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Methamphetamine: A Potential Risk Factor for Neurodegenerative Conditions

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by

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Abstract

Methamphetamine (METH) is a highly addictive substance that persists to be a major drug of abuse within the United States, particularly the Midwest regions. While some of the major health ramifications are well documented, its neurological impact is not fully understood. Neurodegenerative conditions like multiple sclerosis (MS), Parkinson's disease (PD), and amyotrophic lateral sclerosis (ALS) continue to increase in frequency, particularly in the Midwest, yet there is no identifiable cause. The purpose of the study was to determine if METH use can contribute to the development of neurological conditions such as MS, PD, and ALS.

A literature review was conducted in order to find high quality systemic reviews, randomized controlled trials, and meta-analysis evaluating METH and its impact on structural and chemical components of the nervous system and how this may impact the future development of a neurodegenerative condition. Multiple high quality articles were found using PubMed, EMBASE, Clinical Key, and DynaMed.

After reviewing the current literature, it appears the METH use has several major neurotoxic effects resulting in physical and cognitive deficits. There is weak evidence supporting METH use as a risk factor for ALS. Additionally, METH use does not appear to be a risk factor for developing MS but can be a mimicker. It is however a risk factor for developing PD.

Keywords: *methamphetamine, neurodegeneration, neurotoxicity, MRI, multiple sclerosis, Parkinson's disease, amyotrophic lateral sclerosis, attenuation, structural changes.*

Introduction

Methamphetamine (METH) is a highly addictive illicit substance that persists to be a major drug of abuse within the United Sates, particularly the Midwest regions. The purity and potency of METH continues to improve while the cost steadily declines (NDTA, 2018) increasing its availability. However, with increased availability the health problems associated with METH use have become a great concern. Many health ramifications of using methamphetamine are well documented including their effects on psychological wellbeing, dental deterioration, and cardiovascular damage. However, the full scope of health ramifications is still not completely understood. Recent research suggests that METH abuse may lead to neurodegeneration and may be a risk factor for conditions such as multiple sclerosis, Parkinson's disease, and amyotrophic lateral sclerosis. The purpose of this study is to evaluate the effects of METH use regarding neurochemistry and structural changes which may lead to the development or progression of neurodegenerative diseases.

Statement of the Problem

Methamphetamine continues to be a relevant drug problem within communities. Providers are faced with managing these individuals and the wide scope of health problems associated with METH use. While some aspects like behavioral and dental health are commonly addressed, neurodegenerative changes related to METH use are not widely recognized by providers. Therefore, medical providers should be informed on the potential ramifications of METH and its neurodegenerative consequences. This will allow for early recognition and better patient education.

Research Question

Are those who use methamphetamine at an increased risk for developing multiple sclerosis vs those who do not use methamphetamine.

Are those who use methamphetamine, at an increased risk for developing Parkinson's disease vs those who do not use methamphetamine.

Are those who use methamphetamine, at an increased risk for developing amyotrophic lateral sclerosis vs those who do not use methamphetamine.

Literature Review

A comprehensive search was performed using the electronic databases PubMed, EMBASE, Clinical Key, and DynaMed. Specific keywords and MESH terms searched included: *methamphetamine, neurodegeneration, neurotoxicity, MRI, multiple sclerosis, Parkinson's disease, amyotrophic lateral sclerosis, attenuation, structural changes.* Sources were further narrowed with the search parameter full text and consist of randomized control trials, systematic reviews, and meta-analyses. Some articles were excluded based on the abstract and lack of relevancy to this literature review.

Drawbacks to most studies include limited sample sizes, varying exclusionary criteria, and inability to perform controlled studies on human subjects. Due to ethical constraints, most human studies are retrospective in nature which creates potential inconsistencies. In animal studies there were limited samples sizes and in several occurrences a failure to mention number of subjects.

Structural changes in the brain related to methamphetamine use

Brain structures. The brain is primarily composed of grey and white matter. Grey matter contains neuronal cell bodies and allows for all movement, speech, decision making, and other functions to be carried out. White matter within the brain is made up of axons coated with myelin and forms pathways between regions of the brain allowing for adequate communication. As individuals age, the amount of grey and white matter naturally decreases (Salo & Fassbender, 2011; Thompson et al. 2004). Pathology and neuronal insult have been shown to alter the rates and volumes of grey and white matter. A common way to evaluate changes to brain tissue is using MRI imaging. These studies make it possible to measure volume changes in regional areas of the brain and to look for abnormalities.

One abnormality of interest includes white matter hyperintensities (WMH). These lesions appear within the brain as areas of increased brightness when visualized with T2-weight MRI. These lesions are related to vascular damage and changes and have been noted in conditions such as hypertension, diabetes mellitus, cocaine use and opiate abuse. Bae et al. (2006) hypothesized that METH dependent individuals would have increased WMH when compared to similar healthy individuals. They also hypothesized that male METH abusers would have more pronounced effects versus female METH abusers due to the protective effects of estrogen. Since WMH are not unique to METH user, subjects were selected using strict criteria based on METH use/dependence, medical history (excluding individuals with comorbid conditions or history of psychiatric disorder), and exposure to additional abuse related drugs. Chosen individuals were then paired with a healthy comparison subject and subjected to a brain MRI. WMH depth and severity were rated using the Fazekas classification and their location noted.

There were no reported increases in WMH occurrences between female METH abuser and comparison subjects ($p = 0.883$). Male METH abusers had greater prevalence and severity of all WMH versus their healthy comparison subjects ($p < 0.001$). Increased METH abuse also correlated with increased severity of WMH ($p = 0.027$). WMH were most commonly reported in the frontal lobe. These results suggest that females possess a protective factor, possibly estrogen, against METH's damaging effects to brain tissue.

It should be noted that individuals who reported smoking tobacco were not excluded from this study. Nicotine dependence has been shown to cause WMH in past studies and should be taken into consideration. Overall the METH group did have a higher percentage of current tobacco smokers versus the controls. This study had a limited sample size of 33 METH abuses and 32 healthy comparison subjects. Since there were significant differences in WMH between male and females, it may be advantageous to repeat these tests on specific gender groups to further investigate the properties of estrogen and structural brain changes.

Thompson et al. (2004) used MRI to evaluate the relationship between methamphetamine use and structural changes to grey and white matter of the brain, specifically cortical surface and key subcortical structures. Subjects with a history of METH abuse were selected using a strict criteria and age matched to healthy controls. Subjects were also asked to perform a memory task to attempt to identify if structural changes correlated with cognitive impairment.

Significant cortical grey matter deficits were observed in METH abusers, particularly in the areas surrounding the corpus callosum ($p < 0.05$). These changes correlated with glucose metabolism changes found in previous studies. Hippocampal volumes were significantly smaller ($p < 0.01$) with correlating memory deficits ($p < 0.05$) in METH abusers. There was an increased volume of white matter found in METH abusers ($p < 0.01$). This effect could be secondary to

decreased grey matter or potentially could be linked to gliosis, neurodegeneration, or unknown psychiatric disorders.

This article does make several important findings about METH use and changes in brain structure. It additionally shows that memory was impaired and correlated with areas of change. Subject groups were small totaling 22 METH users and 21 comparison individuals. Subjects were predominantly male and were not excluded if they used nicotine products or marijuana. Authors did recognize that marijuana use may impact their results and ran all analyses with and without the individuals reporting its use. Ultimately it was found that it did not affect the results in a statistically significant way. For more accurate results, larger sample sizes, nicotine free individuals, and gender specific trials should be considered.

Given that the neurotoxic properties of methamphetamine can cause structural brain changes and cognitive behavioral modifications, Sabrini et al. (2019) performed a systemic review composed of 29 articles. Prior to this review, most studies focused on a singular type of imaging and had a limited amount of cognitive aspects. Authors of this article collectively evaluated outcomes and correlated measured brain changes and cognitive performance in METH abusers. Structural changes reviewed included regional volume, blood flow, glucose metabolism, neuronal integrity, and activation. Seven specific cognitive domains were evaluated: psychomotor function, working memory, attention cognitive flexibility, inhibitory control, cognitive impulsivity, and risky decision making.

Results showed that in all cognitive domains, METH abusers performed more poorly than the controls. These cognitive changes also positively correlated with brain measure changes including changes to regional density, blood flow, glucose metabolism, fractional anisotropy value, N-acetylasparate level, and activation. A strong associated was found between cognitive

control and changes to the prefrontal cortex and anterior cingulate cortex. These changes tended to improve with protracted abstinence from METH. This potentially indicates that the brain may be able to recover to some degree. A positive correlation was also made between decision making and changes to the prefrontal cortex, anterior cingulate cortex and striatum. These changes may be related to decreased dopamine and changes to the reward system in the brain.

