Review article

Nutrition and Alzheimer's disease: The detrimental role of a high carbohydrate diet

Stephanie Seneff [a], Glyn Wainwright [b], Luca Mascitelli [c],*

[a] Department of Electrical Engineering and Computer Science, MIT Cambridge, MA, USA
[b] Independent Reader of Research, Leeds, United Kingdom
[c] Comando Brigata alpina "Julia", Medical Service, 8 Via S. Agostino, Udine 33100, Italy

ABSTRACT

Alzheimer's disease is a devastating disease whose recent increase in incidence rates has broad implications for rising health care costs. Huge amounts of research money are currently being invested in seeking the underlying cause, with corresponding progress in understanding the disease progression. In this paper, we highlight how an excess of dietary carbohydrates, particularly fructose, alongside a relative deficiency in dietary fats and cholesterol, may lead to the development of Alzheimer's disease. A first step in the pathophysiology of the disease is represented by advanced glycation end-products in crucial plasma proteins concerned with fat, cholesterol, and oxygen transport. This leads to cholesterol deficiency in neurons, which significantly impairs their ability to function. Over time, a cascade response leads to impaired glutamate signaling, increased oxidative damage, mitochondrial and lysosomal dysfunction, increased risk to microbial infection, and, ultimately, apoptosis. Other neurodegenerative diseases share many properties with Alzheimer's disease, and may also be due in large part to this same underlying cause.

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1. Introduction and background

It has been well established that the brain of patients with Alzheimer's disease (AD) is characterized by the build-up of a signature plaque containing an abundance of the protein amyloid-β (Aβ) [1]. As a consequence, hundreds of millions of research dollars are currently being invested by the pharmaceutical industry towards finding and testing drugs that interfere with Aβ synthesis. The assumption is that, by reducing the supply of Aβ, the plaque build-up would be attenuated, and this might avert or delay the disease process.

Eli Lilly recently set up two long-term Phase III trials of the promising new drug, Semagacestat, which had been shown to interfere with Aβ synthesis. However, these trials were abruptly halted in August, 2010, due to clear evidence that they were causing an accelerated deterioration in cognition in the treatment group compared to placebo-based controls [2]. It is possible that the reason for failure may be the late intervention, due to the fact that biomarkers proposed for an early diagnosis [3,4] are not currently used in clinical practice. It may also be the case that the drug is not sufficiently specific to Aβ, and that other drugs with a more specific target may fare better. However, it is conceivable (as we will argue) that Aβ plays a protective role before it succumbs to crystallization and precipitation into plaque.

In the brain, the primary cells in the innate immune response are microglia which, like other tissue macrophages, participate in repair and resolution processes after infection or injury to restore normal tissue homeostasis. These cells are often found near damaged neurons and Aβ plaque deposits. It is unclear whether their role is detrimental or positive, as they appear to eliminate Aβ aggregates via phagocytosis, but they may also contribute to cell death of nearby neurons by releasing neurotoxic cytokines and proteases. Some research efforts have been directed towards the idea of finding ways to inhibit microglial-induced neuron death while avoiding interfering with their role in Aβ clearance [5]. In experiments with cultured microglia, the stress hormone, norepinephrine, has shown some promise in this regard [6,7]. However, this kind of treatment would likely lead to undesirable side effects.

1.1. Early Alzheimer's disease

Some researchers have directed their energies towards understanding very early stage AD, reasoning that any defects appearing early are likely to be more relevant to the true underlying cause. These efforts have borne fruit in at least two related directions.

- Several researchers have noted a strong correlation between insulin resistance in the brain and early AD, suggesting that AD might be considered a neuroendocrine disorder of the brain or so-called "type 3 diabetes" [8,9].
- Others have noted an association of AD with mitochondrial dysfunction [10]. A genetic defect in mitochondrial Complex I genes is associated with a small minority (2%) of AD cases.

It is noteworthy that mitochondrial defects, particularly in Complex I, have been found to be present in all major neurodegenerative diseases,

* Correspondong author: Tel.: +39 0432584044; fax: +39 0432584053.
 E-mail address: lumasci@libero.it (L. Mascitelli).

not just AD, but also Parkinson's disease [11], and amyotrophic lateral sclerosis (ALS) [12,13]. Mitochondrial dysfunction leads to two damaging conditions: insufficient ATP to fuel the cell's energy needs and oxidative damage due to excessive reactive oxygen species (ROS).

