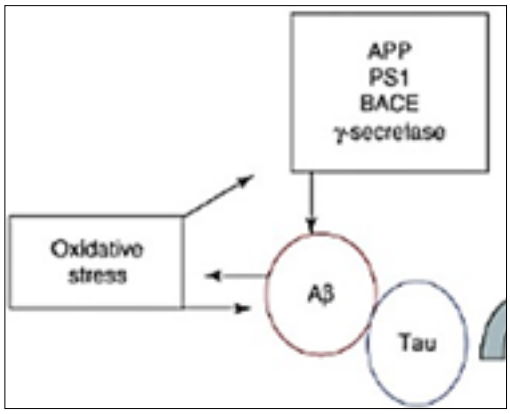
Schematic illustration of the Aβ-amyloid cascade from APP cleavage by secretases to generate Aβ monomers, to plaque formation, via oligomers, protofibrils, and fibrils. Causative factors for neuronal injury are indicated in italic letters under the Aβ pathway. Anti-amyloid agents are also shown in solid white letters above the therapeutic targets in the Aβ pathway [1].

Glutathione (GSH) is the major cellular thiol present in mammalian cells and is critical for maintenance of redox homeostasis [15]. However, current assay systems for glutathione lack application to intact animal tissues. Although reports exist on the quantitative imaging of glutathione in hippocampal neurons and glia in culture using bimane fluorescence, there is an urgent need to map the levels of glutathione in intact brain with cellular resolution (acute tissue slices and live animals) [16]. Glutathione is a major antioxidant system in the mammalian central nervous system (CNS). Abnormalities of GSH metabolism have been associated with many disorders of the CNS, including Parkinson's, Alzheimer's, and Huntingdon's diseases and ischemic/reperfusion injury. Investigation of GSH levels in the CNS generally relies on biochemical assays from cultures enriched for different cell types.

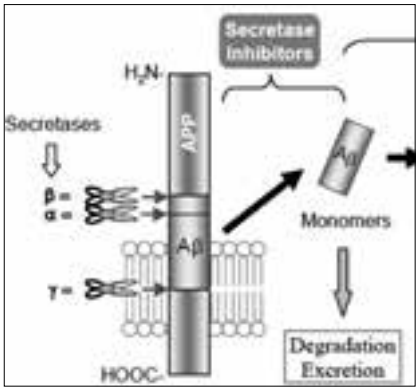
There is Oxidative Damage Is the Earliest Event in Alzheimer Disease [17]. Glutathione S- transfer, commonly abbreviated (GST), refers to a group of enzymes that employ glutathione

in many reactions that contribute to the conversion of many compounds such as therapeutic drugs, carcinogens, and products involved in oxidative stress. The evaluation of the enzyme Glutathione Transferases (GST), a method that may become instrumental in the choosing glutathione Transferases level as a biomarker for Alzheimer's Disease [18,19]. Scientist developed a spectrophotometric assay for the glutathione conjugation and determined specific activities with a range of human GSTs as well as some rat GSTs for comparison. The ubiquitous GST P1-1 showed the highest activities with the 6-halogenopurines, which bodes well for the application of pro-probes for human investigations.

The acceptance of the "oxidative stress" as a major process in the early evolution of neurodegenerative pathology has become a major trend in this field of research.

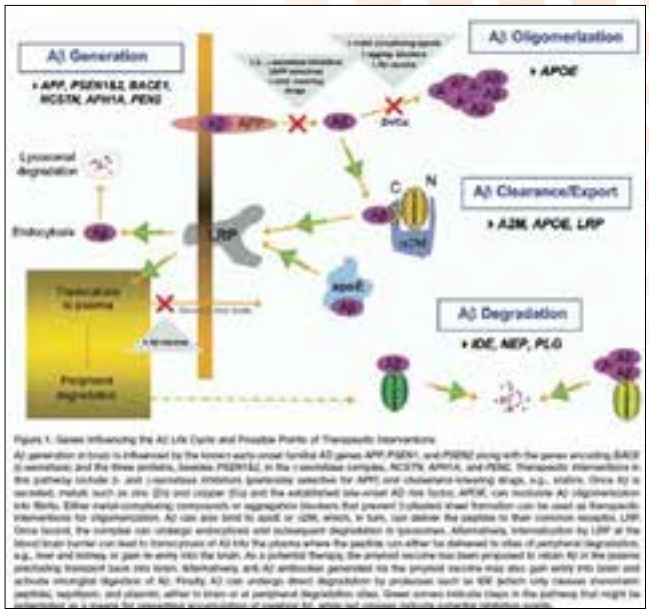


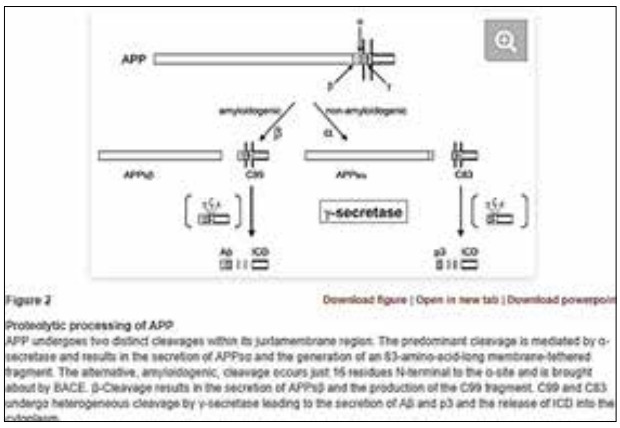
Oxidative stress early stages



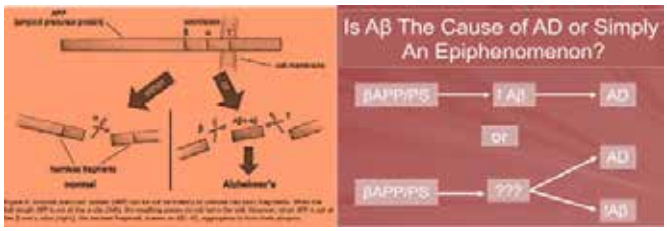
Amyloid hypothesis early stages

Digestion of beta-amyloid precursor protein is supposed to produce fractures that do not harm or harm fractures. There is an evolving consensus seeing amyloid cerebrospinal fluid (Aß) as a key biological marker for the Alzheimer's disease stage of mild cognitive impairment. Aβ is directly involved in the pathogenesis of AD or in close correlation with other primary pathogenic factors. It is produced from amyloid precursor protein (APP) by proteolytic processing dependent on the enzyme 1-closed AP-site and the γ-secretase complex, and is degraded by a wide variety of proteases. This review summarizes targeted proteolytic studies of Aβ in biological fluids and identifies clinical useful markers of Aβ homeostasis in AD disruption.





