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## Cannabis use during pregnancy: Pharmacokinetics and effects on child development

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## ABSTRACT

The broad-based legalization of cannabis use has created a strong need to understand its impact on human health and behavior. The risks that may be associated with cannabis use, particularly for sensitive subgroups such as pregnant women, are difficult to define because of a paucity of dose-response data and the recent increase in cannabis potency. Although there is a large body of evidence detailing the mode of action of  $\Delta^9$ -tetrahydrocannabinol (THC) in adults, little work has focused on understanding how cannabis use during pregnancy may impact the development of the fetal nervous system and whether additional plant-derived cannabinoids might participate. This manuscript presents an overview of the historical and contemporary literature focused on the mode of action of THC in the developing brain, comparative pharmacokinetics in both pregnant and non-pregnant model systems and neurodevelopmental outcomes in exposed offspring. Despite growing public health significance, pharmacokinetic studies of THC have focused on nonpregnant adult subjects and there are few published reports on disposition parameters during pregnancy. Data from preclinical species show that THC readily crosses the placenta although fetal exposures appear lower than maternal exposures. The neurodevelopmental data in humans and animals suggest that prenatal exposure to THC may lead to subtle, persistent changes in targeted aspects of higher-level cognition and psychological well-being. There is an urgent need for well-controlled studies in humans and preclinical models on THC as a developmental neurotoxicant. Until more information is available, pregnant women should not assume that using cannabis during pregnancy is safe.

## 1. Introduction

The history of cannabis use and its impact on human health and society is complicated and rapidly changing (National Academies Press, 2017). Throughout the world, this flowering plant remains the most commonly used illicit drug and there is a strong shift towards the legalization of its medical and recreational use (Azofeifa et al., 2016). According to the 2015 National Survey on Drug Use and Health, 22.2 million Americans currently use cannabis and 2.6 million individuals aged 12 or older tried cannabis for the first time in the last twelve months (Center for Behavioral Health Statistics and Quality, 2016;

Lipari et al., 2015). This sobering statistic translates into approximately 7100 new users each day. Attitudes about cannabis use are changing and this is particularly apparent in adolescents and young adults. With significant profits at stake, the legal cannabis market has implemented selective growing methods to boost psychoactive potency. Over the last two decades, the average THC content of cannabis (potency) has increased from approximately 4% to 12% (ElSohly et al., 2016), but levels as high as 30% have recently been documented in legal cannabis grown for recreational use (American Chemical Society, 2015). The cultivation of cannabis is evolving and dramatic increases in potency make it difficult to understand the health risks that may be associated with

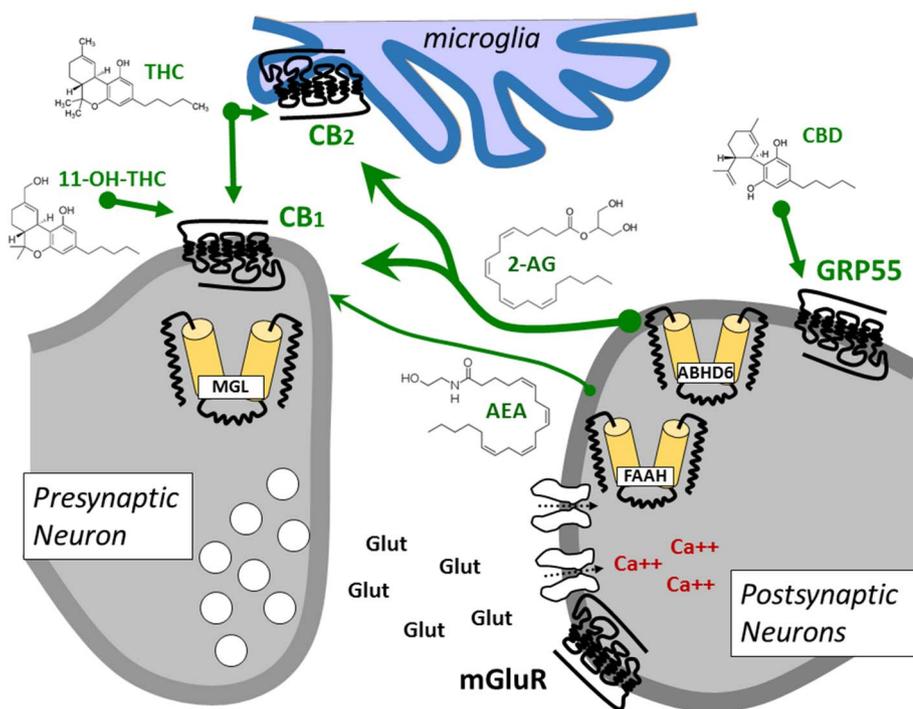
**Abbreviations:** 2-AG, 2-arachidonoyl glycerol; 11-nor-THC-COOH, 11-nor-tetrahydrocannabinol-9-carboxylic acid; 11-OH-THC, 11-hydroxy-tetrahydrocannabinol; 8-OH-THC, 8-hydroxy-tetrahydrocannabinol; CB, cannabinoid; CB<sub>1</sub>R, Type I cannabinoid receptor; CB<sub>2</sub>R, Type II cannabinoid receptor; CBD, cannabidiol; C<sub>max</sub>, maximum concentration; CNS, central nervous system; CYP, cytochrome P450; eCB, endocannabinoid; GD, gestational day; GPCR, G-protein-coupled receptor; ip, intraperitoneal; iv, intravenous; MOA, mode of action; phyto-CB, phyto-cannabinoids; PND, postnatal day; po, oral; sc, subcutaneous; THC,  $\Delta^9$ -tetrahydrocannabinol; UGT, UDP-glucuronosyltransferase

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**Fig. 1.** Endogenous signaling system is used by multiple cell types in the brain and periphery. It encompasses cannabinoid receptors CB<sub>1</sub> and CB<sub>2</sub> (primarily expressed by neurons and immune cells microglia, respectively), the two endocannabinoids anandamide (arachidonoyl ethanolamine, AEA) and 2-AG (2-arachidonoyl glycerol) and the enzymes that produce them (not shown) and inactivate them, namely fatty acid amide hydrolase (FAAH), monoacylglycerol lipase (MGL),  $\alpha/\beta$ -hydrolase domain 6 (ABHD6) and cyclooxygenase 2 (COX2, not shown). Additional molecular components are GPR55 that is modulated by cannabidiol (CBD). Tetrahydrocannabinol (THC) modulates both CB<sub>1</sub> and CB<sub>2</sub>, and 11-OH tetrahydrocannabinol (11-OH-THC) modulates CB<sub>1</sub>.

contemporary use, particularly for sensitive subgroups within the general population.

The increased availability and quality of cannabis, paired with relaxed attitudes about use in younger populations, will likely result in a rise of cannabis use in women of childbearing age. In fact, data collected from 2002 to 2014 in the U.S. indicate that 7.5% of pregnant women between 18 and 25 years of age use cannabis, while the rate of use in all pregnant women is approximately 4% (Brown et al., 2016). This statistic places cannabis firmly in the bull's-eye of public health concerns and suggests that thousands, if not millions, of infants will be prenatally exposed to this chemically-complex compound over the coming decades. Women who become pregnant may continue to use cannabis for a variety of reasons. For example, a survey of women in Vancouver, Canada found that up to 77% of medicinal cannabis use during pregnancy was related to nausea; over 50% of respondents also reported cannabis use to treat a lack of appetite, general pain, insomnia, anxiety, depression and fatigue (Westfall, Janssen, Lucas, & Capler, 2009). Despite knowledge of potential fetal health risks, cannabis use in pregnant women is becoming more commonplace and the need for clear messaging on the safety of use during pregnancy is urgently needed (Mark, Gryczynski, Axenfeld, Schwartz, & Terplan, in press).

Given the possibility of increased use in pregnant women and the fact that cannabis is being widely investigated as a novel treatment for a variety of diseases, including epilepsy, multiple sclerosis and cancer, it is imperative that significant efforts be immediately dedicated to evaluating the potential consequences of exposure for the fetus. For example, the oromucosal spray (Sativex) has been approved in Europe to treat spasticity due to multiple sclerosis. While cannabis use during pregnancy has been studied in humans and several animal model systems, defining the risk of fetal cannabis exposure has been complicated by factors such as concurrent maternal use of drugs with their own toxicity profile and an absence of quantitative markers of cannabis exposure. Studies in human and preclinical model systems are needed to generate mechanistic data on the maternal-fetal kinetics and toxicity of cannabis that can be interpreted within the context of fetal pharmacology and developmental psychobiology. The consequences of prenatal cannabis exposure will not be elucidated without

methodological control of confounding factors such as tobacco/alcohol use and quantitative measurements of exposure to generate critical dose-response information. This review was written to bridge our current understanding of 1) THC pharmacokinetics in adults with a focus on pregnancy 2) the consequences of fetal exposure at the molecular and cellular levels and 3) the effects of prenatal exposure on child neurodevelopmental outcomes from birth through adolescence. The marriage of pharmacokinetics with neurodevelopmental data provides an interdisciplinary framework to generate data-driven messages about fetal risk and highlight directions for future research objectives.

## 2. What is cannabis?

Cannabis is a dioecious plant that grows wild in many tropical parts of the world. It is one of the world's oldest crops and the history of cannabis dates back about 12,000 years (Warf, 2014). Cannabis was widely used in ancient China and records of medical applications appeared about 4000 years ago, originally in relation to its use as a surgical anesthetic. Plant-derived cannabinoids (CB) are referred to as phyto-cannabinoids (phyto-CB) and THC is the most famous, representing the primary psychoactive ingredient produced by the cannabis plant (see Fig. 1). Specific strains of cannabis may also produce high levels of a second phyto-CB, cannabidiol (CBD), often referred to as non-psychoactive. Although not associated with cannabis-induced euphoria or intoxication (Grotenhermen, Russo, & Zuardi, 2017), CBD exposure is psychoactive and affects several brain functions and behaviors, including neuronal activity, seizure incidence and social interactions (Renard, Norris, Rushlow, & Lavolette, 2017; Todd & Arnold, 2016). Accordingly, CBD has been linked to influencing a wide range of clinical outcomes such as epilepsy and neuropsychiatric disorders. Additional phyto-CBs that exhibit a certain level of bioactivity include cannabitol, cannabigerol, and cannabichromine (Rosenthaler et al., 2014; Turner, Williams, Iversen, & Whalley, 2017). These compounds are synthesized by a family of enzymes expressed by the cannabis plant and their biological activity and mechanism of action have not yet been studied in great detail. Thus, the cannabis plant produces a family of structurally related compounds, the phyto-CB, that produce a wide

array of biological effects, many of which remain to be studied.

### 3. Phyto-cannabinoids, cannabinoid receptors and endocannabinoid signaling

The two main phyto-CBs, THC and CBD, and several of their metabolites (e.g. 11-hydroxy- $\Delta^9$ -THC (11-OH-THC, Fig. 1)) bind and differentially activate cannabinoid receptors (Devane, Dysarz, Johnson, Melvin, & Howlett, 1988). Cannabinoid receptors 1 (CB<sub>1</sub>R) are G protein-coupled receptors (GPCR) abundantly expressed in the central nervous system (CNS) by many neuronal and glial cell types (Stella, 2010). A key function of CB<sub>1</sub>R in mature neurons is to modulate the release of neurotransmitters such as glutamate and GABA (Bloomfield et al., 2016; Katona & Freund, 2008, 2012). Central to this review, activation of CB<sub>1</sub>R during neuronal development influence cell proliferation, migration, and differentiation through control of the expression of key factors, including brain-derived nerve factor (Calvignoni, Hurd, Harkany, & Keimpema, 2014; de Salas-Quiroga et al., 2015; Marsicano et al., 2003). CB<sub>1</sub>R are also expressed by cells in the periphery, including in reproductive tissues (Pertwee et al., 2010). Thus, most of the psychoactive responses produced by THC and 11-OH-THC exposure are mediated through CB<sub>1</sub>R expressed by a complex network of neuronal and glial cells in the CNS but circulating phyto-CBs and their metabolites also exhibit significant activity on both the CNS and peripheral tissues.

Cannabinoid receptors 2 (CB<sub>2</sub>R) are closely-related GPCRs of CB<sub>1</sub>R and are expressed by hematopoietic cells, as well as by select neurons and cells in the periphery, including cells in reproductive tissues (Pertwee et al., 2010). CB<sub>2</sub>R also couple to G<sub>i/o</sub> proteins and their activation modulates similar signaling pathways and cell functions as CB<sub>1</sub>R (Pertwee, 2008). Accordingly, both CB<sub>1</sub>R and CB<sub>2</sub>R mediate the biological activity of different phyto-CBs, resulting in pronounced changes in several neural and immune functions.

