A Review of Preclinical Studies on Methamphetamine-Induced Gut Microbiome Alterations

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Basic Definitions

Gut Microbiota	Houses the ensemble of microorganisms such as bacteria, viruses, fungi, and yeasts that live within the gastrointestinal tract (Salavrakos et al., 2020)
Gut Microbiome	Refers to the total genetic material of bacteria in the gastrointestinal tract (Salavrakos et al., 2020)
Gut Dysbiosis	Disturbances in the homeostasis of the microbiota (Forouzan, Hoffman, and Kosten, 2020)
Autophagy-Related Proteins	Autophagy related proteins are essential to maintaining cellular homeostasis and growth. As well as playing a key role in cell death/survival (Chen et al., 2020)
Apoptosis	Form of programmed cell death (Chen et al., 2020)

Prevalence of Substance Use Disorder

Substance use disorder (SUD) is a chronic, pervasive disease that affects approximately 40.3 million Americans aged 12 (SAMHSA, 2021)

FDA approved medications are only available for those with alcohol (AUD) and opioid use disorders (OUD) (Forouzan et al., 2021)

964,000 Americans have methamphetamine use disorder (MUD) within the (Forouzan, Hoffman, and Kosten, 2020)

The Gut Microbiome and SUD

The gut microbiome has between 100-1,000 different identified species (Salavrakos et al., 2020)

It is difficult to determine what a “normal” gut microbiome composition is since it can be impacted by genetics, mode of delivery at birth, race, diet, and environment (Salavrakos et al., 2020)

Alterations in the gut microbiome have been reported in those with AUD and OUD (Salavrakos et al., 2020)

This is a cause of concern given that several channels in the gut microbiome communicate with the brain

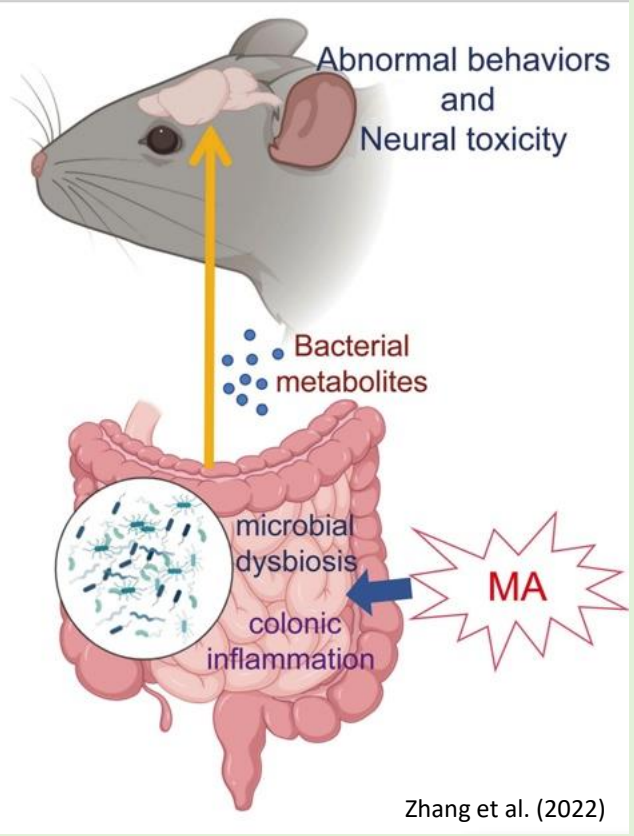
Gut-Brain Interactions

The most recognized gut-brain interactions are

The immune response	The metabolic pathway	The neuroendocrine pathway	The Vagus nerve

All of these interactions can be affected by alterations within the gut microbiome (Salavrakos et al., 2020)

Several studies have demonstrated evidence for gut dysbiosis in AUD and OUD populations. Fewer studies have examined the microbiome alterations associated with meth use



Zhang et al. (2022)

Meth Use Disorder and Affective Disorders

MUD withdrawal is characterized by drug-cravings, physical discomfort, anxiety, stress, and depression (Forouzan et al., 2021)

MUD withdrawal symptoms are also present in affective disorders

Given these similarities in symptoms and the comorbidity of affective disorders and SUD, Forouzan, Hoffman, and Kosten (2020) proposed that they share commonalities within the gut-microbiome

Gut Microbiota Dysbiosis and Probiotics

Dysbiosis has been implicated in several conditions such as SUD. (Forouzan, Hoffman, and Kosten, 2020)

There is relationship between intestinal inflammation and alcohol-induced gut dysbiosis (Forouzan, Hoffman, and Kosten, 2020)

Probiotics have been reported to affect the HPA-axis' stress responses by decreasing corticosteroid levels (Wang et. al, 2016). As well as, affecting levels of neurotransmitters (Wang et al., 2016)

Wang et al. (2016) in a systematic review reported that several probiotic formulations improved behavioral indices of anxiety and depression

Methods

Obtained through Western Michigan University's library databases

Contains five preclinical studies and three literature reviews

Inclusion criteria: Utilized behavioral tests predictive of depression and anxiety or examined indices of neurotoxicity . Additionally, must have addressed gut microbiota dysbiosis and examined the effects of methamphetamines on gut microbiome composition.