1.2. The relationship between cholesterol, ApoE, and AD

The brain represents only 2% of the body's total mass, but contains 25% of the total cholesterol [14]. Cholesterol is required everywhere in the brain as an antioxidant, an electrical insulator (in order to prevent ion leakage), as a structural scaffold for the neural network, and a functional component of all membranes. Cholesterol is also utilized in the wrapping and synaptic delivery of the neurotransmitters. It also plays an important role in the formation and functioning of synapses in the brain [15] (Fig. 1).

The most well known genetic variant leading to an increased risk of AD is Apolipoprotein E-4 (ApoE-4) [16]. Thus, it seems logical that ApoE's functional role would provide useful information towards unraveling a cause for AD. The apolipoproteins play an essential role in the delivery of fat, cholesterol, and antioxidants from the liver to all the cells of the body, via specific receptor-based mechanisms in plasma membranes. Only two apolipoproteins are known to exist in the cerebrospinal fluid: ApoE and Apo-A1 [17,18]. Astrocytes are specialized glial cells which are abundant in the brain, where their essential role centers on the care and feeding of neurons. ApoE is synthesized in large amounts by astrocytes, which allows them to extract the contents of intermediate-density lipoprotein (IDL) and low-density lipoprotein (LDL) particles arriving from the bloodstream.

It had long been thought that the blood brain barrier (BBB) prevented the transport of lipids from the bloodstream into neurons in the brain [19]. However, it has now been shown conclusively that mechanisms exist to promote transport of LDL intact across the BBB via an LDL-receptor mediated mechanism in astrocytes [20]. Furthermore, astrocytes can act to increase the rate of such LDL transcytosis when they are lipid poor. Hence, astrocytes are capable of obtaining cholesterol, fats, and antioxidants directly from LDL in the bloodstream. It has been shown that the ApoE-4 allele is associated with reduced cholesterol uptake by astrocytes in the hippocampus [21]. Defective LDL uptake, as seen in the case of the apoE-4 allele, would logically lead to a depletion in the availability of these nutrients in astrocytes, and hence in their availability to neurons (Fig. 2).

People with the ApoE-4 allele tend to have high serum LDL. In 1998, a team of researchers [22] addressed the question of whether this high cholesterol level might be an attempt on the part of the body to adjust for a poor rate of cholesterol uptake in the brain. They studied 444 men between 70 and 89 years old at the time, for whom there existed extensive records of cholesterol levels dating back to several decades ago. Most significantly, cholesterol levels fell for the men who developed AD prior to their showing AD symptoms. The authors suggested that their high cholesterol might have been a protective mechanism against AD. Indeed, high cholesterol level is positively correlated with longevity in people over 85 years old [23], and in some cases has been shown to be associated with better memory function [24] and reduced dementia [25].

Mounting evidence suggests that a defect in cholesterol metabolism in the brain may play an important role in AD. Significantly, the cerebrospinal fluid of AD patients is substantially depleted in lipoproteins, cholesterol, triglycerides, and free fatty acids, compared to matched controls [26]. Along with depletion of cholesterol and triglycerides, the fatty acid content of cerebrospinal fluid is reduced by an alarming factor of 6.

One of the important roles of cholesterol is to cause the lipoproteins in cell membranes to pack into a tighter molecular configuration [27], such that the fatty acids are protected from exposure to both oxidative damage and invasive pathogens. The neurons in the AD brain are chronically exposed to excessive amounts of glutamate as well as hydrogen peroxide and hydroxyl radicals due to mitochondrial defects. With insufficient fatty acid supplies to repair the damage to the cell wall, they would also become susceptible to penetration by pathogenic microbes [28].

