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REVIEW

An Overview of the Methamphetamine Effect on Male Sexual Behavior and Reproductive System

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Summary

Drug addiction and its effect on the behavior and development of children has become a serious problem in our society. Methamphetamine (MA) is one of the most abused psychostimulants in the Czech Republic, and its abuse is rising worldwide. Previous studies have demonstrated the adverse long-term effects of maternal drug abuse on rat offspring. However, the father's contribution as a parent and donor of half of the genetic information is unclear. Previous studies of other psychostimulant drugs indicate that long-term application of MA to adult male rats may induce changes in their reproductive system and lead to changes in rat pup functional and behavioral development. Therefore, the present review aimed to investigate the effect of MA administration on reproductive toxicity and sexual behavior of adult male rats, as well as the impact of paternal MA exposure on behavioral development and locomotor activity in rat offspring.

Key words

Drug addiction • Methamphetamine abuse • Paternal exposure • Behavioral tests • Testosterone • Spermatogenesis

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Introduction

Methamphetamine (MA) is a psychotropic stimulant that affects the body on a biological, behavioral, and psychological level. In many countries, it is one of the most widely used illicit drugs. Psychostimulant drugs such as MA activate the dopaminergic and serotoninergic pathways of the central nervous system (CNS), which are mainly associated with reward circuits, affective states, sexual behavior, and also in control of motor function and cognition [1]. In humans, MA induces feelings of happiness and pleasure, suppresses anxiety and depression, increases concentration, reduces appetite, and promotes weight loss [2].

Controlling MA abuse has been a significant change over the decades. It is known to be a powerfully addictive drug, and due to its low price and relatively simple production, it has become increasingly popular in our society. The stable crystallized form of the drug was first synthesized in Japan in 1919 by Ogata, which provides the basis for producing the drug on a larger scale [3]. In the 1950s, MA was legally administered to treat depression, and athletes used MA as a permitted stimulant. After its massive expansion in the 1970s and recognition of severe side effects, its production and use were declared illegal [4,5]. Worldwide, the largest group of abusers is concentrated in East and Southeast Asia, where most of the global MA production is located. However, the annual report of the EMCDDA in 2019 considered MA the 4th most abused illegal drug in the Czech Republic after cannabinoids, ecstasy, and hallucinogenic fungus (psilocybin).

Our laboratory has been studying the effect of MA on the mother and her offspring in an animal model since 2002 [6]. Psychostimulants affect the behavior of addicts, increase aggression and disrupt social and maternal behavior [7,8]. These facts highlight the very serious worldwide problem of women abusing MA during pregnancy [9]. While MA abuse has apparent advantages

for drug-addicted pregnant women compared to other drugs, such as helping to maintain lower body weight, reducing appetite, and increasing energy. Prenatal MA exposure causes many negative impacts, because it is a lipophilic molecule that rapidly penetrates membrane structures including placenta, and enter the breast milk of mothers [10]. This may seriously affect the prenatal development of the fetus or postnatal development of the newborn during lactation, respectively [11]. Maternal MA exposure has been shown to cause morphological changes in the brain and altered brain metabolism [12,13]. Previous studies showed that MA administration to pregnant rats impairs postnatal sensorimotor development of pups during the pre-weaning period [14,15,16], affects learning abilities [17], and leads to a higher susceptibility to drug sensitization in adulthood [18].

Previous animal studies demonstrated that prenatal exposure to MA impairs cognition [19,20,21], evokes anxiety-like behavior in offspring [22,23] and affects pain sensitivity later in life [24]. Furthermore, prenatal MA exposure is also associated with increased physiological stress, decreased arousal, and poor quality of movement during the first five days of life [25,26]. Maternal MA abuse has also been shown to affect stress responses in adult animals [27]. Alterations in behavioral patterns manifested in social behavior during adolescence [16] and in aggressive and sexual behavior during adulthood [28]. Previous studies also demonstrated that the administration of MA during pregnancy attenuates the maternal behavior of rat mothers [29,30].

In the general population, however, MA abuse is frequent in both female and male populations, primarily because of its stimulant effects and easy availability. Currently, most MA users are reproductive-age men between 18 and 34 years [31]. While the effect of MA on mothers and offspring has been the subject of detailed research over the past few decades, the effect of MA on fathers and offspring has received far less attention. Therefore, our review aimed to investigate the impact of paternal MA abuse on the behavior of addicted male rats and their offspring.

In our review, we focused mainly on methamphetamine drug addiction and its influence on sexual activity and reproductive toxicity in males. Literature research was based on main keywords: sexual activity, methamphetamine exposure, drug addiction, reproductive toxicity. Our literature sources are from online databases of science journals and biomedical articles. We have drawn from scientific papers published since 1972 until recently published articles.

Effect of MA exposure on male rats

Effect of MA exposure on locomotor activity in male rats

Previous studies found that psychostimulants, such as MA, increase locomotor activity and exploration in unknown environment [33,34,35]. Moreover, our findings confirmed that acute MA (1 mg/kg) exposure significantly increased overall activity (increased locomotion, rearing, grooming, speed average, and distance traveled) and decreased immobility in adult male rats [36]. The increased locomotor activity seen in MA-exposed rats is mainly associated with increased levels of dopamine, especially in the nucleus accumbens [37]. Dopaminergic neurotransmission in the nucleus accumbens and the caudate nucleus mediates MA-induced hyperlocomotion and stereotypy, respectively [38,39,40]. Experimental animal models demonstrate that the effect of MA exposure on locomotor activity is dose-dependent [41]. Acute MA administration increases locomotor activity when administered at lower doses (1 mg/kg) and elicits stereotypic behavior when administered at higher doses (5 mg/kg) [42,43]. In addition, female rats have been shown to be more sensitive to the locomotor activating effect of i.p. MA administration (0.1-3.0 mg/kg) than male rats [44]. Interestingly, previous studies indicate that increased sensitivity to MA exposure is age-related. A study by Zakharova *et al.* demonstrated that daily administration of MA increased locomotor activity in both adolescent and adult rats, with a more significant effect seen in adults [45]. In addition, our previous study showed an interaction between chronic 30-day MA exposure, acute application and time the way that acute MA application increased locomotor activity more in animals treated 30-days with saline than in animals with MA chronic treatment within the 20^{th} and 50^{th} minute of measure [36]. This finding indicates that chronic MA administration decreased the baseline level of locomotor activity in male rats compared to control saline-treated group (SA). Previous study by Segal and Mandell demonstrated that the first amphetamine administration significantly increased locomotor activity in rats; however, this was gradually replaced by progressive increase in stereotypy and decrease in locomotion during 36 days of drug exposure [46]. Moreover, the same impact on locomotor activity has been reported after long-term MA administration [47].

According to these findings, we assume that long-term MA administration impairs locomotor activity in rodents.

Effect of drugs on the sexual behavior of rats

The mechanism of action for MA is based on the release of neurotransmitters, such as dopamine (DA), serotonin (5HT), and noradrenaline (NA) by the central nervous system (CNS) and by blockade of their reuptake from synapses and into synaptic vessels [48,49]. DA in the nigrostriatal tract influences motor activity, in the mesolimbic tract it activates behaviors including copulation, and in the medial preoptic area, DA controls genital reflexes, copulatory patterns, and sexual motivation. In addition, 5HT positively or negatively affects copulatory patterns such as erection and ejaculation by targeting a specific subtype of the 5HT receptors [50]. According to these studies, DA and 5HT strongly influence sexual behavior. Therefore, there is a possibility that MA could mediate sexual activities in drug-exposed males. Some MA users report experiencing enhanced sexual pleasure, sexual confidence, and sexual performance compared to when they were not using MA [51]. A previous study by Winland *et al.* showed that MA enhances sexual motivation in female rats, i.e. MA-treated female rats were less discriminating about how and with whom they mated and were more interested in sex than SA-treated female rats [52]. A study by Frohmader *et al.* demonstrated that MA administration in male rats activates neurons in the brain's mesolimbic system, which regulates sexual behavior [53]. As for amphetamines, experimental animal models have demonstrated that repeated exposure to amphetamine stimulates sexual behavior in naïve male rats regardless of the environment in which the experiment was conducted [54].

