Invited review

The impact of exposure to addictive drugs on future generations: Physiological and behavioral effects

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ABSTRACT

It is clear that both genetic and environmental factors contribute to drug addiction. Recent evidence indicating trans-generational influences of drug abuse highlight potential epigenetic factors as well. Specifically, mounting evidence suggests that parental ingestion of abused drugs influence the physiology and behavior of future generations even in the absence of prenatal exposure. The goal of this review is to describe the trans-generational consequences of preconception exposure to drugs of abuse for five major classes of drugs: alcohol, nicotine, marijuana, opioids, and cocaine. The potential epigenetic mechanisms underlying the transmission of these phenotypes across generations also are detailed. This article is part of a Special Issue entitled 'NIDA 40th Anniversary Issue'.
during pregnancy introduces (e.g. changes in nutritional status, renal function, vascular perfusion, etc.), prenatal substance use models will not be included in the current review. We will, however, include data from studies examining transgenerational effects of female drug use occurring prior to conception. Thus, the current review will examine the effects of parental exposure to drugs of abuse prior to conception on the development of subsequent generations.

2. Alcohol

Currently, alcohol is the most commonly abused drug in the United States. In 2011, the center for disease control estimated that 60% of males and 44% of females engage in chronic alcohol drinking (Edward J. Sondlik et al., 2012). While the neurobehavioral effects of fetal alcohol exposure are well described, less is known about the effects of parental exposure to alcohol prior to conception. It should be noted, however, that reports and writings as early as the 1720’s, during the so-called gin epidemic, observed that both maternal and paternal alcohol use had detrimental effects on offspring (Warner and Rosett, 1975). While few studies have examined maternal alcohol use prior to pregnancy (i.e. in the absence of prenatal use), several findings have demonstrated effects of paternal alcohol exposure on offspring development. Indeed, as early as 1913 animal studies found that offspring sired by alcohol inhaling rats demonstrated malformations, low birth weight, retarded growth and increased neonatal mortality across several generations (Friedler, 1996). Clearly the concept that alcohol use by the father, and not solely his genetic composition, can affect future progeny is not novel. The interest in paternal effects, however, has been reinvigorated by the emergence of the field of epigenetics, with a number of preclinical findings suggesting transgenerational epigenetic effects of paternal alcohol use.

Several of the initial animal studies on paternal alcohol effects focused on basic parameters of reproductive success, such as fertility and fecundity, following exposure to alcohol in peripubertal males. These studies observed a significant reduction in the number of successful pregnancies which decreased from 92% in controls to 75% in naïve females mated with alcohol-drinking sires (Emanuele et al., 2001). Litter size was also substantially reduced (Cicero et al., 1990; Emanuele et al., 2001). It was determined that alcohol exposure during puberty modified sexual maturation and resulted in decreased testes and secondary sex organ weight, eliminated the typical pubertal surge in testosterone, decreased beta-endorphin levels in the hypothalamus, and enhanced testicular oxidative injury (Cicero et al., 1990; Emanuele et al., 2001). Interestingly, offspring of these pubertal alcohol-exposed sires demonstrated similar alterations, including decreased serum testosterone levels, reduced seminal vesicle weights, and lower levels of hypothalamic beta-endorphin (Abel and Lee, 1988; Cicero et al., 1990).

Alcohol-sired offspring also demonstrate abnormalities in development. In an elegantly designed rodent study, Jamerson and colleagues revealed that paternal alcohol exposure that was ongoing, or that had ceased weeks prior to conception resulted in more rapid development of various reflexes, differences in gait, and thicker cortical layers (Jamerson et al., 2004). It was also noted that timing of alcohol exposure in relation to conception impacted the neurobehavioral effects of the offspring (Jamerson et al., 2004). For example, one study found that a single exposure to alcohol just prior to conception resulted in a significant increase in small for gestational age offspring as well as an increase in offspring demonstrating significant malformations (Bielawski and Abel, 1997). Moreover, alcohol-exposed sires also produced offspring displaying increased adrenal cortex and decreased spleen weights (Abel, 1993b). Finally, there is evidence that metabolic and immune functioning may be disrupted in alcohol-sired offspring, given that they display reductions in leptin levels (Emanuele et al., 2001) and a diminished immune response (Berk et al., 1989; Hazlett et al., 1989).

