



MINOCYCLINE, AN ANTIBIOTIC AND A NEUROPROTECTIVE: JUSTIFYING ROLE IN ALZHEIMER'S DISEASE

VIVEK KUMAR SHARMA¹, ASHOK GOYAL², SUBRAHMANYA SARMA GANTI³

¹Govt. College of Pharmacy, Rohru – 171207, Distt. Shimla, Himachal Pradesh, India. E mail: viveksharma_pharma@yahoo.co.in

²Onkar College of Pharmacy, Sajuma, Sunam -148026, Distt. Sangrur, Punjab, India.

³ISF College of Pharmacy, Moga -142001, Punjab, India.

ABSTRACT

Alzheimer's disease is the most common yet partly understood cognitive syndrome that affects mainly the geriatric population. Despite commendable progress in understanding pathogenesis of Alzheimer's disease yet the drugs available provide symptomatic relief only and do not stop progression of the disease. This has forced and motivated the researchers to keep exploring the newer molecules and out of which Minocycline has grabbed maximum attention from scientific community as it appears astounding and fascinating at the same time that one drug alone can attenuate the severity of diseases like stroke, cerebral ischemia, multiple sclerosis, spinal-cord injury, Parkinson's disease, Traumatic brain injury, Huntington's disease, Amyotrophic lateral sclerosis and Alzheimer's disease. The status of Minocycline has changed from a simple antibiotic to a versatile neuroprotective molecule. The role of Minocycline in alleviation of several neurological disorders is increasingly being recognized and explored. Despite slow but steady progress in understanding the mechanism of action of Minocycline in several neurological disorders the status and role of Minocycline is still debated. In this review, we sum up the evidences for the Minocycline in Alzheimer's disease and discuss the mechanisms by which minocycline affects a range of pathological features in AD.

Key words: Alzheimer's disease, Minocycline, CNS.

INTRODUCTION

During most of the 20th century, neurodegenerative diseases remained among the most enigmatic disorders of medicine¹. Alzheimer's disease (AD) is the most common neurodegenerative disease in developed countries². AD is an progressive³, multifactorial⁴, etiologically heterogeneous form of brain failure⁵ with common clinical and pathological features including inexorable deterioration of memory and intellect⁶ followed by aphasia, agnosia, apraxia and behavioral changes⁷.

Neuropathological hallmarks include, neuropil threads, Hirano's bodies, granulovacuolar bodies and cerebral amyloid angiopathy⁸ neocortical neurofibrillary tangles (NFTs)⁷, accumulation of senile lipofuscin pigment (SP)⁹ caused by the extra cellular deposition of Amyloid β Protein ($A\beta$) in neuritic plaques¹⁰ neuronal loss¹¹ and psychotic disturbances¹².

All these changes are attributed to vulnerability of particular memory-focused synapses to degeneration¹³. The cognitive decline in AD is accompanied by neuronal atrophy and loss, mainly in the cortex, hippocampus and amygdala¹⁴ with progression of disease due to severe cortical dysfunction, patient becomes demented, aphasic, disorientated, immobile and emaciated. Pneumonia or urinary infections are the common causes of death⁸.

Amyloid plaques and Neurofibrillary tangles (NFTs) are most important neuropathological hallmark of Alzheimer's disease¹⁵. NFTs are intraneuronally generated aggregates of paired helical filaments (PHFs), which are assembled from hyperphosphorylated forms of the microtubule-associated protein tau¹⁶. Abnormal Hyperphosphorylation of tau may impair its microtubule-binding ability and thus lead to their aggregation into paired helical filaments (PHFs)¹⁷.

$A\beta$ peptides are proteolytic cleavage products of the $A\beta$ precursor protein (APP) sequentially processed by α and γ secretases¹⁸. Deposition of $A\beta$ in AD brain occurs in either diffuse or fibrillar forms. In addition to parenchymal plaques, fibrillar $A\beta$ deposition in the cerebral vasculature, a condition known as cerebral amyloid angiopathy (CAA), is commonly found in AD¹⁹.

Molecular mechanism of $A\beta$ -mediated cell death probably involves oxidative stress, free radicals²⁰, neuroinflammation and apoptosis of Neurons⁹, a situation to which oligodendrocytes (OLGs) are particularly susceptible because their glutathione (GSH) content is low and they have a high concentration of iron, thus presenting an

impaired ability to scavenge oxygen radicals²¹. Through the interaction with caspase-3 and caspase-8, $A\beta$ can induce the apoptosis of neurons and be cytotoxic⁹. Furthermore, $A\beta$ possesses an increased capability for damaging cholesterol rich membranes, such as those found in OLGs and myelin²².