Cannabinoid receptor activity is regulated by endocannabinoids (eCB), anandamide (AEA) and 2-arachidonoyl glycerol (2-AG), two signaling lipids that are produced and inactivated by distinct lipases and hydrolases, respectively (Castillo, Younts, Chávez, & Hashimoto, 2012; Piomelli, 2003). Increased cannabinoid receptor activation allows for rapid modulation of neuronal functions by modifying synaptic circuits (Katona & Freund, 2008). eCBs are produced on-demand by many different cell types in response to increased activity (typically associated with increases in intracellular calcium) via lipase activation that releases the eCBs from their membrane precursors (Di Marzo et al., 1994; Stella & Piomelli, 2001; Stella, Schweitzer, & Piomelli, 1997). Biological inactivation of eCBs occurs in two steps: a rapid uptake into cells via active transport, followed by FAAH-mediated hydrolysis for anandamide (Cravatt et al., 2001; Fu et al., 2012; McKinney & Cravatt, 2005) and hydrolysis or chemical modification of 2-AG by mono acyl glycerol lipase (MGL),  $\alpha/\beta$ -hydrolase domain 6 (ABHD6) and cyclooxygenase 2 (COX2) (see Fig. 1) (Dinh et al., 2002; Dinh, Kathuria, & Piomelli, 2004; Karlsson, Contreras, Hellman, Tornqvist, & Holm, 1997; Nomura et al., 2011; Tornqvist & Belfrage, 1976). Thus, members of the eCB signaling system include CB<sub>1</sub>R and CB<sub>2</sub>R, anandamide and 2-AG, and enzymes that produce and inactivate these two main eCBs. Note that both anandamide and 2-AG modulate the activity of additional targets that play fundamental roles in cell function involved in development and regulation of homeostasis and metabolic pathways. For example, anandamide is a low-efficacy agonist of TRPV1 and GPR55, and high concentrations of 2-AG activate peroxisome proliferator-activated receptor- $\alpha$  (PPAR $\alpha$ ) and PPAR $\gamma$  (Abood, Sorensen, & Stella, 2012; Pertwee et al., 2010). Thus, eCB hydrolyzing enzymes represent cardinal molecular hubs within lipid signaling networks that control the levels and action of eCBs on their various targets (Marrs & Stella, 2007; Stella, 2012).

While THC remains the most famous phyto-CB thus far, the pharmacological activity of CBD is being studied in greater detail and was

recently shown to modulate several non-CB<sub>1</sub>/CB<sub>2</sub> receptors. For example, emerging in vitro evidence suggests that CBD might modulate CB<sub>1</sub>R through an allosteric site (Laprairie, Bagher, Kelly, & Denovan-Wright, 2015). A large body of work based on mouse genetics studies show that CBD modulates the activity of GPR55, a GPCR expressed by cells in both CNS and peripheral tissue (Ross, 2009; Sharif & Abood, 2010). As mentioned above, much less is known about the mechanism of action of cannabinol (CBN), cannabigerol (CBG), and cannabichromine (CBC).

Over 50 years of medicinal chemistry efforts have led to the development of several classes of synthetic cannabinoids that exhibit remarkable potency and selectivity profiles at the various targets mentioned above. For example, the indole-based compound, JWH-018, is a high potency selective agonist at CB<sub>1</sub>R (Atwood, Huffman, Straiker, & MacKie, 2010). In recent years, the human use of recreational cannabis-based products that contain indole-based cannabinoids (such as JWH-018 in *K2* and *Spice*) has greatly increased and its impact on human health is only starting to be reported. Concerning reports indicate that JWH-018 is associated with a toxicity profile that is radically different from phyto-CBs and characterized by renal failure and seizures (Buser et al., 2014; Lapointe et al., 2011). One should note, however, that this indole-based compound was developed from a different chemical scaffold and exhibits a different selectivity profile to the rest of cannabinoids, including THC. Therefore, it is not surprising that its toxicity profile may be different from the rest of cannabinoids. Together, this evidence depicts a complex bioactivity profile linked to the use of cannabinoids that is due to their polypharmacological activity at multiple targets that regulate cardinal physiological functions.

### 4. eCB signaling in development

The eCB signaling system plays an overarching regulatory role during the initial stages of embryo development, implantation and ensuing prenatal development and differentiation. It undergoes a drastic switch in function from the prenatal determination of cell fate to the homeostatic regulation of metabolic pathways and transmission in the mature CNS. The functional role of CB<sub>1</sub>R and CB<sub>2</sub>R during this early stage of embryogenesis is not well understood but is likely linked to their ability to control cell proliferation and differentiation (Galve-Roperh et al., 2013).

During brain development, the proliferation and asymmetric division of neural progenitor cells, as well as their positioning and molecular diversification into neuronal and glial progenies, are all tightly regulated by both morphogenetic signaling molecules that contribute to building complex tissues. CB<sub>1</sub>R and CB<sub>2</sub>R are expressed by divergent pluripotent cells, including neuronal stem and progenitor cells where they differentially regulate cell proliferation and differentiation (Galve-Roperh, Palazuelos, Aguado, & Guzmán, 2009; Maccarrone, Guzmán, Mackie, Doherty, & Harkany, 2014). For example, activation of CB<sub>1</sub>R expressed by neural stem cells isolated from embryos enhances differentiation into neurons without affecting astrocytes and oligodendrocytes, as evidenced by increased neurite outgrowth and expression in neuronal markers (de Salas-Quiroga et al., 2015). By contrast, activation of CB<sub>1</sub>R expressed by post-natal radial and neuronal stem cells controls differentiation in the adult brain by promoting astroglial differentiation of newly born cells (Aguado et al., 2005, 2006). Given that CB<sub>1</sub>R expressed by neural progenitor cells in the developing forebrain regulates the ratio of neurons to astroglial cells in areas such as the hippocampus and cerebral cortex, changes in CB<sub>1</sub>R during this critical period are likely to influence the connectivity of these brain regions (Berghuis et al., 2007; Maccarrone et al., 2014). Evidence suggests that focal eCB gradients are probably generated to control the direction of cell migration (Miller & Stella, 2008; Tortoriello et al., 2014). Accordingly, the downregulation of DAGL $\alpha$  and DAGL $\beta$  expression (enzymes involved in 2-AG release from membranes) following neuronal specification is likely to represent an essential step to increase the reliance of

post-mitotic neurons on extracellular 2-AG produced by pyramidal cells in the cortical plate and thus function as positional cues (Tortoriello et al., 2014). Additional molecular mechanisms involving eCB signaling that regulate directed migration include the tropic action of growth factors. Indeed, specific receptor tyrosine kinases are transactivated following stimulation of CB<sub>1</sub>R, and conversely CB<sub>1</sub>R are sensitized by growth factors (e.g. BDNF) (Marsicano et al., 2003). In line with these studies, 2-AG signaling through CB<sub>1</sub>Rs represents an essential effector molecular step of neurotrophic factor signaling (De Chiara et al., 2010; De March et al., 2008; López-Gallardo et al., 2012). This body of work identified a novel physiological role of eCB signaling system in providing signaling cues involved in the regulation of neural stem and progenitor cell differentiation and ensuing function. These studies also highlight the importance of determining how prolonged activation of CB<sub>1</sub>R in neural progenitors by phyto-CBs intake impact newly born cells.

## 5. Impact of cannabinoids on the developing brain

The developing brain undergoes substantial structural remodeling that makes it particularly vulnerable to the harmful effects of bioactive ingredients (Chambers, Taylor, & Potenza, 2003; Crews, He, & Hodge, 2007). Such remodeling occurs in many brain areas involved in vital neuronal function, including sensory inputs and the control of body temperature, as well as higher-order cognitive processes such as learning, memory and decision making (Wise, 2004). CB<sub>1</sub>R signaling modulates long-range neuronal connectivity, including corticofugal connectivity (Katona & Freund, 2008; Tortoriello et al., 2014). Accordingly, THC administration to pregnant mice during a restricted time window interferes with subcerebral projection neuron generation, thereby altering corticospinal connectivity and producing long-lasting alterations in the fine motor performance of the adult offspring (de Salas-Quiroga et al., 2015).

Mechanistically, such impairments are reminiscent of those elicited by the genetic ablation of CB<sub>1</sub>R and accordingly regimented THC administration to pregnant mice leads to down-regulation of CB<sub>1</sub>R signaling through desensitization (Berghuis et al., 2007; Castelli et al., 2007; Keimpema, Mackie, & Harkany, 2011; Vitalis et al., 2008). This impairment of long-range neuronal connectivity occurs for dorsal telencephalic glutamatergic neurons but not for forebrain GABAergic neurons (Mulder et al., 2008). Importantly, repeated CB<sub>1</sub>R activation during sensitive periods of CNS development affects the expression and functionality of multiple receptors, including dopaminergic receptors that are critically involved in higher cognitive functions (Renard et al., 2017). In mice, in utero exposure to THC leads to CB<sub>1</sub>R activation and neuronal rewiring through the degradation of the molecular effector, SCG10/stathmin 2, known to regulate microtubule dynamics in axons (Tortoriello et al., 2014). A telling example is provided by results showing that CB<sub>1</sub>Rs activation on the axonal surface induces repulsive growth cone turning and eventual collapse in in vitro model systems (Harkany, Keimpema, Barabás, & Mulder, 2008; Harkany, Mackie, & Doherty, 2008; Maccarrone et al., 2014). The molecular mechanism of CB<sub>1</sub>R-mediated cytoskeletal instability in growth cones involves select signaling pathways, including RHO-family GTPases, RAS, and PI3K–AKT–β-catenin signaling (Alpár et al., 2014; Díaz-Alonso et al., 2012; Maccarrone et al., 2014). Accordingly, THC exposure leads to ectopic formation of filopodia and alterations in axon morphology, together limiting the computational power of neuronal circuits involved in high cognitive function in affected offspring (Cristino & Di Marzo, 2014; Tortoriello et al., 2014).

An interesting cellular component of the impact of THC on brain development that has not been studied in detail is whether repeated activation of cannabinoid receptors expressed by glial cells will also lead to their down-regulation or desensitization and whether this response might affect normal brain maturation and result in persisting impairments. In accordance with the tripartite synapse hypothesis,

which states an involvement of astrocytes in synaptic transmission, peri-synaptic astrocytes that express MAGL should form a barrier limiting 2-AG spread and action on its targets to 20–100 μm and not beyond its immediate site of action (Maccarrone et al., 2014; Metna-Laurent & Marsicano, 2015; Navarrete & Araque, 2010; Oliveira da Cruz, Robin, Drago, Marsicano, & Metna-Laurent, 2016; Stella, 2010). This MAGL expression pattern is likely to both limit axonal spread in the prospective internal capsule and help delineate migratory routes for CB<sub>1</sub>R-expressing neurons, such as exemplified by cortical interneurons (Alpár et al., 2014; Keimpema et al., 2010; Wu et al., 2010). Thus, pharmacological manipulation of eCB signaling and its hijack by phyto-CB during crucial periods of synaptogenesis and/or postnatal pruning might precipitate or predispose an individual to neuropsychiatric disease-like phenotypes.

## 6. Pharmacokinetics of THC in humans

There is strong evidence that THC pharmacology has considerable impacts on neuronal signaling and development and as such, it is plausible to hypothesize that THC exposures during pregnancy could lead to long-term changes in neuronal development. However, a critical component in understanding the potential consequences of cannabis consumption during pregnancy is the duration of exposure, the overall magnitude of exposure and the extent to which the fetus and fetal brain are exposed to THC after maternal cannabis consumption. While the pharmacokinetics (PK) of THC and its metabolites have been studied in adult humans following intravenous (iv), oral and inhalation administration (see Table 1), little is known about the changes in cannabis PK during pregnancy and the maternal-fetal transfer and fetal PK of THC. In addition, it is possible that the route of cannabis consumption (oral, inhalation, and different ways of smoking) will have an impact on the overall fetal toxicity.