Authors did an excellent job in making correlations between behavior, cognition, function, and structural brain structure changes. This is the first systemic review on the topic of METH use and changes to structure and function of the brain. This study was limited on the total number of articles used. There was also a total of six different types of imaging modalities used between various studies. All articles had small sample sizes making for limited data. In several instances there was an overlap of subjects used and overlap of authors making for a potential bias. There was also variation in subject screening criteria, many of which did not exclude nicotine use. Inconsistencies in documentation of amount of METH use, length of abstinence (if any), or cumulative years of use was also present. Lastly, not all studies were performed against a control group of non-METH users.

Methamphetamine use has been associated with higher risk-taking behavior and alterations in decision making secondary to changes in the reward pathway. This pathway is essential for reinforcing behaviors needed for survival. Key brain structures involved with the reward pathway including the stratum, cerebral cortex (insula), thalamus, and the basal ganglia. An increase in the major transmitter, dopamine, reinforces behaviors by interpreting the action as pleasurable in the brain. Methamphetamine use significantly increases the amount of dopamine released causing a downregulation of total dopamine receptors in the brain. May et al. (2013) hypothesized that individuals whom were methamphetamine dependent would show alterations

in brain activation and structure, specifically in areas involved with the reward pathway and interoception (how one interprets an external feeling). Subjects consisted of 25 recently abstinent METH abusers and 17 comparison controls. Individual's was subjected to functional magnetic resonance imaging and asked to complete a simple task while experiencing a soft touch stimulus to the forearm or hand. Images were obtained and compared between the two groups. Individuals were also asked to rate the sense of pleasantness regarding soft touch.

METH individuals showed lower activation in bilateral anterior insula, bilateral dorsal striatum, left middle frontal gyrus, and bilateral thalamus to soft touch verses the controls. There was a positive correlation between lower anterior insula activation had faster reactions times ($p = 0.02$). Additionally, those with lower striatal activation had increased pleasure ratings to soft touch stimuli ($p = 0.05$.) These results indicate that those using METH do have alterations to the reward pathways in the brain and is supported by alterations in neural responses and the interoceptive stimuli.

Subjects were intensively screened and excluded for potential complicating factors including pre-existing psychiatric conditions, comorbid health problems, and additional illicit drug use. This resulted in a small sample size which was predominantly male. However, one factor that was not excluded was the use of nicotine. The METH group had a significantly higher proportion of nicotine users which could impact results. The total number of tactile stimulations was limited to forty repetitions in a duration of 840 seconds narrowing the power for statistical analyses.

Nerve structures. Methamphetamine use has neurotoxic properties potentially causing damage to the myelin sheath of nerves resulting in reduced nerve impulses. Possible mechanisms of neurotoxicity include oxidative stress, neuroinflammation, dopamine imbalances, and mitochondrial dysfunction. Manifestations of this may present as muscle weakness, loss of coordination, or changes in sensation. However, the extent to which METH use has on motor aspects of the brain is still poorly understood. Flavel et al. (2012) investigated the relationship between stimulant use (predominantly METH) and its effects on motor cortical a corticospinal excitability.

Subjects were divided into three groups: stimulant users, cannabis users, and healthy controls. Individuals then underwent transcranial magnetic stimulation (TMS). A target location on the hand was utilized. An electromyography response and motor evoked potentials (MEP) were then recorded. There was a significantly larger MEP recorded for stimulant users for both relaxation ($p = 0.045$) and muscle contraction ($p < 0.001$). Prolonged MEP latency and increased muscle activity during activity was also apparent in stimulant users. There was no significant difference in resting motor threshold between stimulant and controls ($p < 0.009$). Interestingly, the average duration of abstinence from stimulants was over two years. This indicates that changes from stimulants are most likely not acute effects but instead long-term consequences. This article supported the authors hypothesis that stimulant users do exhibit detrimental changes to the motor cortical and corticospinal excitability.

The stimulant group was predominantly METH users but did also include several individuals with history of ecstasy use. It was also found that most stimulant users tested positive for cannabis. Authors attempted to correct for this by using a cannabis only control group and excluded individuals who used cannabis within twelve hours of testing. Results were also limited

by using only one muscle. More muscle should be tested to ensure consistency across muscle groups. Again, the total number of subjects was also limited (52 total, 26 stimulant users, 9 cannabis users, and 17 healthy controls). To strengthen this study, length of abstinence should be further investigated.

It is being established that chronic METH use can lead to motor disorders such as choreoathetoid movements and punding (prolonged purposeless movements). Most articles have focused on brain regions related to the reward pathway. Other regions, including those related to movement, have been far less studied. Huang et al. (2017) aimed to establish if METH use in humans could alter plasticity of MEP and inhibit physiological motor learning.

A total of 56 male METH and 35 healthy comparisons were used for this study. To reduce potential acute effects of METH use, all subjects had been abstinent for at least two weeks. Subjects then underwent TMS and MEP procedures. Additionally, individuals were asked to learn and repeat a simple task. Results were significant for diminished motor cortical plasticity ($p < 0.05$) and impaired leaning abilities in METH users ($p < 0.05$). This indicates that METH use is detrimental to motor and learning abilities.

This article had a substantial group of METH users but failed to screen for depression, other potential psychological conditions, use of prescription medications acting on the central nervous system, or use of other illicit drugs in the past. There were repeated trails of TMS, MEP, and motor learning which were separated by adequate time, reducing the likelihood of erroneous values. However, they only measured the results of TMS and MEP in one isolated muscle vs multiple locations.

Changes in locomotor activities secondary to METH use stem from decreased dopaminergic terminals, decreased dopamine transporters, reduced dopamine production in the striatum, and reduced tyrosine hydroxylase (TH). While many of these changes remain into prolonged abstinence, there is some degree of measurable recovery. The mechanism behind the repair pathways are poorly understood. One of the proposed theories is regeneration of striatal fibers. The most frequently seen striatal fibers are type I. These are characterized by their very fine mesh appearance. Additional striatal fibers include types II-IV, all varying by thickness and generalized shapes. Granado et al. (2018) evaluated the pathway for partial recovery of TH and neurons in the striatum by evaluating they type of striatal fibers. Additionally, they attempted to answer if striatal fiber changes correlate with locomotor changes.

Mice were given METH in either a single high dose (30mg/kg) or three lower doses of 5 or 10mg/kg in 3-hour intervals. Mice were then terminated on day 1 or 3 of abstinence and tissues collect/examined. All results were compared to control mice who received saline injections. Results showed that all mice receiving METH had significantly decrease in striatum fibers one day post-METH ($p < 0.001$). By day three, while all METH subgroups had some recovery in striatal fibers numbers, however they remained fewer than that of controls. Striatal fiber changes were seen most predominantly as a depletion of type I fibers of METH mice. Reductions in type I fibers were most prominent on day 1. On day three, new axon terminals with compensatory sprouting of embryonic axon buds were noted. New growth of fibers were confirmed with the use of CAP-43, a marker of new dopaminergic axon terminals. Locomotor skill were the most reduced on day 1 of METH mice; horizontal movement $p < 0.001$, time/distance $p < 0.05$. By day three, locomotor had recovered to some degree; horizontal movement $p < 0.001$, time/distance $p > 0.05$. These studies show that with METH use, striatal

fibers were acutely impacted one day post METH but by day three axonal sprouting was extensive. New growth was confirmed with markers. Locomotor improvements also correlated with new axonal growth. While Granado et al. (2018) was able to isolate distinct changes in striatum fibers, the results were poorly reported with minimal p values. They also fail to disclose how many mice were evaluated in total. Group numbers at one-point state n= 4-6 mice per group. While this evidence supports repair of neurotoxic effects from METH use, it has not yet been demonstrated on humans.

Chemical changes in the brain related to methamphetamine use

Dopamine. Many studies involving methamphetamine have involved the analysis of dopamine. Dopamine is an essential neurochemical component of the brain's reward system. If something positive or pleasurable occurs, an increase of dopamine is released reinforcing the behavior. It has been hypothesized that a dysregulation of dopamine may be an underlying factor in addictive behavior. It is not clear however, exactly how METH affects these pathways. Ashok et al. (2017) performed a systemic review and meta-analysis to comprehensively review methamphetamine's effect on the dopaminergic system to better comprehend data to consider potential treatments.

A total of 31 articles with comparable dopaminergic measures were selected. In all studies either cocaine or methamphetamine was the stimulant abused. Average abstinence from stimulants was five days to three weeks. There were two studies evaluating effects of METH and dopamine release. Both resulted in decreased dopamine release when compared to healthy controls; standard mean differences of -1.05 (95% CI, -1.76 to -0.34) and -0.40 (95% CI, -1.11 to 0.32). In nine studies, there was a significant reduction in dopamine transporter availability in those using METH; effect size -1.47 (95%CI, -1.83 to -1.10; $P < 0.001$). Seven studies evaluated

dopamine receptor availability and found an overall reduction in D2/D3 receptor availability in those using METH; effect size -0.81(95%CI, -1.12 to -0.49; P <0.001). Four studies evaluated vesicular monoamine transporter 2 (VMAT2) showing conflicting data. No studies were found evaluating the ability to synthesis dopamine in METH users. These results conclude that there is a generalized effect of dopaminergic down-regulation associated with METH use. This could imply that those using METH may need to increase the dose of METH to maintain increased levels of dopamine to have positive reward. Due to the artificially high levels of dopamine, there is a natural reduction in dopamine release.