MA abuse is commonly associated with sexually compulsive behavior [55] and increased sexual risk behavior in humans [53]. An animal study by Bolin and Akins demonstrates that chronic pre-exposure to MA impairs sexual motivation but not sexual performance [56]. However, MA is also associated with decreased sexual function, as chronic MA abuse results in an inability to reach full erection, delayed ejaculation, and orgasm [53]. A study by Kuiper *et al.* reports that MA administration leads to maladaptive sexual behavior in rats, which was associated with alterations in neural activation of the brain [57]. Studies by Frohmader *et al.* found that MA administration in male rats impairs sexual motivation and performance in a dose-dependent matter [53]. Low doses of MA did not disrupt sexual functions, and acute MA administration failed to impair sexual interest and activity.

Moreover, they [58] found that MA pretreatment did not affect the expression of sexual behavior; however, the association between MA and mating was essential for the development of compulsive sexual behavior and changes in sex and drug reward systems. Despite the above-cited works, studies using chronic MA exposure to examine male rodent sexual behavior are still lacking. Our previous results also showed no effect of MA on sexual activity (n[MA]=8, n[SA]=8) and correlated with previous findings that MA exposure does not affect sexual motivation and performance in pre-treated male rats [36].

Effect of drug exposure on spermatogenesis

Rats are considered pubertal until 50 days of age, when spermatozoa are first found in the tail of the epididymis. Sperm production increases up to age 75 days, and testicular weight increases until 100 days of age. Sperm reserves in the tail of the epididymis are not maximal until 100 days of age. Therefore, Wistar rats should not be considered sexually mature until 100 days. Sexually mature rats have testes weighing 3-7 g, and the average sperm concentration ranged from 152.5-230.0×10^7 spermatozoa/ml [59].

Drugs such as cocaine, amphetamines, and cannabinoids may adversely affect the quality and quantity of sperm and result in infertility of drug users [60]. Previous animal studies have demonstrated the adverse effect of cocaine abuse on reproduction and spermatogenesis in males [61]. A study by George *et al.* also found that the significant toxic effect of cocaine on spermatogenesis is attributed to the ischemic effect of cocaine. Cocaine, which enhances noradrenaline and adrenaline release, induces intense vasoconstriction. Moreover, it has been demonstrated that chronic cocaine administration increases germ cell apoptosis [62]. Other studies have shown the negative impact of cocaine and its metabolites on Sertoli cell function in male rats [63]. Cocaine-induced apoptosis in sperm cells is significantly increased 15 days after the first administration of the drug [62], reaches a maximum 30 days after application, and persists until day 90. It has been known since the 1980s that ethanol and acetaldehyde (the first ethanol metabolite) are potent Leydig cell toxins [64]. Moreover, it has been experimentally determined that alcohol abuse reduces cytosine methyltransferase mRNA levels in

sperm cells of addicted males (fathers), leading to spermatogenesis malfunctions [65]. Opioids are also known to impair spermatogenesis. Rats chronically treated with tramadol (a commonly prescribed effective opiate painkiller) exhibited degenerative histological changes in the seminiferous tubules, the Sertoli cells, and the Leydig cells [66]. It was shown that human spermatozoa express μ-, δ-, and κ-opioid receptors located on the head, middle region, and tail of sperm [67].

Other addictive substances such as nicotine, cannabis, and amphetamines also alter spermatogenesis by inducing oxidative stress and subsequent apoptosis in testicular tissue. The study by Condorelli *et al.* showed that nicotine is responsible for all the negative impacts of cigarette smoke on sperm [68]. Chronic, intensive marijuana usage has been associated with oligospermia in 35 % of adult men who provided semen samples [69]. Human spermatozoa express cannabinoid 1 (CB1) receptors, and *in vitro* studies exposing human spermatozoa to marijuana extracts have demonstrated decreased sperm motility, viability, and function [70,71,72]. Daily administration of synthetic THC derivates leads to a significant reduction in sperm count and daily sperm production and a reduction in the number of Sertoli cells in rats [73].

A previous study also demonstrated that MA administration significantly decreases cell proliferation and increases apoptosis in rat spermatogonia and primary spermatocytes [74]. A study by Saberi *et al.* demonstrated the adverse effects of MA exposure (after 7 and 14 days) on rat testes structure and spermatogenesis [75]. A study by Montagnini *et al.* demonstrated that repeated administration of methylphenidate (Ritalin) during childhood to early adulthood interfered with testicular functions in adult rats [76]. Moreover, a study by Gonzalez *et al.* showed the potential role of the local dopaminergic system in psychostimulant-induced testicular pathology [77]. The above studies show that drug exposure may induce reduced sperm cell production and leads to changes in sexual behavior. Several studies have reported that MA administration induces apoptosis of spermatogenic cells, lower sperm quality, as well as damage to Leydig cells and their functions [78,79,80]. Recent animal studies indicate that MA exposure leads to changes in GABAergic activity, which is involved in the proliferation of Leydig cells, testosterone production, and spermatogenesis [80]. A study by Saberi *et al.* demonstrated that MA abuse causes a significant decrease in the number of seminiferous tubule cells and lower sperm production in the MA-treated group of rats compared to controls [75]. Additionally, MA abuse in a dose-dependent manner showed detrimental effects on male reproductive functions, including impaired sperm parameters and sperm chromatin/DNA integrity [81]. Our unpublished data examined the effect of the long-term abuse of MA on rat spermatogenesis. The total sperm amount per milliliter (ml) was evaluated by using a bright field microscope and counted in a Bürker hemocytometer (Fig. 1).

The analysis of spermatogenesis in adult male rats exposed to MA did not show any significant differences in sperm production compared to control group. However, our results indicate that both treated groups had lower sperm levels compared to physiological level of non-treated healthy male rats (Fig. 2). The average amount of spermatozoa/ml in MA-treated group was $125.5\pm11.32\times10^7$/ml (Mean ± SEM) and in SA-treated group was $132.7\pm5.432\times10^7$/ml (Mean ± SEM). If we found reduced sperm concentration only in the MA group, we could argue that this was a drug effect; however, since the same reduction was also seen in the control group exposed to SA injections, then the explanation may be due to the effect of stress induced by repeated injections.

In animals, social stress, high altitude, surgery or injections, and immobilization stress were shown to affect body weight, testosterone levels, and copulatory behavior with variable effects on testicular morphology [82]. In addition, a study by Drude *et al.* showed that mice that received a single intraperitoneal injection of harmless saline had an increased corticoid stress response to a second saline injection [83]. Thus, we suggest that the same effect occurred in our experiment too.