Studies excluded examined drugs other than meth

Results

Study	Animal	Behavioral Assays	Meth (dosage, duration)	Behavioral Outcomes	Neurotoxicity	Gut Microbiome Alterations
Zhang et al., 2022	Wild Type C57BL/6 male mice (8-10 weeks)	Locomotor Sensitization (open field test)	2mg/kg - 24hr Intervals	Locomotor ↑ Sensitization	DAT, TH, and MAOA ↑	Microbial Diversity ↓
		Light-Dark Test (measuring anxiety)	6-days	Anxiety-Like Behaviors ↑	Beclin1, Atg5, and LC3-II proteins ↑	*Bacteroidetes*, A*ctinobacteria, and unclassified K_norank* ↑
		Tail Suspension Test (measuring depression)		Immobility ↑	Apoptosis related proteins (p53, Bax, caspase 3, and cleaved caspase 3) ↑	*Firmicutes, Proteobacteria, Tenericutes, and* ↓ *Deferribacteres*
		Forced Swim Test (measuring depression)		Immobility ↑		Expression of TLR4, MyDD88, NF-kB, and NLRP3 ↑
Forouzan et al., 2020	16 Sprague-Dawley male rats (60-90 days old)	Open Field Test (measuring anxiety)	2mg/kg (twice-daily)	Anxiety-like Behaviors ↑	N/a	*Actinobacteria* on Meth day 7, 14, and 24h cessation ↑
		Elevated Plus Maze (measuring anxiety)	14 days of Meth treatment	No significant difference between groups was found		*Allobaculum*, B*ifidobacterium*, and L*actobacillus* ↑ (abundance of these genera returned to baseline after 7 days of cessation)
		Forced Swim Test (measuring depression)	Behavioral Assays conducted initally with Vehicle and then during cessation from Meth	Immobility ↑		Significant microbial community differences were observed ↑ on Meth day 7, 15, and 24h cessation of Meth. These differences dissipated by 96h post Meth cessation

Results Cont.

Study	Animal	Behavioral Assay	Meth (dosage, duration)	Behavioral Outcomes	Neurotoxicity	Gut Microbiome Alterations
Chen et al., 2021	14 BALB/c male mice (6-8 weeks old)	N/a	One injection per day for 6 days as follows; 1.5, 4.5, and 7.5 On days 7-8, mice exposed to binge dose involving four successive injections of 10.0mg/kg at two-hour intervals for two sucessive days	N/a	MAOA ↑ TH ↓ Beclin1, Agt5, and ↑ LC3II protiens	Inflammation factors in the colonic mucosa (overexpression of TLR4, MyDD88 and Nf-kB proteins) ↑ Microbial Diversity ↑ *Bacteroidetes* and F*irmicutes* ↓ *Proteobacteria* and A*ctinobacteria* ↑ Relative abundance of F*usobacteriaceae*, L*actobacillacea*, and P*revotellaceae* ↓
Yang et al., 2020	22 Sprague-Dawley male rats	Conditioned Place Preference Assessment (measuring the rewarding effects of the drug)	2mg/kg (one injection per day) Alternate injections of Meth or Saline for 9 days Pre-treated with anti-biotics	Preference for Meth- ↑ paired chamber	N/a	Gut dysbiosis in rats pre- treated with anti-biotics ↑ Abundance of *Akkermansia, Allobaculum, and Olsenella* ↑ Abundance of *Acetivibrio* ↓ Abundance of *Butyricimonas* ↑
Ning et al., 2017	16 Sprague-Dawley male rats	Conditioned Place Preference Assessment (measuring the rewarding effects of the drug)	1mg/kg (one injection per day) Alternate injections of Meth or Saline for 14 days	Preference for Meth- ↑ paired chamber	N/a	Microbial Diversity ↑ Abundance of *Ruminococcaceae, Bacillus, CetoBacterium, Fusobacteria,* and *Aeromonas* ↑ Abundance of *Phascolarctobacterium* ↓

Methods of Analysis

Collected fecal samples and conducted 16s rRNA sequencing to determine the fecal microbial composition

Reported Simpson and Shannon indices to draw conclusions about microbial diversity (Chen et al., 2021; Forouzan, Hoffman, and Kosten, 2020; Ning et al., 2017; Yang et al., 2020; Zhang et al., 2022). Chen et al. (2021)

Performed ANOVAs and applicable t-tests to compare experimental and control groups (Chen et al., 2021; Forouzan, Hoffman, Kosten, 2020; Ning et al., 2017; Yang et al., 2020; Zhang et al., 2022).

Measured neurotoxicity which was achieved by Western Blot Analysis (Chen et al., 2021; Zhang et al., 2022).

Discussion

Methamphetamine can produce gut dysbiosis and subsequent neurotoxicity

Reported increases in anxiety and depressive-like behavior following methamphetamine treatment. They also all reported methamphetamine-induced changes in the gut microbial composition.

Since probiotics decrease inflammation and anxiety/depression in patients with IBS, they may curb the negative effects of MUD such as anxiety, depression, and impulsivity (Wang et al., 2016)

Discussion Cont.

Impulsivity contributes to relapse

Future studies should evaluate the effects of methamphetamine on impulsivity as well as examine whether probiotics can attenuate these effects

Additional research should be conducted on the health-related effects of microbial diversity

Accurate assessment of relevant behavioral outcomes is essential to measure the effects of methamphetamine-induced gut dysbiosis and the possible effects of probiotics

Conclusion

Significant relationship between exposure to meth and gut dysbiosis as well as anxiety and depression-like behaviors in preclinical behavioral assessments

Based on these pre-clinical findings, future research is warranted to evaluate probiotic treatments with human SUD populations

The research methods must be translated to human populations

Questions/Comments?

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