This is the first article to compile information regarding METH use and associated changes to dopamine regulation. There is also excellent discussion as to potential mechanisms for dopamine changes and adequate comparisons of data available between METH, cocaine, and healthy subjects. There were several limitations. In all 31 studies, there were limited sample sizes. There is also some variation in subjects regarding inclusion/exclusion criteria including nicotine and alcohol. Another limiting factor is the lack of data and research on dopamine release. Future studies with larger sample sizes and by identification of differences in abstinence time vs dopamine alterations should be considered in the future.

In animal studies, METH use has been shown to be toxic to dopamine terminals. It has been established that dopamine levels are impacted by METH in humans also, however, the exact mechanism in which this occurs is still unclear. Additionally, it is unclear if changes in leads to cognitive changes. Volkow et al. (2001) evaluated if METH use alters dopamine transporters in the brain, specifically in the striatum and cerebellum. The second component of the study was to observe the relationship between brain changes and cognitive effects.

A total of 15 METH users and 18 matched healthy controls were used for this study. All individuals were administered a dopamine cell terminal marker (d-threo-methylphenidate) and subjected to a PET scan. Images were then compared. Additionally, all subject underwent a five-part neuropsychological evaluation measuring locomotor function and cognition. Results showed there was a significant decrease in dopamine transporter availability in the caudate and putamen of METH users versus the healthy controls; ($p < 0.0001$ for both). Individuals using METH over longer periods of time had decreased dopamine transporter levels in the caudate ($p < 0.05$). No correlation was seen between last METH use and dopamine transporter level ($p > 0.70$). All METH users had poorer neuropsychological evaluations when compared to controls; timed gait $p < 0.05$, grooved pegboard $p < 0.05$, word recall $p < 0.005$, delayed recall $p < 0.01$, immediate recall $p < 0.05$. This study demonstrates that individuals using METH had dopamine transporter reduction resulting in impaired locomotor tasks and memory. It also demonstrates that even people whom have been abstinent for at least 11 months had significant deficits, indicating effects could be for prolonged periods of time. These results were similar to those pertaining to dopamine transporter levels in subjects with Parkinson's disease.

While Volkow et al. (2001) isolated their proposed questions, the sample size was very limited and included mixed male and females study groups. This study would have potentially benefited by finding more subjects and running statistics on gender groups. Individuals using nicotine were also not excluded from this study potentially impacting the results. Future studies need to be performed to determine if dopamine transporter reduction is secondary to terminal damage, down regulation, or both.

It is proving to be evident that METH has a strong impact on overall dopamine levels and dopamine terminals. Another critical component involving dopamine pathways are the dopamine

transporters (DAT) which can be found in the caudate nucleus and the putamen. These areas of the brain are involved with motor skills. The rate limiting enzyme needed for the formation of dopamine is tyrosine hydroxylase (TH). Significant reductions of TH have been associated with movement disorders such as dystonia, Parkinson's disease, and Huntington's disease (Ares-Santos et al. 2013). If TH levels are preserved, another potential cause of decreased dopamine production is neurodegeneration of dopaminergic cell bodies in the substantia nigra causing decreased striatal dopamine levels.

Ares-Santos et al. (2013) evaluated whether METH causes neurodegeneration of dopaminergic cell bodies in the substantia nigra and if so, the mechanism of degeneration. Additionally, they tested varying regimens and dosages of METH and the impact on the striatum and substantia nigra with regards to time. In this study, mice were given a single high dose of METH or multiple lower doses in intervals. Mice were then terminated at varying intervals and tissue and brain studies were performed. Tyrosine hydroxylase marked with TH-immunoreactive (TH-ir) was evaluated first. Overall TH-ir was reduced in the striatum of METH verses the control ($P<0.001$). Mice receiving multiple lower doses had greater reductions than a single high dose ($P<0.001$). Over time it was noted that in METH mice TH-ir levels did increase but remained significantly lower that control mice ($p<0.001$). At the same time, silver staining in the striatum correlated with the changes of TH-ir indicating a strong terminal degeneration. In METH mice, significant loss of TH-ir cell bodies was noted in the substantia nigra (reduction of 22%, $P<0.001$). Peak degeneration occurred between 1-3 days. Thirty days post METH administration, there was no recovery of TH-ir neurons in the substantia nigra. Locomotor activity was severely impacted in METH mice when compared to controls ($P<0.001$). Effects predominantly occurred between day 1-3 post METH. After seven days, most mice recovered

normal locomotor activity. Overall this evidence show that METH can cause prolonged detrimental effects on dopaminergic cell bodies of the substantia nigra and can also destroy dopaminergic terminal in the striatum. All mice treated with METH had increases in body temperature versus controls ($p < 0.001$) indicating that neurotoxicity may also cause hyperthermia.

This study was very strong regarding depth of testing. They successfully were able to distinguish neurodegenerative effects to dopaminergic neurons in the substantia nigra and striatum of mice. There was correlating locomotor difficulties. This potentially strengthens the idea that METH use may increase the potential for developing Parkinson's disease. Mice were the subjects for this study; however it is never mentioned how many mice were actually included in this trial. To strengthen this study, the period of abstinence period should be extended beyond 30 days.

Microglia. Microglia cells are specialized macrophages that survive in the central nervous system (CNS). Their primary role is to scavenge and remove degenerating and damaged neurons. It has been long established that microglia cells become activated when there are neurotoxic injuries to the CNS and do not exacerbate neuronal injury. Increased microglia have been associated with non-neurodegenerating conditions via circulating blood containing inflammatory properties, lipopolysaccharides, and damage associated molecular proteins. This implies that if a substance degraded the blood brain barrier, there would be vascular leakage and microglia activation of the damaged vessels and their surround tissues. Bowyer et al. (2016) attempted to determine what the microglia response is to METH on brain vasculature (specifically the septum, hippocampus, and thalamus) and the surrounding tissues.

A total of ninety-five rats were used for this study. Rats were given saline or METH (varying amounts) every two hours for a total of four injections. Body temperatures were recorded hourly. All rats were terminated on either day 1 or 3 and brain tissue preserved and tested. Results showed that majority of METH rats experienced hyperthermia requiring cooling protocol to avoid death. Rats with increased doses of METH had higher temperatures. There was significantly elevated levels of Iba1 (marker used to identify microglia) in several brain regions which correlated with amount of time body temperature were >41.7 degrees Celsius; septum $r=0.781$, $p < 0.0008$; hippocampus r-0.850, $p < 0.00001$; thalamus $r=0.927$, $p < 0.00001$; parietal cortex $r=0.625$, $p < 0.014$. A positive correlation was also noted with increased body temperatures >41.7 degrees Celsius and FJc (marker used to identify neuronal, axonal, and nerve terminal degeneration) in the thalamus ($r=0.714$, $p = 0.0061$). This indicates there was neurodegeneration and a loss of cell bodies in the thalamus. Microglia activation was present in the vascular endothelium and in adjacent tissue in the septum, medial dorsal hippocampus, piriform cortex, and thalamus; all regions previously identified as having vascular leakage with METH use. There was no evidence of neuronal damage in the septum or hippocampus (negative FJc) even with activated microglia.

Bowyer et al. (2016) established that microglia do respond to vascular damage, even in the absence of neurodegeneration (as with the septum and hippocampus). Vascular damage was more likely to occur in these two areas with multiple METH administrations and with prolonged elevated temperatures. Drawbacks to this study include the fact that subjects are rats instead of humans. The ethical components of this study would not allow for human studies. Long term effects should be studied to further understand METH effects on microglia and vasculature.

Duration of microglial activation has not been deeply investigated. Most study subjects have relatively short abstinence periods leaving long term effects unknown. Additionally, it remains unclear if microglia cell activation is neuro-protective or if it contributes to neurodegeneration. Sekine et al. (2008) attempted to locate areas of microglia activation in subjects with protracted abstinence.

Twelve abstinent METH users (used for >6 years and abstinent for approximately 2 years) were selected and matched by age, gender, and education level to a control subject. All subjects underwent a positron emission tomography with a radiotracer for activated microglia. Five areas of the brain were then evaluated and compared. Microglia activation was significantly higher in METH users versus controls; midbrain $p = 0.008$, striatum $p = 0.004$, thalamus $p = 0.002$, orbitofrontal cortex $p = 0.006$, insular cortex $p = 0.006$. There were no correlations made between length of METH use and microglial activation. There was a significant negative correlation between length of abstinence and microglial activation in three brain areas; midbrain $p = 0.004$, striatum $p = 0.008$, thalamus $p = 0.001$. These results show METH users have significant microglial cell activation when compared to controls. Microglial cell activation also is present for at least two years into abstinence, although with time neurons may be able to recover.