Recent population studies have confirmed a correlation between low blood serum cholesterol and both dementia and Parkinson's disease. A study published in 2007 compared three elderly population groups: subjects with dementia, subjects with depression, and controls [29]. They found that those with dementia and depression had significantly lower serum cholesterol levels than the controls. Another study looking at Parkinson's disease among the elderly showed that those with the lowest LDL had 3.5 times the risk of Parkinson's disease compared to those with the highest serum LDL levels [30].
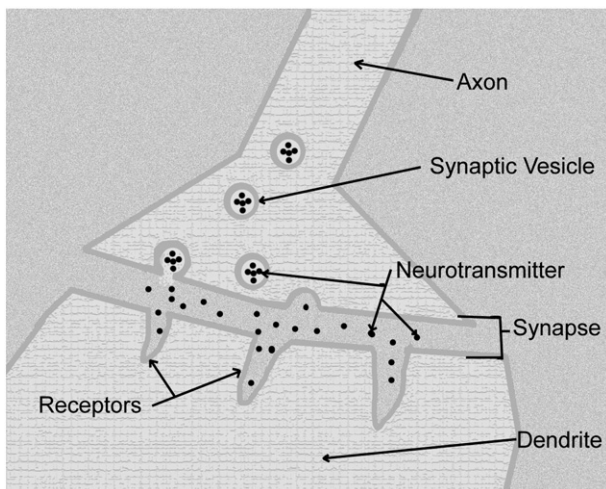

Fig. 1. The neurotransmitter synapse. [The synaptic vesicles are containers of neurotransmitter in a membrane of lipids and cholesterol. Both the vesicle and neuron membranes are a mixture of lipids and cholesterol supporting the synaptic mechanisms. On receiving the trigger signal from the axon, the vesicle membranes fuse with synapse membrane, releasing the neurotransmitter into the synapse where it migrates to the dendrite receptors. This exocytotic release of neurotransmitter depends upon the presence of cholesterol-rich domains in the vesicle and synaptic membranes].
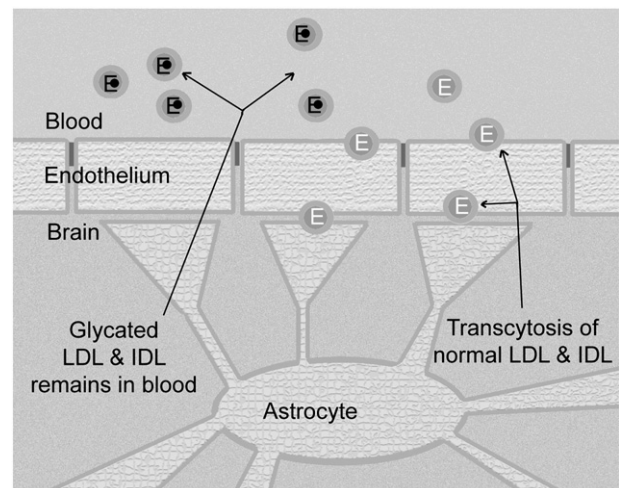

Fig. 2. An astrocyte receiving LDL by transcytosis across the blood brain barrier. [Normal ApoE labelled LDL (E) will be recognized by receptors and transported intact across the endothelium of the blood brain barrier. If the ApoE label is damaged by glycation (E·), it will remain in the bloodstream until it is scavenged by adipocytes].

2. Alzheimer's disease: the effect of a long-standing exposure to glucose and oxidizing agents

Cell membranes need constant maintenance to repair and replace their fats and cholesterol subsequent to oxidative damage. The strong influence of Ancel Keys [31], beginning in the 1960s, has led to dietary avoidance of fats and cholesterol along with over-zealous prescription of cholesterol-reducing medications over the same decades in which there has been a parallel rise in AD prevalence. Although this epidemiological coincidence is not proof, it gives weight to underlying research showing a possible link between cholesterol depletion and neuronal failure [32].

The process likely begins with the development of defects in the supply of cholesterol and fats to neurons, progressing to extensive oxidative damage, mitochondrial defects, and a shift of metabolism towards utilizing sources other than glucose and minimizing mitochondrial involvement. In the end stage, neurons can no longer transmit signals effectively, and apoptosis is the best option to avoid damaging neighboring cells.

A crucial role is played by astrocytes, the glial cells that supply cholesterol and fats to neurons. A consequence of excess exposure to glucose and oxidizing agents may lead to an initial damage due to glycation and oxidation of ApoE in astrocytes. The subsequent cascade begins with defects in the transmission of neural signals, progressing to mitochondrial dysfunction, insulin resistance, and the increased synthesis of Aβ. The dual roles of Aβ are to stand in for cholesterol and to redirect energy management from the mitochondria towards alternative cytoplasmic solutions, in order to reduce potential oxidative and glycation damage to active proteins and membranes. As such problems escalate, a neuron is eventually unable to function in its intended role (neurotransmission) without exposing itself and neighboring neurons and glial cells to dangerously high levels of oxidative agents. The microglia will then program the defective neuron for cell death. By this time, the proteasome and the lysosome systems have usually been so irreversibly damaged by oxidative exposure and insufficient ATP generation that the cell undergoes apoptosis. A legacy of complex protein debris is left in place.