Effect of drug exposure on testosterone levels in males

Testosterone is the primary androgenic steroid hormone. The hypothalamic-pituitary-gonadal axis regulates sexual behavior in male rats. Gonadotropin-releasing hormone (GnRH) released from the hypothalamus initiates the release of luteinizing hormone (LH), which in turn stimulates the release of testosterone from the testes [84,85]. Male rats reflexively release testosterone when they smell or mate (ejaculatory release) with a novel receptive female. LH is elevated 10 min after exposure to a female and is followed by a release of

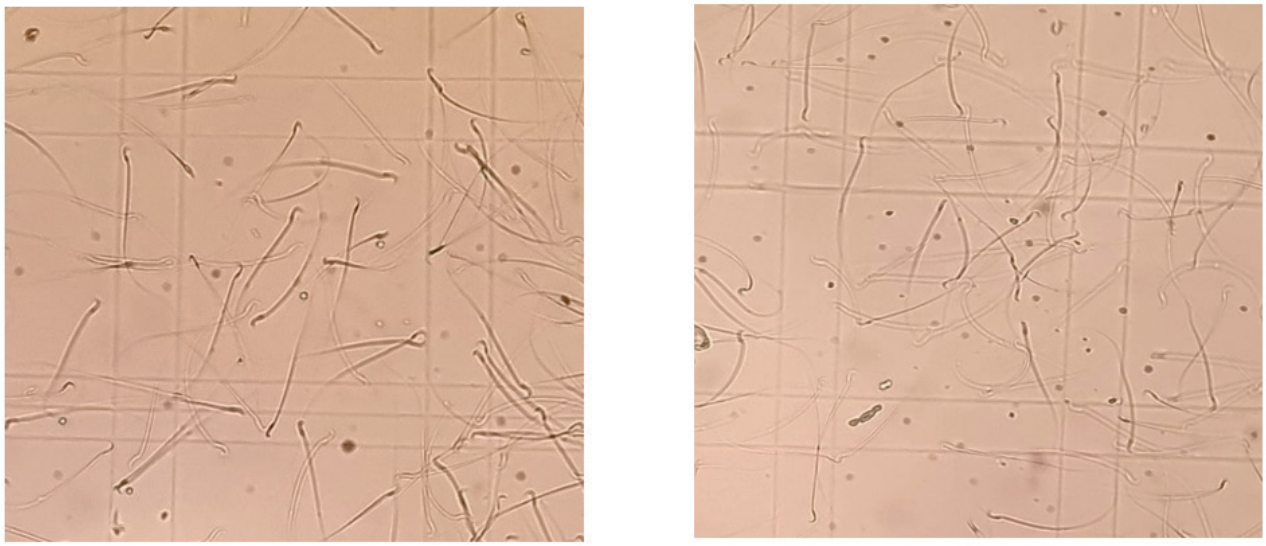
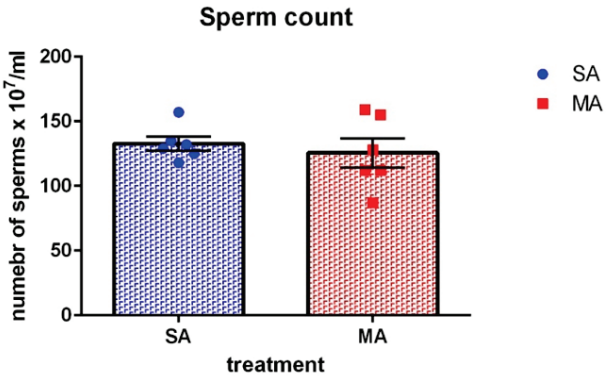


Fig. 1. Sperm count in light microscope MA-treated group (left) SA-treated group (right).



NUMBER OF COUNTED SPERMS [×10^7/ml]	
SA	**MA**
118	**87**
135	**112**
132	**112**
125	**128**
129	**155**
157	**159**

Fig. 2. Chronic MA administration did not influence the sperm production of MA-exposed males compared to saline controls. MA=methamphetamine, SA=saline. Values are mean ± SEM (n[MA]=6, n[SA]=6).

testosterone about 30 min after exposure [84,85,86]. GnRH secretion in adulthood is pulsatile, with the highest testosterone peaks during the early morning hours [87]. Although, the circulating serum testosterone concentration decreases with age, mainly due to the attrition of Leydig cells and declined secretion of hypothalamic GnRH [88,89].

Testosterone and its metabolites initiate male sexual behavior by acting on key brain regions. Aromatization of testosterone to estradiol in the medial preoptic area of the hypothalamus is essential for initiating copulation, while dihydrotestosterone, another testosterone metabolite, is crucial for controlling genital reflexes [84,90].

Plasma testosterone concentrations in adult male rats and female rats during estrus and proestrus were determined to be 5.71±0.84, 1.24±0.29, and 0.80±0.36 ng/ml, respectively [91]. A study by Lee *et al.* found a rise in testosterone levels from PD 25 that persisted until sexual maturity [92]. Moreover, a stepwise rise occurred to reach levels in excess of 230 ng/100 ml between PD 70-80. Another study found that plasma levels varied significantly during the day, with the acrophase occurring between 9.00-13.00 h [93]. The plasma levels of testosterone are elevated during sexual intercourse. However, an animal study by Shulman and Spritzer concluded that when male rats have daily sexual interactions, sexual behavior tends to show cyclic changes, and testosterone levels are significantly elevated only on the first day of sexual interaction [84].

Alcohol, opioids, and anabolic steroids can reduce testosterone production in males, thus interfering with testicular and/or hypothalamic-pituitary function [94]. Van Thiel *et al.* showed that alcohol-abusing rats had testosterone levels reduced by 50 % compared to the control group [64]. Moreover, *in vitro* studies demonstrated that rat testes perfused with ethanol and acetaldehyde showed reduced production and secretion of

testosterone. Decreased testosterone level in heavy drinkers is linked to reduced Leydig cell production and increased androgen metabolism. Alcohol also induces the aromatase enzyme that catalyzes the conversion of testosterone in estradiol [95]. Opioid-induced androgen deficiency (OPIAD) is a well-known syndrome characterized by decreased testosterone levels, reduced libido and muscle mass, fatigue, and osteopenia [96]. However, the inhibitory effects of opioid drugs on the hypothalamic–pituitary–testicular axis and, thus, on testosterone production, have been described for over 40 years [97,94]. Anabolic-androgenic steroids are drugs frequently used by athletes, amateur athletes, and bodybuilders to improve sports performance [94]. Anabolic-androgenic steroids suppress gonadotropin release from the pituitary gland by a negative feedback mechanism that results in down-regulation of gonadotropins and decreased secretion of testosterone [98].

It has also been demonstrated that nicotine and its metabolites inhibit multiple steps in testosterone biosynthesis [94]. Chronic administration of nicotine in male rats leads to a reduction in testosterone levels [99]. The decreased testosterone levels seem to return to physiological levels after nicotine cessation, indicating the potentially reversible effects of nicotine on Leydig cell function [100]. Chronic, intensive marijuana usage in humans has been associated with decreased serum testosterone levels in a dose-dependent manner [69].

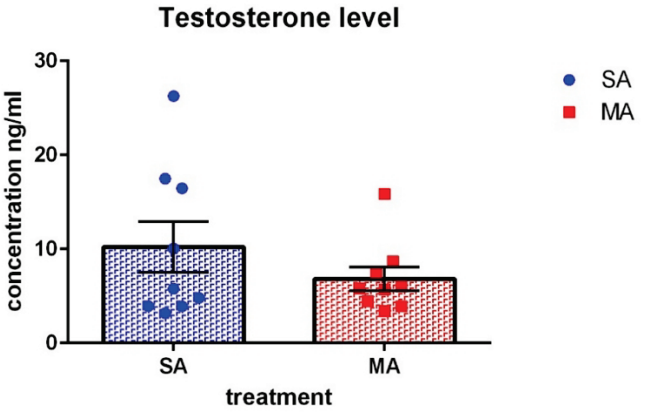
However, a recent population study on 1500 men indicates no differences in serum testosterone levels among marijuana users compared to non-users [101]. Surprisingly, the chronic administration of cocaine to rats (15 mg/kg for 100 days) did not induce changes in testosterone levels [61]. However, in another study, male Wistar rats receiving low doses, i.p. injections of cocaine, showed an increase in testosterone concentration, but the same effect was not demonstrated with high doses of cocaine [102].