The absorption pathways between smoked and edible cannabis products are distinctly different. Following oral administration, THC absorption is typically > 90% and not affected by formulation (Parikh, Kramer, Khurana, Smith, & Vetticaden, 2016). Studies have shown that the bioavailability of THC is limited (10–20% when ingested in gelatin capsules (Wall, Sadler, Brine, Taylor, & Perez-Reyes, 1983) and 6 ± 3% when ingested in a chocolate cookie (Ohlsson et al., 1980)) due to significant liver first pass metabolism. In contrast, smoked cannabis is not subject to liver first pass metabolism. Loss of THC in side stream smoke and in the butt of the cigarette, as well as loss via pyrolysis, result in low absorption of THC from smoking (Grotenhermen, 2003) and overall highly variable bioavailability of 2–56% (mean 18 ± 6%) (Huestis, 2007; Ohlsson et al., 1980).

An important difference between oral and smoked cannabis is also the rate of THC absorption (see Table 1). Following smoking, THC is rapidly absorbed and its peak concentrations (C<sub>max</sub>) are reached within minutes (Kauert, Ramaekers, Schneider, Moeller, & Toennes, 2007; Ohlsson et al., 1980). In comparison, absorption from oral capsules is considerably slower and maximum THC concentrations are reached 1–3 h after dosing (Ahmed et al., 2015; Ohlsson et al., 1980; Schwilke et al., 2009). As expected from the faster rate of absorption from smoked cannabis, the average C<sub>max</sub> values for THC following smoking are somewhat higher than those observed after oral consumption if similar THC content is consumed (see Table 1). The average peak concentrations of THC in serum reached after smoking cannabis cigarettes containing 18.2 mg (0.25 mg/kg body weight) and 36.5 mg (0.5 mg/kg body weight) of THC were 48 μg/L and 79 μg/L (Kauert et al., 2007). In a study in occasional users who smoked a cannabis cigarette with 4% THC (20 mg dose) with tobacco, the average C<sub>max</sub> was 25.8 ± 42.9 μg/L with an average time to maximum concentration of 0.2 h. (Marsot et al., 2016). The range of individual peak concentrations (1.6–160 μg/L) emphasizes the large inter-individual variability in THC exposures. After oral dosing in daily cannabis users, the C<sub>max</sub> of THC was 16.5 μg/L after 20 mg THC orally, (Schwilke et al.,

**Table 1**  
Summary of measured maximum plasma concentrations ( $C_{max}$ ) and times to reach maximum concentrations ( $t_{max}$ ) for THC and its two main metabolites, 11-OH-THC and 11-nor-THC-COOH following different routes of administration and exposure.

Route of administration	Dose	THC				11-OH-THC				11-nor-COOH-THC				Reference
		$C_{max}$ ( $\mu\text{g/L}$ )	$t_{max}$ (h)	AUC ( $\mu\text{g}\cdot\text{h/L}$ )	$t_{1/2}$ (h)	$C_{max}$ ( $\mu\text{g/L}$ )	$t_{max}$ (h)	AUC ( $\mu\text{g}\cdot\text{h/L}$ )	$t_{1/2}$ (h)	$C_{max}$ ( $\mu\text{g/L}$ )	$t_{max}$ (h)	AUC ( $\mu\text{g}\cdot\text{h/L}$ )	$t_{1/2}$ (h)	
Intravenous	0.053 mg/kg	81-641	0.1	117	1.2	9.1	0.1	-	36.7	1	-	-	Naef, Russmann, Petersen-Felix, and Brenneisen (2004)	
Intravenous	2.2-4 mg	85	0.2	201-280	29-36	3.8	0.33	15-33	24	1	-	25-55	Wall et al. (1983)	
Intravenous Smoking	5 mg Single 5.9% THC cigarette	161-316 28.3	0.05 0.5	72 89.3	- -	3.9 3.9	- 0.5	- 16.5	47	0.5	-	352	Ohlsson et al. (1980) Lee et al. (2015)	
Smoking	Single 11% THC cigarette	112	0.25	88	1.6	16.9	0.25-0.5	31 <sup>a</sup>	133	0.25-0.75	367 <sup>a</sup>	5.5 <sup>a</sup>	Toennes et al. (2011)	
Smoking	0.25 mg/kg	48	0	23	-	2.5	0-0.75	4.6	13.6	0-0.5	39.9	-	Kauert et al. (2007)	
Smoking	0.5 mg/kg	79	0	40	-	3.6	0-0.25	8	23.4	0.25	66.8	-	Kauert et al. (2007)	
Smoking	19 mg	33-118	0.05	17	-	-	-	-	-	-	-	-	Ohlsson et al. (1980)	
Smoking	29.3 mg	135	0.2	76	-	9.2	0.4	24 <sup>a</sup>	30	0.8	90 <sup>a</sup>	-	Hunault et al. (2008)	
Smoking	49.1 mg	203	0.2	113	-	16.4	0.4	37 <sup>a</sup>	60	0.8	150 <sup>a</sup>	-	Hunault et al. (2008)	
Smoking	69.4 mg	231	0.2	150	-	15.8	0.4	40 <sup>a</sup>	54	0.7	161 <sup>a</sup>	-	Hunault et al. (2008)	
Smoking	Ad libitum for > 5 days	83.5	0.2	170	-	14.2	0.28	59.1	155	0.52	1034	-	Lee et al. (2015)	
Pulmonary	0.053 mg/kg	18.9	0.33	20	0.77	1.4	0.66	-	10.0	2	-	-	Naef et al. (2004)	
Sublingual	5 mg	2.3	1.1	3.93	4.2	3.1	1.4	8.7	-	-	-	-	Klumpers et al. (2012)	
Oromucosal spray	10.8 mg	2.94	1	9.9	4.68	3.38	1.4	21.6	-	-	-	-	Stott et al. (2013)	
Oral solution	4.25 mg	1.8	1.5	3.75	5.41	2.53	1.5	10.1	-	-	-	-	Parikh et al. (2016)	
Oral capsule	5 mg	2.2	1	3.85	2.68	3.28	1.6	13.3	-	-	-	-	Parikh et al. (2016)	
Oral tablet	5 mg	2.92	1	3.15	1.2	4.7	1.2	10.8	-	-	-	-	Klumpers et al. (2012)	
Oral tablet	6.5 mg	4.43	0.6	4.8	1.3	5.9	0.8	14.1	-	-	-	-	Klumpers et al. (2012)	
Oral tablet	8 mg	4.69	0.75	6.3	1.3	6.1	1.3	18.1	-	-	-	-	Klumpers et al. (2012)	
Oral tablet	0.75 mg	0.41	1.5	2.21	5.1	0.56	1.0	3.86	-	-	-	-	Ahmed et al. (2015)	
Oral tablet	1.5 mg	1.0	1.0	4.66	5.1	1.2	1.8	8.92	-	-	-	-	Ahmed et al. (2015)	
Oral	15-20 mg	9.4-14	1.75-2.5	150-266	25	5.9-6.6	1.75-2	-	68-89	2	-	25-37	Wall et al. (1983)	
Oral cookie	20 mg	4.4-11	1-1.5	32.7	-	-	-	-	-	-	-	-	Ohlsson et al. (1980)	

<sup>a</sup> Only partial AUC and half-life were collected.

2009). In elderly patients the  $C_{max}$  was, 0.41  $\mu\text{g/L}$  after 0.75 mg oral dose and 1  $\mu\text{g/L}$  after a 1.5 mg oral dose (Ahmed et al., 2015). Collectively, the observed differences in the various studies in rate and extent of absorption and in first pass metabolism between oral and smoked cannabis may ultimately contribute to different toxicity profiles, especially if toxicity is related to peak concentrations and/or if overall metabolite exposure contributes to fetal pharmacology.

The site of action of THC's psychoactive effects is in the CNS and hence distribution to the site of action is critical for effects. Indeed, THC distributes extensively into tissues with a steady state volume of distribution ( $V_{ss}$ ) of 523–626 L (7.5–8.9 L/kg) (Hunt & Jones, 1980; Wall et al., 1983). The distribution of THC into the brain is, however, delayed and the volume of distribution of the central compartment after iv bolus is estimated between 2.6 L (0.04 L/kg) (Hunt & Jones, 1980) and 22.8 L (0.33 L/kg) (Ohlsson et al., 1980). Following iv administration, the maximum rated psychological “high” is reached at 15 min after the dose (Ohlsson et al., 1980). This corresponds to the time required to reach distribution equilibrium (minutes to an hour) (Hunt & Jones, 1980; Ohlsson et al., 1980) at the site of action and results in a hysteresis loop that describes the relationship between THC concentrations in plasma and the observed pharmacological effect with time (see also Grotenhermen, 2003). After peak effects are reached, the effects decline slowly due to the long terminal half-life between 20 and 57 h (Hunt & Jones, 1980; Lemberger, Axelrod, & Kopin, 1971) suggesting prolonged exposures and pharmacological effects even after single use.

In humans, THC is extensively metabolized (see Fig. 2) with a

systemic clearance of 12–36 L/h (Hunt & Jones, 1980; Wall et al., 1983). The clearance is somewhat restricted by plasma protein binding (THC unbound fraction of 3%). The majority of THC clearance in humans is thought to be hepatic, although metabolism of THC exists in the gut mucosa, lung and heart, at least in preclinical species (Grotenhermen, 2003). After iv dosing in humans, < 0.05% of the THC dose is recovered as unchanged  $\Delta^9$ -THC in urine or feces as the vast majority of THC is eliminated as metabolites either in urine (20%) or via biliary secretion of the metabolites in feces (25–40%) (Hunt & Jones, 1980; Lemberger et al., 1971; Wall et al., 1983). Over 80 metabolites of THC have been identified to date, but only some of these metabolites are quantitatively important and pharmacologically active, including 11-OH-THC, 11-nor- $\Delta^9$ -THC-9-carboxylic acid (11-nor-THC-COOH) and 8-OH- $\Delta^9$ -THC (see Fig. 2) (Dinis-Oliveira, 2016; Grotenhermen, 2003).

11-OH- $\Delta^9$ -THC is even more pharmacologically active than THC (Christensen et al., 1971) but the activity of 8-OH-THC is not known and 11-nor-THC-COOH is not a potent cannabinoid. In vitro and in vivo data suggest that 11-OH-THC is formed predominantly by CYP2C9 while 8-OH-THC is mainly formed by CYP3A4 (Bland, Haining, Tracy, & Callery, 2005; Bornheim, Lasker, & Raucy, 1992; Stott, White, Wright, Wilbraham, & Guy, 2013; Watanabe, Yamaori, Funahashi, Kimura, & Yamamoto, 2007). The 11-nor-THC-COOH is formed from 11-OH-THC by microsomal alcohol dehydrogenase enzymes (Narimatsu et al., 1988). Both 11-OH-THC and 11-nor-THC-COOH undergo glucuronidation by UGT1A9 and UGT1A10 (11-OH-THC) and UGT1A1 and