Alternative splitting b synuclein enzymes, digestion of beta-amyloid precursor protein [20]. Is it a secondary effect or byproduct that arises from but does not causally influence a process, in particular.



Such defects are considered to the postulated initiation of the cascade leading to Metabolic abnormities In Alzheimer's Disease to confirm the characteristics of metabolic abnormalities in Alzheimer's Disease, regional metabolic activity summarized in tennis of quantitative cerebral glucose metabolic rate.

Introduction

Glutathione (GSH) is required for many critical cell processes, buy plays a particularly key role in the care and regulation of the thiol-redox status of the cell. GSH is the most important endogenous antioxidant and plays an important role in the

detoxification of xenobiotics and their metabolites, as well as in the maintenance of the intracellular redox balance.

With current estimates of 36 million people affected worldwide. Alzheimer's Disease (AD) - a disorder characterized by impossible progressive impairment of memory and other cognitive functions - is the most prevalent form of dementia. Therefore, identifying biological markers that can serve as a reliable surrogate for the onset of the disease disorder and its progression is of paramount importance for information on intervention. Oxidative stress, which is a common denominator of a number of pathophysiological events associated with AD, appears to be a major factor in the pathogenesis and progression of AD. Alzheimer's Disease (AD) represents one of the great and unresolved medical needs facing society during this thousand years. Despite considerable work over the last quarter. There are no drugs that attack the pathophysiology underlying the Disease. One of the cardinal characteristics of counting Disease is the placement of platelets composed of peptides (3-amyloid, A, s) in the brain. Especially in areas related to cognition and memory overproduction of A that appears to be directly neurotoxic. Can be detected in the earliest stages of AD and. in fact.

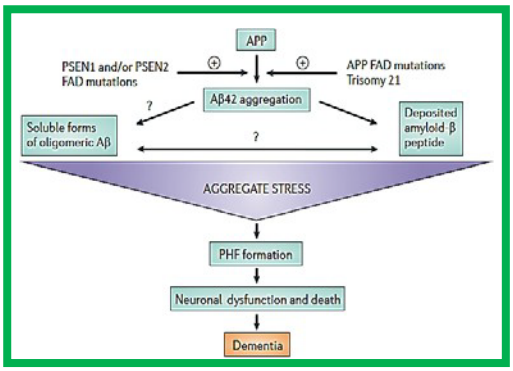
Before cognitive function can be discerned a father is extracted from a protein from its flesh. Amyloid precursor protein (APP), by proteolytic processing at its N- and C-terminals by enzymes α,β- and γ-secretase enzymes [21,22].

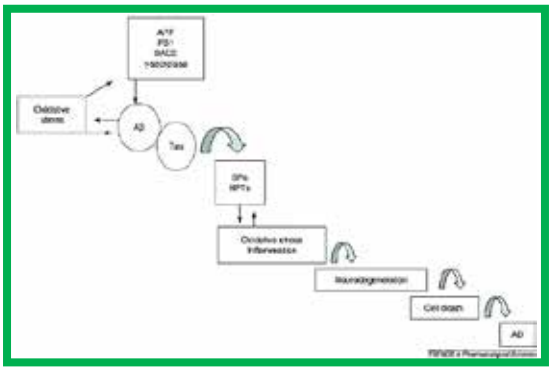
Two main hypotheses are today directing neurodegenerative diseases research. The Amyloid hypothesis and the oxidative stress mechanism. Although the accumulation of data supports the Amyloid beta aggregation hypothesis, in conclusion, the oxidative stress hypothesis (inflammation) of AD is very much alive and viable, but a great deal of work needs to be done to design future studies and appropriate clinical trials that will conclusively establish the role of oxidative stress in AD pathogenesis.

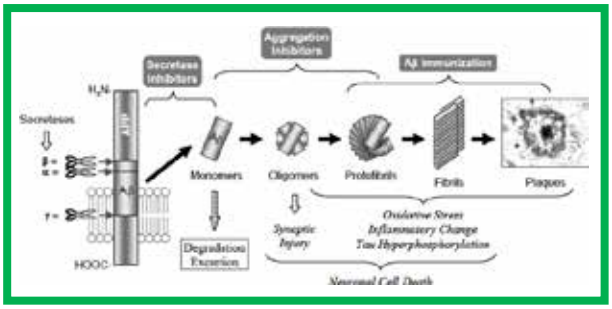
One high hurdle is the lack of a quantitative instrumental method to diagnose and follow the Disease from initial stages. The development of new drugs depends a lot of such a helpful device.

Alzheimer's Disease is one of the many neurodegenerative disorders that are tormenting many, older people. It is in the unflavored situation where many theories on what initiates the cascade of events and when exactly it started to affect the life of the ill people [23].

Today, only a post mortem analysis of the patient brain can diagnose 100% the Alzheimer's Disease.







Amyloid Beta Hypothesis Oxidative stress Hypothesis Glutathione [24, 25]

Schematic illustration of the Aβ amyloid cascade from APP cleavage by secretases to generate Aβ monomers, to plaque formation, via oligomers, protofibrils, and fibrils. Causative factors for neuronal injury are indicated in italic letters under the Aβ pathway. Anti-amyloid agents are also shown in solidwhite letters above the therapeutic targets in the Aβ pathway [9].

The oxidative (inflammatory) step Is the Earliest Event in Alzheimer Disease [26,27]. Observations indicate that increased oxidative damage is an early event in AD that decreases with disease progression and lesion formation. These findings suggest that AD is associated with compensatory changes that reduce damage from reactive oxygen. The activities and expression of several antioxidant enzymes such as Cu/Zn- and Mn-superoxide dismutase, glutathione peroxidase, glutathione reductase, and catalase have been studied in AD and could be in part responsible for the decrease in oxidative damage we observed.

"Clinical criteria for the diagnosis of AD include dementia established by clinical examination and neuropsychological testing, deficits in two or more areas of cognition, progressive worsening of memory and other cognitive functions, no disturbance in consciousness, onset between ages 40 and 90, and absence of systemic disorders or other brain disease to account for the progressive cognitive decline. A diagnostic laboratory test for AD has not been found and AD remains a diagnosis of exclusion. A definitive diagnosis cannot be made without neuropathological confirmation.

Two neuropathological criteria are available for the diagnosis of AD. The major microscopic alterations in AD are SP and NFT formation, selective neuron loss and shrinkage, synapse loss, neuropil thread formation, and amyloid antipathy"[28]. Decrease in glutathione is also a major event that is associate with neurodegenerative (Alzheimer's, Parkinson's, as examples) diseases [29]. Most antioxidant defenses (SOD, GSH-PX, a-tocopherol) do not seem to be substantially changed in the aging brain, but glutathione (GSH). Concentration and the glutathione redox index are lowered [30].