This study did well at excluding subjects for psychiatric conditions, nicotine use, history of additional illicit drug use, and comorbid conditions. This is one of few studies that examines METH users with protracted abstinence. Limitations of the study include a small sample size and failure to state gender. Further studies are needed for determining if microglial activation is neuro-protective or contributory to neurodegeneration.

Reactive oxygen species. Methamphetamine use stimulates a significant release of dopamine in the reward pathway of the brain, resulting in a reinforcement of the behavior.

However, this type of dopamine release creates negative effect including the release of reactive oxygen species (ROS) via mitochondrial dysfunction. These ROS in turn contribute to neurodegeneration. Increased ROS have been noted in conditions such as Parkinson's disease, multiple sclerosis, amyotrophic lateral sclerosis, and Alzheimer's. The most common clinical feature seen with increased ROS is abnormal behavior and increase locomotor activity. Studies involving cocaine have found that ROS contribute to addictive behavior; this relationship has not been investigated with METH. It has recently been established that two inhibitors of ROS (a-phenyl-N-ter-butyl nitrone; PBN & 4-hydroxy-2,2,6,6-tetramthylpiperidine-1-oxyl; TEMPOL) may be able to prevent mitochondrial oxidative damage and addictive behaviors. Jang et al. (2016) evaluated the effects of ROS in METH induced locomotor difficulties and study its involvement with the reward pathway and addiction. Additionally, they tested the abilities of PBN and TEMPOL to reduce addictive behavior and improve locomotor abilities.

This study involved two parts, locomotion and addiction. For locomotion, rat movements were recorded to establish a base line. Rats were administered saline, PBN, or TEMPOL ten minutes prior to METH or saline. Movements were then recorded and compared. Results showed METH rats had increased locomotion versus saline rats ($p < 0.001$). METH rats received PBN and TEMPOL had reduced locomotor activity ($p < 0.01$; $p < 0.001$). Saline rats receiving PBN and TEMPOL had changes in locomotor activity.

Addiction properties were evaluated by training rats to self-administer METH via lever system; rats given choice to pull active lever which deliver METH or to pull inactive lever which delivered no METH. Total number of METH lever pulls were recorded. METH rats were then treated with saline, PBN, or TEMPOL and lever pulls recorded. Rats were sacrificed and brain material evaluated. Rats given PBN or TEMPOL had fewer infusions of METH when compared

to saline controls ($p < 0.001$; $p < 0.08$). Active lever pulls also decreased in a dose dependent manner (PBN $p < 0.001$; TEMPOL $p < 0.05$). There was no significant difference in inactive lever pulls in either PBN, TEMPOL, or saline groups. PBN and TEMPOL was also tested on METH free rats also lever trained except was given food instead of METH. Neither PBN, TEMPOL, or saline altered number of lever pulls. METH rat brain tissue showed increase antibody marker 8-OHG verses saline rats indicating oxidative stress ($p = 0.034$). Most oxidative stress was in the neurons of the nucleus accumbens (key component to reward system) sparing the astrocytes, microglia, and oligodendrocytes. Use of TEMPOL significantly reduced the release of dopamine into the nucleus accumbens ($p = 0.01$). These results ultimately determine that PBN and TEMPOL decrease locomotion and addictive properties of METH, TEMPOL decreases dopamine release, and that ROS does help mediate/enforce the effects of METH on the reward system.

This study was well structured. It could be strengthened by increasing the number of rats per experiment; sample sizes were frequently ten or less. Duration of action of PBN and TEMPOL was also not explored, it is unclear how long either can effectively reduce cravings.

N-acetyl-aspartate. N-Acetyl-aspartate (NAA) is a metabolite marker, that can be measured by proton magnetic resonance spectroscopy (MRS), to identify neuronal viability in the brain. My-inositol (MI) is another common brain metabolite associated with glial cell damage. Low NAA and high MI levels have been associated with other neurodegenerative conditions such as Alzheimer's disease, multiple sclerosis, schizophrenia, and traumatic brain injury. Past studies, regarding METH's neurodegenerative properties, have used these two metabolites to confirm neuronal damage. However, these studies focused on locating damage without respect to total dosage, gender, length of abstinence, or correlated clinical outcomes.

Sung et al. (2007) attempted to identify differences in NAA and MI levels of grey and white matter between METH abusers and healthy controls. Additionally, they hypothesized that total length of abstinence and cumulative dose of METH would correlate with amounts of NAA and MI found.

Thirty METH abusers were selected and grouped based on length of abstinence (greater or less than 6 months) and lifetime amount of METH used (greater or less than 100g). Healthy controls were then gender and aged matched (20 total controls used). All subjects underwent a proton MRS and levels of NAA, MI, and several other metabolites were measured/compared. Overall, there was only marginal differences in total concentrations of grey matter NAA between METH users and controls ($p = 0.053$). No significant difference was found in concentrations of gray matter NAA and length of abstinence in METH users ($p = 0.32$). However, within the METH subgroups, NAA concentration in the specific area of the midfrontal gray matter was correlated negatively to cumulative METH dose ($p = 0.05$) and positively to length of abstinence (0.03). Length of abstinence had no impact on total NAA concentrations in frontal white matter but, a significant difference was seen between cumulative METH dose and controls ($p = 0.004$); larger doses had lower NAA levels. This trend was most apparent when comparing male METH users to controls ($p = 0.009$). No statistics were compared to female groups due to insufficient sample size. MI was also found to be significantly increased in the frontal white matter of METH users versus controls ($p = 0.02$) but had no correlation between abstinence duration or total amount of METH used.

Sung et al. (2007) established a relationship between frontal white matter NAA concentration and total METH dose, but not abstinence length. No relationship was seen between midfrontal grey matter NAA concentration and total METH or abstinence length. This study was

limited by a small sample size and gender differences. Subjects were also not screened for HIV and not excluded if they had a history of nicotine use. Ultimately extensive research is still needed to determine if prolonged abstinence influences neurotoxicity from METH.

Relationship between methamphetamine and multiple sclerosis

Multiple sclerosis background. Multiple sclerosis is an idiopathic inflammatory disease of the CNS resulting in demyelination of the white matter of the brain and spinal cord. These changes manifest themselves as locomotor changes. The cause of MS is still unknown but is thought to be related to environmental exposures, sunlight, diet, and other modifiable risk-factors. In the last 40 years there has been a 27% increase in the incidence of MS in North America (Wallin et al. 2019). This equates out to 309 occurrences per 100,000 people. Most individuals are diagnosed in the second and third decade of life (DynaMed Plus, 2019). The McDonald Criteria is used to assist in make the diagnosis of MS but ultimately is a diagnosis of exclusion. This criteria states there must be evidence of CNS damage (WMH) in two or more parts of the brain, spinal cord, or optic nerves with evidence of dissemination in time. These incidents must also occur in the absence of fever, infection, and other potential neurologic mimickers including; amyotrophic lateral sclerosis, nutritional deficiencies, alcohol, and other illicit substances. A supportive test that can be used to diagnose, but is not required, is the presence of CSF oligoclonal bands. If there is an absence of lesions with respect to time via MRI, an absence of CSF oligoclonal bands, or potential mimicking comorbidity, then an alternative diagnosis must be considered.

Corticospinal excitability and evoked potentials. MRI has been one of the main tools used to diagnose and monitor disease the progression of MS yet, it is unable to accurately predict disease progression regarding locomotor changes. A potential adjunctive tool that could be used

in predicting disability outcomes is evaluation of peripheral and centrally evoked potentials. This evaluation would better assess corticospinal excitability from the central nervous system to the peripheral nervous system. Individuals with MS should have changes in several conduction pathways due to partial demyelination of the corticospinal tracts or by lesions in the motor cortices. In theory, changes to corticospinal excitability should correlate with level of disability. Neva et al. (2016) evaluated if these changes in corticospinal excitabilities correlated to clinical disability level in individuals with MS.

A total of twenty-six individuals with relapsing-remitting MS (RRMS) and eleven healthy controls underwent an electromyography (EMG) of the extensor carpi radialis muscles bilaterally. TMS elicited MEPS were then recorded and compared. Results showed there was significant differences between MS individuals and controls for MEP duration ($p < 0.001$), MEP latency ($p = 0.002$), resting motor threshold ($p < 0.022$), active motor threshold ($p = 0.012$) and cortical silent period onset ($p = 0.011$). Hemisphere differences were only noticed in MEP latency ($p = 0.002$). MS individuals had greater latency in the left hemisphere versus the right ($p = 0.015$). These results contributed to associating levels of disability with corticospinal excitability changes.