In vivo and *in vitro* studies have demonstrated that amphetamine exposure decreases testosterone concentrations [103]. As a matter of MA, the results are very inconsistent. Animal study by Lin *et al.* showed that chronic administration of MA for 15, 30, 60, and 90 days significantly decreased total testosterone secretion compared to the control treatment [79]. Also, other researchers reported that illicit use of MA decreased plasma testosterone concentrations [78]. However, there are also studies showing the opposite. A study by Heidari-Rarani *et al.* showed that 14 days of MA increased serum testosterone levels in adult male rats [104]. Furthermore, a study by Yamamoto *et al.* demonstrated that serum testosterone concentrations showed a biphasic change after MA exposure in mice [105]. An initial significant decrease was followed by an increase, which 48 h after drug injection showed testosterone levels higher than the control group. In contrast with previous studies, our unpublished data demonstrated that chronic MA exposure did not influence testosterone concentrations compared to the saline controls. Furthermore, it is of interest that our raw testosterone data showed differences in measured values of testosterone concentration among the same treatment group. We suggest that dominance-subordinate relationships between male rats could influence these differences. During the MA application period (30 days), male rats were housed 2 per cage. It has been demonstrated that adult male rats living together form dominance relationships, with one dominant and the others adopting subordinate roles [106]. Animal studies on rats demonstrate that testosterone plays a primary role in intermale social aggression and dominant behavior and that castration, thus the loss of testosterone, is typically accompanied by a loss of social dominance. Therefore, we suggest that the variety of measured testosterone levels shown in Figure 3 could have been influenced by animal hierarchy [107].

The above-mentioned studies present very controversial findings about the effect of drugs administered to male rats and the effect on their reproductive system and sexual behavior. We suggest that more detailed studies are needed to thoroughly investigate dose-dependent responses and other factors that may play a role in the possible effect of MA on male reproductive performance. An overview of the studies investigating the effect of drugs on male rats is summarized in Table 1.

Effect of paternal drug exposure on offspring

There are only few studies examining the impact of paternal drug exposure on rat pup development and behavior in adulthood. A study by Abel *et al.* showed that paternal cocaine administration leads to increased hyperactivity and behavioral changes in rat pups [108]. Other studies report that paternal cocaine exposure affects the birth weight of offspring, with birth weights being decreased, increased, or unchanged [108,109,61,110]. Relative to neurocognitive outcomes, a study by Le *et al.* showed that the offspring of addicted male rats had



Concentration of Testosterone [ng/ml]	
SA	MA
3.198	3.406
10.064	15.844
4.801	5.783
5.783	8.694
17.486	3.906
3.938	4.459
16.456	5.709
26.25	6.004
3.924	7.553

Fig. 3. Chronic MA exposure did not result into significant changes of testosterone levels in adult male rats. MA=methamphetamine, SA=saline. Values are mean ± SEM (n[MA]=9, n[SA]=9).

Table 1. Effect of drugs on male rats.

Effect of drugs on male rats	Treatment	Number of individuals	Effect of drug	Publication
Locomotor activity	Methamphetamine	[MA]=8, [SA]=8	Acute MA↑ Chronic MA↓	Mihalčíková *et al.*
	Amphetamine	[AMP]=8, [SA]=8	Acute AMP↑ Chronic AMP↓	Segal and Mandell
Sexual behavior	Methamphetamine	[MA]=8, [SA]=8	No effect	Mihalčíková *et al.*
	Methamphetamine	[MA]=8, [SA]=8	No effect	Frohmader *et al.*
	Methamphetamine	[MA]=27, [SA]=26	↓	Kuiper *et al.*
Spermatogenesis	Cocaine	[COC]=33, [SA]=16	↓	George *et al.*
	Alcohol	n=13	↓	Bielawski *et al.*
	Opioids	n=15	↓	Abdellatief *et al.*
	Nicotine	n=10	↓	Condorelli *et al.*
	THC	n=6	↓	Lewis *et al.*
	Methamphetamine	[MA]=6, [SA]=6	No effect	Mihalčíková *et al.*
	Methamphetamine	[MA]=7, [SA]=7	↓	Alavi *et al.*
	Methamphetamine	[MA]=20, [SA]=20	↓	Saberi *et al.*
Testosterone	Alcohol	n=40	↓	Van Thiel *et al.*
	Amphetamine	n=7	↓	Tsai *et al.*
	Marijuana	n=20	↓	Kolodny *et al.*
	Methamphetamine	[MA]=32, [SA]=32	↓	Lin *et al.*
	Methamphetamine	[MA]=9, [SA]=9	No effect	Mihalčíková *et al.*

increased cocaine self-administration [111]. Studies by Bielawski *et al.* found that paternal alcohol abuse results in offspring malformations and reduced fetal weight [65]. Another study by Dalterio *et al.* showed that paternal THC (delta-9-tetrahydrocannabinol) exposure significantly impairs the development of rat pups [112]. On the other hand, a study by Levin *et al.* demonstrated that paternal THC exposure does not significantly impact the clinical health of rat offspring, including litter size, sex ratio, pup birth weight, survival, and growth. However, it results in neurocognitive alterations with increased habituations of locomotor activity and decreased attentional function of offspring in adulthood [113].

The above few studies present very conflicting findings about the effect of different drugs administered to male rats and the effect on their offspring. However, what

is the effect of MA is not yet known. Our recent study was the first examining the effect of paternal MA exposure on development and behavior of rat offspring [114].

Behavioral experiments during development

Our recent data did not show any significant effects of paternal MA exposure on sensorimotor development of pups [114] indicating that paternal MA administration does not result in such a severe impairment of offspring development compared to paternal cocaine and cannabinoid exposure as shown in studies of Abel *et al.*, George *et al.*, Dalterio *et al.* [108,61,112]. Moreover, these data showing no paternal MA effect are in contrast to the effects induced by maternal MA administration, which showed that after maternal MA exposure, the surface righting reflex (on PD 1-5) was slowed [115,116], the Negative geotaxis on PD 9 was unchanged [116], and performance on the Rotarod, but not Bar holding, was impaired on PD 23. Thus, it seems paternal MA exposure does not influence the sensorimotor development of rat pups, as does maternal MA exposure. The explanation may be that while maternal exposure can directly affect the development of pups (since MA crosses the placenta and enters breast milk during lactation) [10,32], paternal exposure would need to change the genetics of the pup, which does not appear to occur.

Effect of paternal drug exposure on the locomotor activity of offspring

Regarding the effect of paternal drug exposure on locomotor activity of their offspring, previous animal studies are inconsistent in their outcomes. A study by Killinger *et al.* showed that spontaneous locomotor activity after paternal cocaine exposure was unaffected [110]. A study by Fisher *et al.* reported increased locomotor activity after paternal cocaine exposure [109], and a study by Levin *et al.* showed that paternal THC treatment does not affect the spontaneous locomotion of offspring [113]. However, it increases habituation during locomotor activity. In addition, another study with paternal alcohol exposure showed that the locomotor activity of rat offspring increased by 30 % compared to controls [117]. The inconsistent outcomes of previous studies could be the result of different experimental conditions as well as related to varying drug doses, methods of administration, and duration of the exposure period. Our previous results indicate that paternal MA exposure does not affect the locomotor activity and exploratory behavior of offspring in adulthood. Our findings agree with previous studies [21] showing that maternal MA exposure during gestation did not affect the baseline level of locomotor activity in adult offspring, while acute MA treatment increased it [33,34]. The increased overall activity in the Laboras test (locomotor activity test), induced by an acute MA application of 1 mg/kg, was mainly associated with increased levels of dopamine, especially in the nucleus accumbens [37]. An overall result of studies investigating the effect of paternal drug exposure on offspring is summarized in Table 2.

Table 2. Effect of paternal drug exposure on offspring.

Effect of paternal drug exposure on offspring	Treatment	Number of individuals	Effect of drug	Publication
Sensorimotor development	Methamphetamine	n[SA]=109, n[MA]=124	No effect	Mihalčíková *et al.*
Locomotor activity	Cocaine	n[COC]=134, n[SA]=23	No effect	Killinger *et al*.
	Cocaine	n[COC]=8, n[SA]=9	↑	Fisher *et al.*
	THC	n[THC]=8, n[SA]=9	No effect	Levin *et al.*
	Alcohol	n=10	↑	Ledig *et al.*
	Methamphetamine	[MA]=8, [SA]=8	No effect	Mihalčíková *et al.*

Conclusions

Much has been published about the adverse long-term effects of maternal drug abuse and its impact on their offspring. However, the father's role as parent and donor of half of the genetic information is unclear. While some studies testing the effect of alcohol and nicotine, but also the effect of some drugs such as cocaine or THC show an effect on male sexual behavior and the effect of paternal exposure on offspring, our results

testing the effects of MA did not show such a substantial effect. Although we found no significant effects of paternal exposure to MA and its effect on offspring, our study was significant in that it was one of the first to investigate whether paternal exposure to MA has similar adverse effects on offspring development as maternal exposure to MA.