Looking At Glutathione in the Brain

The event in which polypeptides are cleaved in the inner brain is the creator of a chain of other events that brings about the formation of fibrils and plaques that finally kill the neurons in the brain of the patients. This process is going on for many tears, decades. The first stages have very little expression on the behavior, memory of the sick. Under thesis circumstances.

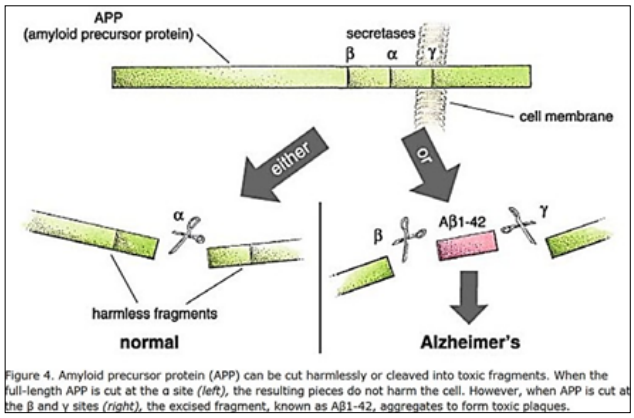


Figure 4. Amyloid precursor protein (APP) can be cut harmlessly or cleaved into toxic fragments. When the full-length APP is cut at the α site *(left)*, the resulting pieces do not harm the cell. However, when APP is cut at the β and γ sites *(right)*, the excised fragment, known as Aβ1-42, aggregates to form toxic plaques.

What's wrong with an Alzheimer's patient? There are about half a dozen different genetic circumstances that can trigger the disease, and probably others that are not currently known. They all lead to the same molecular pathology - the formation of aggregates of an "unfolded" fraction of a normal protein. This normal protein, the amyloid precursor protein (or APP) [a-b], is embedded in the outer membrane of cells in a variety of tissues. During its normal function, the APP is cut into segments, or peptides, at three specific sites targeted by α-, β-, and γ-secretase enzymes, respectively. During the development of Alzheimer's disease, the APP protein is cleaved at the β and γ sites, resulting in a fracture that folds itself into a sticky, self-accumulating form. This peptide can have between 39 and 43 amino acids, given the different variability of the β-secretase cleavage site. Not all variants are produced in equal amounts - what is known as Aβ1 - 40 is most common - and some forms are worse than others, with the most toxic peptide being Aβ1-42. This fraction includes the first 42 amino acids after the ß-secretase site and easily forms insoluble lumps in the brain. These aggregates are toxic and aggressively lead to dysfunction of nearby brain cells and their resulting death and removal. Once these brain cells disappear, there is currently no way to replace them [31[a-b]].

Disruption of glutathione homeostasis and changes in glutathione-dependent enzyme activity are increasingly involved in the induction and progression of neurological diseases, including Alzheimer's Disease, Parkinson's and Huntington's, amyotrophic atherosclerosis, and Friedrich's ataxia [32].

Various lines of evidence suggest that the operating system in the brain (oxidative stress) is an underlying factor underlying the etiology of AD. GSH levels have been consistently proven to reflect operating system status. Furthermore, the literature reviewed so far reveals a strong correlation between pathology and counting and reduced GSH levels. These findings spurred the development of tests for GSH levels as a biological marker for AD. A number of methodologies have been developed to evaluate GSH levels in peripheral biological samples, such as blood. Recent advances in technology have also enabled non-invasive in vivo measurement of GSH directly in different brain regions using MRS. We discuss recent findings from studies using different GSH measurement methodologies and evaluate their relative potential to serve as a reliable measure of GSH levels.

Comments on new blood test suggestions for accurate early detection of Alzheimer's can be found, samples available online [33].

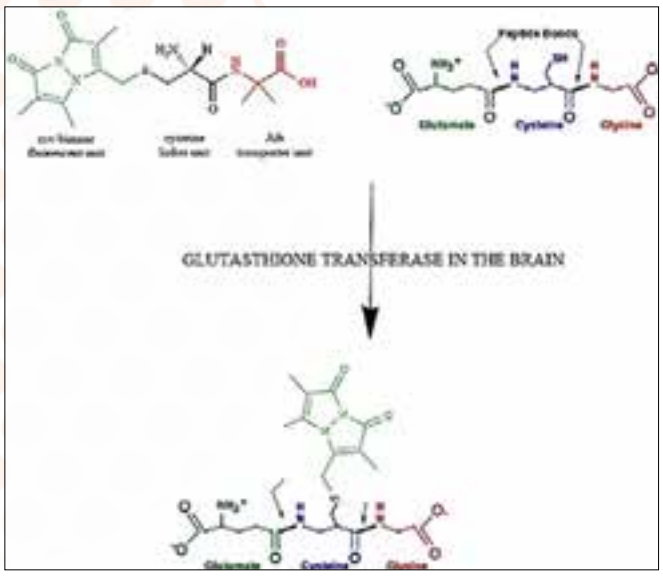
We suggest measuring a "biphotonic laser scanning microscopy" (TPLSM) for direct measurement of glutathione (GSH) as a combination of its S-Bimane glutathione (GSB) in blood samples after AIB-CYS-BIMANE injection (there is an analogue to S- bimanylmercaptoacetic acid as), Followed by sleep of the animal [34]. A Decrease in GSH is indicative for Oxidative Stress (OS) , In the past, the presence of an efflux system in mouse cerebral micro vessel endothelial cells was examined in vitro by using a fluorescent glutathione-bimane (GS-B) conjugate [35,36]. Oxidative stress and the diminishing Glutathione because of this early process suggest that Glutathione can be viewed as a molecular whistleblower for the Alzheimer's Disease [37].

It was found (Post Mortem) that in certain regions of the brain, GSH reduction in these regions correlated with decline in cognitive functions [38]. Receiver operator characteristics analyses evidenced that hippocampal GSH robustly discriminates between mild cognitive impairment (MCI) and healthy controls with 87.5% sensitivity, 100% specificity, and positive and negative likelihood ratios of 8.76/.13, whereas cortical GSH differentiates MCI and AD with 91.7% sensitivity,

100% specificity, and positive and negative likelihood ratios of 9.17/.08.

Conclusion

The present study provides compelling in vivo evidence that estimation of GSH levels in specific brain regions (with magnetic resonance spectroscopy) constitutes a clinically relevant biomarker for MCI and AD. Glutathione relates to neuropsychological functioning in mild cognitive impairment [39].