2.1. Advanced glycation end-products impede supply of cholesterol and fats to neurons

A diet high in processed carbohydrates and low in fats results in a rapid rise in blood glucose levels following meals. Over time, this may lead to insulin resistance and diabetes [33]. Serum proteins that are exposed to high levels of glucose become impaired due to a process called glycation, resulting in the appearance of a diverse group of modified proteins known collectively as advanced glycation end-products (AGEs). Fructose, a sugar increasingly in use in processed foods, also because of economical constraints, is estimated to be ten times more reactive than glucose in inducing glycation [34]. With the widespread availability of high fructose corn syrup as a sweetening agent, the Western diet is associated with much higher risk of AGE damage to proteins. Hemoglobin A1c, the protein whose serum level is the standard blood test for diabetes, is a prototypical AGE product.

Fructose is far more damaging than glucose as a reducing agent, leading to diverse AGE products. For example, an experiment involving feeding rats controlled diets containing either fructose, glucose, or sucrose demonstrated that the fructose-fed rats were worse off on many indicators of glycation damage [34]. Serum levels of fructose, fructosamine and glycated hemoglobin were all significantly higher for the fructose-fed rats. Increased damage to collagen was also evident. We hypothesize that AGE damage to LDL leads to defects in receptor-based uptake of LDL by astrocytes in the brain, causing them to become cholesterol and fat depleted, and therefore stressed.

A set of proteins that are critically affected by glycation are the apolipoproteins. ApoB, the main apolipoprotein in LDL, is rich in lysine, an amino acid that is especially susceptible to glycation [35]. Furthermore, glycation, but especially fructation, of lysine is enhanced in diabetes [36], and small LDL particles are more susceptible than large ones. Glycation disturbs the uptake of apoE-containing lipoproteins by interfering with the heparan sulfate proteoglycan pathway [37]. Most significant for our hypothesis is the interference with the supply to astrocytes, which then places a burden on them to synthesize cholesterol in order to keep up with the needs of the neurons they supply. The synthesis of cholesterol is a difficult process involving twenty-five to thirty steps, and thus imposes increased energy demands on the astrocytes. This then leads to an increase in exposure to damage due to both glucose and oxygen, which, over time, will impair their ability to produce an adequate supply for the neurons they maintain.

It has been shown that patients with type-2 diabetes are at two to five times increased risk to AD [38,39]. It has been proposed that neuron dysfunction in diabetes might be due to insufficient cholesterol availability [40]. Through mouse studies, it was found that serum insulin levels influence the ability of both neurons and astrocytes to synthesize cholesterol, with reduced insulin (due to diabetes) leading to reduced cholesterol synthesis. It was therefore proposed that this reduction in cholesterol bioavailability would lead to reduced neurotransmission at the synapse. Diabetics with the ApoE-4 allele are at greater risk than predicted, meaning that these two factors interact [41]. ApoE-4 is defective in taking up cholesterol from blood serum, and glycation can only make this situation worse.

Glycation is now viewed as being an important contributor to AD risk [42]. The brains of AD patients contain high levels of AGEs, and these contribute to amyloid plaque deposition [43]. ApoE is highly susceptible to glycation, and it has been shown that excess glycated ApoE is present in the cerebrospinal fluid in Alzheimer's patients [44]. ApoE-4 shows a three-fold greater AGE-binding activity than ApoE-3 [45]. This suggests that glycation of ApoE may be an early step in Alzheimer's cascade, and that the increased affinity of ApoE-4 may be a significant contributor to increased risk to AD.

With increased exposure to glucose, multiple proteins in both astrocytes and neurons are susceptible to glycation damage. A glycated protein suffers from a loss of function, increased susceptibility to oxidative damage, and increased resistance to degradation and disposal [46]. It has been shown that the Aβ plaque characteristic of AD is consistently associated with AGEs, and that these damaged proteins can be detected even in neuronal microscopic slices where Aβ is absent, suggesting that glycation is an early hallmark of AD [47].