Conflict of Interest

There is no conflict of interest.

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References

1. Frost DO, Cadet JL. Effects of methamphetamine-induced neurotoxicity on the development of neural circuitry: a hypothesis. Brain Res Brain Res Rev 2000;34:103-118. https://doi.org/10.1016/S0165-0173(00)00042-4
2. Logan BK. Methamphetamine - Effects on Human Performance and Behavior. Forensic Sci Rev 2002;14:133-151.
3. Panenka WJ, Procyshyn RM, Lecomte T, MacEwan GW, Flynn SW, Honer WG, Barr AM. Methamphetamine use: a comprehensive review of molecular, preclinical and clinical findings. Drug Alcohol Depend 2013;129:167-179. https://doi.org/10.1016/j.drugalcdep.2012.11.016
4. EMCDDA EM and ADDICTION CFDAD. Statistical bulletin 2009. Lisbon.
5. Mravčík V, Vorel F, Zábranský T. Drugs and fatal traffic accidents in the Czech Republic. Cent Eur J Public Health 2007;15:158-162. https://doi.org/10.21101/cejph.a3429
6. Šlamberová R. Review of long-term consequences of maternal methamphetamine exposure. Physiol Res 2019;68(Suppl 3):S219-S231. https://doi.org/10.33549/physiolres.934360
7. Šlamberová R, Macúchová E, Nohejlová-Deykun K, Schutová B, Hrubá L, Rokyta R. Gender differences in the effect of prenatal methamphetamine exposure and challenge dose of other drugs on behavior of adult rats. Physiol Res 2013;62(Suppl 1):S99-S108. https://doi.org/10.33549/physiolres.932593
8. Holubová A, Poništ S, Jurčovičová J, Šlamberová R. Different Oxytocin Responses to Acute Methamphetamine Treatment in Juvenile Female Rats Perinatally Exposed to Stress and/or Methamphetamine Administration. Front Physiol 2019;10:305. https://doi.org/10.3389/fphys.2019.00305
9. Marwick C. NIDA seeking data on effect of fetal exposure to methamphetamine. JAMA 2000;283:2225-2226. https://doi.org/10.1001/jama.283.17.2225-JMN0503-2-1
10. Dattel BJ. Substance abuse in pregnancy. Semin Perinatol 1990;14:179-187.
11. Smith LM, Chang L, Yonekura ML, Grob C, Osborn D, Ernst T. Brain proton magnetic resonance spectroscopy in children exposed to methamphetamine in utero. Neurology 2001;57:255-260. https://doi.org/10.1212/WNL.57.2.255
12. Abar B, LaGasse LL, Wouldes T, Derauf C, Newman E, Shah R, Smith LM, Arria AM, Huestis MA, DellaGrotta S, Dansereau LM, Wilcox T, Neal CR, Lester BM. Cross-national comparison of prenatal methamphetamine exposure on infant and early child physical growth: a natural experiment. Prev Sci 2014;15:767-776. https://doi.org/10.1007/s11121-013-0431-5
13. Acuff-Smith KD, Schilling MA, Fisher JE, Vorhees CV. Stage-specific effects of prenatal d-methamphetamine exposure on behavioral and eye development in rats. Neurotoxicol Teratol 1996;18:199-215. https://doi.org/10.1016/0892-0362(95)02015-2
14. Hrubá L, Schutová B, Šlamberová R, Pometlová M, Rokyta R. Effect of methamphetamine exposure and cross-fostering on sensorimotor development of male and female rat pups. Dev Psychobiol 2009;51:73-83. https://doi.org/10.1002/dev.20346
15. Šlamberová R, Pometlová M, Charousová P. Postnatal development of rat pups is altered by prenatal methamphetamine exposure. Prog Neuropsychopharmacol Biol Psychiatry 2006;30:82-88. https://doi.org/10.1016/j.pnpbp.2005.06.006
16. Ševčíková M, Petríková I, Šlamberová R. Methamphetamine exposure during the first, but not the second half of prenatal development, affects social play behavior. Physiol Res 2020;69:319-330. https://doi.org/10.33549/physiolres.934230

17. Macúchová E, Nohejlová-Deykun K, Šlamberová R. H.2-Does prenatal and adult methamphetamine exposure affect spatial learning of female rats? Behav Pharmacol 2013;24:e60-e61. https://doi.org/10.1097/01.fbp.0000434881.23436.53
18. Šlamberová R, Schutová B, Matejovská I, Bernášková K, Rokyta R. Effects of a single postnatal methamphetamine administration on NMDA-induced seizures are sex- and prenatal exposure-specific. Naunyn Schmiedebergs Arch Pharmacol 2009;380:109-114. https://doi.org/10.1007/s00210-009-0427-7
19. Hrebíčková I, Ševčíková M, Macúchová E, Šlamberová R. How methamphetamine exposure during different neurodevelopmental stages affects social behavior of adult rats? Physiol Behav 2017;179:391-400. https://doi.org/10.1016/j.physbeh.2017.07.009
20. Macúchová E, Nohejlová K, Ševčíková M, Hrebíčková I, Šlamberová R. Sex differences in the strategies of spatial learning in prenatally-exposed rats treated with various drugs in adulthood. Behav Brain Res 2017;327:83-93. https://doi.org/10.1016/j.bbr.2017.03.041
21. Šlamberová R, Macúchová E, Nohejlová K, Štofková A, Jurčovičová J. Effect of amphetamine on adult male and female rats prenatally exposed to methamphetamine. Prague Med Rep 2014;115:43-59. https://doi.org/10.14712/23362936.2014.5
22. Schutová B, Hrubá L, Pometlová M, Deykun K, Šlamberová R. Cognitive functions and drug sensitivity in adult male rats prenatally exposed to methamphetamine. Physiol Res 2009;58:741-750. https://doi.org/10.33549/physiolres.931562
23. Macúchová E, Šlamberová R. Does prenatal methamphetamine exposure sensitization to drugs in adulthood? Physiol Res 2017;66(Suppl 4):S457-S467. https://doi.org/10.33549/physiolres.933803
24. Yamamotová A, Hrubá L, Schutová B, Rokyta R, Šlamberová R. Perinatal effect of methamphetamine on nociception in adult Wistar rats. Int J Dev Neurosci 2011;29:85-92. https://doi.org/10.1016/j.ijdevneu.2010.08.004
25. LaGasse LL, Wouldes T, Newman E, Smith LM, Shah R, Derauf C, Huestis MA, ET AL. Prenatal methamphetamine exposure and neonatal neurobehavioral outcome in the USA and New Zealand. Neurotoxicol Teratol 2011;33:166-175. https://doi.org/10.1016/j.ntt.2010.06.009
26. Smith LM, Lagasse LL, Derauf C, Grant P, Shah R, Arria A, Huestis M, ET AL. Prenatal methamphetamine use and neonatal neurobehavioral outcome. Neurotoxicol Teratol 2008;30:20-28. https://doi.org/10.1016/j.ntt.2007.09.005
27. Holubová A, Štofková A, Jurčovičová J, Šlamberová R. The effect of neonatal maternal stress on plasma levels of adrenocorticotropic hormone, corticosterone, leptin, and ghrelin in adult male rats exposed to acute heterotypic stressor. Physiol Res 2016;65(Suppl 5):S557-S566. https://doi.org/10.33549/physiolres.933530
28. Hol T, Van den Berg CL, Van Ree JM, Spruijt BM. Isolation during the play period in infancy decreases adult social interactions in rats. Behav Brain Res 1999;100:91-97. https://doi.org/10.1016/S0166-4328(98)00116-8
29. Šlamberová R, Charousová P, Pometlová M. Maternal behavior is impaired by methamphetamine administered during pre-mating, gestation and lactation. Reprod Toxicol 2005;20:103-110. https://doi.org/10.1016/j.reprotox.2004.11.010
30. Šlamberová R. Drugs in pregnancy: the effects on mother and her progeny. Physiol Res 2012;61(Suppl 1):S123-S135. https://doi.org/10.33549/physiolres.932357
31. Zábranský T. Methamphetamine in the Czech Republic. J Drug Issues 2007;37:155-180. https://doi.org/10.1177/002204260703700108
32. Rambousek L, Kačer P, Syslová K, Bumba J, Bubeníková-Valešová V, Šlamberova R. Sex differences in methamphetamine pharmacokinetics in adult rats and its transfer to pups through the placental membrane and breast milk. Drug Alcohol Depend 2014;139:138-144. https://doi.org/10.1016/j.drugalcdep.2014.03.023
33. Glatt SJ, Bolaños CA, Trksak GH, Crowder-Dupont C, Jackson D. Prenatal cocaine exposure alters behavioral and neurochemical sensitization to amphetamine in adult rats. Neuropharmacology 2000;39:599-610. https://doi.org/10.1016/S0028-3908(99)00181-1
34. Hall DA, Stanis JJ, Marquez Avila H, Gulley JM. A comparison of amphetamine- and methamphetamine-induced locomotor activity in rats: evidence for qualitative differences in behavior. Psychopharmacology (Berl) 2008;195:469-478. https://doi.org/10.1007/s00213-007-0923-8
35. Schutová B, Hrubá L, Rokyta R, Šlamberová R. Gender differences in behavioral changes elicited by prenatal methamphetamine exposure and application of the same drug in adulthood. Dev Psychobiol 2013;55:232-242. https://doi.org/10.1002/dev.21016