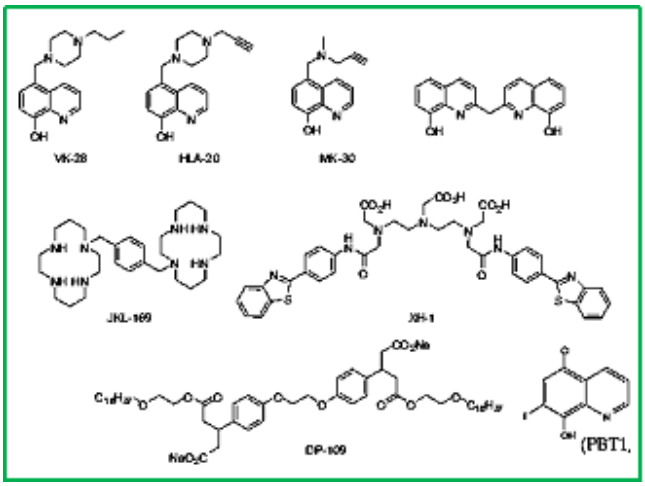
The Staining of Glutathione in the living brain and then determining the content in the blood

Oxidative Stress Hypothesis -Neuropathic diseases [40]

To defend against free radicals, living organisms have learned over time to generate antioxidants and repair enzymes to remove and/or repair molecules that are oxidized. A few enzymatic antioxidants are synthesized by cells. These include Cu/Zn- and MN-superoxide dismutase (SOD) methionine sulfoxide reductase [41]. Other none-enzymatic antioxidants and metal chelators. Researchers pointed out that there is a crucial role of metal ions in neurodegeneration, it masy become the basis for a promising therapeutic strategy, a chelation therapy could be a valuable therapeutic approach, since metals are a pharmacological target for the rationale design of new therapeutic agents directed towards the treatment of neurodegeneration [42,43].

Metal Ions

Metal ion chelators have been suggested as potential therapies for diseases involving metal Ion imbalance. Neurodegeneration is an excellent target for exploiting the metal chelator approach to therapeutics. In contrast to the direct chelation approach in metal ion overload disorders, in neurodegeneration the goal seems to be a better and subtle modulation of metal ion homeostasis, aimed at restoring ionic balance. Thus, moderate chelators able to coordinate deleterious metals without disturbing metal homeostasis are needed. To date, several chelating agents have been investigated for their potential to treat neurodegeneration, and a series of 8-hydroxyquinoline analogues showed the greatest potential for the treatment of neurodegenerative diseases.



Chemical structure of chelators tested in AD

A series of 8-hydroxyquinoline analogues (VK-28, HLA-20 and MA-30) have shown the greatest potential for the treatment of several neurodegenerative diseases and one of these compounds, clioquinol (PBT1), reached the pilot phase II clinical trial, which suggests that clioquinol improves cognition and lowers plasma levels of Ab42 in some patients.

The regional distributions of iron, copper, zinc, magnesium, and calcium in parkinsonian brains were compared with those of matched controls. In mild Parkinson's Disease (PD), there were no significant differences in the content of total iron between the two groups, whereas there was a significant increase in total iron and iron in substantia nigra of severely affected patients. Although marked regional distributions of iron, magnesium, and calcium were present, there were no changes in magnesium, calcium, and copper in various brain areas of PD [44].

Oxidative Stress-Glutathione and Glutathione transferases as Biomarker

Alzheimer's Disease (AD) is the most generic A type of degenerative disorder with dementia. In its Spanish form, AD results from a combination of genetic factors with various Afghan events. Among them, oxidative metabolic reactions and their by-products have been consistently affected in the pathogenesis of AD and represent the biological basis for the "oxidative stress hypothesis" of AD. Many studies demonstrate that various biological markers of mediating events with increased oxidative stress in the AD brain. Brain glutathione levels - a new Alzheimer's disease [45].

Metal Ions, pH, and Cholesterol Regulate the Interactions of Alzheimer's Disease Amyloid-β (Aβ) Peptide with Membrane Lipid. The interaction of Aβ peptides with the lipid matrix of neuronal cell membranes plays a key role in the pathogenesis of Alzheimer's Disease. By using EPR and CD spectroscopy, it

was found that in the presence of Cu^{2+} or Zn^{2+}, pH, cholesterol, and the length of the peptide chain influenced the interaction of these peptides with lipid bilayers. In the presence of Zn^{2+}, Aβ40 and Aβ42 both inserted into the bilayer over the pH range 5.5–7.5, as did Aβ42 in the presence of Cu^{2+}. In a comprehensive research work, scientists noticed that a significantly lower glutathione content was present in pooled samples of putamen, globus pallidus, substantia nigra, nucleus basalis of Meynert, amygdaloid nucleus, and frontal cortex of PD brains with severe damage to substantia nigra, whereas no significant changes were observed in clinicopathologically mild forms of PD [36]. The quantitative imaging of glutathione in hippocampal neurons and glia in culture using mono- or chloro-bimane refers to the Kosower and collaborators work. There is Oxidative Damage Is the Earliest Event in Alzheimer Disease [46,47]. Gutathione S-transferase, commonly abbreviated as GST, refers to a group of enzymes that employ glutathione in many reactions that contribute to the conversion of many compounds such as therapeutic drugs, carcinogens, and products involved in oxidative stress. Glutathione is an essential metabolic molecule produced in the liver of humans and animals. GST (Glutathione S-transferase) from a family of detoxifying enzymes contains a lot of micro-cytokine and mitochondrial proteins, which make up considerable parts of the enzyme's body. They exist in prokaryotes and macriotics, where they play the role of speeding up different responses and at the same time receive xenobiotic and endogenous substrates. Each of the eukaryotic species has multiple GST isoenzymes that are bound to cytosols and membranes. Each exhibits catalytic and non-catalytic binding properties.

The is decreased glutathione transferase activity in brain and ventricular fluid in Alzheimer's Disease [48]. Thiols in general, which are components of many proteins and simple molecules, such as glutathione (GSH) and cysteine (Cys), play an important role in the cellular antioxidant defense system. 1 GSH is the most abundant intracellular nonprotein thiol (1-10 mM). 2 It has a pivotal role in maintaining the reducing environment in cells and acts as the redox regulator because thiols exist in redox equilibrium between sulfhydryl and disulfide forms.3-5 Intracellular thiol levels change dramatically in the response to oxidative stress. 1 Thus, the quantitative detection of intracellular thiols is of great importance for investigating cell functions. Blood-Brain Barrier-Penetrating 6-Halogenopurines Suitable as Pro-Probes for Positron Emission Tomography are Substrates for Human Glutathione Transferases [49]. Scientist developed a spectrophotometric assay for the glutathione conjugation and determined specific activities with a range of human GSTs as well as some rat GSTs for comparison. The ubiquitous GST P1-1 showed the highest activities with the 6- halogenopurines, which bodes well for the application of pro-probes for human investigations.