2.2. The decline of myelin and neuronal membrane integrity

Oligodendrocytes surrounding axons produce a continuous stream of myelin to wrap and protect the axons from impulse leakage, overheating and oxidative damage [27]. The cholesterol in the myelin is highly susceptible to oxidation damage. Oxidized cholesterol is water soluble and escapes into the surrounding fluid, requiring its replacement with new cholesterol from the oligodendrocyte. The oligodendrocyte needs a constant supply of cholesterol from the ApoE fraction in the cerebrospinal fluid. As defects in ApoE due to glycation and oxidation damage interfere with the supply chain, axon damage may follow, and this will lead to impaired signal transmission. Several studies have indicated that cholesterol-deficient brain cells are less capable of withstanding neurological insults [48].

The depletion of cholesterol leads to loss of both myelin and membrane functions [49]. The cell membranes in the brain will also become deficient in essential unsaturated fats, such as omega-3 fats derived in the diet especially from fish oils, with further functional failure of important metabolic processes. Oxidative damage to the fatty acids in the membrane cannot be easily repaired due to an

insufficient supply of replenishments mediated via ApoE. The loss of integrity in the cell membrane caused by cholesterol depletion [50] leads to loss of the membrane potential, a run-away energy burn to rebuild the loss [27], and the production of Aβ proteins to plug the damage [51].

3. Impaired glutamate homeostasis

Impaired glutamate homeostasis is often associated with neuro-degenerative diseases [52,53]. Glutamate is arguably the most important neurotransmitter. It is secreted into the synapse to enable signal transport from one neuron to another. Neurotransmitter release at the synapse requires membrane fusion. Cholesterol has been shown to increase lipid mixing in the synapse by a factor of 5, at physiological concentrations [54]. Membrane-bound cholesterol is present in high concentration in the synapse, where it plays an important role in fusion. Cholesterol depletion of synaptic vesicles leads to a loss of membrane ion gradient and a subsequent leakage of glutamate out of the vesicle [55]. In vitro experiments involving chemical depletion of cholesterol in the rat synaptosome have demonstrated an excess of glutamate accumulating in the synapse as a consequence of cholesterol depletion, likely due to a decreased ability of synaptic vesicles to uptake and retain the transmitter [56].

Astrocytes are responsible for reuptake of excess glutamate from the synapse, and it has been demonstrated that cholesterol deficiency leads to increased synthesis and reduced degradation of glutamate [57]. Oxidatively damaged Aβ has also been shown to suppress reuptake of glutamate from the synapse by astrocytes [58]. This is a reasonable reaction, as ion leakage in synaptic vesicles and inefficiencies in membrane fusion would reduce transmission rates, necessitating higher concentrations of glutamate to compensate. Insufficient cholesterol and fatty acid repair would also impair myelin sheath, leading to signal leakage during transport through axons. This would further enhance the need for higher glutamate concentrations in the synapse to compensate for anticipated signal leakage in transport.

Glutamate is a powerful oxidative agent, and has been shown to induce oxidative stress in neurons [59]. So the increased glutamate required for proper function under cholesterol-starved conditions necessitates increased risk to oxidative damage, while at the same time insufficient cholesterol makes the cell membranes more susceptible to oxidative damage.

This places a great deal of stress on the cell, as the process of transmitting signals through defective media exposes it to toxic elements.

4. Emergency responses of neurons towards alternative fuels

4.1. ApoE and Aβ

In this section, we develop several arguments for how Aβ establishes a reduction in dependence on mitochondrial activities, with the goal, in our view, of reducing the risk of oxidative damage to the cell. Researchers have recently found that ApoE in the astrocytes controls the levels of Aβ in neurons: whether it induces an increase or a decrease in Aβ concentrations depends upon the status of lipids in the cell [60–62]. ApoE enhances Aβ clearance when it is lipidated; i.e., when sufficient fatty acids are present. When it is unattached, ApoE increases the concentration of Aβ. ApoE also plays a crucial role in bringing fats and cholesterol into the astrocyte from the bloodstream and in delivering it to neurons either directly or via the cerebrospinal fluid. It is likely that ApoE serves as a sensor of cholesterol levels, and would be able to draw Aβ into play when cholesterol is deficient.

The mechanism by which snails manage a reduced energy state during estivation [63] is highly relevant to our discussion. Snails enter estivation (similar to hibernation) during harsh environmental conditions. In a study of mitochondrial function during estivation, it was determined that the snails are able to simultaneously reduce their consumption of oxygen and decrease the ion gradient across the mitochondria, such that the same efficiency (ratio of ATP produced over oxygen consumed) is maintained as in normal conditions, but with an overall reduction in the total amount of nutrients consumed and the total amount of ATP produced. We argue that Aβ induces a similar strategy in neurons.