36. Mihalčíková L, Ochozková A, Šlamberová R. Effect of methamphetamine exposure on sexual behavior and locomotor activity of adult male rats. Physiol Res 2019;68(Suppl 3):S339-S346. https://doi.org/10.33549/physiolres.934357
37. Bubeníková-Valešová V, Kačer P, Syslová K, Rambousek L, Janovský M, Schutová B, Hrubá L, Šlamberová R. Prenatal methamphetamine exposure affects the mesolimbic dopaminergic system and behavior in adult offspring. Int J Dev Neurosci 2009;27:525-530. https://doi.org/10.1016/j.ijdevneu.2009.06.012
38. Kelly PH, Iversen SD. Selective 6OHDA-induced destruction of mesolimbic dopamine neurons: abolition of psychostimulant-induced locomotor activity in rats. Eur J Pharmacol 1976;40:45-56. https://doi.org/10.1016/0014-2999(76)90352-6
39. Lucot JB, Wagner GC, Schuster CR, Seiden LS. The effects of dopaminergic agents on the locomotor activity of rats after high doses of methylamphetamine. Pharmacol Biochem Behav 1980;13:409-413. https://doi.org/10.1016/0091-3057(80)90247-6
40. Wallace TL, Gudelsky GA, Vorhees CV. Methamphetamine-induced neurotoxicity alters locomotor activity, stereotypic behavior, and stimulated dopamine release in the rat. J Neurosci 1999;19:9141-9148. https://doi.org/10.1523/JNEUROSCI.19-20-09141.1999
41. Rivière GJ, Byrnes KA, Gentry WB, Owens SM. Spontaneous locomotor activity and pharmacokinetics of intravenous methamphetamine and its metabolite amphetamine in the rat. J Pharmacol Exp Ther 1999;291:1220-1226.
42. Kelly PH, Seviour PW, Iversen SD. Amphetamine and apomorphine responses in the rat following 6-OHDA lesions of the nucleus accumbens septi and corpus striatum. Brain Res 1975;94:507-522. https://doi.org/10.1016/0006-8993(75)90233-4
43. Cho AK, Segal DS. *Amphetamine and its Analogs: Psychopharmacology, Toxicology, and Abus*e. Academic Press, 1994, 503 p.
44. Schindler CW, Bross JG, Thorndike EB. Gender differences in the behavioral effects of methamphetamine. Eur J Pharmacol 2002;442:231-235. https://doi.org/10.1016/S0014-2999(02)01550-9
45. Zakharova E, Leoni G, Kichko I, Izenwasser S. Differential effects of methamphetamine and cocaine on conditioned place preference and locomotor activity in adult and adolescent male rats. Behav Brain Res 2009;198:45-50. https://doi.org/10.1016/j.bbr.2008.10.019
46. Segal DS, Mandell AJ. Long-term administration of d-amphetamine: progressive augmentation of motor activity and stereotypy. Pharmacol Biochem Behav 1974;2:249-255. https://doi.org/10.1016/0091-3057(74)90060-4
47. Nazari A, Perez-Fernandez C, Flores P, Moreno M, Sánchez-Santed F. Age-dependent effects of repeated methamphetamine exposure on locomotor activity and attentional function in rats. Pharmacol Biochem Behav 2020;191:172879. https://doi.org/10.1016/j.pbb.2020.172879
48. Rothman RB, Baumann MH, Dersch CM, Romero DV, Rice KC, Carroll FI, Partilla JS. Amphetamine-type central nervous system stimulants release norepinephrine more potently than they release dopamine and serotonin. Synapse 2001;39:32-41. https://doi.org/10.1002/1098-2396(20010101)39:1<32::AID-SYN5>3.0.CO;2-3
49. Kish SJ, Fitzmaurice PS, Boileau I, Schmunk GA, Ang LC, Furukawa Y, Chang LJ, ET AL. Brain serotonin transporter in human methamphetamine users. Psychopharmacology (Berl) 2009;202:649-661. https://doi.org/10.1007/s00213-008-1346-x
50. Hull EM, Muschamp JW, Sato S. Dopamine and serotonin: influences on male sexual behavior. Physiol Behav 2004;83:291-307. https://doi.org/10.1016/j.physbeh.2004.08.018
51. Semple SJ, Grant I, Patterson TL. Female methamphetamine users: social characteristics and sexual risk behavior. Women Health 2004;40:35-50. https://doi.org/10.1300/J013v40n03_03
52. Winland C, Haycox C, Bolton JL, Jampana S, Oakley BJ, Ford B, Ornelas L, ET AL. Methamphetamine enhances sexual behavior in female rats. Pharmacol Biochem Behav 2011;98:575-582. https://doi.org/10.1016/j.pbb.2011.03.001
53. Frohmader KS, Bateman KL, Lehman MN, Coolen LM. Effects of methamphetamine on sexual performance and compulsive sex behavior in male rats. Psychopharmacology (Berl) 2010;212:93-104. https://doi.org/10.1007/s00213-010-1930-8