There were several limitations to this study including eight individuals whom could not participate in the study due to TMS stimulus output being greater than the maximum stimulator allowed output. These individuals were able to partake in neurophysiologic data collection however, it did reduce an already limited sample size for corticospinal excitability testing. Subjects were also mixed gender and did not account for nicotine use or medications potentially impacting muscle tone.

While MRI is the standard imaging modality for the diagnosis or MS, there are limitations. Individuals can have clinically silent T2-weighted brain lesions (50-70% of patients) and disease progression that presents without inflammatory infiltration, which is not visible with MRI. In these situations, a secondary modality is needed to evaluate and project disease progression. Giffroy et al. (2016) evaluated the effectiveness of using evoked motor potentials (EP) to predict projected disability status.

A retrospective study was performed on 100 MS patients. Individuals had to have confirmed MS using the McDonald criteria, have a base line Expanded Disability Status Scale (EDSS) with EP examination, and a follow up EDSS at least three years later with repeat EP examination. Evoked motor potentials were recoded from a total of four sides (both upper and lower limbs). Results were then scored based on severity and classified under a global EP score (overall worst score possible is 30). Average follow up time observed was 6.3 years. Results showed that both EDSS and global EP had significantly worsened with time ($p < 0.0001$). It was determined that global EP score was found to be a significant prognostic value ($p = 0.0012$). A global EP score of 17/30 proved to be an adequate cut-off point to identify patients at high risk for faster disease progression and worsening clinical outcome. This study confirms that the use of EP may be a useful diagnostic and disease monitor tool in addition to MRI. Additionally, it may allow for early intervention and aggressive treatment.

Limitations to this study include its retrospective design. This allows for the possibility for variable that are unknown. These results were also not controlled for individuals receiving any disease modifying therapies. Further studies would be needed to explore the relationship between MRI findings and EP results.

Relationship between methamphetamine and Parkinson's disease

Parkinson's disease background. Parkinson's disease (PD) is the second most common neurodegenerative condition affecting approximately one million people in the United States today. This number has more than doubled since 1990 making it one of the fastest growing problems in the medical community (Marras et al. 2018). Interestingly, Minnesota and North Dakota has the highest concentration of cases in the nation. This progressive neurodegenerative condition is characterized by bradykinesia, a resting tremor, rigidity, ataxia, and cognitive decline. Symptoms occur secondary to progressive death of dopamine producing cells in the substantia nigra (DynaMed Plus, 2019). What causes PD is largely unknown but is thought to be link to environmental factors, specifically exposure to pesticides, agricultural occupations, and consumption of well water. Diagnosis is based on symptoms and has no confirmatory tests while a patient is alive. Post-mortem a diagnosis can be confirmed via evaluation of the substantia nigra. Due to the negative effect that METH has on dopamine production and the substantia nigra, it is reasonable to question if its use could contribute to the development of PD. Additionally, this relationship should be further explored due to the frequency of anhydrous ammonia and other agricultural components used to produce METH.

Incidence of Parkinson's disease amongst methamphetamine users. Methamphetamine use contributes to destruction to dopamine terminals in the striatum. This results in cognitive and locomotor difficulties like that of Parkinson's disease (PD) albeit, at a lower magnitude. This poses the question of if METH users have an increased risk of parkinsonism or PD. Callaghan et al. (2010) hypothesized that individuals with diagnosed

METH use disorder would have a higher risk of subsequent admission diagnoses of PD versus a control group.

Inpatient medical records from 1990-2000 from a California hospital were assessed for individuals 50 years or older with qualifying ICD-9 codes and followed for up to ten years. Those with METH use disorder diagnosis were matched 5:1 with controls (appendicitis) and assessed for subsequent admission and diagnosis of PD. Individuals were excluded from the METH groups if there was ICD-9 codes for prior or current parkinsonism, additional illicit drug/alcohol use disorders (other than METH), or HIV. Control group had similar exclusion criteria for selection with the addition of no prior METH use disorder ICD-9 code. A total of 38,939 appendicitis and 1,863 METH diagnosis occurred. Of the appendicitis diagnosis, 9,315 individuals could be matched to METH individuals. To verify the control appendicitis group, incidence rates of PD were compared to that of a health maintenance organization in another part of the state. This showed a nonsignificant incidence rate between the groups ($p = 0.28$) verifying appendicitis as successful representation of the general population for a comparison group. The METH group showed a significant increase in subsequent admission diagnosis of PD versus the appendicitis control ($p = 0.019$). This suggests that METH users may have an increased risk of PD verses the general population not using METH.

There were several limitations to this study. The basis of these results is dependent on accurate ICD-9 code input. The validity of the codes used cannot be verified. The ICD-9 coding also does not distinguish between METH and other amphetamine use. Validity in the diagnosis of PD can also be difficult since it can only be confirmed by neuropathology. Additionally, only one ICD-9 code was used for PD potentially underestimating the occurrence of parkinsonism conditions. Lastly, the ICD-9 for nicotine use, which is more common in METH users and is

neuroprotective against PD, was not excluded potentially altering outcomes. Repeat studies with more regulated ICD-9 codes, more inclusive in age, and gender specific use would be recommended in the future to verify results.

A similar study was performed by Curtin et al. (2015) who aimed to verify a relationship between METH and incidence of PD. The study was a retrospective long term follow up of a statewide population based in Utah. Additionally, factors of gender, age, and nicotine status were added into the study. Individuals were selected from a statewide population pool which included inpatient records from 26 hospitals and 215 outpatient clinics. This ultimately accounted for 80% of all patient encounters in the state of Utah. Patient records from 1996-2011 with ICD-9 codes of interest were targeted. A total of three cohorts were compared; METH users 4,935, cocaine users 1,867, and controls matched 5:1 to substance users based on gender and age. Individuals were excluded if they had a pre-existing diagnosis of HIV, PD or parkinsonism essential tremors (PT), or illicit drug/alcohol use other than the mono-drug isolated for in the cohorts. Results showed a significant increase in PD/PT diagnosis in the METH group verses the control ($p < 0.0001$). Females METH users versus female controls had an increased occurrence when compared to male METH users verses male controls ($p < 0.0001$, $p < 0.001$ respectively). However, there was no significant difference between male and female METH users and risk of PD/PT. There was no statistical significance between cocaine users and controls for PD/PT occurrence. Increased PD/PT was found in METH users compared to cocaine users ($p = 0.01$) even when adjusted for nicotine use ($p =0.01$). This study ultimately supports that use of METH increases the risk of PD/PT when compared to cocaine users and healthy controls.

Even with a statewide population base, results were still limited due to overall subsequent diagnoses of PD/PT being uncommon (30 METH/49 matched controls, 4 cocaine/22 matched

controls). It is also difficult to confirm a diagnosis of PD in patients due to confirmatory testing constraints. Lastly, subjects were not excluded if concurrently using antipsychotic medications; these medications can potentially produce extra-pyramidal symptoms mimicking PD/PT. Future studies should be performed to determine if there is a gender factor in risk of PD/PT development in conjunction with METH use.

Methamphetamine vs dopamine vs Parkinson's disease. The hallmark characteristic of PD is substantially reduced levels of dopamine secondary to the loss of the entire dopamine containing neuron. Additional dopamine markers that are also significantly reduced in PD include dopamine transporters, dopamine metabolites, and vesicular monoamine transporter 2 (VMAT2). Changes can predominantly be seen in the putamen followed by the caudate. Kish et al. (2016) performed a systemic review to evaluate if METH use effected dopamine neurons in the brain and how that compared to known data of individuals with PD.

Dopamine levels in the postmortem brains of human users have only been measured in one study. This study did show that METH users had significantly lower levels of dopamine in the caudate and putamen versus controls ($p < 0.05$). However, there was a substantially greater loss of dopamine in the postmortem brains of individuals with PD. Loss was greatest in the putamen of PD individuals whereas METH users had greater loss in the caudate. Dopamine transporters in METH users were found to be reduced in eight different studies. VMAT2, a transporter for monoamine neurotransmitters from neuronal cytoplasm to storage vesicles, was evaluated in five separate studies. Collectively, there was little to no changes in VMAT2 levels in METH users. Three studies were found regarding gliosis, a marker of brain neurotoxicity and damage. Results were conflicting with no significant data available.

Overall this systemic review concluded that METH users had significant reductions in dopamine and dopamine transporters in the caudate and putamen when compared to controls. However, it was not as significant of a depletion as those with PD. There is still limited evidence if a reduction in dopamine is caused by damage or total loss of dopamine neurons. There were several limitations to this study, including a very limited number of available studies. Studies that have been performed had a small sample size with varying testing modalities and inclusionary criteria.