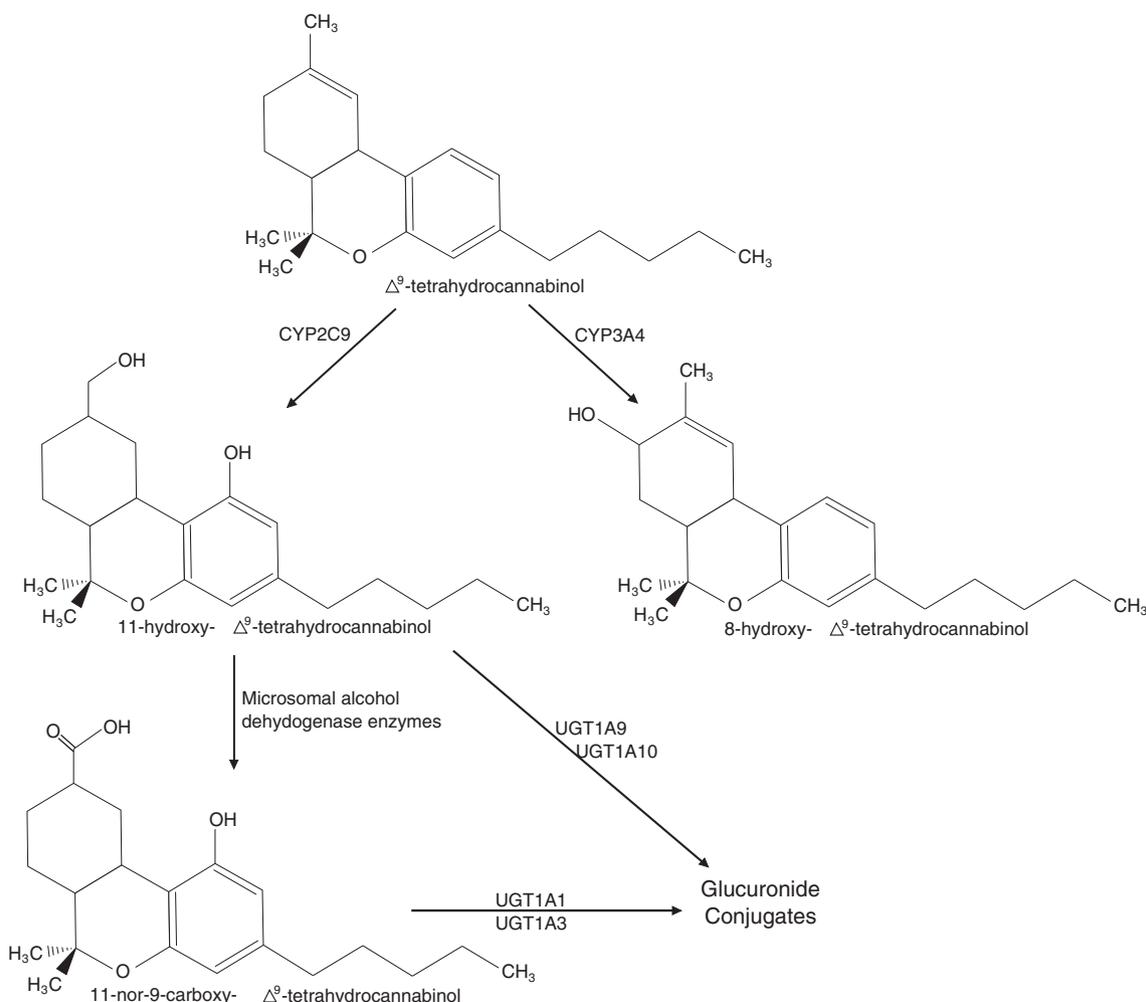


Fig. 2. In humans, THC is metabolized predominantly by CYP2C9 to 11-OH-THC and subsequently by CYP and alcohol dehydrogenase enzymes to 11-nor-THC-COOH. CYP3A4 forms primarily the 8-OH-THC metabolite in humans, which is a minor elimination pathway for THC. 11-OH-THC and 11-nor-THC-COOH are glucuronidated by UGT1A family enzymes.

UGT1A3 (11-nor-THC-COOH) (Mazur et al., 2009). As the 11-nor-THC-COOH, together with its acyl glucuronide conjugate, account for the majority (30–65%) of THC elimination (Glaz-Sandberg et al., 2007), altered CYP and UGT activity, as occurs during pregnancy, may significantly alter THC and metabolite exposures and pharmacology. Of note, the route of THC consumption will also alter the exposures to the metabolites. Following iv administration of THC or smoking of cannabis, 11-OH-THC plasma concentrations are much lower than those of THC (Grotenhermen, 2003; Wall et al., 1983) declining with the same half-life as THC (25–33 h). By contrast following oral THC administration, 11-OH-THC plasma concentrations can exceed those of THC but decline with a shorter half-life (12h) than THC suggesting that most of 11-OH-THC is formed during first pass in the liver (Grotenhermen, 2003; Wall et al., 1983). As 11-OH-THC has been suggested to penetrate the brain much better than THC, it is possible that 11-OH-THC contributes to the pharmacological effects observed after oral cannabis or THC consumption. Because of the low clearance of 11-nor-THC-COOH (5.5 L/h), it is the main circulating compound following any route of THC administration (Glaz-Sandberg et al., 2007). Overall, these data show that the route of consumption of THC may result in distinctly different exposures and pharmacology. Further research is needed to determine the role of peak concentrations, duration of exposures and role of metabolites in THC pharmacology and toxicity.

### 6.1. THC pharmacokinetics in preclinical species

The disposition of THC has been studied in several animal model systems, including mice, rats, rabbits, dogs and nonhuman primates (Freudenthal, Martin, & Wall, 1972; Garrett & Hunt, 1977; Ginsburg, Hrubá, Zaki, Javors, & McMahon, 2014; Leuschner, Harvey, Bullingham, & Paton, 1986). These species generally reproduce the distribution kinetics observed in humans characterized by a relatively rapid distribution phase and long terminal half-life predominantly driven by the extensive distribution of THC into adipose tissue. Unfortunately, due to low analytical sensitivity in the early studies, and oftentimes insufficient duration of sample collection, the terminal half-lives of THC have not yet been thoroughly characterized in most species. A terminal half-life of 8 days was reported in dogs based on radiolabeled THC (Garrett & Hunt, 1977) while terminal half-lives ranging from 41 h to 76 h were documented in rabbits (Leuschner et al., 1986). In rodents, the terminal half-life of petroleum ether extractable THC related radioactivity was 21 h (Klausner & Dingell, 1971). It is likely that these terminal half-lives measured based on radioactivity include both THC and its non-polar metabolites and hence may provide misleading estimates of the half-life of THC in different species. As such, these half-lives should be interpreted with caution and not compared across species.

In contrast to human studies, information on THC metabolite disposition in preclinical species is sparse. The formation of 11-OH-THC is largely conserved across species as a major metabolite and consistent with data in humans, rat CYP2C enzymes (CYP2C11 in males and CYP2C6 in females) predominantly form the 11-OH-THC (Narimatsu et al., 1990, 1992). THC is almost exclusively eliminated via metabolism in rats, and similar to what is observed in humans, only 10% of the dose is excreted in urine. The majority of the iv dose of THC is found in feces as metabolites of THC (Klausner & Dingell, 1971). The role of biliary secretion in rats was confirmed using the isolated perfused rat liver model in which 90% of the perfused THC radioactivity was secreted into the bile. Studies in rats also suggested that enterohepatic circulation of THC metabolites may occur (Klausner & Dingell, 1971). Whether the metabolite exposures in rats reflect those observed in humans is not known. In nonhuman primates (rhesus macaque monkeys) exposed to 0.1 mg/kg iv dose, the plasma disposition of THC is similar to humans and the human metabolites 11-OH-THC and 11-nor-THC-COOH are also detected. 11-nor-THC-COOH is present in the monkey plasma at similar concentrations as 11-OH-THC, but unlike in

humans, the concentrations of 11-nor-THC-COOH are lower than THC in monkey plasma. The difference could be due to either lower efficiency of 11-nor-THC-COOH formation in the monkey compared to humans or higher clearance of the 11-nor-THC-COOH in the monkey. These minor species differences are, however, unlikely to affect the validity of the model species as 11-nor-THC-COOH is an inactive metabolite. Together, these studies show that we still have only a basic understanding of THC disposition and metabolism in both human and relevant preclinical model systems and further studies are urgently needed in this area.

### 6.2. THC pharmacokinetics during pregnancy and maternal-fetal distribution

While THC and its metabolites can be reliably detected in meconium and infant urine as an indicator of maternal cannabis use in humans, very few studies have been conducted to evaluate maternal-fetal disposition of THC and its metabolites following maternal smoking or oral consumption of THC products. In addition, infant exposure to THC via passive smoking or via breastmilk has not been systematically assessed. THC is highly lipophilic (logP 6.97 for THC, 5.7 for 11-OH-THC and 5.2 for 11-nor-THC-COOH) and as such readily crosses the placenta but fetal exposure may be limited to some degree by active transport in the placenta. Observational studies in a small number of humans have demonstrated that THC distributes into the fetal compartment and readily crosses the placenta, although the overall extent of fetal distribution in humans has not been evaluated (Blackard & Tennes, 1984; Boskovic, Klein, Woodland, Karaskov, & Koren, 2001). Interestingly, based on meconium and hair samples of monozygotic and dizygotic twins, it was shown that THC levels in dizygotic twins can be discrepant (i.e. observed in one fetus but not in the other). This result suggests that placental and possibly fetal factors are the predominant factors controlling fetal THC exposures at constant maternal exposure.

While some epidemiological studies have incorporated maternal measures of exposure in the design of their study, none have generated data on the relationship between maternal and fetal measurements of cannabis exposure. Some initial information on this topic has been provided by a study of Spanish women undergoing voluntary pregnancy terminations (Falcon et al., 2012). In this study, samples of maternal hair, placenta and fetal tissue were collected from participants and THC and THC-COOH were detected in maternal hair. Placental THC tissue concentrations averaged  $196.8 \pm 110.1$  ng/g (range 101.1–432.8), while the mean THC level in fetal remains was  $118.5 \pm 97.9$  ng/g (range from 3.9 to 281.7). These results document the transplacental passage of THC during the embryonic and fetal period but are not sufficient to establish a quantitative relationship between maternal cannabis exposure and fetal cannabis levels.

To date, no studies on the disposition of THC in humans during pregnancy have been conducted. Based on the significant contribution of CYP2C9 and CYP3A4 to THC metabolism and their known increase in expression during pregnancy (Isoherranen & Thummel, 2013), one could suggest up to 2-fold increase in THC clearance during pregnancy and potentially a decrease in THC exposure for a given consumption level. However, one cannot easily predict that maternal or fetal exposures to the metabolites of THC are also increased during pregnancy as a result of enhanced CYP activity, as we still do not know how pregnancy may change the metabolite glucuronidation and clearance. Hence, more studies are needed to mechanistically characterize the elimination pathways of THC and its metabolites in humans and to predict whether pregnancy-mediated changes in maternal THC disposition are of clinical significance.

In preclinical model systems of human developmental exposure, fetal concentrations are generally lower than those observed in the mother and may be dependent on route of administration. In pregnant mice, fetal liver concentrations of THC and its metabolites were similar to maternal liver based on autoradiograms that were taken 2 h after iv

dosing of THC (Freudenthal et al., 1972). When THC was administered orally, a similar analysis yielded higher concentrations in the fetus than maternal liver. However, presence of THC metabolites may confound these measurements. In contrast, in pregnant female dogs administered 0.5 mg/kg THC iv, maternal brain concentrations of THC related radioactivity were 3-fold higher than those observed in the fetal brains 30 min after dosing, although overall distribution of THC to maternal and fetal brain regions was similar (Martin, Dewey, Harris, & Beckner, 1977). In pregnant rats given chronic oral doses of THC at 15 mg/kg or 50 mg/kg throughout gestation, fetal THC concentrations reached only 9–13% of the maternal concentrations and fetal exposure to THC increased proportionately to the dose (Hutchings, Martin, Gannagaris, Miller, & Fico, 1989). However, due to the distribution kinetics of THC to the fetus and the potential slow elimination from the fetus, these single point concentrations may not reflect the overall fetal exposure. In pregnant ewes (~130 days gestation) exposed via inhalation to cannabis smoke containing 3.2% THC, maternal THC concentrations peaked rapidly at 10 min after inhalation (Abrams et al., 1984) while the concentrations in fetal blood peaked at 90 min. Consistent with the rodent data, the fetal concentrations remained lower than those in the mothers during the 24 h study indicating potential placental efflux transport of THC. In 3 pregnant rhesus macaque monkeys administered iv doses of 0.3 mg/kg THC, distribution to the fetal compartment was faster than that observed in the ewes (Bailey, Cunny, Paule, & Slikker, 1987) and the fetal plasma THC concentrations peaked at 15 min after iv administration. Consistent with all other species, the fetal concentrations reached only 28% of the concentrations observed in maternal plasma at this sampling time point. **Alarming, the half-life of THC in the fetal compartment appears longer than that in the dam with fetal THC concentrations reaching those of maternal plasma at 180 min after dosing** (Bailey et al., 1987). Unfortunately, this study does not provide data on longer periods of sampling which prevents full pharmacokinetic analysis of this pattern. However, this study measured fetal tissue THC concentrations and found that the thymus, adrenals and bile had the highest THC concentrations at 3 h after THC dosing to the pregnant dam (about 5 times those observed in fetal plasma) (Bailey et al., 1987). In other tissue samples, such as fetal brain, liver and kidney, THC concentrations were either similar or slightly higher than those observed in fetal plasma. Together, these results indicate a broad distribution of THC to fetal tissues. The studies in rhesus macaques also show that 11-nor-THC-COOH is not detectable in the fetal plasma and that the maternal concentrations of 11-nor-THC-COOH were lower than those of THC up to 180 min post-exposure.