54. Fiorino DF, Phillips AG. Facilitation of sexual behavior and enhanced dopamine efflux in the nucleus accumbens of male rats after D-amphetamine-induced behavioral sensitization. J Neurosci 1999;19:456-463. https://doi.org/10.1523/JNEUROSCI.19-01-00456.1999
55. Rawson RA, Washton A, Domier CP, Reiber C. Drugs and sexual effects: role of drug type and gender. J Subst Abuse Treat 2002;22:103-108. https://doi.org/10.1016/S0740-5472(01)00215-X
56. Bolin BL, Akins CK. Methamphetamine impairs sexual motivation but not sexual performance in male Japanese quail. Exp Clin Psychopharmacol 2009;17:10-20. https://doi.org/10.1037/a0014505
57. Kuiper LB, Frohmader KS, Coolen LM. Maladaptive Sexual Behavior Following Concurrent Methamphetamine and Sexual Experience in Male Rats is Associated with Altered Neural Activity in Frontal Cortex. Neuropsychopharmacology 2017;42:2011-2020. https://doi.org/10.1038/npp.2017.1
58. Frohmader KS, Lehman MN, Laviolette SR, Coolen LM. Concurrent exposure to methamphetamine and sexual behavior enhances subsequent drug reward and causes compulsive sexual behavior in male rats. J Neurosci 2011;31:16473-16482. https://doi.org/10.1523/JNEUROSCI.4013-11.2011
59. Kempinas WG, Lamano-Carvalho TL. A method for estimating the concentration of spermatozoa in the rat cauda epididymidis. Lab Anim 1988;22:154-156. https://doi.org/10.1258/002367788780864547
60. Verstegen RHJ, Wang G, Langenberg-Ververgaert KPS, Ren LY, Nulman I. Paternal exposure to recreational drugs before conception and its effect on live-born offspring: A scoping review. Birth Defects Res 2020;112:970-988. https://doi.org/10.1002/bdr2.1702
61. George VK, Li H, Teloken C, Grignon DJ, Lawrence WD, Dhabuwala CB. Effects of long-term cocaine exposure on spermatogenesis and fertility in peripubertal male rats. J Urol 1996;155:327-331. https://doi.org/10.1016/S0022-5347(01)66654-0
62. Li H, Jiang Y, Rajpurkar A, Dunbar JC, Dhabuwala CB. Cocaine induced apoptosis in rat testes. J Urol 1999;162:213-216. https://doi.org/10.1097/00005392-199907000-00070
63. Zhang H, Loughlin KR. The effect of cocaine and its metabolites on Sertoli cell function. J Urol 1996;155:163-166. https://doi.org/10.1016/S0022-5347(01)66583-2
64. Van Thiel DH, Gavaler JS, Cobb CF, Santucci L, Graham TO. Ethanol, a Leydig cell toxin: evidence obtained in vivo and in vitro. Pharmacol Biochem Behav 1983;(Suppl 1):317-323. https://doi.org/10.1016/0091-3057(83)90193-4
65. Bielawski DM, Abel EL. Acute treatment of paternal alcohol exposure produces malformations in offspring. Alcohol 1997;14:397-401. https://doi.org/10.1016/S0741-8329(97)87951-7
66. Abdellatief RB, Elgamal DA, Mohamed EE. Effects of chronic tramadol administration on testicular tissue in rats: an experimental study. Andrologia 2015;47:674-679. https://doi.org/10.1111/and.12316
67. Agirregoitia E, Valdivia A, Carracedo A, Casis L, Gil J, Subiran N, Ochoa C, Irazusta J. Expression and localization of delta-, kappa-, and mu-opioid receptors in human spermatozoa and implications for sperm motility. J Clin Endocrinol Metab 2006;91:4969-4975. https://doi.org/10.1210/jc.2006-0599
68. Condorelli RA, La Vignera S, Giacone F, Iacoviello L, Vicari E, Mongioi L, Calogero AE. In vitro effects of nicotine on sperm motility and bio-functional flow cytometry sperm parameters. Int J Immunopathol Pharmacol 2013;26:739-746. https://doi.org/10.1177/039463201302600317
69. Kolodny RC, Masters WH, Kolodner RM, Toro G. Depression of plasma testosterone levels after chronic intensive marihuana use. N Engl J Med 1974;290:872-874. https://doi.org/10.1056/NEJM197404182901602
70. Schuel H, Burkman LJ, Lippes J, Crickard L, Mahony MC, Giuffrida A, Picone RP, Makriyannis A. Evidence that anandamide-signaling regulates human sperm functions required for fertilization. Mol Reprod Dev 2002;63:376-387. https://doi.org/10.1002/mrd.90021
71. Rossato M, Ion Popa F, Ferigo M, Clari G, Foresta C. Human sperm express cannabinoid receptor Cb1, the activation of which inhibits motility, acrosome reaction, and mitochondrial function. J Clin Endocrinol Metab 2005;90:984-991. https://doi.org/10.1210/jc.2004-1287
72. Whan LB, West MC, McClure N, Lewis SE. Effects of delta-9-tetrahydrocannabinol, the primary psychoactive cannabinoid in marijuana, on human sperm function in vitro. Fertil Steril 2006;85:653-660. https://doi.org/10.1016/j.fertnstert.2005.08.027

73. Lewis SE, Paro R, Borriello L, Simon L, Robinson L, Dincer Z, Riedel G, Battista N, Maccarrone M. Long-term use of HU210 adversely affects spermatogenesis in rats by modulating the endocannabinoid system. Int J Androl 2012;35:731-740. https://doi.org/10.1111/j.1365-2605.2012.01259.x
74. Alavi SH, Taghavi MM, Moallem SA. Evaluation of effects of methamphetamine repeated dosing on proliferation and apoptosis of rat germ cells. Syst Biol Reprod Med 2008;54:85-91. https://doi.org/10.1080/19396360801952078
75. Saberi A, Sepehri G, Safi Z, Safi Z, Razavi B, Jahandari F, Divsalar K, Salarkia E. Effects of Methamphetamine on Testes Histopathology and Spermatogenesis Indices of Adult Male Rats. Addict Health 2017;9:199-205.
76. Montagnini BG, Silva LS, dos Santos AH, Anselmo-Franci JA, Fernandes GS, Mesquita SdeF, Gerardin DC. Effects of repeated administration of methylphenidate on reproductive parameters in male rats. Physiol Behav 2014;133:122-129. https://doi.org/10.1016/j.physbeh.2014.05.016
77. González CR, González B, Matzkin ME, Muñiz JA, Cadet JL, Garcia-Rill E, Urbano FJ, Vitullo AD, Bisagno V. Psychostimulant-Induced Testicular Toxicity in Mice: Evidence of Cocaine and Caffeine Effects on the Local Dopaminergic System. PLoS One 2015;10:e0142713. https://doi.org/10.1371/journal.pone.0142713
78. Nudmamud-Thanoi S, Thanoi S. Methamphetamine induces abnormal sperm morphology, low sperm concentration and apoptosis in the testis of male rats. Andrologia 2011;43:278-282. https://doi.org/10.1111/j.1439-0272.2010.01071.x
79. Lin JF, Lin YH, Liao PC, Lin YC, Tsai TF, Chou KY, Chen HE, Tsai SC, Hwang TI. Induction of testicular damage by daily methamphetamine administration in rats. Chin J Physiol 2014;57:19-30. https://doi.org/10.4077/CJP.2014.BAB155
80. Kaewman P, Nudmamud-Thanoi S, Thanoi S. GABAergic Alterations in the Rat Testis after Methamphetamine Exposure. Int J Med Sci 2018;15:1349-1354. https://doi.org/10.7150/ijms.27609
81. Sabour M, Khoradmehr A, Kalantar SM, Danafar AH, Omidi M, Halvaei I, Nabi A, Ghasemi-Esmailabad S, Talebi AR. Administration of high dose of methamphetamine has detrimental effects on sperm parameters and DNA integrity in mice. Int J Reprod Biomed 2017;15:161-168. https://doi.org/10.29252/ijrm.15.3.161
82. McGrady AV. Effects of psychological stress on male reproduction: a review. Arch Androl 1984;13:1-7. https://doi.org/10.3109/01485018408987495
83. Drude S, Geissler A, Olfe J, Starke A, Domanska A, Schuett C, Kiank-Nussbaum C. Side effects of control treatment can conceal experimental data when studying stress responses to injection and psychological stress in mice. Lab Anim (NY) 2011;40:119-128. https://doi.org/10.1038/laban0411-119
84. Shulman LM, Spritzer MD. Changes in the sexual behavior and testosterone levels of male rats in response to daily interactions with estrus females. Physiol Behav 2014;133:8-13. https://doi.org/10.1016/j.physbeh.2014.05.001
85. Nyby JG. Reflexive testosterone release: a model system for studying the nongenomic effects of testosterone upon male behavior. Front Neuroendocrinol 2008;29:199-210. https://doi.org/10.1016/j.yfrne.2007.09.001
86. Pfaus JG, Kippin TE, Centeno S. Conditioning and sexual behavior: a review. Horm Behav 2001;40:291-321. https://doi.org/10.1006/hbeh.2001.1686
87. Lord C, Sekerovic Z, Carrier J. Sleep regulation and sex hormones exposure in men and women across adulthood. Pathol Biol (Paris) 2014;62:302-310. https://doi.org/10.1016/j.patbio.2014.07.005
88. Basaria S. Reproductive aging in men. Endocrinol Metab Clin North Am 2013;42:255-270. https://doi.org/10.1016/j.ecl.2013.02.012
89. Celec P, Ostatníková D, Hodosy J. On the effects of testosterone on brain behavioral functions. Front Neurosci 2015;9:12. https://doi.org/10.3389/fnins.2015.00012
90. Hull EM, Dominguez JM. Sexual behavior in male rodents. Horm Behav 2007;52:45-55. https://doi.org/10.1016/j.yhbeh.2007.03.030
91. Falvo RE, Kaltenbach CC, Pancoe WL. Determination of testosterone concentration in the plasma of normal and androgen-sterilized female rats, using a competitive protein binding technique. Neuroendocrinology 1972;10:229-234. https://doi.org/10.1159/000122092
92. Lee VW, de Kretser DM, Hudson B, Wang C. Variations in serum FSH, LH and testosterone levels in male rats from birth to sexual maturity. J Reprod Fertil 1975;42:121-126. https://doi.org/10.1530/jrf.0.0420121
93. Heywood LH. Testosterone levels in the male laboratory rat: variation under experimental conditions. Int J Androl 1980;3:519-529. https://doi.org/10.1111/j.1365-2605.1980.tb00140.x