4.2. Aβ redirects cell metabolism away from aerobic oxidation of glucose

Aβ has been implicated, particularly in its oligomer form, as a causative agent in so-called type 3 diabetes [64]: the development of insulin resistance in neurons. Our view is that pathologies in the Alzheimer's brain necessitate a redirection of metabolism towards exploiting substrates other than glucose. One powerful effect of insulin is to stimulate hydrogen peroxide generation in the mitochondria, in anticipation of launching the citric acid cycle for oxidative metabolism [65,66]. However, hydrogen peroxide is a potent oxidizing agent, which will further damage susceptible membranes. Hence, by suppressing insulin receptors, the cell would reduce the risk of oxidative damage.

A further reason for Aβ oligomers to suppress insulin is to allow Insulin Degrading Enzyme (IDE, also known as insulysin) to redirect itself towards degrading damaged Aβ, a skill it also possesses [67]. In parallel, Aβ reduces the supply of oxygen by causing blood vessels to constrict [68]. Finally, it has been shown that Aβ interferes with the glucose transporter protein, GLUT3, which would result in a reduced ability to import glucose [69]. With these reductions in place (of oxygen, of glucose, and of insulin), a reduced ion gradient will still be able to maintain efficient energy production, while ameliorating the problem of hydrogen ion leaks across the membrane; i.e., a strategy similar to that used by snails during estivation [63]. However, the amount of ATP produced will be correspondingly reduced.

At the same time, Aβ initiates measures to increase the production of ATP in the cytoplasm from alternative food sources besides glucose. An enzyme which binds to Aβ, and is found in increased amounts in Alzheimer's patients, is ABAD (amyloid-β-peptide-binding alcohol dehydrogenase) [70]. This is a very versatile peptide which can metabolize a broad array of substrates, including alcohols and ketone bodies. Curiously, while it is present in both the endoplasmic reticulum (cytoplasm) and the mitochondria, it appears to be far more effective in the cytoplasm than in the mitochondria [71], for reasons that remain elusive, but may be related to Aβ-binding (Fig. 3).

In a review paper on glucose metabolism and AD, Schubert [8] argues that Aβ may serve a neuroprotective role against oxidative stress by redirecting glucose metabolism away from oxidative phosphorylation in the mitochondria and towards the so-called pentose shunt. This pathway, which takes place anaerobically in the cytoplasm, generates additional reducing power in the form of NADPH, which serves to protect the cell from oxidation damage due to excessive ROS. In in vitro experiments exposing cortical neurons to Aβ, it was shown that the neurons increased their production of a rate-limiting enzyme in the pentose-shunt pathway by more than a factor of 4.

4.3. Ketone bodies and ketogenic diet

A ketogenic diet has been found to be therapeutic in AD patients [72,73]. It involves an extremely high fat diet, with up to 88% of calories derived from fats. This benefit may be likely due in part to the bioavailability of a plentiful supply of fats to repair damaged membranes. However, this diet leads to the generation of a significant concentration of ketone bodies in the blood serum, which can be used as an alternative fuel to glucose (metabolized by ABAD). In a mouse model, it was found that ketosis leads to a greater production of
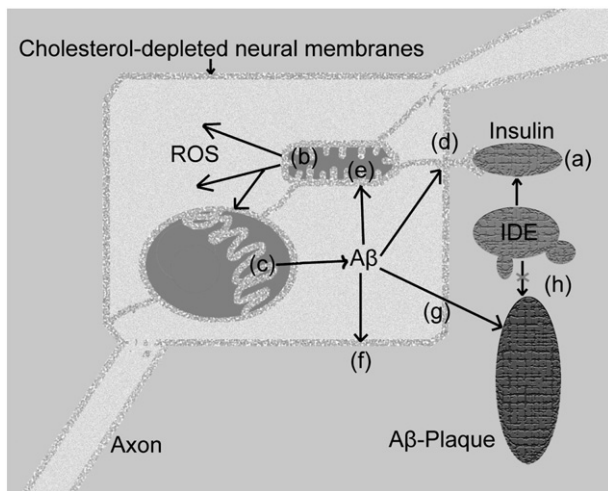
Fig. 3. A cholesterol-depleted neuron producing Aβ. [(a) An activated insulin receptor stimulates aerobic oxidation of glucose; (b) reactive oxygen species (ROS) released by excessive overrun of aerobic oxidation due to ion leakages in membranes; (c) ROS stimulate protective Aβ production in ER and Golgi; (d) Aβ protectively suppresses the insulin receptor stimulus; (e) Aβ interferes with the neuron's glucose transport (GLUT3); (f) Aβ substitutes for depleted cholesterol assisting membrane integrity; (g) Aβ plaques accumulate with ROS induced neuron death (apoptosis); and (h) raised insulin levels (type II diabetes) reduces IDE availability for normal Aβ-plaque removal].