These preclinical animal studies show that THC concentrations in the fetus appear lower than in the mother and thus suggest that active transport (active efflux) in the placenta limits fetal exposure to THC. However, most of the studies are limited to single time points and further detailed studies are needed to carefully characterize the rate and extent of THC distribution into the fetus. For example, it is critical to understand whether peak concentrations in the fetal brain are similar to those observed in the mother and whether the exposure in the fetus is prolonged in comparison to the mother even after single doses of cannabis. In addition, mechanistic studies are needed to identify placental transporters responsible for THC transport to support our understanding of potential interindividual variability in THC distribution into the fetus and to identify individuals at high-risk for fetal THC toxicities. Variability in maternal-fetal THC disposition and transport may be partially responsible for inconsistent reports of neurodevelopmental effects from studies of THC exposure during pregnancy, outlined in detail below.

## 7. Impact of prenatal cannabis exposure on neurodevelopmental outcomes in humans

There are numerous publications focused on the reproductive and developmental effects of cannabis. Excellent review articles describing

a range of reproductive effects in both males and females have been published recently and will not be reviewed here (Brents, 2016; du Plessis et al., 2015).

Early chemical or drug exposure can result in subtle injuries to the developing CNS that are expressed as changes in postnatal development (Bellinger, Matthews-Bellinger, & Kordas, 2016; Grant & Rice, 2008). Over the past four decades, a number of prospective studies have found changes in the developmental trajectory of children prenatally exposed to cannabis. The demographic characteristics of subjects in these studies as well as exposure and outcome measures are summarized in Table 2. Most studies were conducted in urban environments with economically-disadvantaged families. The most common metric to estimate use of cannabis during pregnancy is maternal self-report on frequency of use (e.g. # joints/day), while fewer studies have collected biological samples to more accurately estimate real-world levels of exposure. **Much of what is known about maternal cannabis use and child development is based on data collected from 3 longitudinal birth cohort studies; the Ottawa Prenatal Prospective Study, the Maternal Health Practices and Child Development Project and the Generation R Study** (McLemore & Richardson, 2016), but other longitudinal and cross-sectional studies **focused on the developmental neurotoxicity of this compound have also made important contributions.** **The wide variation in cannabis potency and individual smoking habits make the interpretation of the developmental literature challenging but a careful review reveals certain common themes surrounding the fetal risks associated with prenatal exposure.** Accordingly, we have separated the developmental outcomes of cannabis exposure into four domains: **physical growth/maturation, neonatal behaviors, cognition and psychological health/adaptive behavior.**

As mentioned above, a frequently used approach to the measurement of cannabis exposure is maternal self-report of use during pregnancy. This approach, while commonly employed, may not provide an accurate evaluation of in utero exposure due to underreporting of drug use by pregnant women (Garg et al., 2016). Reluctance to report cannabis use is commonly linked to feelings of guilt, the fear of being arrested and concern over repercussions in child custody cases. This makes it difficult to characterize biologically-based dose-response relationships for cannabis-related developmental effects. Few studies have collected biological samples to augment maternal self-report estimates of use and for those that have, the information has been primarily used to determine incidence of drug exposure, not dose-response relationships. The laboratory analysis of cannabis exposure from biological mediums most often relies on samples of maternal urine and blood (Musshoff & Madea, 2006), but more recently, maternal hair, placenta and fetal meconium have been utilized (Falcon et al., 2012).

### 7.1. Physical growth and maturation

In utero exposure to cannabis does not typically result in congenital birth defects (Linn et al., 1983; van Gelder et al., 2010; Warner, Roussos-Ross, & Behnke, 2014) and there is no phenotypic signature of this compound in newborns. Effects on physical growth at birth and during the neonatal period have been reported in some studies (see below) but not others (Bada, Reynolds, & Hansen, 2006; Conner, Carter, Tuuli, Macones, & Cahill, 2015; van Gelder et al., 2010). In a study of maternal cannabis use and effects on fetal growth, decrements in birthweight and neonatal head circumference were associated with prenatal exposure but only when data were restricted to women with a positive urine assay for cannabis (Zuckerman, Amaro, & Cabral, 1989). When maternal self-report data were used for analysis, no significant relationship between cannabis exposure and early growth was detected. In a retrospective records study, maternal use of cannabis, as determined by either self-report or a positive urine assay for THC, was associated with decrements in fetal growth (e.g. small-for-gestational age) and an increase of 54% in neonatal intensive care unit admissions (Warshak et al., 2015). This investigation is particularly noteworthy for

**Table 2**  
Results from human epidemiological studies of in utero cannabis exposure. Number of children is the number of original enrolled participants. Exposure typically measured by self-described use or number of joints/times smoked per week. † indicates significant increase in outcome measure. ↓ indicates significant decrease in outcome measure. ↔ indicates no significant change in outcome measure. SES: socioeconomic status; NICU: neonatal intensive care unit.

Study name	Location	# of children	Maternal demographics	Exposure	Growth	Behavior	Cognition	Physical health/adaptive behavior
<b>Ottawa Prenatal Prospective Study (OPPS)</b> Fried (1980) Fried, Buckingham, & Von Kulmiz (1983); Fried et al. (1987); Fried, O'Connell, et al. (1992), Fried, Watkinson, et al. (1992), Fried et al. (1997, 1998, 1999, 2001), Fried et al. (2003) Fried & O'Connell (1987) Fried & Watkinson (1988, 1990, 2001) Fried & Smith (2001) Smith et al. (2006) Smith et al. (2016)	Ottawa, CA	190	Middle SES Predominantly Caucasian/white	Irregular ( $\leq 1$ ) Moderate (2-5) Heavy ( $> 5$ )	↓ birthweight (neonate) ↔ growth (13-18 years) ↔ pubertal milestones (13-18 years)	↓ stimuli habituation (newborn) ↑ tremors (newborns) ↑ startle (newborn)	↓ verbal/memory processing (3-4, 9-12, 13-18 years) ↔ verbal/memory processing (5-6 years) ↔ reading/language (9-12 years) ↔ IQ (9-12, 13-18 years) ↓ attention (9-12 years) ↓ visual analysis/hypothesis testing (9-12, 13-18 years) ↓ impulse control (9-12 years) ↑ activity in frontal, occipital, parahippocampal gyri and cerebellum (8-22 years) ↓ activity in right anterior and middle frontal gyri (8-22 years)	
<b>Maternal Health Practices and Child Development Project (MHPDCD)</b> Scher et al. (1988) Richardson, Day, & Taylor (1989), Richardson et al. (1995), Richardson et al. (2002) Dahl et al. (1995) Day et al. (1994), Day et al. (2011) Goldschmidt et al. (2000); Goldschmidt et al. (2004); Goldschmidt et al. (2008), Goldschmidt, Richardson, Larkby, & Day (2016) Gray et al. (2005) Leech et al. (2006)	Pittsburgh, PA, USA	763	Lower SES Half sample Caucasian/white, half black Most single	Light (0-2.8) Moderate (2.8-7.0) Heavy use ( $> 7.0$ )	↔ growth (5-6 years)	↑ nighttime arousal (newborns, 3 years) ↓ time sleeping (newborn, 3 years) ↔ reflexes (newborn)	↑ cognitive performance/IQ (1-2, 3, 5-6 years) ↓ attention (3-4 years) ↓ verbal-memory processing (3-4 years)	↑ depression symptoms (6-10 years) ↑ anxiety symptoms (6-10 years) ↔ drug use (15-18 years) ↓ impulse control (6-10 years) ↑ hyperactivity (6-10 years) ↑ delinquent behavior (6-10, 11-16 years) ↑ aggressive behavior in females (18 months) ↑ thickness of frontal cortex (6 years)
<b>Generation R</b> Jaddoe et al. (2008, 2010) El Marroun et al. (2009), El Marroun et al. (2011), El Marroun et al. (2015) Zuckerman et al. (1989) Astley & Little (1990)	Rotterdam, NL  Boston, MA, USA Seattle, WA, USA	4000  1226 136	Middle-upper SES Predominantly Dutch/European; multi-ethnic Most married  Middle SES Predominantly Caucasian/white Most married Middle to slightly above average SES Predominantly Caucasian/white Most married Low-Ave SES Mostly nonwhite	Not defined  Not defined # days used during pregnancy and lactation  < 1 × /week ≥ 1 × /week	↓ birthweight (newborn) ↓ head circumference (fetal) ↓ birthweight (newborn)  ↓ birthweight (newborn) ↓ birth length (newborn)		↓ attention in females (18 months) ↓ thickness of frontal cortex (6 years)  ↔ early cognition (1-2 years)	
<b>Avon Longitudinal Study of Parents and Children</b> Fergusson et al. (2002) Zammit et al. (2009) Mathews et al. (2014)	Avon, UK	12,129	Middle to slightly above average SES Predominantly Caucasian/white Most married Low-Ave SES Mostly nonwhite	Light (0-2.8) Heavy use ( $> 7.0$ )	↓ birthweight (newborn) ↓ foot length (newborn)			↑ Tourette syndrome (12 years) ↔ psychosis-like symptoms (12 years)
Hurd et al. (2005)	Pittsburgh, PA, USA	139	Low SES Predominantly African	Mean = 2.8				
Noland et al. (2005)	Cleveland, OH, USA	330						↓ attention (4 years)

(continued on next page)

Table 2 (continued)

Study name	Location	# of children	Maternal demographics	Exposure	Growth	Behavior	Cognition	Physical health/ adaptive behavior
van Gelder et al. (2010)	United States	5871	American Mostly single Lower SES Predominantly Caucasian/white and nonwhite Hispanic	Not defined	↔ birthweight (newborn) ↔ gestational age (newborn)			
Frank et al. (2014)	Boston, MA, USA	157	Lower-Middle SES Predominantly non- Hispanic black	Light and heavy users				↔ early cannabis use (18 years) ↔ substance abuse disorders (18 years)
Saurel-Cubizolles et al. (2014)	France	13,454	Middle SES Predominantly Caucasian/white	Less than once a month vs once a month or more	↓ birthweight (newborn)			
Vamer et al. (2014)	United States	3224	Middle SES	Positive toxicology assay	↑ stillbirths (newborn)			
Conner et al. (2015)	St Louis, MO, USA	8106	Lower SES Predominantly non- Hispanic black	At least once during pregnancy vs did not use	↔ birthweight (newborn)	↔ APGAR score (newborn)		
Warshak et al. (2015)	Cincinnati, OH, USA	6488	Predominantly non- Hispanic black	Not defined	↓ birthweight (newborn)	↑ NICU admissions (newborn)		

its enrollment criteria and women who used tobacco during pregnancy were not included in the study population. Because subjects were classified only as cannabis users or nonusers, it is not possible to glean information about dose-response relationships for these effects. Exposure-related changes in early growth were also detected in a study where fetal meconium, maternal self-report and urine were collected from women undergoing voluntary saline-induced abortions (Hurd et al., 2005). The anthropometric examination of fetuses of varying gestational ages revealed a significant exposure-related decrement in fetal foot length, a standard marker of physical maturation at birth. This effect was observed as early as mid-gestation (weeks 17–22) and statistical trends in the data showed that offspring of women who were heavy users of cannabis during early pregnancy (~1 or more joints/day) were most likely to be affected. In contrast, a study of over 8000 women that relied on either self-report or a positive THC urine screen to determine fetal exposure found no relationship between maternal cannabis use during pregnancy and a composite score of neonatal morbidity composed of birthweight, APGAR score (health status of newborn immediately after birth) and umbilical artery pH levels (Conner et al., 2015).

Changes in physical growth and development have also been documented in studies relying solely on measures of maternal self-report to estimate cannabis use. Ultrasound images collected from thousands of pregnant women demonstrated that maternal cannabis use was not related to adverse neonatal outcomes, such as perinatal death, but was associated with small but detectable reductions in birthweight and fetal head circumference (El Marroun et al., 2009). Changes in weight and growth trajectories were primarily observed in infants whose mothers reported using cannabis on a weekly or daily basis. In separate studies using maternal self-report, the use of cannabis during pregnancy has been linked to an increased risk of having a small-for-gestational age infant (Saurel-Cubizolles, Prunet, & Blondel, 2014) and reductions in birthweight (Fergusson, Horwood, & Northstone, 2002).

Our reading of the literature on prenatal cannabis exposure and early physical development indicates mixed results. Despite some research supporting a significant relationship between cannabis use during pregnancy and decrements in fetal growth, there is no strong evidence that cannabis has a long-term negative impact on physical maturation (Fried & O'Connell, 1987). Longitudinal tracking of children with a history of prenatal cannabis exposure revealed normal physical growth trajectories at the time of school entrance (age 5–6) and during adolescence (Day, Richardson, Geva, & Robles, 1994; Fried, Watkinson, & Gray, 1999) and key pubertal milestones such as age at menstruation in females and shaving in males were also not affected (Fried et al., 1999; Fried, James, & Watkinson, 2001).