94. Duca Y, Aversa A, Condorelli RA, Calogero AE, La Vignera S. Substance Abuse and Male Hypogonadism. J Clin Med 2019;8:732. https://doi.org/10.3390/jcm8050732
95. Gordon GG, Southren AL, Vittek J, Lieber CS. The effect of alcohol ingestion on hepatic aromatase activity and plasma steroid hormones in the rat. Metabolism 1979;28:20-24. https://doi.org/10.1016/0026-0495(79)90163-X
96. Hsieh A, DiGiorgio L, Fakunle M, Sadeghi-Nejad H. Management Strategies in Opioid Abuse and Sexual Dysfunction: A Review of Opioid-Induced Androgen Deficiency. Sex Med Rev 2018;6:618-623. https://doi.org/10.1016/j.sxmr.2018.04.003
97. Fronczak CM, Kim ED, Barqawi AB. The insults of illicit drug use on male fertility. J Androl 2012;33:515-528. https://doi.org/10.2164/jandrol.110.011874
98. La Vignera S, Condorelli RA, Cannarella R, Duca Y, Calogero AE. Sport, doping and female fertility. Reprod Biol Endocrinol 2018;16:108. https://doi.org/10.1186/s12958-018-0437-8
99. Kavitharaj NK, Vijayammal PL. Nicotine administration induced changes in the gonadal functions in male rats. Pharmacology 1999;58:2-7. https://doi.org/10.1159/000028262
100. Oyeyipo IP, Raji Y, Bolarinwa AF. Nicotine alters male reproductive hormones in male albino rats: The role of cessation. J Hum Reprod Sci 2013;6:40-44. https://doi.org/10.4103/0974-1208.112380
101. Thistle JE, Graubard BI, Braunlin M, Vesper H, Trabert B, Cook MB, McGlynn KA. Marijuana use and serum testosterone concentrations among U.S. males. Andrology 2017;5:732-738. https://doi.org/10.1111/andr.12358
102. Rodriguez MC, Sanchez-Yague J, Paniagua R. Effects of cocaine on testicular structure in the rat. Reprod Toxicol 1992;6:51-55. Erratum in Reprod Toxicol 1992;6:283. https://doi.org/10.1016/0890-6238(92)90020-T
103. Tsai SC, Chiao YC, Lu CC, Doong ML, Chen YH, Shih HC, Liaw C, Wang SW, Wang PS. Inhibition by amphetamine of testosterone secretion through a mechanism involving an increase of cyclic AMP production in rat testes. Br J Pharmacol 1996;118:984-988. https://doi.org/10.1111/j.1476-5381.1996.tb15496.x
104. Heidari-Rarani M, Noori A, Ghodousi A. Effects of methamphetamine on pituitary gonadal axis and spermatogenesis in mature male rats. Zahedan J Res Med Sci 2014;16:37-42.
105. Yamamoto Y, Yamamoto K, Hayase T. Effect of methamphetamine on male mice fertility. J Obstet Gynaecol Res 1999;25:353-358. https://doi.org/10.1111/j.1447-0756.1999.tb01176.x
106. Pellis SM, Pellis VC, McKenna MM. Some subordinates are more equal than others: Play fihgting amongst adult subordinate male rats. Aggress Behav 1993;19:385-393. https://doi.org/10.1002/1098-2337(1993)19:5<385::AID-AB2480190508>3.0.CO;2-#
107. Albert DJ, Walsh ML, Gorzalka BB, Siemens Y, Louie H. Testosterone removal in rats results in a decrease in social aggression and a loss of social dominance. Physiol Behav 1986;36:401-407. https://doi.org/10.1016/0031-9384(86)90305-7
108. Abel EL. Paternal behavioral mutagenesis. Neurotoxicology 1989;10:335-345. https://doi.org/10.1016/0892-0362(88)90016-5
109. Fischer DK, Rice RC, Martinez Rivera A, Donohoe M, Rajadhyaksha AM. Altered reward sensitivity in female offspring of cocaine-exposed fathers. Behav Brain Res 2017;332:23-31. https://doi.org/10.1016/j.bbr.2017.05.054
110. Killinger CE, Robinson S, Stanwood GD. Subtle biobehavioral effects produced by paternal cocaine exposure. Synapse 2012;66:902-908. https://doi.org/10.1002/syn.21582
111. Le Q, Yan B, Yu X, Li Y, Song H, Zhu H, Hou W, ET AL. Drug-seeking motivation level in male rats determines offspring susceptibility or resistance to cocaine-seeking behaviour. Nat Commun 2017;8:15527. https://doi.org/10.1038/ncomms15527
112. Dalterio S, Steger R, Mayfield D, Bartke A. Early cannabinoid exposure influences neuroendocrine and reproductive functions in male mice: I. Prenatal exposure. Pharmacol Biochem Behav 1984;20:107-113. https://doi.org/10.1016/0091-3057(84)90110-2
113. Levin ED, Hawkey AB, Hall BJ, Cauley M, Slade S, Yazdani E, Kenou B, ET AL. Paternal THC exposure in rats causes long-lasting neurobehavioral effects in the offspring. Neurotoxicol Teratol 2019;74:106806. https://doi.org/10.1016/j.ntt.2019.04.003
114. Mihalčíková L, Ochozková A, Šlamberová R. Does paternal methamphetamine exposure affect the behavior of rat offspring during development and in adulthood? Physiol Res 2021;70(Suppl 3):S419-S430. https://doi.org/10.33549/physiolres.934814

115. Hrubá L, Schutová B, Šlamberová R, Pometlová M. Does cross-fostering modify the impairing effect of methamphetamine on postnatal development of rat pups? Prague Med Rep 2008;109:50-61.
116. Malinová-Ševčíková M, Hrebíčková I, Macúchová E, Nová E, Pometlová M, Šlamberová R. Differences in maternal behavior and development of their pups depend on the time of methamphetamine exposure during gestation period. Physiol Res 2014;63(Suppl 4):S559-S572. https://doi.org/10.33549/physiolres.932925
117. Ledig M, Misslin R, Vogel E, Holownia A, Copin JC, Tholey G. Paternal alcohol exposure: developmental and behavioral effects on the offspring of rats. Neuropharmacology 1998;37:57-66. https://doi.org/10.1016/S0028-3908(97)00185-8