acetyl-CoA in the forebrain, which would allow neurons to consume relatively less glucose, while achieving an increased production of glutamine and GABA, important nutrients for neurotransmission [74].

4.4. Lactate

Aβ also catalyzes the production of lactate dehydrogenase [75]. The first step in glucose metabolism is to convert glucose to pyruvate, which releases a small amount of ATP. Ordinarily, pyruvate would be further broken down in the mitochondria, while consuming oxygen. However, lactate dehydrogenase allows pyruvate to be converted into lactate through anaerobic fermentation. Lactate can then be further processed by neurons to yield additional ATP.

An interesting theory regarding lactate proposes that astrocytes shuttle lactate to neurons during periods of intense activity, to supply an alternative fuel source to glucose [8]. Indeed, lactate is interchangeable with glucose to support oxidative metabolism in cortical neurons [76]. Under this hypothesis, astrocytes convert pyruvate to lactate (utilizing lactate dehydrogenase) and shuttle the lactate to neurons, which could generate ATP from it, thus further reducing their dependency on glucose metabolism in the mitochondria. This would result in reduced exposure to glucose in the neuron, thus decreasing susceptibility to glycation damage. Lactate supply directly from the blood serum is not an option for neurons due to the BBB.

5. Infection and compromised immune response

Pathogens are likely to flourish in the bloodstream if excess glucose and AGE debris are abundant. An increased prevalence of bacterial infection is associated with diabetes, and this is likely due to the ability of AGEs to suppress superoxide production by immune system cells [77]. Indeed, there is evidence that AD may also be related to an increased likelihood of infective agents appearing in the brain [78]. A recent study showed that AD patients had a significantly higher concentration of an antibody against *Helicobacter pylori*, a bacterium that is common in the digestive system, in both their cerebrospinal fluid and their blood, compared to non-AD controls [79]. *Helicobacter pylori* was detected in 88% of the AD patients but only 47% of the controls. In an effort to treat the AD

patients, the researchers administered a potent combination of antibiotics, and assessed the degree of mental decline over the next 2 years [80]. For 85% of the patients, the infection was successfully routed, and for those patients, cognitive improvement was also detected after 2 years had elapsed.

Chlamydia pneumoniae is a very common bacterium, estimated to infect 40–70% of adults. But there is a big distinction between a bacterium being in the bloodstream and making its way into the inner sanctum of the brain. Another study tested for *Chlamydia pneumoniae* in post-mortem samples from various regions of the brains of AD patients and non-AD controls. They found that 17 out of 19 AD brains tested positive for the bacterium, whereas only 1 out of 19 brains from the control group tested positive [81].

Many other infective agents, both viruses and bacteria, have been found to be associated with AD, including herpes simplex virus, picornavirus, Borna disease virus, and spirochete [28]. One proposal was that a particular bacteriophage, a virus that infects the bacterium *Chlamydia pneumoniae*, might be responsible for AD [82]. The authors argued that the phages might make their way into the mitochondria of the host cell and subsequently initiate AD.

Interestingly, recent research has demonstrated that Aβ has antimicrobial activity [83]. *In vitro* experiments demonstrated that Aβ can defend against eight common and clinically relevant microorganisms.

6. Late-stage Alzheimer's disease: neuronal apoptosis

As more and more damage is incurred by the cell membranes, without sufficient replenishment of the supplies of fats and cholesterol to repair them, an increase in ion leakage across all membranes leads to further depletion of ATP and further exposure to pathogens and oxidative damage. Over time, the accumulated depletion of ATP leads to lysosomal dysfunction, likely because of an inability to maintain a sufficiently acidic pH for the digestive enzymes to work properly. In parallel, the further glycation and oxidation of Aβ convert it into an oligomeric complex with both decreased function and reduced susceptibility to degradation by the lysosomes. Once a sufficient percentage of the cell membrane has been compromised due to oxidative damage, it incurs rapid calcium influx and subsequent apoptosis. The protein plaques and tangles of AD are unrecyclable debris that remains in place after cell death.