As humans age, it is natural to have a reduction in total number of dopamine neurons. One of the leading theories behind natural neuron death is oxidative stress. Oxidative stress could impact dopamine neurons in utero but may not become apparent until later in life. Average age for onset of PD is 50 years. Exposure to METH can induce oxidative stress potentially accelerating or adding to the cumulative effects. This in theory would mean METH use can aid in the development of PD. Morrow et al. (2011) evaluated the susceptibility of dopamine neurons by exposing in utero, young, and adult monkeys to METH. Monkeys were exposed to a total of four doses of METH over the course of two days. Subjects were then terminated, and brain tissue examined.

All subjects given METH had reduced TH-ir (marker of active dopamine neurons) when compared to controls ($p < 0.0001$). Dopamine loss was greater in the caudate nucleus and putamen versus the nucleus accumbens ($p = 0.006$). Interestingly, when evaluating the striatal sub-regions of young monkeys exposed to METH, there was no significant reduction in dopamine versus the adult METH group ($p < 0.0001$). Plasma levels of METH (three hours post METH administration) were the highest in young monkeys followed by in utero fetuses and adults (in utero vs young = $p < 0.0001$, young vs adult = $p = 0.0025$). Levels of GDNF (glial-

derived neurotrophic factor, a protein that promotes neuron survival) were similar in young and adult monkeys three hours after a single dose of METH. After seven days, there was a significantly increase in GDNF in young monkeys' verses adults ($p < 0.0001$).

These results show that METH does reduce dopamine neuron levels, however the total impact varied with age. Fetal and adult monkeys were more vulnerable to the effects of METH while young monkeys had compensation, even though METH doses were all the same. How dopamine neurons were protected in young monkeys is not understood but may have to deal with increased GDNF. This study provided previously unknown information about METH effects on dopamine with respect to age. Limitations to the study include using animal models. While results showed interesting data, there is no certainty that this translates into a human model. This study would be difficult to replicate with human subjects.

Methamphetamine vs oxidative stress vs Parkinson's disease. Dopamine neurons are under continual oxidative stress which contributing to their potential degeneration and death. This type of oxidative stress occurs naturally with the aging process, adding to why most cases of PD don't occur until the fifth decade of life. The oxidative stress secondary to METH use has been theorized to accelerate the development of PD yet, it remains unclear if METH use causes total dopamine neuronal death or just damage. By better understanding this process, it would allow for potential treatment and preventative therapies. However, studies historically have been limited due to a requirement of living human brain tissue. Lotharius et al. (2005) were able to overcome this by using human in vitro models of PD. They then attempted to answer if METH causes neurodegeneration or dopamine neuron death, like that in PD. Additionally, they evaluated several potentially protective substances against this type of damage.

Human in vitro cells, when exposed to METH for seven days, showed selective destruction of dopamine neurites but spared dopaminergic cell bodies. In order to obtain complete neuronal death, increased levels of iron had to be added. This was significant in that it shows METH alone does not cause complete neuronal cell death. With the additions of increased iron, a normal characteristic of PD, cell death was possible. When an iron chelating agent was added to samples, neuronal protection was observed ($p < 0.001$). A combination of METH/Fe to human in vitro cells showed evidence of neuronal cell death as soon as 12 hours post drug exposure ($p < 0.05$). After 60-72 hours, complete cell death was obtained within a sample ($p < 0.01$). Samples treated with antioxidants, CEP1347 (inhibitor of mixed-lineage kinases, a promoter of neuronal cell death), and iron chelating all successfully reduced neuronal damage ($p < 0.001$), indicating neuroprotective attributes and pharmacological potential.

Lotharius et al. (2005) concluded that MA could contribute but may not cause PD independently. In order to complete total dopamine neuron death, increased iron was required (another hallmark of PD). It also shows oxidative stress is key to neuronal deterioration. However, there are components potentially available to prevent this damage. Limitations to this study include being only able to use cell models. These results may be altered in a living model.

Alpha-synuclein. A hallmark of PD is the increased presence of misfolded alpha-synuclein (AS) proteins called Lewy bodies. These misfold proteins are markers for neurodegeneration and can only be identified in post-mortem examination. It has been found there is a direct correlation between levels of neuronal loss/motor dysfunction and increased levels of AS. Exactly how these misfolded proteins form is poorly understood. Some of the current theories include increased levels of reactive oxygen species (ROS) caused by increased iron levels and exposure to environmental factors. Currently it is believed that METH could be a

contributing factor to the increased levels of AS. The exact mechanism of how this occurs remains unclear. Jiang et al. (2014) used rats exposed to METH to investigate the underlying epigenetic mechanism causing structural changes to AS in the substantia nigra.

Rats were exposed to daily dosing of METH for five days. On days six, ten, and fourteen, locomotor activity was tested along with brain tissue samples. Results showed that rats exposed to METH exhibited poorer balance and decreased movements beginning at day 6 ($p < 0.01$, and $p < 0.01$, respectively). METH rats also had significantly increased AS and cytokines in the substantia nigra compared to controls (both $p < 0.01$). This implied METH caused increased inflammation and motor dysfunction. Epigenetic evaluation showed decreased binding site occupancy in two regions of the Snca promotor region (Snca is responsible to increased expression of AS) of the substantia nigra ($p < 0.01$). This implies that changes to these two regions may play an essential role in the upregulation of AS.

Overall, this study further confirms that METH use contributes to increased AS production which could lead to an increased risk of developing PD. Limitations to this study include study subjects being mice. There is no guarantee that results will be similar in human subjects. Additionally, there is no mention of how many rats were a part of each group.

Tavassoly et al. (2012) utilized nanopore analysis to evaluate the interactions between METH and AS. They theorized that METH may bind to the N-terminus of AS causing a conformational change consistent with the development of Lewy body formation, as seen in PD. Alpha-synuclein was combined with diluted METH and allowed to rest. Conformation changes were then observed via nanopore technology. Results showed that when AS and METH were combined, translocation events occurred at the N-terminus of AS. This conformation change increased the number of “bumping events” that occurred contributing to misfolding occurrences.

Ultimately this showed a potential relationship between increased PD occurrences amongst METH user. Limitations to this study include that it is an isolated reaction outside of the human body. There was no evidence to suggest a potential dose or frequency of METH use needed to obtain these results in a human subject. While this does provide a potential link, more research is needed on a larger scale.

Relationship between methamphetamine and amyotrophic lateral sclerosis

Amyotrophic lateral sclerosis background. Amyotrophic lateral sclerosis (ALS) is a rapid neurodegenerative condition most commonly occurring between the fifth and seventh decade of life. Deterioration of motor neurons in the brain stem and spinal cord results in symptoms like progressive muscle weakness, muscle spasticity, dysphagia, and cognitive changes (DynaMed Plus, 2019). There are two established types of ALS, familial (FALS) and sporadic (SALS). FALS, which is hereditary only accounts for 5-10% of all cases (DynaMed Plus, 2019). The remainder of cases are sporadic meaning there is no clear identifiable cause. ALS is diagnosed based on a thorough neuro exam and exclusion of other possible mimickers. Unfortunately, from time of diagnosis life expectancy is only 2-5 years (DynaMed Plus, 2019). Early treatment is ideal to slow disease progression however, this remains challenging due to unknown causes or contributing factors. In recent studies, it has been found that neurodegeneration from METH use has a similar mechanism of action to that of neurodegeneration related to ALS.

Reactive oxygen species. Sporadic type ALS has no readily identifiable cause however, a specific gene mutation to Cu/Zn superoxide dismutase (SOD1) has been identified in familial type. These types of mutations suggest that oxidative damage may contribute to the development

of ALS. Ferrante et al. (2002) attempted to answer if oxidative damage could contribute to neurodegeneration and disease progression in both FALS and SALS.

Tissue samples were obtained from postmortem individuals and subjected to immunohistochemistry (cell staining) and biochemical oxidative markers evaluations. A total of 8 FALS (with and without known mutation), 16 SALS, 11 disease control, and 28 normal matched controls were used for this study. Results showed that individuals with SALS have a significantly increased level of oxidative damage markers; OHdG $p < 0.05$ and protein carbonyl $p < 0.001$. Individuals with FALS, both with and without known mutations, had no significant changes in oxidative markers. Immunohistochemistry results showed significant increase in several oxidative markers for both SALS and FALS groups when compared to controls (hemeoxygenase-1, malondialdehyde, OHdG). Damage was most apparent in the ventral horn neurons. Ultimately, Ferrante et al. (2002) were able to identify evidence for oxidative damage that may contribute to neurodegeneration and progression of both SALS and FALS. Immunohistochemistry changes in both SALS and FALS groups and isolated biochemical changes in the SALS group, suggests that oxidative damage may occur via different pathways.