In terms of behavioral effects, both increases and decreases in activity have been noted in offspring of alcohol-consuming sires (Abel, 1989a,b; 1993a, b; Abel and Lee, 1988), with the direction of these effects mediated by a number of factors including the level of alcohol consumption, the time between exposure and conception (Jamerson et al., 2004), and the age at the time of testing (Abel, 1989a). Alterations in behavioral activity following amphetamine were also noted in male offspring (Abel, 1993a), with some evidence suggesting that increased activity was dependent on the cholinergic system (Abel, 1994). Potential modifications in the cholinergic system of alcohol-sired offspring are notable given that deficits in learning and memory have also been reported. For example, offspring sired by alcohol treated males demonstrated impairments in spatial learning (Wozniak et al., 1991) and had increased latencies to reach a choice point in a T-maze (Abel, 1994; Abel and Lee, 1988). In addition to deficits observed in males, alcohol-sired female offspring showed impaired performance in a two-way shock avoidance learning task (Abel and Tan, 1988). Finally, offspring of alcohol-consuming sires demonstrated decreased grooming, as well as decreased immobility in a forced swim test, an effect that was rescued by imipramine and propranolol and exacerbated by yohimbine and metergoline (Abel, 1991a, b; Abel and Biliztke, 1990). Together, these results indicate a detrimental behavioral phenotype of paternal alcohol consumption on both male and female offspring.

Examining epigenetic parental effects in human populations can be difficult. Human studies have, however, revealed correlations between parental drinking behavior and offspring initiation and drinking patterns. Thus, heavy paternal drinking or heavy-episodic drinking in both parents predicts earlier onset of offspring drinking as well as heavier drinking (Vermeulen-Smit et al., 2012). Children born with characteristics associated with fetal alcohol syndrome whose mother’s did not drink but fathers were alcoholics have also been observed (Lemoine et al., 2003). Additionally, there is evidence that sons of early-onset (adolescent) alcoholic fathers perform more poorly on tests of verbal intelligence and attention than late onset (adult) alcoholics (Tarter et al., 1989). While this may suggest that adolescence, or periods of significant neural and endocrine development, are particularly important in determining the effects of paternal alcohol exposure, one cannot exclude the possibility that early-onset drinking may be a marker of genetic vulnerability or additional comorbid pathologies. Finally, in addition to deficits in cognition, attention, and visuospatial capacity observed in children of alcoholic fathers, increased hyperactivity has also been noted (Goodwin et al., 1975), although the relationship between hyperactivity and paternal alcohol consumption remains equivocal (Knopik et al., 2009). Much less work has been done examining the effects of maternal preconception alcohol consumption. However, a decreased birth weight has been observed in children of alcoholic women that abstain from use during pregnancy (Little et al., 1980; Livy et al., 2004; Ramsay, 2010). While human studies remain more difficult to interpret than preclinical research due to many aspects that are impossible to control, it is important that both lines of research continue in order to fully understand the scope of preconception alcohol use and abuse.

3. Nicotine

Nicotine is the second most abused substance in the United States. In 2011 it was reported that 21.6% of adult men and 16.5% of
adult women smoke regularly (Agaku et al., 2011). It is clear from both human epidemiological and preclinical research models that exposure to tobacco smoke in utero is harmful to development and can result in low birth weight, sudden infant death syndrome, and serious behavioral issues in the offspring (Abbott and Winzer-Serhan, 2012). Examining the indirect effects (i.e. non-prenatal exposure) in humans is more challenging because it is difficult to distinguish the indirect epigenetic effects of paternal or maternal nicotine abuse on the offspring from potential exposure to second-hand smoke or post-partum. Keeping that in mind, some studies have shown increased risk of spontaneous abortion associated with paternal smoking (Blanco-Munoz et al., 2009; Venners et al., 2004). However, others have shown no association (Chatenoud et al., 1998; Windham et al., 1992, 1999). Moreover, paternal smoking is a factor associated with anorectal malformations in humans in multiple studies (Zwink et al., 2011). Human epidemiological studies also indicate that early paternal smoking (onset before age 11) is associated with greater body mass index in male, but not female, offspring (Pembrey, 2010; Pembrey et al., 2006).