TETRACYCLINES - INVESTIGATING ROLE IN ALZHEIMER'S DISEASE

The tetracyclines, which were discovered in the 1940s, are a family of antibiotics that inhibit protein synthesis by preventing the attachment of aminoacyl-tRNA to the ribosomal acceptor (A) site. Tetracyclines are broad-spectrum agents, exhibiting activity against a wide range of gram-positive and gram-negative bacteria, atypical organisms such as chlamydiae, mycoplasmas, rickettsiae, and protozoan parasites. The favorable antimicrobial properties of these agents and the absence of major adverse side effects have led to their extensive use in the therapy of human and animal infections. Chlortetracycline and oxytetracycline, both discovered in the late 1940s, were the first members of the tetracycline group to be described. These molecules were products of *Streptomyces aureofaciens* and *S. rimosus*, respectively²³.

Minocycline is a member of the tetracycline family of broad-spectrum antibiotics²⁴. It has superior tissue penetration into the brain and cerebrospinal fluid²⁵. During recent years, Minocycline have been shown to exert biological effects that are completely separate and distinct from their antimicrobial action. These effects include: inhibition of matrix metalloproteases²⁶, tumor-induced angiogenesis²⁷, malignant cell growth and depression of oxygen radical release from polymorphonuclear neutrophils. Experimental and clinical studies indicate that minocycline may be beneficial in treatment of other disorders too including neurological disorders.

MINOCYCLINE: EVIDENCES IN ALZHEIMER'S DISEASE

Minocycline is a semi synthetic tetracycline that has been in use for over 30 years to treat pneumonia and acne vulgaris, infections of the skin, genital, and urinary systems²⁸ and rheumatoid arthritis²⁹. Minocycline is a small (495 kDa), highly lipophilic molecule capable of crossing the blood-brain barrier. Minocycline penetrates the CSF of human beings better than doxycycline and other tetracyclines. Minocycline is readily absorbed from the gut after oral ingestion and, because of its low propensity to produce antibiotic resistance; it is commonly used in the management of chronic conditions such as acne and rosacea³⁰. In addition to its antibiotic properties,

minocycline has been reported to have neuroprotective effects in various experimental models of cerebral ischemia³¹, traumatic brain injury³², amyotrophic lateral sclerosis (ALS)³³, Parkinson's diseases (PD)³⁴, kainic acid treatment³⁵, Huntington's disease (HD)³⁶, and multiple sclerosis³⁷. Additionally, Minocycline was reported to attenuate white matter damage in a rat model of chronic cerebral hypoperfusion³⁸. The capacity of minocycline to alleviate disease for several neurological disorders in animals is increasingly being recognized. Indeed, that one drug alone can attenuate the severity of variety of disorders is astounding. In addition to the neurological disorders described above role of Minocycline in Alzheimer's treatment is being looked at.

OXIDATIVE STRESS, EXCITOTOXICITY AND MINOCYCLINE

Oxidative stress appears to be the most important factor for cell death in neurodegeneration and is one of several processes that contribute to AD pathophysiology³⁹. It induces several other key events like protein aggregation, mitochondrial dysfunction and glutamate excitotoxicity and all of them combinedly contribute to the death of neurons⁴⁰. A β not only induces oxidative stress and neuronal death but enhances glutamate-induced excitotoxicity. Excessive amount of glutamate would cause NMDA receptor mediated Ca²⁺ overload, disrupt homeostasis, and induce neuronal death⁴¹. Minocycline has been shown to protect against excitotoxicity caused by N-methyl-d-aspartate (NMDA) in vitro⁴².

NEUROINFLAMMATION, AD AND MINOCYCLINE

Inflammation plays an integral role in Alzheimer's disease development and may precede plaque and tangle formation⁴³. Inflammatory components related to AD neuroinflammation include brain cells such as microglia and astrocytes⁴⁴, the classic and alternate pathways of the complement system, the pentraxin acute-phase proteins, neuronal-type nicotinic acetylcholine receptors (AChRs), peroxisomal proliferator-activated receptors (PPARs), as well as cytokines and chemokines⁴⁵.

Inhibition of chronic neuroinflammation, particularly of microglial activation, has been suggested to be a practical strategy in the treatment of neurodegenerative diseases including Alzheimer's disease⁴⁶.

Activated microglia and reactive astrocytes, often associated with extracellular A β deposits, contribute not only to the production of cytokines, chemokines, reactive oxygen species, and neurotoxic substances, but also to neurotrophic factors⁴⁷. The relationship between this cell activation and the cognitive dysfunction in A β depositing diseases is not well understood, partially because of the complex responses generated by activated inflammatory cells. Minocycline, a tetracycline derivative with anti-inflammatory properties that crosses the blood-brain barrier, has been shown to suppress the activation of cultured human and mouse microglia stimulated with A β ⁴⁸.