7.2. Neonatal behaviors

Neurobehavioral effects of in utero cannabis exposure have been detected in some studies during the newborn period. Infants born to moderate and heavy users of cannabis during pregnancy (≥ 2 joints/week, maternal self-report) showed increased tremors/startles and poorer habituation to visual stimuli (Fried, 1980; Fried, Watkinson, Dillon, & Dulberg, 1987). The authors note that these behavioral findings are consistent with a mild narcotic withdrawal syndrome and may portend exposure-related changes in CNS functioning. Some women who decreased use or quit cannabis during pregnancy showed a reduced risk of delivering an infant with clinical symptomatology. Gestational cannabis exposure has also been associated with changes in postnatal cortical activity. Specifically, a study of neonatal electroencephalography (EEG) sleep patterns found that in utero exposure to cannabis was associated with increased body movements and decreased time in a quiet sleep state (Scher, Richardson, Coble, Day, & Stoffer, 1988). This effect was most widely observed in infants born to women who used cannabis on a daily basis. When children in this cohort reached 3 years of age, a similar pattern of EEG sleep disturbances was

documented (Dahl, Scher, Williamson, Robles, & Day, 1995). These results suggest that the neurophysiological mechanisms that control infant/toddler arousal and sleep cycling may be disrupted by cannabis use during pregnancy. This behavioral change in affected infants may reflect subtle chemical injury to the brain stem, particularly in neurons that comprise the raphe nuclei. The long-term significance of these effects, if any, is unknown.

### 7.3. Cognition

Learning and memory are perhaps the most consequential outcome measures in developmental cannabis research but studies are relatively few and findings are inconsistent. With few exceptions (e.g. Noland et al., 2005), the central limitation of studies investigating neurocognitive endpoints is their methodological reliance on maternal self-report of cannabis use during pregnancy to estimate fetal exposure. Several studies focused on early cognitive outcomes have reported that maternal cannabis use during pregnancy was not related to performance on infant tests of mental development (Astley & Little, 1990; Fried & Watkinson, 1988). Other studies however, have reported a significant decline in early cognitive performance (Richardson, Day, & Goldschmidt, 1995). The reduction in test scores for 9 month-old infants with the highest levels of maternal cannabis use (> 1 joint per day) was a disquieting 10 points, providing some evidence for dose-related effects in early mental test performance. When these infants were re-evaluated at 19 months using the same exam, fetal THC exposure was no longer related to language and cognitive scores.

During the preschool period of development (3–4 years), results from child assessment studies have demonstrated that prenatal cannabis exposure is related to adverse effects on sustained attention, short-term memory and verbal processing, although it is important to note that decrements in performance were frequently subtle and limited in scope (Day et al., 1994; Fried & Watkinson, 1990; Noland et al., 2005). At school age (5–6 years), one prospective, birth-cohort study found no evidence of an adverse effect of prenatal cannabis exposure on any cognitive outcome, including global intelligence quotient (IQ) scores (Fried, O'Connell, & Watkinson, 1992). Additional testing with these children did, however, reveal small deficits in sustained attention and increased levels of impulsivity and hyperactivity (Fried, Watkinson, & Gray, 1992). The number of lapses in attention (omission errors) during a vigilance task was greatest in children born to heavy users of cannabis during pregnancy (> 6 joints/week). In contrast, a separate longitudinal study found that heavy maternal cannabis use during pregnancy (~1 or more joints/day) was associated with diminished scores on a standardized IQ test at age 6, including deficits in short-term memory processing, and the effects varied by trimester of exposure (Goldschmidt, Richardson, Willford, & Day, 2008).

By middle childhood and adolescence, a pattern of neurocognitive results highlights the resiliency of global IQ and the possible sensitivity of attention and memory to prenatal cannabis exposure. Between 9 and 12 years of age, the data suggest that fetal cannabis exposure is not associated with composite IQ scores or performance on broad-based reading and language exams (Fried, Watkinson, & Gray, 1998; Fried, Watkinson, & Siegel, 1997). However, the heavy use of cannabis during pregnancy (~1 or more joints/day) has been linked with decreased scores on tests of academic achievement, impulse control, visual analysis/hypothesis testing and learning/memory in exposed children (Fried et al., 1998; Goldschmidt, Richardson, Cornelius, & Day, 2004; Richardson, Ryan, Willford, Day, & Goldschmidt, 2002). Longitudinal tracking of a birth cohort through adolescence (13–16 years) demonstrated that global IQ scores remain unaffected by fetal cannabis exposure but certain aspects of cognition, particularly those related to sustained attention and visual working memory, may continue to be negatively impacted (Fried, Watkinson, & Gray, 2003).

Two cannabis research programs have paired behavioral protocols with in vivo visualization of the brain using functional magnetic

resonance imaging (fMRI) (Smith et al., 2016; Smith, Fried, Hogan, & Cameron, 2004, 2006). Prenatal cannabis exposure in subjects ranging in age from 8 to 22 was not related to decrements in performance on a visuospatial cognitive task but fMRI scans revealed increased neural activity in the frontal gyri, parahippocampal gyrus, occipital gyrus and cerebellum and decreased activity in the right inferior and middle frontal gyri in exposed subjects. Brain imaging techniques were also utilized in a study of 6 year old children to investigate cannabis-related changes in brain morphology (El Marroun et al., 2015). Using MRI technology to compare prenatally exposed and nonexposed children, no differences in brain volume were detected but there were significant differences in cortical thickness. While the mechanism and functional significance of these findings remains unknown, thicker cortices in the frontal regions of both hemispheres suggest exposure-driven changes in the maturation of the frontal cortex.

A collective examination of the body of knowledge on fetal cannabis exposure and childhood neurocognitive development suggests that heavy maternal use of cannabis during pregnancy does not result in a reduction in global IQ but rather, may act to diminish performance on tasks that require the harnessing and implementation of executive function skills; a top-down set of cognitive processes that are used to manage attention, exert inhibitory control and plan goal-directed behavior (Fried & Smith, 2001). Functional losses in executive function skills may place exposed children at a disadvantage for long-term success in school, the community and the workplace (Diamond & Lee, 2011).

### 7.4. Psychological health and adaptive behavior

On the continuum of cannabis-related developmental neurotoxicity, there is growing evidence that psychological health may be particularly vulnerable to the adverse effects of in utero exposure. A study of infant social behavior demonstrated that maternal cannabis use during pregnancy was related to a significant increase in aggressive behavior and attentional problems in 18 month-old girls (El Marroun et al., 2011). In middle childhood, prenatal exposure was predictive of damaging or maladaptive behaviors such as increases in hyperactivity, impulsivity and delinquent behavior (Goldschmidt, Day, & Richardson, 2000). In children born to heavy cannabis users (~1 or more joints/day), the risk of scoring in the borderline clinical range for delinquent behavior was 2.4 times that of children born to nonusers. Increased reporting of depressive symptoms and anxiety has also been documented in children with a history of heavy prenatal cannabis exposure during the first trimester (Gray, Day, Leech, & Richardson, 2005; Leech, Larkby, Day, & Day, 2006).

A similar pattern of results has been observed in adolescence where rates of delinquency varied by prenatal exposure history (41% non-exposed, 50% light to moderate exposure and 61% heavy exposure) (Day, Leech, & Goldschmidt, 2011). It is useful to note that cannabis-exposed children who expressed depressive symptoms at age 10 were at the highest risk of reporting delinquent behaviors during puberty. In separate studies focused on mental health and adaptive behavior during young adulthood, maternal cannabis use during pregnancy was not predictive of non-clinical psychopathology (Zammit et al., 2009) but was related to an increased risk for diagnosis of Tourette syndrome or chronic tic disorder (Mathews et al., 2014). Recent studies have suggested that prenatal exposure predicts the early onset of cannabis use in young adults (22 years of age), but this effect was primarily observed in subjects born to heavy users (~1 or more joints/day) (Sonon, Richardson, Cornelius, Kim, & Day, 2015). While an intriguing result, a positive relationship between prenatal cannabis exposure and the early onset of cannabis use was not found in a study that utilized both maternal self-report and infant meconium to measure levels of gestational exposure (Frank et al., 2014).

**Table 3**  
 Summary of results from preclinical animal studies of prenatal and perinatal THC exposure. ↑ indicates significant increase in outcome measure. ↓ indicates significant decrease in outcome measure. ↔ indicates no significant change in outcome measure. GD: gestational day; im: intramuscular; iv: intravenous; ip: intraperitoneal; PND: postnatal day; po: oral administration; PTZ: phenothiazine; sc: subcutaneous.

Study	Species/strain	n	Dose	Route	Age at dose	Physical growth and maturation	Cognition	Emotionality and adaptive behavior	Physical activity
Gianusos and Abbatiello (1972)	Wistar rats	75	250 mg/kg cannabis extract	sc	GD 8–11		↓ learning/memory via Lashley III maze (PND 65)		
Borgen et al. (1973)	Wistar rats	48	10 mg/kg THC	sc	GD 10–12	↓ birthweight (PND 0)			↑ locomotion via open field (PND 9) ↔ locomotion via elevated plus maze (PND 21) ↓ locomotion via open field (PND 7) ↔ locomotion via open field (PND 1, 14)
Fried (1976)	Wistar rats	32	~19 mg/kg	Inhalation	GD 1–19	↓ birthweight (PND 0) ↓ bodyweight (PND 0–100) ↔ postnatal mortality			
Abel (1984)	Long Evans hooded rats	100	10 and 15 mg/kg crude cannabis extract	po	GD 3–23	↓ birthweight (PND 0) ↔ litter size (PND 0) ↑ postnatal mortality (PND 21)			
Abel (1984)	Long Evans hooded rats	10–14/test	50–150 mg/kg THC	po	GD 1–23	↓ birthweight (PND 0)			↔ rotarod performance (PND 36)
Asch and Smith (1986)	Rhesus macaques	47	2.5 mg/kg	im	GD 156–163	↔ birthweight (PND 1)			
Brake et al. (1987)	Wistar rats	38	15 and 50 mg/kg THC	po	GD 8–22				↔ locomotion via open field (PND 2–32) ↔ nipple attachment (PND 2–14)
Hutchings et al. (1987), Hutchings et al. (1989), Hutchings, Brake, et al. (1991), Hutchings, Fico, et al. (1991)	Wistar rats	35	15 and 30 mg/kg THC	po	GD 2–22	↔ birthweight (PND 1) ↑ postnatal mortality		↔ startle response	
Navarro et al. (1995)	Wistar rats	40/test	Hashish w/ ~20 mg/kg THC	po	GD 5–PND 1	↔ weight (PND 1–70) ↔ litter size (PND 1)			↑ locomotion via open field (PND 1, 20)
Rubio et al. (1995)	Wistar rats	40/test	5 mg/kg THC	po	GD 5–PND 1				↑ locomotion via open field (PND 70) ↑ locomotion in females via elevated plus maze (PND 70) ↑ immobility via open field (PND 70) ↓ locomotion via open field (PND 70)
Moreno et al. (2003)	Wistar rats	192	0.1–2 mg/kg THC	po	GD 5–PND 24				
O'Shea and Mallet (2005)	Wistar rats	12	5 mg/kg THC	sc	PND 4–14	↓ bodyweight (PND 4–52)	↔ motivation and learning via spatial discrimination (PND 59) ↓ working memory via delayed alternation (PND 59)		

(continued on next page)

Table 3 (continued)

Study	Species/strain	n	Dose	Route	Age at dose	Physical growth and maturation	Cognition	Emotionality and adaptive behavior	Physical activity
Campolongo et al. (2007)	Wistar rats	23–28	5 mg/kg THC	po	GD 15–PND 9	↔ birthweight (PND 1) ↔ litter size (PND 1) ↔ postnatal mortality	↓ long-term memory via inhibitory avoidance (PND 80) ↓ short term memory via social discrimination (PND 80)	↑ anxiety/fear via open field activity (PND 90) ↓ anxiety via social interaction (PND 90) ↔ depression via forced swim test (PND 90)	
Newsom and Kelly (2008)	Long-Evans hooded rats (males only)	24	2 mg/kg THC	sc	2 × /day GD 1–22 and PND 2–10	↔ birthweight (PND 1) ↔ postnatal mortality		↑ anxiety via ultrasonic vocalizations (PND 12) ↑ anxiety via elevated plus maze (PND 80) ↓ social play (PND 35)	↔ locomotion via elevated plus maze (PND 80)
Trezza et al. (2008)	Wistar rats	50	5 mg/kg THC	po	GD 15–PND 9	↔ birthweight (PND 1) ↔ litter size (PND 1) ↔ postnatal mortality	↓ attention (PND 55) ↓ learning/memory in males via active avoidance (PND 45)		↓ locomotion activity when challenged with amphetamines (PND 60)
Silva et al. (2012)	Sprague Dawley rats	40–96	0.15 mg/kg THC	iv	Before conception to GD 21				↓ motor skills activity (PND 60) ↓ time to seizures when administered PTZ (PND 60)
de Salas-Quiroga et al. (2015)	WT mice	10–24	3 mg/kg THC	ip	GD 12.5–16.5				
Benevenuto et al. (2017)	Balb/C mice	20	5 mg/kg cannabis	Inhalation	Daily from conception	↓ fetal weight (GD 18.5) ↑ placental weight (GD 18.5) ↑ number male pups (GD 18.5) ↓ male fetal:placental weight ratio (GD 18.5)			

## 8. Insights from experimental work with preclinical species

Preclinical animal model systems provide an important bridge between brain and behavior, allowing for the identification of neural pathways and processes that underlie postnatal changes in exposed offspring (Thompson, Levitt, & Stanwood, 2009). One of the most important aspects of preclinical work is the ability to control many of the confounding environmental and genetic factors that can adversely affect neurodevelopmental outcomes in human populations; a condition that is essential to determining the independent contribution of drug exposure (Fried, 2002). Most investigations have used rodent models to study oral or subcutaneous (sc) routes of exposure but a small number of investigations have employed inhalation or intravenous dosing (see Table 3). THC doses in preclinical studies range from 0.1 to 150 mg/kg.