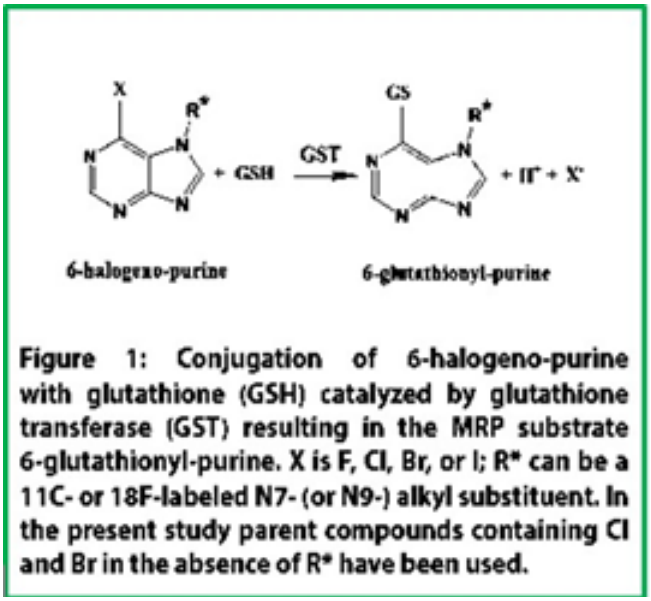


Figure 1: Conjugation of 6-halogeno-purine with glutathione (GSH) catalyzed by glutathione transferase (GST) resulting in the MRP substrate 6-glutathionyl-purine. X is F, Cl, Br, or I; R* can be a 11C- or 18F-labeled N7- (or N9-) alkyl substituent. In the present study parent compounds containing Cl and Br in the absence of R* have been used.

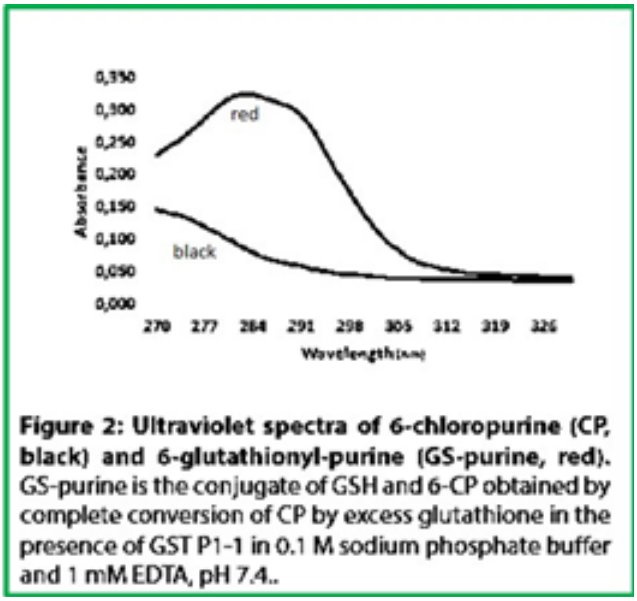


Figure 2: Ultraviolet spectra of 6-chloropurine (CP, black) and 6-glutathionyl-purine (GS-purine, red). GS-purine is the conjugate of GSH and 6-CP obtained by complete conversion of CP by excess glutathione in the presence of GST P1-1 in 0.1 M sodium phosphate buffer and 1 mM EDTA, pH 7.4..

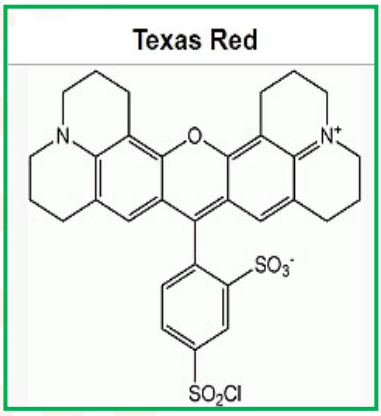
Glutathione transferase assay

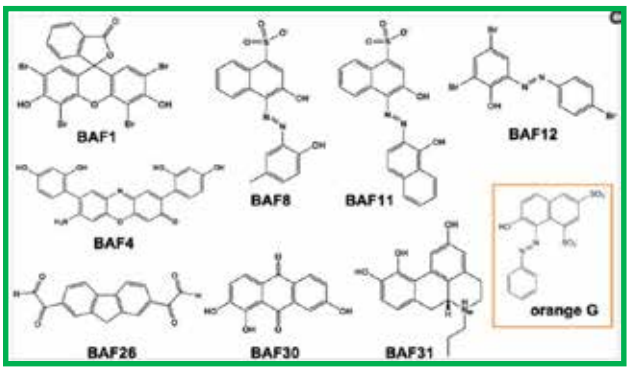
Since bromo- (or chloro-) bimanes were shown to have a very useful and sensitive application in reacting with thiols to produce fluorescent labelling, attempts were made to stain brain tissues [50]. Glands and galea with the direct use of the halo-bimanes [51]. The enzymes of the Glutathione S-Transferases family may become instrumental in choosing glutathione Transferases level as a biomarker for Alzheimer's Disease [32,52].

Slicing brain in the laboratory serves research abundantly. However. Dealing with living brains is the way to go. In respect to the biogenesis of brain compounds, Nedergaard and coworkers found out in mice that while seeping, the waste is excreted from the brain via the spinal fluids and then transported in the blood system to the regular way the living organism gets rid of such wastes [53,54]. Scientist observe the decrease of free SH groups inproteins extracted from the hippocampus of AD patients provides additional evidence for increased oxidative damage of proteins in a vulnerable region of the AD brain [55].

Staining Amyloid-beta

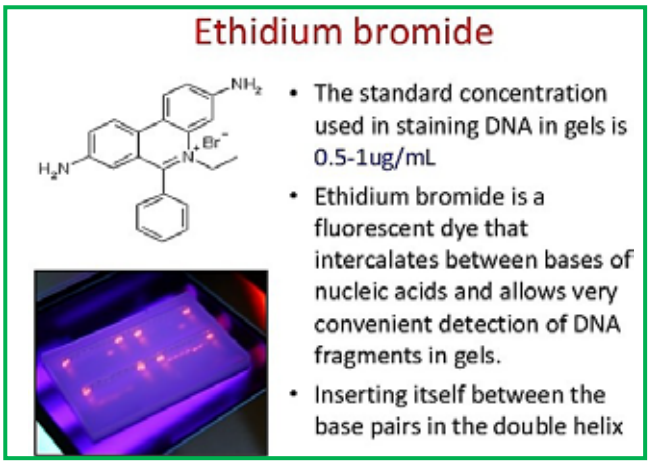
In contrast, Use of Thioflavin derivative resolve individual Aβ plaques and cerebrovascular amyloid in living microscopy. Future studies will include imaging amyloid load in transgenic mice using newly developed high-resolution micro PET, a technology that will provide a direct transition to PET imaging studies in human subjects [56,57].