In various experimental models of AD, Minocycline suppressed microglial production of Interleukins (IL), IL-1beta, IL-6, TNF, and NGF in vitro as well as APP transgenic mice^{46, 48}. In addition, minocycline attenuated cholinergic cell loss, glial activation, and transcription of downstream pro-inflammatory mediators and mitigated the cognitive impairment, induced by mu p75-saporin, a novel immunotoxin that mimics the selective loss of basal forebrain cholinergic neurons and induces cognitive impairment in mice. These reports showed that minocycline exerts neuroprotective effects based on its anti-inflammatory actions in AD experimental animal models⁴⁹.

APOPTOSIS, AD AND MINOCYCLINE

Apoptosis and the release of apoptotic factors is a common mechanism of neurodegeneration in Alzheimer's disease. Minocycline reduces apoptosis of neurons and oligodendrocytes in various neural insults⁵⁰, and alleviates necrotic cell death⁵¹. Minocycline stabilizes mitochondrial membranes and inhibits the mitochondrial permeability transition-mediated release of cytochrome c into the cytosol⁵², which is a potent stimulus for the activation of caspases 9 and 3 and the induction of apoptosis. The

stabilisation of mitochondrial membranes also reduces the release into the cytoplasm of other factors that trigger both caspase-dependent and caspase-independent apoptotic pathways, including apoptosis inducing factor and Smac/Diablo. Minocycline also upregulates the anti-apoptotic factor Bcl-2, which then accumulates in mitochondria to antagonise the pro-apoptotic Bcl-2 family members.⁵²

PARP ACTIVATION IN AD AND MINOCYCLINE

Increased Oxidative stress in Alzheimer's disease lead to fragmentation of nuclear DNA, in a much higher proportion of neurons, oligodendrocytes, astrocytes and microglia^{53,54} which then activates the poly (ADP-ribose) polymerase (PARP) a nuclear enzyme which physiologically participates in DNA repair. When DNA damage is mild, PARP is thought to be involved in the maintenance of chromatin integrity. This DNA repair enzyme is over expressed in Alzheimer's disease⁵⁴. Over activation of PARP after cellular insults can lead to cell death caused by depletion of the enzyme's substrate b-nicotinamide adenine dinucleotide and of ATP⁵⁵, leading to resultant decrease in the rate of glycolysis, electron transport and ATP formation.

Extensive PARP-1 activation can, in addition, lead to neuronal death through mechanisms linked to NAD depletion and release of apoptosis inducing factor from the mitochondria. PARP-1 activation is a key mediator of neuronal death during excitotoxicity, ischemia, and oxidative stress. PARP-1 is emerging as an important activator of caspase-independent cell death. PARP-1 generates the majority of long, branched poly (ADP-ribose) (PAR) polymers following DNA damage. Overactivation of PARP-1 initiates a nuclear signal that propagates to mitochondria and triggers the release of AIF. AIF then shuttles from mitochondria to the nucleus and induces peripheral chromatin condensation, large-scale fragmentation of DNA and, ultimately, cytotoxicity⁵⁶.

The activities of the pro-apoptotic proteins caspase 3 and poly (ADP-ribose) polymerase (PARP) are increased in brain cells during normal ageing too⁵⁷. The occurrence of rapid mitochondrial depolarization by NO in hippocampal neurons and energy depletion soon follows, and the facile conclusion is that decreased production of ATP is entirely responsible. However, NO also may increase ATP hydrolysis by the cell, particularly by activation of PARP⁵⁸. Minocycline has a direct inhibitory effect on PARP-1 at submicromolar concentrations and has been found to be neuroprotective. Minocycline protect neurons against PARP-1-mediated toxicity at submicromolar concentrations and was found to be a highly potent inhibitor⁵⁹.

To date, the primary CNS mechanism implicated in minocycline neuroprotection is the drug's highly potent inhibitory effect on microglial activation, which is achieved by blocking the phosphorylation of p38 and the translocation of 5-Lipoxygenase into the nucleus, thereby preventing the release of cytokines and the induction of inflammation⁶⁰⁻⁶².

In addition current evidences show that Minocycline ameliorates stroke-induced behavioral deficits, prevents the ischemic cell death, via an anti-apoptotic mechanism⁶³. The safety and therapeutic efficacy of minocycline and its robust neuroprotective effects during acute ischemic stroke make it an appealing drug candidate for stroke therapy⁶³.

CONCLUSION

In this review the evidences proves Minocycline to be a good target in finding a cure for Alzheimer's disease treatment as it appears to interact with multiple factors including oxidative stress, neuroinflammation, PARP, Apoptosis, Amyloid β , Excitotoxicity and Caspases. The present review advocates thorough, planned and extensive efforts to verify all the claims.

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