An oral dose of 5 mg/kg THC in rats is thought to correspond to moderate levels of drug exposure in humans (Garcia-Gil, Romero, Ramos, & Fernandez-Ruiz, 1999). Because comprehensive reviews of the behavioral and neuroendocrine effects of prenatal cannabis exposure in preclinical models are available (e.g. Campolongo, Trezza, Ratano, Palmery, & Cuomo, 2011), the present discussion of the rodent and primate literature is focused on studies which bring translational value to the human research findings outlined above.

### 8.1. Physical growth and maturation

The effects of THC on fetal growth and neonatal outcomes were among the first investigational topics to be addressed in preclinical modeling research. In a study of chronic oral THC exposure (2.4 mg/kg/day) that was undertaken in pregnant macaque monkeys, THC readily crossed the placenta but was not related to changes in fetal growth or infant birthweight (Asch & Smith, 1986). In an early and ground-breaking longitudinal research program using rats, gravid animals were exposed to THC via gastric intubation (15 or 50 mg/kg/day) on gestation day (GD) 2–22 and offspring were evaluated on a series of neurodevelopmental metrics (Hutchings, Morgan, Brake, Shi, & Lasalle, 1987). While there were significant increases in offspring mortality at both doses, broad-based behavioral testing (including the rest-activity cycle, latency to attach to a nipple and ontogeny of locomotor activity) did not reveal adverse effects of early THC exposure (Brake, Hutchings, Morgan, Lasalle, & Shi, 1987; Hutchings et al., 1987, Hutchings, Brake, et al., 1991, Hutchings, Fico, Banks, Dick, & Brake, 1991; Hutchings, Gamagaris, Miller, & Fico, 1989). In a recent inhalation mouse study, a dose of ~0.5 mg/kg/day THC smoke from GD 5.5 to 17.5 produced deficits in fetal growth and reduced birthweights in cannabis-exposed offspring (Benevenuto et al., 2017). Pups with a history of gestational THC exposure showed a surprising 9.9% drop in birthweight and significant decrements in the weight of lungs, brain, thymus, and liver. In general, male fetuses appeared more susceptible to cannabis-related disruptions in early physical growth. These results suggest that low-dose exposure to cannabis for periods as little as 5 min a day via inhalation can have a compromising effect on fetal development.

### 8.2. Cognition

Cognitive endpoints have also been evaluated in animals prenatally exposed to THC and like results from human studies, the evidence is evolving. While some results with animal models suggest adverse effects on learning and memory, it is important to remember that there are multiple studies that did not identify prenatal cannabis exposure as a risk factor for short- or long-term neurocognitive effects (e.g. Abel, 1984). The original discovery work in this field was plagued by methodological and translational issues that detracted from the overall significance of the experimental results, including the role of maternal toxicity in producing false-positives on behavioral assessments in exposed offspring (Hutchings & Dow-Edwards, 1991). With that said, impairments in learning abilities were among the earliest reported

effects of prenatal cannabis exposure (Fried, 1976; Gianutsos & Abbatello, 1972). More contemporary research with preclinical species has demonstrated that maternal oral exposure to 5 mg/kg/day THC during pregnancy produces measurable deficits in learning and short-term olfactory memory in exposed offspring (Campolongo et al., 2007). Cognitive impairments in exposed animals were accompanied by changes in cortical gene expression that suggest alterations in glutamatergic neurotransmission. In a recent mouse study targeting early CNS development, gravid mice were exposed to intraperitoneal (ip) injections of 3.0 mg/kg/day THC from GD 12.5 to 16.5 and offspring showed reductions in skilled motor activity and an increased vulnerability to seizures (de Salas-Quiroga et al., 2015). The authors theorize that fetal THC exposure may impede the normal development of corticospinal connectivity and increase seizure susceptibility by interfering with CB<sub>1</sub>R-dependent regulation of both glutamatergic and GABAergic neuron development.

Consistent with preclinical findings on physical growth and maturation, the effects of developmental THC exposure on cognition are more pronounced in males. Offspring of rats exposed to 0.15 mg/kg/day iv injections of THC throughout gestation showed reduced performance on a test of learning and long-term memory with the most pronounced deficits occurring in male offspring (Silva, Zhao, Popp, & Dow-Edwards, 2012). This investigation is commendable for tracking animals from weaning through adulthood and the findings suggest emergent memory processes may be particularly vulnerable to perturbation from in utero exposure. Additional evidence of cognitive-based impairments was obtained in a rat study using 5 mg/kg/day THC sc on GD 4–14, a period of major synaptogenesis and analogous to the 3rd trimester in humans (O'Shea & Mallet, 2005). As adults, exposed animals committed more errors and took longer to reach a level of proficient performance on a test of spatial learning and memory. Neurocognitive testing across studies with rats and mice have identified cognitive effects in exposed offspring that align with cognitive findings from prenatally exposed children, providing corroboration that memory-in-action, or working memory, may be the seat of cannabis-induced cognitive impairment.

### 8.3. Emotionality and adaptive behavior

Given that prenatal cannabis exposure may be associated with increased anxiety and depressive symptomatology in children, emotionality is an important outcome measure in studies with animals. Auditory startle in adulthood, a sensitive measure of CNS functioning, was not impaired in rat pups exposed to either 15 or 50 mg/kg/day THC via gastric intubation on GD 2–22 (Hutchings et al., 1991). Conversely, results from a longitudinal study of 2.5–5 mg/kg/day oral THC exposure from GD 15 to Postnatal day 9 in rats demonstrated that perinatal exposure to THC was associated with an increased number of adverse effects including ultrasonic distress calls from pups, inhibited social behavior during adolescence and anxiety-like symptoms during adulthood (Trezza et al., 2008). These findings led investigators to hypothesize that developmental exposure to cannabinoids may exert long-term effects on select brain regions that control emotional development and cognition (Trezza et al., 2012). Interestingly, changes in adult social behavior expressed as increased interactions with peers have been reported after gestational and early postnatal exposure to 2 mg/kg/day THC sc (Newsom & Kelly, 2008). The theoretical premise that cannabinoid exposure may impact the modulation of emotional states, including sociability, is bolstered by two research findings 1) CB<sub>1</sub>R are highly expressed in brain regions that regulate anxiety (e.g. cortex, hippocampus, lateral septum, nucleus accumbens, amygdala) and 2) the eCB signaling system controls the release of several neurotransmitters (e.g. serotonin, dopamine) involved with emotionality (Campolongo et al., 2011).

#### 8.4. Physical activity

Findings from modeling studies suggest that fetal exposure to THC may produce transient effects on postnatal physical activity (i.e. body movement). In rodent studies using oral doses of 5 or 10 mg/kg/day THC during gestation, increases (Borgen, Davis, & Pace, 1973; Mereu et al., 2003; Navarro, Rubio, & Rodriguez de Fonseca, 1995), decreases (Fried, 1976) and no changes (Brake et al., 1987; Trezza et al., 2008) in psychomotor activity levels have been documented. While results are inconsistent, some data suggest that prenatal exposure to cannabinoids may impact the development of brain areas involved in motor behavior, a finding that is relevant to the increased hyperactivity and impulsivity observed in exposed children and adolescents. Oral administration of THC from GD 5 to Postnatal day 24 in gravid rats led to significant changes in offspring physical activity levels at doses as low as 0.1 mg/kg (range 0.1–2 mg/kg/day) (Moreno, Trigo, Escuredo, Rodriguez de Fonseca, & Navarro, 2003). Offspring perinatally exposed to THC spent significantly more time in an immobile state and displayed reduced levels of activity in an open field test apparatus when compared to controls.

#### 9. Conclusions and directions of future research objectives

Much of the research conducted thus far on the mechanisms of THC toxicity has focused on understanding the MOA of THC and role of the eCB signaling system in the mature brain. Much less is known about the MOA mediating the effects of THC on the developing brain and whether glial cells play a role in the impact of THC and how this is influenced by additional phyto-CB. Almost nothing is known about the impact of phyto-CB on the developing brain when exposure occurs in combination with other drugs. There is a strong need to establish the fetal neurotoxic effects of phyto-CB exposure to properly evaluate the safety profile of cannabis use during pregnancy. In addition, a key gap in translating preclinical findings of cannabis toxicity to humans is the lack of detailed knowledge of the pharmacokinetics and maternal-fetal transfer mechanisms of THC and its metabolites in humans and in animal models. Detailed pharmacokinetic studies and quantitative modeling of cannabis pharmacokinetics are needed to develop methods that allow interspecies scaling and determination of human maternal and fetal exposures from spot urine or blood samples. It is also notable that no studies have been conducted to evaluate the potential fetal toxicity of the metabolites of THC and such studies are critically needed.

In terms of neurodevelopmental effects, the current evidence is inconsistent but certain patterns can be gleaned from the data. Cannabis does not act as a classical teratogen and is not associated with morphological abnormalities at birth. Fetal exposure has been associated with changes in physical growth and maturation early in life but long-term growth, including pubertal milestones, are unaffected. Global intelligence scores in children with a history of in utero cannabis exposure are typically not affected but aspects of cognition involved with executive functioning (e.g. attention, inhibitory control, planning) can be negatively impacted. Effects of exposure also include higher levels of depression and anxiety during adolescence, suggesting that psychological outcomes may be particularly sensitive to the disrupting influence of gestational cannabis exposure. Results from preclinical modeling studies have confirmed that in other mammalian species, fetal exposure to THC does not result in changes in long-term physical growth but may negatively impact certain aspects of cognition and heighten the occurrence of behaviors that are consistent with anxiety. While the neurodevelopmental effects of in utero cannabis exposure are subtle, they are persistent and have been observed in more than one species. Our overall conclusion is that there is a public health need for well-controlled scientific studies to elucidate the pattern of neurotoxicity that may be associated with fetal exposure and until such time more information is available, pregnant women should not assume that it is safe to use cannabis during pregnancy.

#### Conflict of interest statement

The authors declare that there are no conflicts of interest.