7. Discussion

AD is a devastating disease, in terms of both mental anguish and health care costs. Epidemiological indicators suggest that the incidence of AD in the U.S. and likely the Western world is currently increasing at an alarming rate [84,85], and disproportionately with the increase in the aged population. In the last decade, significant research money has been invested in trying to understand the underlying cause(s) of AD.

It has long been recognized that the accumulation of Aβ plaque in the extracellular matrix is a signature of late-stage AD [86]. Much research has been directed towards finding therapies that interfere with the synthesis of Aβ, but thus far with disappointing results. A growing minority of researchers are proposing that Aβ plaque is an effect rather than a cause: that the true dysfunction is an inability to degrade proteins; i.e., defects in the proteasome and/or the lysosome [87]. While we agree with this analysis of the end stage of AD, we propose that Aβ, rather than being a principal causative agent, may play a role in defenses against AD. In our view, lysosomal dysfunction is a late-stage effect occurring long after mitochondrial dysfunction has led to extensive oxidative damage to important proteins such as Aβ.

In this paper, we have highlighted that AD may also be caused by a deficiency in the supply chain of cholesterol, fats, and antioxidants to the brain. We have provided much evidence of the importance of these nutrients to brain function, and have shown that AD patients are deficient in cholesterol and fats in the cerebral spinal fluid. We

suggest that a diet high in high-glycemic index carbohydrates, particularly fructose, and relatively low in fats and cholesterol, leads to a destructive process that begins with glycation damage to blood serum lipid supply as well as to proteins important to the metabolism of astrocytes. The impairment of astrocytes is accelerated by the excess energy requirements associated with increased cholesterol synthesis in the absence of a sufficient supply from the bloodstream. Neurons begin to suffer damage as well, once the astrocytes can no longer supply adequate amounts of cholesterol, fats and antioxidants.

Aβ synthesis may represent a protective mechanism that redirects metabolism away from glucose and away from the mitochondria, in order to decrease the rate of further decline due to glycation and oxidative damage. Increased production of glutamate is necessary to maintain signal transport in the face of cholesterol deficiency, yet glutamate causes further oxidative damage. Eventually, further maintenance of the cell is counterproductive, and it undergoes apoptosis. Once a sufficient number of cells are destroyed in this way, major cognitive decline is manifested.

Learning points

- The amyloid-β present in Alzheimer's plaque may not be causal, since drug-induced suppression of its synthesis led to further cognitive decline in the controlled studies performed so far.
- Researchers have identified mitochondrial dysfunction and brain insulin resistance as early indicators of Alzheimer's disease.
- ApoE-4 is a risk factor for Alzheimer's disease, and ApoE is involved in the transport of cholesterol and fats, which are essential for signal transduction and protection from oxidative damage.
- The cerebrospinal fluid of Alzheimer's brains is deficient in fats and cholesterol.
- Advanced glycation end-products (AGEs) are present in significant amounts in Alzheimer's brains.
- Fructose, an increasingly pervasive sweetening agent, is ten times as reactive as glucose in inducing AGEs.
- Astrocytes play an important role in providing fat and cholesterol to neurons.
- Glycation damage interferes with the LDL-mediated delivery of fats and cholesterol to astrocytes, and therefore, indirectly, to neurons.
- ApoE induces synthesis of Aβ when lipid supply is deficient.
- Aβ redirects neuron metabolism towards other substrates besides glucose, by interfering with glucose and oxygen supply and increasing bioavailability of lactate and ketone bodies.
- Synthesis of the neurotransmitter, glutamate, is increased when cholesterol is deficient, and glutamate is a potent oxidizing agent.
- Over time, neurons become severely damaged due to chronic exposure to glucose and oxidizing agents, and are programmed for apoptosis due to highly impaired function.
- Once sufficiently many neurons are destroyed, cognitive decline is manifested.
- Simple dietary modification, towards fewer highly-processed carbohydrates and relatively more fats and cholesterol, is likely a protective measure against Alzheimer's disease.

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