Aside from these few studies looking at the effect of paternal smoking on offspring, numerous studies have found evidence that male smoking affects multiple fertility factors in the male. Thus, paternal tobacco use is associated with defects in the tail of the spermatoozon (Ozug et al., 2005) and decreases in sperm density, motility, and morphology (Sofikitis et al., 1995). Another study found no differences in sperm concentration or motility but decreased semen volume (Pasqualotto et al., 2006). Taken together, these studies point toward decreases in male fertility and or embryo viability which suggests potential negative effects in the offspring. To date, we found only one animal study which found that paternal nicotine smoke exposure did not affect development or any behavioral parameters measured (Gaworski et al., 2004). More animal work is needed to determine if nicotine exposure and smoking has transgenerational epigenetic effects.

4. Cannabinoids

Marijuana is the most widely abused illicit substance in the United States. Of particular concern is the high percentage of adolescents that engage in marijuana use. It is estimated that almost 50% of teenagers have used marijuana and approximately 9% use marijuana heavily (~20 days out of the month) (Metlife, 2011). Marijuana use by adolescents may be particularly problematic as developing systems may be more vulnerable to the impact of exogenous cannabinoids (Rice and Barone, 2000). Abuse by adolescent females is of particular concern as the endogenous cannabinoid system is important for reproductive physiology, with CB1 receptors as well as endogenous cannabinoids expressed in the ovaries, uterine endometrium, and other peripheral endocrine tissue (Bari et al., 2011). Thus, cannabinoid exposure during this critical period could result in lasting modifications in female reproduction, and might impact future offspring. To examine the potential effects on offspring, adolescent females were exposed to cannabinoids during adolescence. They were then maintained drug-free for over 3 weeks and bred during adulthood. Using this model, it was shown that both adolescent and adult male offspring of adolescent-exposed dams exhibit greater sensitivity to morphine-induced conditioned place preference than the control animals, even in the absence of any direct in utero exposure (Byrnes et al., 2012). Moreover, unpublished data from the Byrnes laboratory shows that female offspring demonstrate enhanced expression of morphine-induced locomotor sensitization accompanied by increased expression of the mu opioid receptor in the nucleus accumbens.

While no studies to date have examined the effect on the offspring of cannabinoid exposure in the sire prior to conception, available evidence suggests that disruptions in endocrine functioning resulting from cannabinoid exposure may cause deleterious effects in subsequent generations. For example, tetrahydrocannabinol (THC) disrupts gonadal functions by depriving testicular cells of energy reserves and stimulating androgen-binding protein secretion which leads to oligosperma in chronic cannabis smokers. THC also interferes with production of prolactin, luteinizing hormone (LH), and follicle stimulating hormone (FSH), which causes reduced testosterone production in the testes (Banerjee et al., 2011; Harclerode, 1984; Husain and Khan, 1985). It has also been suggested that acute cannabinoid treatment affects the quality and quantity of spermatozoa produced by the testis (Harclerode, 1984). Animal models have demonstrated that cannabinoid administration suppresses gonadal steroids, growth hormone, prolactin, thyroid hormone and activates the HPA-axis (Banerjee et al., 2011; Brown and Dobs, 2002). These effects have been shown to be mediated by the binding of exogenous cannabinoids to the endogenous receptor in or near the hypothalamus (Brown and Dobs, 2002). However, the effects in humans have been inconsistent likely due to the development of tolerance (Brown and Dobs, 2002). Finally, in humans, two case controlled studies found an increased risk for congenital heart defects associated with paternal marijuana use (Ewing et al., 1997; Wilson et al., 1998). Clearly, there is a large gap in the literature examining the effects of marijuana use preconception on future generations.