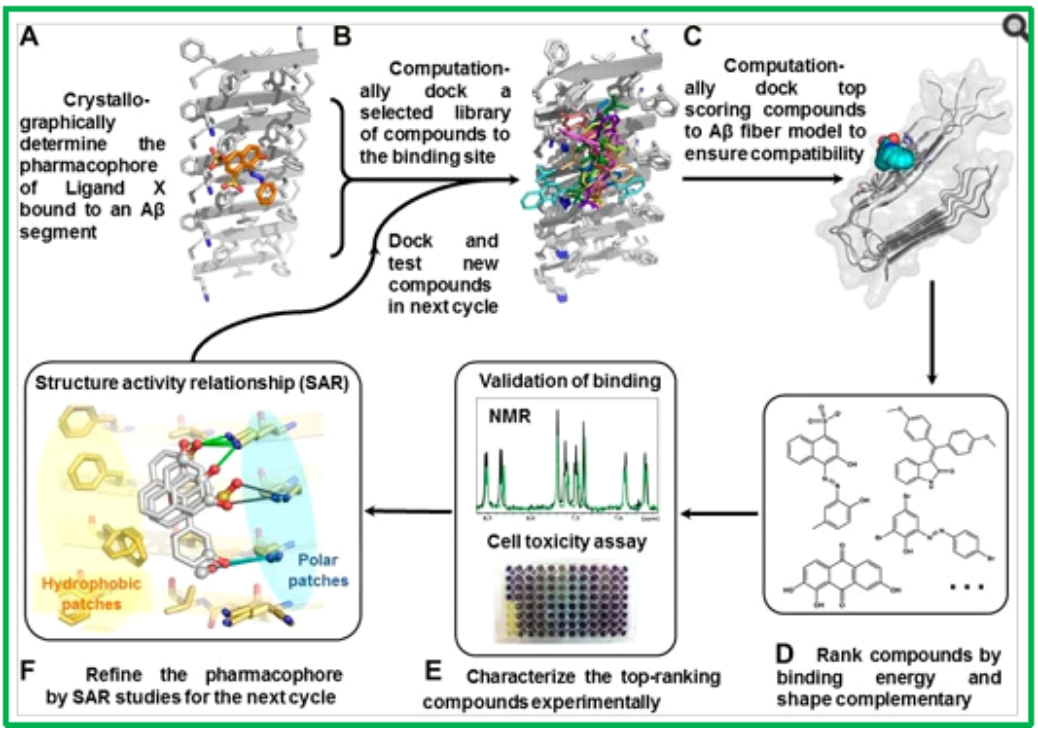




Diversified chemical structures of 8 active BAF compounds that reduce Aβ toxicity.

Orange G in an orange box is also displayed for comparison.

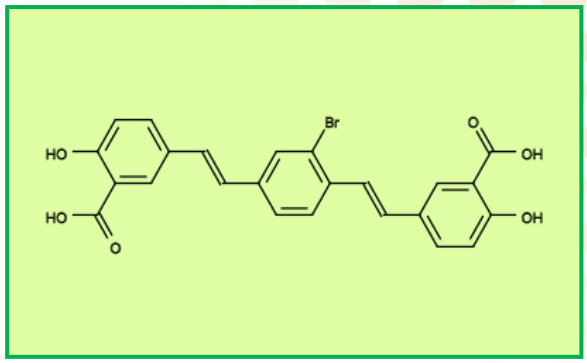




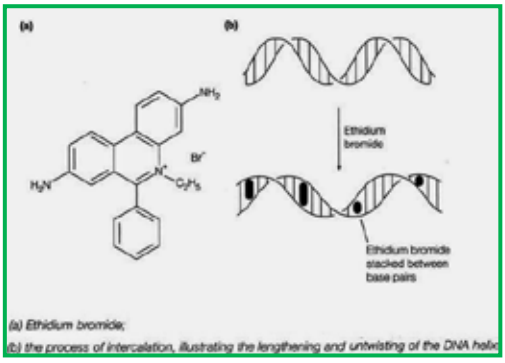
Structure-based identification of small compound inhibitors of Aβ toxicity [52].

A typical commercial reagent for amyloid staining is "Amylo-Glo" [58-61]:

Compound [53]: "Amylo-glo"; specification: Styrylbenzene derivative; Appearance: Yellow solution; Molecular Weight: 392; Filter system for visualizing: UV Ethibium Bromide: EtBr, 2,7-Diamino-10-ethyl-6-phenylphenanthridinium bromide; Appearance: light red-orange solution; Molecular Weight: 394.32. 392; Filter system for visualizing: UV Purity: Thin layer chromatography using alumina plates and a solvent system of ethanol and water (3:1) revealed the presence of two fluorescent isomers. No amount of starting material was detected. Biol. activity: Excitation Peak for Amylo-Glo: 334 Emission Peak: 533 nm - unbound, 438 nm when bound to amyloid.