This study was limited with a small sample size and a lack of excluding criteria. Postmortem individuals were not excluded for comorbidities, drug history, medication or nicotine use. Additionally, while evidence of oxidative damage is present, it remains unclear as to how this damage occurs. There was also no report on severity of illness at time of death.

Methamphetamine can produce neurodegeneration by increasing the amount of reactive oxygen species (ROS) causing mutations to proteins. In past studies, it has been established that the CuZn-superoxide dismutase 1 (SOD1) gene is essential in reducing the toxicity of METH on dopamine neurons. When there is a mutation to the SOD1 gene, there is a significant increase in

neurotoxicity predominantly effecting motor neurons of the spinal cord and is linked to the development of FALS. However, recent studies have shown that other regions of the brain are also affected. Ferrucci et al. (2008) hypothesized that a mutated SOD1 gene and METH share a common mechanism for neurotoxicity. They also hypothesize that defective SOD1 genes may contribute to the neurotoxic effects of METH.

Brain and spinal cord tissue samples were taken from mice expressing the human G93A CuZn SOD1 mutation and were compared to SOD1 mutation negative littermates. Samples showed that mice with SOD1 mutation had a significantly decreased amount of motor neurons in the ventral horn (classic location in those with ALS) when compared to mutation negative mice. Remaining neurons had signs of deterioration including swollen cytoplasm, vacuolation, loss of neuronal processes, and enhanced reactive astrogliosis. These remaining neurons do resemble that of previously described MA exposed dopamine neurons. Immunohistochemical cells staining also showed evidence of accumulations of ubiquitin and SOD1 in the motor cortex, brainstem, and spinal cord of SOD1 mice. SOD1 mutation free mice had no increased areas of cell staining. Ubiquitin and SOD1 are substrates of the autophagic pathway potentially implying that there is a defective autophagic pathway resulting in abnormal accumulations of proteins.

Ferrucci et al. (2008) concluded with the suggestion that all neurodegenerative diseases may share dysfunction of autophagy. This study did confirm that there is abnormal build up of SOD1 and ubiquitin in neuro tissue of FALS mice. Additionally, they found damaged neurons with similar characteristics to METH exposed dopamine neurons. Limitations to this study included having no METH mice or using METH mice as a comparison for results. This ultimately resulted in making more hypotheses and questions than answers.

Methamphetamine use results in massive amounts of dopamine being released into the striatum. As the increased dopamine degrades, it produces increased oxygen free radicals which in turn make reactive oxygen species (ROS). Peroxynitrite, composed of an oxygen free radial and nitric oxide (NO), is one of the main ROS produced and is attributed to neurotoxicity. The biomarker 3-nitrotyrosine (3-NT) can be used to identify levels of peroxynitrite in the tissues. It has been suggested in recent articles that increased CuZn superoxide dismutase (SOD-Tg) and decreased NO production may result in protection against neurotoxicity.

Imam et al. (2008) further investigated the relationship between NO radicals and METH neurotoxicity. Mice were divided into three groups; nNOS knock-out (lack the gene for NO production), SOD-Tg (increased production of SOD gene), and mutation free control. All mice were exposed to METH and terminated 72 hours later. Brain tissue samples were then taken and compared to control mice given saline. Results showed a significant increase of 3-NT in control METH mice ($p < 0.05$). There was no significant increase in 3-NT in nNOS, SOD-Tg, or saline control mice. There was a significant reduction in dopamine in METH control mice ($p < 0.05$). There was no significant reduction of dopamine in nNOS and SOD-Tg METH mice. This concluded that peroxynitrite does play a significant role in neurotoxicity related to METH use and free radicals. It was also found that mice with nNOS and increased SOD levels had neuroprotection against METH use.

While these results are significant, they were performed on animal models with no guaranteed translation into human subjects. It also fails to disclose how many mice were tested. This study could be strengthened by repeating it with longer MA abstinence period to determine if results or temporary or permanent. Potentially this could impact some studies for FALS due to the relationship with the SOD gene.

Inflammatory markers. In recent mice studies, it was found that inflammatory markers presented prior to physical manifestation of ALS. Kuhle et al. (2009) evaluated serum and cerebrospinal fluid (CSF) for proinflammatory chemokines indicating inflammation in those with ALS. Samples of serum and CSF from 20 human subjects with ALS were compared to 20 subjects with non-inflammatory neurological diseases (ranged from vertigo, PD, low back pain etc.). A total of nine chemokines were evaluated. Results showed there were significantly increased levels of the chemokines MCP-1 and IL-8 in the CSF of ALS subjects ($p = 0.013$, $p = 0.035$, respectively). IL-8, MCP-4, and eotaxin were significantly elevated in the serum of ALS subjects ($p = 0.055$, $p = 0.465$, $p = 0.011$ respectively). There were no significantly elevated levels of any chemokines in subjects with non-inflammatory neurological diseases. A trend of higher reported CSF-MCP-1 in ALS subjects correlated with a shorter period between symptoms onset and diagnosis ($p = 0.075$).

These results indicate that there are specific inflammatory markers in the CSF and serum of patients with ALS. It was also important to note that ALS subjects with faster disease progression had higher levels of CSF MCP-1. This study was limited with a small sample size for both ALS and non-inflammatory groups. There was no mention of potential use of anti-inflammatory medications in any subjects. There was also no comparison for any other inflammatory conditions. Further studies would need to be performed to determine the value of chemokine markers for treatment/diagnostic purposes of ALS.

Individuals with ALS become weak rapidly inhibiting them from participating in simple motor task evaluations. Without the ability to map brain changes related to functional abilities, the potential alternative testing would involve a passive activation of the brain. Choi et al. (2010) assessed motor circuitry in rats with familial ALS (FALS) by using a pharmacologic MRI and

amphetamines to activate the brain. It was also hypothesized that a passive pharmacologic challenge with amphetamines could be used as a potential marker for motor neuro dysfunction in ALS.

Rats with FALS were divided into three groups; longitudinal (followed from pre-symptomatic to post-symptomatic), pre-symptomatic, and post-symptomatic. Pre-symptomatic and post-symptomatic rats were injected with amphetamine once and longitudinal rats received two doses; one pre-symptomatic and one post-symptomatic). Rats then underwent MRI experiments after their amphetamine dose and were compared to amphetamine control rats without FALS. Imaging showed that all pre-symptomatic rats were indistinguishable from the control rats. Symptomatic rats had significant changes to the sensorimotor cortex ($p < 0.05$), M2 motor cortex ($p < 0.05$), thalamus ($p < 0.01$), and insula ($p < 0.05$) when compared to controls. These changes were similar in longitudinal rats excluding the possibility to amphetamine sensitization changes. Ultimately this provided evidence that amphetamine use can provide an alternative pathway to stimulate the motor system passively. This becomes applicable for individuals with ALS who cannot perform motor functions.

Limitations to this study include a small sample size of rats (total of 7). While these results may have been profound, it would be recommended to repeat the experiment with a much larger sample size. Additionally, this article did not aim to evaluate the effects of amphetamine on ALS. Instead it used amphetamine to distinguish areas affects by ALS. It does provide insight that ALS and amphetamine use stimulates/effects similar areas of the brain, thus producing locomotor difficulties.

Discussion

Structural and chemical changes summary

This review has demonstrated that METH has significant neurotoxic properties to structural and chemical components of the brain and nervous system. Structures impacted the most are those related to the reward pathway including the stratum, cerebral cortex (insula), thalamus, and the basal ganglia (May et al, 2003; Thompson et al, 2004). Structural abnormalities most frequently seen include white matter hyperintensities. Salo & Fassbender (2011) and Thompson et al. (2004) both established that this abnormality was more frequently seen in individuals using METH versus abstinent controls. A higher proportion of white matter hyperintensities (WMH) were seen in the frontal lobes and were of greater frequency and severity in the male gender (Thompson et al, 2004; Sabrini et al, 2019). Structural abnormalities also correlated with cognitive and physical manifestations. Physical manifestations seen included choreoatheroid movements, punding, and ataxia. Symptoms were most apparent within three days of METH use (Granado et al, 2018). Cognitive learning, reasoning, impulsivity, and risky decision making were all significantly impacted by METH use as seen in the systemic review by Sabrini et al. (2019). Flavel et al. (2012), Huang et al. (2017), and Granado et al. (2018) also established that the myelin sheath of nerves was negatively impacted by METH. Prolonged latency in muscle activity, ataxia, and dyskinesia were most frequently seen. There also appeared to be a correlation between increased frequency/dose of METH with worsening symptoms and performance. Those with a protracted abstinence had improvement and resolution of physical manifestations; however, WMH and impaired nerve impulses remained for years, indicating there may be long-term consequences. All studies had small sample sizes and frequently allowed

the use of nicotine and marijuana potentially impacting results. Further studies should also be performed on gender specific outcomes due to an apparent protective factor of estrogen.