5. Morphine

The significant impact of maternal opioid use on fetal outcomes and offspring neurodevelopment has been an area of interest for decades (Hutchings, 1982; Malanga and Kosofsky, 1999; Vathy, 2002). In both substance abusing women and animal models of prenatal substance use the fetus is directly exposed to opioids, limiting the applicability to a discussion on transgenerational epigenetic processes. To avoid direct fetal exposure, animal models were developed that expose both males and females to opioids preconception and then examine the effects on their offspring. Such studies have primarily examined the impact of exposure to morphine during adolescence. In male rats, morphine administered throughout adolescent development resulted in significant modifications in sexual maturation in the males themselves. When these males were mated to drug naïve females several weeks after morphine exposure, they produced smaller litters and as adults their offspring demonstrated significant modifications in endocrine parameters, including sex specific changes in adrenal weights, luteinizing hormone and hypothalamic β-endorphin (Cicero et al., 1991). As all that the male contributed in this model was sperm, such findings strongly suggest that prior exposure to opioids can induce transgenerational epigenetic effects even in the absence of continuing use.

Similar findings have been revealed using a female rat model. In those studies, females were exposed to an intermittent, increasing dose regimen of morphine during early-mid adolescence. All animals were then drug-free for several weeks prior to mating with drug naïve males. In these studies there were no significant differences in any postnatal parameter (i.e. litter size, weight, sex ratio). When offspring were examined as adults, however, a number of significant effects have been observed. These include sex-specific changes in social and emotional behaviors (Byrnes et al., 2011; Johnson et al., 2011) as well as alterations in their response to morphine (Byrnes, 2005; Byrnes et al., 2011). More recent findings using this model also observed significant changes in the functional response to dopamine agonists coupled with increased levels of
dopamine- and opioid-related gene expression in the nucleus accumbens. Of note, these effects were observed in both the first (F1) and second (F2) generation, suggesting multigenerational epigenetic effects triggered by adolescent exposure (Byrnes et al., 2013).

The mechanisms underlying such intergenerational transmission are unknown and perhaps more complicated when considering maternal transmission. Exposure to drugs of abuse, even when experienced prior to conception, may alter the prenatal hormonal milieu, thereby modifying the developmental trajectory of offspring. Alternatively, or perhaps additionally, modifications in maternal care as a result of prior exposure to opioids, could impact the development of offspring. Indeed, morphine administered both prior to parturition or during active mothering significantly alters maternal care (Bridges and Grimm, 1982; Mann et al., 1991; Miranda-Paiva et al., 2001; Slamberova et al., 2001). Moreover, non-genomic transmission via subtle variations in maternal care have been demonstrated repeatedly in animal models (McLeod et al., 2007; Szyf et al., 2007; Weaver, 2007; Weaver et al., 2004). Thus, one possible mechanism of transmission may be alterations in the pre- and/or postnatal environment.

Alternatively, exposure to opioids could directly affect epigenetic germine cells in the exposed male and female. Such effects could alter early embryogenesis and impact a number of developmental processes. These effects could then be passed forward via alterations in behavior/physiology of the next generation or via direct epigenetic inheritance. Additional studies are needed to determine the mechanisms underlying transmission in both males and females exposed to opioids. Nonetheless, the nature of the observed offspring effects, including dysregulation of the hypothalamic–pituitary–adrenal axis, increased sensitivity to opioids, and blunted response to dopamine agonists, all suggest that parental exposure to opioids prior to conception may increase the risk of substance use in their future offspring.

### 6. Cocaine

In terms of parental cocaine administration, the clinical and preclinical literatures have focused primarily on the developmental effects of prenatal cocaine exposure. In that regard, preclinical studies relative consistently show disrupted cortical and hippocampal development in the offspring as well as impaired executive function and memory following in utero cocaine exposure (Dow-Edwards, 2011; Lidow, 2003; Malanga and Kosofsky, 2003). Epigenetic mechanisms appear to play a role in these changes in that male mice exposed to cocaine prenatally had altered patterns of DNA methylation in hippocampal pyramidal neurons (Novikova et al., 2008), which could underlie impaired sustained attention and spatial working memory observed in these offspring (He et al., 2006b). While interesting, studies of this sort are directly examine the effect of cocaine on development. Moreover, the dams administered both prior to parturition or during active mothering significantly alters maternal care (Vassoler et al., 2013). Taken together, these findings indicated that voluntary paternal ingestion of cocaine reprograms the germline resulting in enhanced BDNF expression in the mPFC among male progeny, which appears to confer resistance to the reinforcing effects of cocaine. This result should be interpreted cautiously. It is as yet unknown if these male offspring have impairments in reward and motivational circuits that underlie the decreased self-administration behavior.