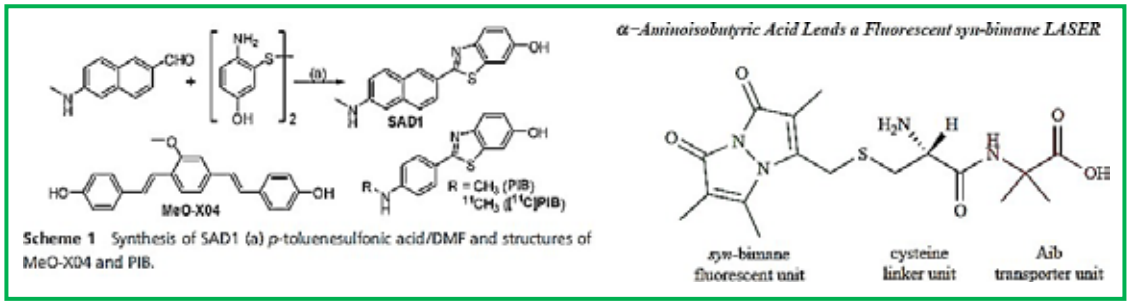


Styrylbenzene derivative and Ethidium Bromide



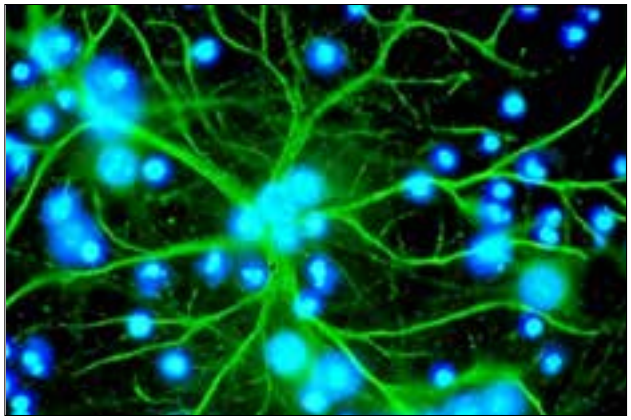
(a) Ethidium bromide;
(b) the process of intercalation, illustrating the lengthening and untwisting of the DNA helix

Today, two agents (see below) that might be useful as probes to detect their biomarkers of neurodegenerative diseases were prepared by two groups. Recently, such small molecules that could be supplied to the brain via the blood streams (abdominal or tail injection) have been reported by an Israeli and a Korean groups and introduced to the inner brain by crossing the blood brain barrier (BBB)[62,63].



Scheme 1 Synthesis of SAD1 (a) *p*-toluenesulfonic acid/DMF and structures of MeO-X04 and PIB.

It is still a great challenge to use either biomarker, β-amyloids or Glutathione or other that are produced in the brain, probably in the hippocampus gland in the early events of the neurodegenerative Disease.



The Green Branches of An Astrocyte, one of Several Kinds of Glial Cells, Surrounded By Blue Nuclei of other Cells (Credit: Karin Pierre, Institut De Physiologie, Unil, Lausanne. Via Wikimedia Commons)[64]

Recently one may find many advertisements of blood tests for Alzheimer early diagnostics. The area is very active in this respect. Since there is no cure for Alzheimer's Disease, but medications, sensory therapy and more that can help its symptoms. And to get the full benefit of the treatments, early diagnosis is important. Learn more about Alzheimer's diagnosis and treatments. The Rowan University [65] announced that Blood test for Alzheimer's shows 100% accuracy in early trials.

Scientist are looking for frontiers were new biomarkers foe the neurodegenerative diseases exist. Smell[66] and vision[67] are connecting directly through nerves into the brain.

TAU induced diseases – High molecular weight proteins

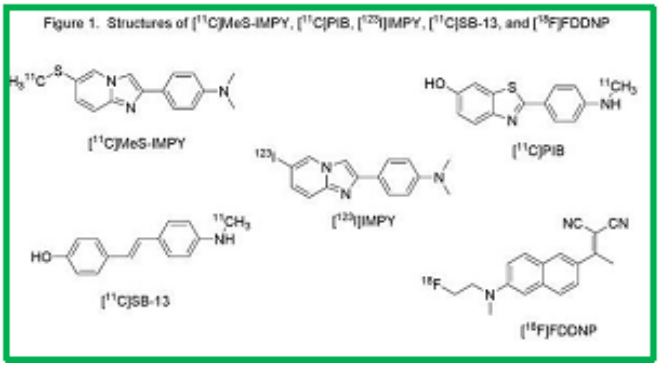
Amyloid senile plaques and tau neurofibrillary tangles are neuropathological hallmarks Of Alzheimer's disease,

Parkinson's disease, which accumulates in the cerebral cortex areas in people with mild cognitive impairment who are at risk for Alzheimer's disease. Non-invasive methods for detecting these abnormal proteins are potentially beneficial Development of surrogate markers for drug detection and diagnosis.

Tao protein is a protein (MAP) attached to a highly soluble micro-tube. In humans, these proteins are found primarily in neurons compared to non-neuronal cells. One of the main functions of the Tao is to change the stability of the axon micro-tubes. MAPs of the other nervous system may perform similar functions, as suggested by take-out mice that did not show abnormalities in brain development - possibly due to compensation for the lack of beta by other MAPs. [10] Tao does not exist in dendrites and is active mainly in the distal parts of the axons where it provides micro-tubular stabilization but also flexibility as needed. This is in contrast to MAP6 (STOP) proteins in the proximal parts of the axons, which essentially lock the microtubules and MAP2 which stabilizes the microtubules in dendrites.

PET of amyloid brain and tau has been shown to have a mild cognitive impairment (FDDNP-PET scan) can differentiate between people with mild cognitive impairment.

For people with Alzheimer's disease and those without cognitive impairment. 18F-PET based on the application of 2- (1- {6 - [(2- [fluorine-18] fluoro-ethyl) (methyl) amino] -2-naphthyl} - ethylene) melonionitrile (FDDNP) is the emission tomography of First positron (PET) molecular [68,69].



Reagents applied in PET in brain Disorder analysis imaging probe to visualize Alzheimer's Disease (AD) pathology in living humans. This technique is potentially useful as a noninvasive method to determine regional cerebral patterns of amyloid plaques and tau neurofibrillary tangles.

PET of Brain Amyloid and Tau in Mild Cognitive Impairment are indicative [70]. Are TAU-based Therapies for Alzheimer's Disease: Wave of the Future? [71].

Focus Points [72]

Tao protein is essential for proper synaptic and nerve function.

- Tao dysfunction has been replaced by the pathogenesis of Alzheimer's disease.
- Tao-based therapies for Alzheimer's disease are currently being tested. A combination of Tao-based therapies with amyloid-based therapies may be necessary to effectively treat Alzheimer's disease. APOE (genetics, apolipoprotein E) predicts the P but not the pathology of Tao Alzheimer in normal cognitive aging.

Typically, parallel PET analysis was performed with 11C and 18F positron sources, 11C-PIB and 18F-FDlutemetamol were tested. The absorption of 18F-flutemetamol can be easily quantified using an uptake ratio of the reference area after 80 min and provides good discrimination between AD patients and cancer and health. Results of these 1 steps justify further pursuit of 18F-flutemetamol as a biomarker for count-related amyloidosis, with wider availability for clinical and research purposes than the "parent molecule", 11C-PiB

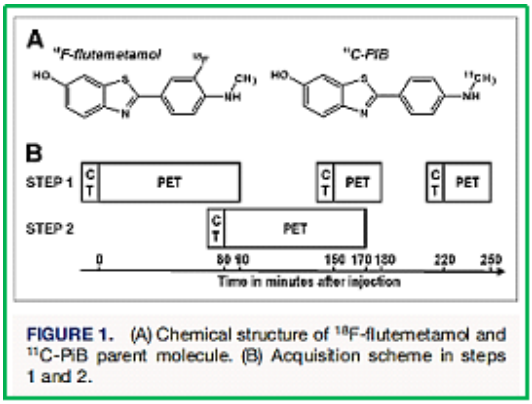


FIGURE 1. (A) Chemical structure of ^{18}F-flutemetamol and ^{11}C-PiB parent molecule. (B) Acquisition scheme in steps 1 and 2.