Chemically, METH had the greatest impact on dopaminergic structures. Every use of METH creates a massive release of dopamine. This activates the reward pathway and signals a pleasurable response thus reinforcing the behavior. This large release of dopamine then creates down regulation of dopamine receptors/dopamine transporters (Ashok et al. 2017). From the neurotoxic properties of METH, there is evidence supporting more than just downregulation of dopamine. In multiple studies involving humans and mice, there was evidence of dopamine terminal damage and an overall loss of dopamine producing cells. When subjects were exposed to various levels of METH, there was a decline the dopaminergic markers TH, DAT, and VMAT2 (Ares-Santos et al. 2013; Ashok et al. 2017; Kish et al. 2016; Morrow et al. 2011). It was concluded that while some of the decline in dopamine can be attributed to downregulation, the primary MOA is impairment/destruction of dopamine terminals (Ares-Santos et al. 2013; Ashok et al. 2013; Morrow et al. 2011; Volkow et al. 2001). Areas affected the most include the striatum, cerebral cortex (insula), thalamus, and basal ganglia (May et al. 2013; Sabrini et al. 2019). Loss of dopamine structures and severity of symptoms were directly linked to dose/duration of METH. The significant increase in dopamine also produces a significant increase in ROS via mitochondrial dysfunction, like damage frequently seen in PD, MS, and ALS. Microglial activation indicating inflammation of neurological structures was made apparent in two separate studies (Bowyer et al. 2016; Sekine et al. 2008). Microglial activation was most frequently seen in the thalamus, striatum, midbrain, and insular cortex. Microglial activation remained in METH subjects for up to two years into abstinence.

Volkow et al. (2001) established in human studies that there was a significant amount of physical and neuropsychological decline post METH use. Individuals abstinent for at least 11 months continued to have declined cognitive function. In studies with mice, there was strong evidence that mice receiving METH had substantial changes to the striatum and substantia nigra resulting in significant changes to locomotor activity. The greatest locomotor changes were seen between days 1-3 post METH use and lasted up to 7 days. After this period there was a resolution of physical manifestations, however, long-term dopaminergic changes remained (Ares-Santoes et al. 2013; Granado et al. 2018).

Are those who use methamphetamine at an increased risk for developing multiple sclerosis vs those who do not use methamphetamine.

Overall, there is a lack of research to confidently state that METH use can cause MS. There is ample evidence to suggest that METH use can be a mimicker of MS. The McDonald Criteria clearly states that there must be evidence of CNS damage (WMH) in two or more parts of the brain, spinal cord, or optic nerves, with evidence of dissemination in time, and all comorbid conditions possibly mimicking MS must be ruled out. A supportive test that can be used is a lumbar puncture testing for the presence of CSF oligoclonal bands (OCB). This test is not required for a diagnosis but has a specificity (87%) and sensitivity (92%) to MS (when compared to mimickers). A negative value cannot rule out the diagnosis of MS. To date, it appears that METH has not been shown to cause the presence CSF oligoclonal banding, however, I was unable to find research or data confirming this statement. As Bae et al. (2006), Thompson et al. (2004), and Sabrini et al. (2019) found, METH causes WMH that are indifferent from those seen in MS on MRI. These abnormalities can persist for a prolonged period. It can also be assumed that if an individual is using METH more than once, it could create WMH with

evidence of dissemination in time. There are also several common mechanisms of action for neurodegeneration including ROS (Jang et al. 2016). Neurological symptoms are strikingly similar between METH and relapsing-remitting MS. To establish a relationship between METH use and MS, epidemiological studies should be performed. At this time, it is not reasonable to conclude that METH use is a direct risk factor for developing MS. However, this may be reasonable to conclude that METH use can mimic MS. Providers should be aware that symptomatic individuals with a history of METH use should not be given the diagnosis of MS without a thorough evaluation and evidence of prolonged abstinence from METH.

Are those who use methamphetamine, at an increased risk for developing Parkinson's disease vs those who do not use methamphetamine.

There is strong evidence that METH use may be a risk factor for developing Parkinson's disease. PD pathophysiology includes progressive death of dopamine producing cells in the substantia nigra. The decline in dopamine produces progressive bradykinesia, resting tremor, rigidity, ataxia, and cognitive decline. While the exact cause of PD is unknown, it is thought to be linked to environmental exposures. In individuals using METH, there is evidence that similar pathophysiology is occurring. In multiple studies involving humans and mice, there was evidence of dopamine terminal damage and an overall loss of dopamine producing cells. When subjects were exposed to various levels of METH, there was a decline in the dopaminergic markers TH, DAT, and VMAT2 (Ares-Santos et al. 2013; Ashok et al. 2017; Kish et al. 2016; Morrow et al. 2011). While some of the decline in dopamine can be attributed to downregulation, evidence suggests the primary mechanism of action is impairment/destruction of dopamine terminals (Ares-Santos et al. 2013; Ashok et al. 2013; Morrow et al. 2011; Volkow et al. 2001). It should also be noted that while there is a similar damage/loss of dopamine levels

between METH users and PD, PD loss is slightly greater. Areas affected the most include the striatum, cerebral cortex (insula), thalamus, and basal ganglia (May et al. 2013; Sabrini et al. 2019). Loss of dopamine structures and severity of symptoms were directly linked to dose/duration of METH.

There were several identifiable molecular mechanisms of neurodegeneration found to be similar between PD and METH use. Reactive oxygen species are essential to the development of Lewy bodies in individuals with PD. In subjects using METH, identical ROS were successful in creating the necessary conformational changes to proteins to make Lewy bodies (Jiang et al. 2014; Tavassoly et al. 2012). Lotharius et al (2005) identified iron levels as one key difference between Lewy body formation in PD and METH users. It was found that to achieve complete dopamine neuron death in PD, iron levels need to be elevated. In METH users, iron levels are not elevated.

The pattern of physical symptoms/manifestations do vary slightly. In individuals with PD, their physical and cognitive functions are steadily declining with no periods of improvement. In individuals using METH, physical symptoms reached their peak within the first three days and would resolve/improve with abstinence. Cognitive decline was also greatest immediately post METH use and only recovered marginally. (Ares-Santos et al. 2013; Jang et al. 2016; Volkow et al. 2001). While physical manifestations did resolve, in two separate epidemiological investigations, it was found that those exposed to METH had a higher reported incidence of PD later in life, versus those without METH use (Callaghan et al. 2010; Curtin et al. 2015). This implies that METH use can cause initial damage to dopaminergic structures and may also contribute to long term damage. This could be further exacerbated with natural aging which also contributes to loss of dopaminergic structures.

For future studies, a possible link between METH use and PD that should be further explored is their relationship with agricultural chemicals. The cause of PD remains unknown but is theorized it could be linked to agricultural chemical exposure. One of the critical components/byproducts of METH are agricultural chemicals. Interestingly, PD has the most reported cases in MN and ND, and METH use is most reported in the Midwest. To prove this relationship, more investigative studies are needed. At this time, there is ample evidence that the use of METH could be a risk factor for future development of PD.

Are those who use methamphetamine at an increased risk for developing amyotrophic lateral sclerosis vs those who do not use methamphetamine.

There is limited evidence available linking METH use and ALS. There is evidence that neurotoxic properties from METH can cause a SOD1 gene mutation, like in individuals with FALS (Ferrucci et al. 2008). These mutations occur through the damaging properties of ROS, including peroxynitrite (Imam et al. 2008). While physical manifestations look similar, there are different disease progressions. ALS/FALS has a rapid degeneration occurring over 2-5 years (DynaMed Plus, 2019). Individuals using METH have an immediate onset of symptoms with eventual resolution of physical manifestations if abstinence is sustained. While there is a common mutation to the SOD1 gene, there is a lack of evidence that METH could be considered a risk factor for ALS/FALS at this time. Future epidemiologic studies should be performed to evaluate this relationship.

Applicability to Clinical Practice

With the available information provided in the literature review, the primary care provider can have a better comprehensive understanding of the implications of

methamphetamine use and neurodegeneration. METH use is detrimental to dopaminergic structures, particularly those related to the reward pathway. Damage to these structures will result in cognitive and physical symptoms. Physical manifestations will be more apparent immediately after and up to one-week post METH use and could include ataxia, impaired fine motor skills, and punding. Providers should be familiar with the McDonald criteria and understand that METH could be a potential mimicker for MS. A true diagnosis of MS cannot be definitively be made from the MRI and symptoms alone. In an individual with METH use in their history, CSF oligoclonal bands or sufficient abstinence would be required. Providers should also be aware that METH use can contribute as a risk factor for PD. In patients with a history of METH, education should be provided and a higher acuity of PD like symptoms should be present. There is still a lack of evidence to confidently conclude that METH use may increase the risk of ALS. Collectively it should be understood that METH has significant neurotoxic properties leading to a cascade of detrimental physical and cognitive outcomes.

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