We examined paternal transmission in order to avoid the influence of in utero cocaine exposure and the potential influence of prior cocaine experience by dams on maternal behavior. It is possible that even the relatively brief exposure to a cocaine-experienced male during breeding might have an impact on maternal behavior. We examined licking/grooming and other maternal behaviors and found no differences between the dams bred with cocaine-experienced sires relative to controls (Vassoler et al., 2013). A critical unanswered question is whether this cocaine resistance phenotype is present in the F2 generation (i.e. the grandoffspring of cocaine-experienced sires). Since the sperm of the sire was exposed to cocaine, it cannot be assumed that the heritability is transgenerational. Indeed, as suggested above, it is possible that cocaine binding to the spermatozoa could allow for transport of
cocaína en el desarrollo del embrión (Yazigi et al., 1991). En el caso de la cocaína, se ha demostrado que tiene efectos profundos en generaciones posteriores. Sin embargo, el fenómeno de transgeneracional hereditabilidad es un tema que no está claramente establecido. Es posible que se deba a que solo los varones muestren esta hereditabilidad en experimentos, como se observó en la segunda generación (Becker, 2008; Quinones-Jenab and Jenab, 2010). Es posible que se deba a que la cocaína no ingresa a la célula fertilizada (Yazigi et al., 1991). En el caso de la cocaína, es necesario que los experimentos se extiendan a generaciones futuras para evitar la transmisión de cambios epigenéticos que puedan alterar las diferencias observadas en los descendientes de fumadores.

La cocaína se considera un factor de riesgo importante para el aprendizaje y la memoria y altera el rendimiento en la conducta de los descendientes, lo que sugiere la necesidad de cambios en la política y la implementación de nuevas campañas para concienciar sobre la importancia de evitar el consumo de drogas que puedan afectar a las futuras generaciones.

7. Conclusões

A ideia de que o ambiente de uma geração pode impactar o fenótipo de futuras gerações não é nova. Jean-Baptiste Lamarck propôs uma teoria de evolução que considerava o ambiente como fator de reprodução em hereditariedade. Seu trabalho foi amplamente criticado e ignorado por falta de mecanismo e para favor de Darwin's eloquent description of "survival of the fittest" (Burkhardt, 2009). No entanto, parece que alguns de seus ideais têm sido recentemente reavaliados e explicados pelo fenômeno de epigenética. A maneira como as exposições ambientais de uma geração podem influenciar e afetar futuras gerações é de importância crítica para o entendimento do comportamento humano e evolução.

Este estudo demonstrou que existem várias consequências para os descendentes de pais expostos a drogas, mesmo em ausência de exposição direta ao feto (sumarizado em Tabela 1). A ideia de que os toxinas ambientais interferem com a conduta de pais com um parente que tenha sofrido um impacto maior no descendente requer mais pesquisa e deve ser destacada. Em conclusão, uma pesquisa recente encontrou que os efeitos de consumo de drogas em pais ou mães têm um impacto maior do que uma interação parental (Walden et al., 2007). Isso sugere a necessidade de mudanças de política e campanhas para aumentar a consciência sobre a exposição de futuras gerações.

Os resultados apresentados sugerem a possibilidade de que a exposição a substâncias de abuso possa produzir alterações epigenéticas que resultam em alterações profundas no comportamento e na fisiologia subsequente. No entanto, para um efeito verdadeiramente genético, essas alterações devem ser confirmadas em futuras gerações para evitar a exposição de informações genéticas. Quando isso é observado, a geração subsequente importante para justificar mais pesquisa definir o impacto de alterações epigenéticas associadas com o uso de drogas de abuso e os mecanismos subjacentes a esses efeitos.

Referências