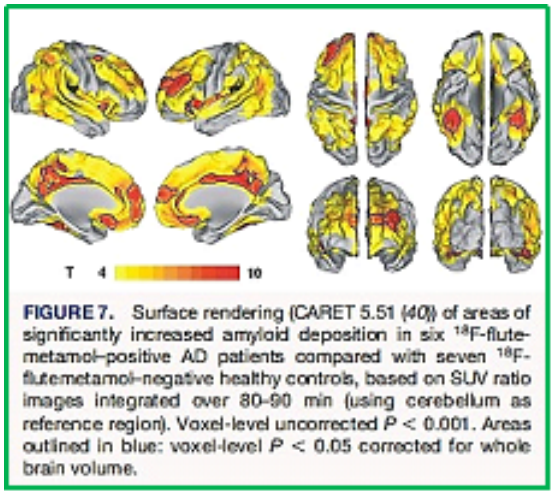
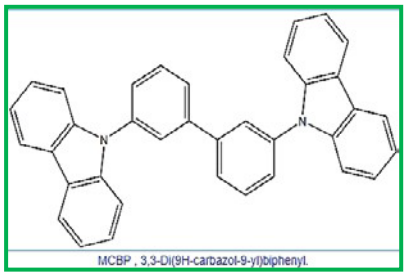
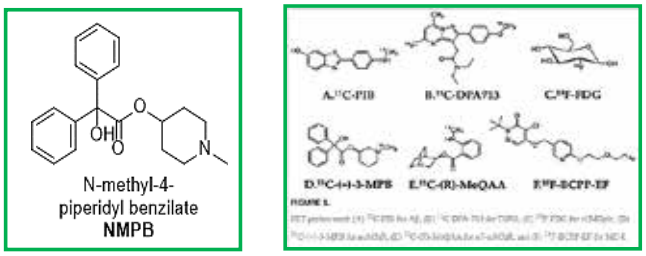


FIGURE 7. Surface rendering (CARET 5.51 (40)) of areas of significantly increased amyloid deposition in six ^{18}F-flute-metamol–positive AD patients compared with seven ^{18}F-flutemetamol–negative healthy controls, based on SUV ratio images integrated over 80–90 min (using cerebellum as reference region). Voxel-level uncorrected $P < 0.001$. Areas outlined in blue: voxel-level $P < 0.05$ corrected for whole brain volume.

The testing of two positron sources for testing amyloids in a living AD patient reagent and results [52].



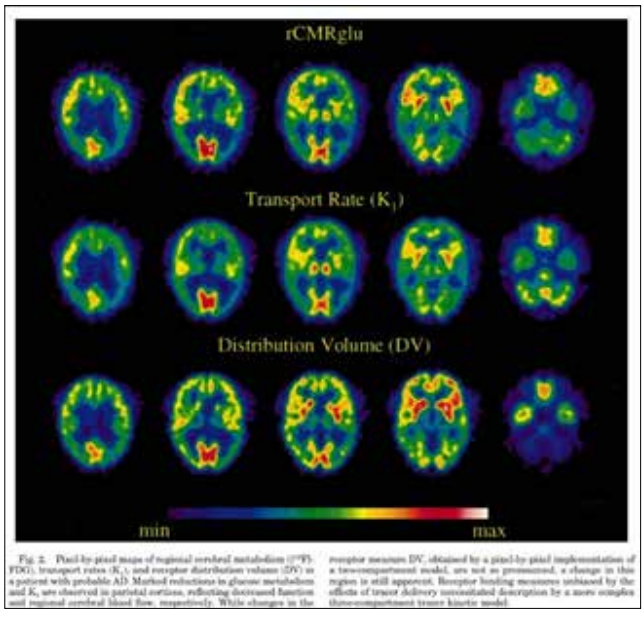


Chemicals structures of agents applied in PET analyses

Frequency of people with binding potential of the High Mean Cortex (MCBP)

For PIB, age ranged from 0% aged 45-49 years to 30.3% aged 80-88 years. Decreased levels of CSF Aβ42 appear to start earlier (18.2% aged 45-49 years) and increase with age at higher frequencies (50% aged 80-88 years) compared to the increase in MCBP. There is an effect of gene dose for the APOE4 genotype, with larger MCBP increases and larger decreases in CSF Aβ42 with an increased number of APOE4 alleles. People with APOE2 have no increase in MCBP with age and having higher CSP Aβ42 levels than people without the APOE2 allele. No APOE4 or APOE2 effect on CSF tau or ptau181.

Mild cognitive impairment is a transitional stage between normal aging and Alzheimer's disease. A recently published study shows that the prevalence of mild cognitive impairment, characterized by a cognitive decline without impairing the ability to perform activities of daily living, is 19% among people under 75 and 29% among those 85 years. Or older. PET of amyloid and tau brain with mild cognitive impairment is recommended.



A typical result in the Alzheimer brain research: Assessment of Muscarinic Receptor Concentrations in Aging and Alzheimer Disease With [11C] NMPB and PET [73].

EXPLORING SENSES (NOSE AND EYE) AS MARKERS FOR NEUROPATHY (Disease or dysfunction of one or more peripheral nerves, typically causing numbness or weakness.)

Abbreviations

EPC : Endothelial Progenitor Cells
DWI : diffusion-weighted imaging
FLAIR : fluid attenuated inversion recovery
ICAM : intercellular adhesion molecule
VCAM : vascular adhesion molecule
TNF : tumor necrosis factor
MMP : matrix metalloproteinase
TIMP : tissue inhibitor of matrix metalloproteinase
ET : endothelin
IL : interleukin
NIHSS : National Institutes of Health Stroke Scale
WBC : white blood cells
RBC : red blood cells
Hb : hemoglobin
Ht : hematocrit
INR : International Normalized Ratio
SBP : systolic blood pressure
DBP : diastolic blood pressure
ACE : angiotensin convertase enzyme
ARB : angiotensin receptor blockers
rtPA : recombinant tissue plasminogen activator
ETOH : chronic alcohol consumption
GST : glutathione S-transferase
MCB :monochlorobimane
GSH CDNB, 1-chloro-2,4-dinitrobenzene
MCI : Mild Cognitive Impairment

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