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#### References

- Abel, E. L. (1984). Effects of  $\Delta^9$ -THC on pregnancy and offspring in rats. *Neurobehavioral Toxicology and Teratology*, 6(1), 29–32.
- Abood, M., Sorensen, R., & Stella, N. (Eds.). (2012). *endoCANNABINOIDS: Actions at non-CB1/CB2 cannabinoid receptors* (24th ed.). Springer Science & Business Media.
- Abrams, R. M., Cook, C. E., Davis, K. H., Niederreither, K., Jaeger, M. J., & Szeto, H. H. (1984). Plasma delta-9-tetrahydrocannabinol in pregnant sheep and fetus after inhalation of smoke from a marijuana cigarette. *Alcohol and Drug Research*, 6(5), 361–369.
- Aguado, T., Monory, K., Palazuelos, J., Stella, N., Cravatt, B., Lutz, B., ... Galve-Roperh, I. (2005). The endocannabinoid system drives neural progenitor proliferation. *The FASEB Journal*, 19(12), 1704–1706. <http://dx.doi.org/10.1096/fj.05-3995fje>.
- Aguado, T., Palazuelos, J., Monory, K., Stella, N., Cravatt, B., Lutz, B., ... Galve-Roperh, I. (2006). The endocannabinoid system promotes astroglial differentiation by acting on neural progenitor cells. *The Journal of Neuroscience*, 26(5), 1551–1561. <http://dx.doi.org/10.1523/JNEUROSCI.3101-05.2006>.
- Ahmed, A. I. A., Van Den Elsen, G. A. H., Colbers, A., Kramers, C., Burger, D. M., van der Marck, M. A., & Olde Rikkert, M. G. M. (2015). Safety, pharmacodynamics, and pharmacokinetics of multiple oral doses of delta-9-tetrahydrocannabinol in older persons with dementia. *Psychopharmacology*, 232(14), 2587–2595. <http://dx.doi.org/10.1007/s00213-015-3889-y>.
- Alpár, A., Tortoriello, G., Calvigioni, D., Niphakis, M. J., Milenkovic, I., Bakker, J., ... Harkany, T. (2014). Endocannabinoids modulate cortical development by configuring Slit2/Robo1 signalling. *Nature Communications*, 5, 4421. <http://dx.doi.org/10.1038/ncomms5421>.
- American Chemical Society (2015). Legalizing marijuana and the new science of weed [press release]. Retrieved May 3, 2017, from <https://www.acs.org/content/acs/en/pressroom/newsreleases/2015/march/legalizing-marijuana-and-the-new-science-of-weedvideo.html?ga=1.115036224.695999133.1470076540>.
- Asch, R. H., & Smith, C. G. (1986). Effects of delta 9-THC, the principal psychoactive component of marijuana, during pregnancy in the rhesus monkey. *The Journal of Reproductive Medicine*, 31(12), 1071–1081.
- Astley, S. J., & Little, R. E. (1990). Maternal marijuana use during lactation and infant development at one year. *Neurotoxicology and Teratology*, 12(2), 161–168. [http://dx.doi.org/10.1016/0892-0362\(90\)90129-Z](http://dx.doi.org/10.1016/0892-0362(90)90129-Z).
- Atwood, B. K., Huffman, J., Straiker, A., & MacKie, K. (2010). JWH018, a common constituent of “Spice” herbal blends, is a potent and efficacious cannabinoid CB1 receptor agonist. *British Journal of Pharmacology*, 160(3), 585–593. <http://dx.doi.org/10.1111/j.1476-5381.2009.00582.x>.
- Azofeifa, A., Mattson, M. E., Schauer, G., McAfee, T., Grant, A., & Lyerla, R. (2016). National estimates of marijuana use and related indicators - National Survey on Drug Use and Health, United States, 2002–2014. *Morbidity and Mortality Weekly Report. Surveillance Summaries*, 65(11), 1–28. <http://dx.doi.org/10.15585/mmwr.ss6511a1>.
- Bada, H. S., Reynolds, E. W., & Hansen, W. F. (2006). Marijuana use, adolescent pregnancy, and alteration in newborn behavior: How complex can it get? *The Journal of Pediatrics*, 149(6), 742–745. <http://dx.doi.org/10.1016/j.jpeds.2006.10.049>.
- Bailey, J. R., Cunny, H. C., Paule, M. G., & Slikker, W. (1987). Fetal disposition of  $\Delta^9$ -tetrahydrocannabinol (THC) during late pregnancy in the rhesus monkey. *Toxicology and Applied Pharmacology*, 90(2), 315–321. [http://dx.doi.org/10.1016/0041-008X\(87\)90338-3](http://dx.doi.org/10.1016/0041-008X(87)90338-3).
- Bellinger, D. C., Matthews-Bellinger, J. A., & Kordas, K. (2016). A developmental perspective on early-life exposure to neurotoxicants. *Environment International*, 94, 103–112. <http://dx.doi.org/10.1016/j.envint.2016.05.014>.
- Benevenuto, S. G., Domenico, M. D., Martins, M. A. G., Costa, N. S., de Souza, A. R. L., Costa, J. L., ... Veras, M. M. (2017). Recreational use of marijuana during pregnancy and negative gestational and fetal outcomes: An experimental study in mice. *Toxicology*, 1(376), 94–101. <http://dx.doi.org/10.1016/j.tox.2016.05.020>.
- Berghuis, P., Rajnecik, A. M., Morozov, Y. M., Ross, R. A., Mulder, J., Urbán, G. M., ... Harkany, T. (2007). Hardwiring the brain: Endocannabinoids shape neuronal connectivity. *Science*, 316(5828), 1212–1216. <http://dx.doi.org/10.1126/science.1137406>.
- Blackard, C., & Tennes, K. (1984). Human placental transfer of cannabinoids. *New England Journal of Medicine*, 311, 797.
- Bland, T. M., Haining, R. L., Tracy, T. S., & Callery, P. S. (2005). CYP2C-catalyzed delta (9)-tetrahydrocannabinol metabolism: Kinetics, pharmacogenetics and interaction with phenytoin. *Biochemical Pharmacology*, 70(7), 1096–1103. <http://dx.doi.org/10.1126/science.1137406>.

- Bloomfield, M. A., Ashok, A. H., Volkow, N. D., & Howes, O. D. (2016). The effects of  $\Delta^9$ -tetrahydrocannabinol on the dopamine system. *Nature*, 539(7629), 369–377. <http://dx.doi.org/10.1038/nature20153> (Review. PMID: 27853201).
- Borgen, L. A., Davis, W. M., & Pace, H. B. (1973). Effects of prenatal  $\Delta^9$ -tetrahydrocannabinol on the development of rat offspring. *Pharmacology Biochemistry and Behavior*, 1, 203–206.
- Bornheim, L. M., Lasker, J. M., & Raucy, J. L. (1992). Human hepatic microsomal metabolism of  $\Delta^1$ -tetrahydrocannabinol. *Drug Metabolism and Disposition*, 20(2), 241–246.
- Boskovic, R., Klein, J., Woodland, C., Karaskov, T., & Koren, G. (2001). The role of the placenta in variability of fetal exposure to cocaine and cannabinoids: A twin study. *Canadian Journal of Physiology and Pharmacology*, 79(11), 942–945. <http://dx.doi.org/10.1139/y01-080>.
- Brake, S. C., Hutchings, D. E., Morgan, B., Lasalle, E., & Shi, T. (1987). Delta-9-tetrahydrocannabinol during pregnancy in the rat II. Effects on ontogeny of locomotor activity and nipple attachment in the offspring. *Neurotoxicology and Teratology*, 9(1), 45–49. [http://dx.doi.org/10.1016/0892-0362\(87\)90069-9](http://dx.doi.org/10.1016/0892-0362(87)90069-9).
- Brents, L. K. (2016). Marijuana, the endocannabinoid system and the female reproductive system. *Yale J. Biol. Med.* 89(2), 175–191 (eCollection 2016 Jun. Review. PMID: 27354844).
- Brown, Q. L., Sarvet, A. L., Shmulewitz, D., Martins, S. S., Wall, M. M., & Hasin, D. S. (2016). Trends in marijuana use among pregnant and nonpregnant reproductive-aged women, 2002–2014. *JAMA*, 72(12), 1235–1242. <http://dx.doi.org/10.1001/jama.2016.17383>.
- Buser, G. L., Gerona, R. R., Horowitz, B. Z., Vian, K. P., Troxell, M. L., Hendrickson, R. G., ... Leman, R. F. (2014). Acute kidney injury associated with smoking synthetic cannabinoid. *Clinical Toxicology*, 52(7), 664–673. <http://dx.doi.org/10.3109/15563650.2014.932365>.
- Calvigioni, D., Hurd, Y. L., Harkany, T., & Keimpema, E. (2014). Neuronal substrates and functional consequences of prenatal cannabis exposure. *European Child & Adolescent Psychiatry*, 23(10), 931–941. <http://dx.doi.org/10.1007/s00787-014-0550-y>.
- Campolongo, P., Trezza, V., Cassano, T., Gaetani, S., Morgese, M. G., Ubaldi, M., ... Cuomo, V. (2007). Perinatal exposure to delta-9-tetrahydrocannabinol causes enduring cognitive deficits associated with alteration of cortical gene expression and neurotransmission in rats. *Addiction Biology*, 12(3–4), 485–495. <http://dx.doi.org/10.1111/j.1369-1600.2007.00074.x>.
- Campolongo, P., Trezza, V., Ratanio, P., Palmery, M., & Cuomo, V. (2011). Developmental consequences of perinatal cannabis exposure: Behavioral and neuroendocrine effects in adult rodents. *Psychopharmacology*, 214(1), 5–15. <http://dx.doi.org/10.1007/s00213-010-1892-x>.
- Castelli, M. P., Paola Piras, A., D'Agostino, A., Pibiri, F., Perra, S., Gessa, G. L., ... Pistis, M. (2007). Dysregulation of the endogenous cannabinoid system in adult rats prenatally treated with the cannabinoid agonist WIN 55,212-2. *European Journal of Pharmacology*, 573(1–3), 11–19. <http://dx.doi.org/10.1016/j.ejphar.2007.06.047>.
- Castillo, P. E., Younts, T. J., Chávez, A. E., & Hashimoto, Y. (2012). Endocannabinoid signaling and synaptic function. *Neuron*, 76(1), 70–81. <http://dx.doi.org/10.1016/j.neuron.2012.09.020>.
- Center for Behavioral Health Statistics and Quality (2016). Results from the 2015 National Survey on Drug Use and Health: Detailed tables. Rockville, Maryland. Retrieved from <https://www.samhsa.gov/data/sites/default/files/NSDUH-DeT-Tab-2015/NSDUH-DeT-Tab-2015/NSDUH-DeT-Tab-2015.pdf>.
- Chambers, R. A., Taylor, J. R., & Potenza, M. N. (2003). Developmental neurocircuitry of motivation in adolescence: A critical period of addiction vulnerability. *American Journal of Psychiatry*, 160(6), 1041–1052. <http://dx.doi.org/10.1176/appi.ajp.160.6.1041>.
- Christensen, H. D., Freudenthal, R. I., Gidley, J. T., Rosenfeld, R., Boegli, G., Testino, L., ... Wall, M. E. (1971). Activity of  $\Delta^8$ - and  $\Delta^9$ -tetrahydrocannabinol and related compounds in the mouse. *Science*, 172(3979), 165–167.
- Conner, S. N., Carter, E. B., Tuuli, M. G., Macones, G. A., & Cahill, A. G. (2015). Maternal marijuana use and neonatal morbidity. *American Journal of Obstetrics and Gynecology*, 213(3), 422.e1–422.e4. <http://dx.doi.org/10.1016/j.ajog.2015.05.050>.
- Cravatt, B. F., Demarest, K., Patricelli, M. P., Bracey, M. H., Giang, D. K., Martin, B. R., & Lichtman, A. H. (2001). Supersensitivity to anandamide and enhanced endogenous cannabinoid signaling in mice lacking fatty acid amide hydrolase. *Proceedings of the National Academy of Sciences*, 98(16), 9371–9376. <http://dx.doi.org/10.1073/pnas.161191698>.
- Crews, F., He, J., & Hodge, C. (2007). Adolescent cortical development: A critical period of vulnerability for addiction. *Pharmacology Biochemistry and Behavior*, 86(2), 189–199. <http://dx.doi.org/10.1016/j.pbb.2006.12.001>.
- Cristino, L., & Di Marzo, V. (2014). Fetal cannabinoid receptors and the “dis-joint-ed” brain. *The EMBO Journal*, 33(7), 665–667. <http://dx.doi.org/10.1002/embj.201488086>.
- Dahl, R. E., Scher, M. S., Williamson, D. E., Robles, N., & Day, N. L. (1995). A longitudinal study of prenatal marijuana use: Effects on sleep and arousal at age 3 years. *Archives of Pediatrics & Adolescent Medicine*, 149(2), 145. <http://dx.doi.org/10.1001/archpedi.1995.02170140027004>.
- Day, N. L., Leech, S. L., & Goldschmidt, L. (2011). The effects of prenatal marijuana exposure on delinquent behaviors are mediated by measures of neurocognitive functioning. *Neurotoxicology and Teratology*, 33(1), 129–136. <http://dx.doi.org/10.1016/j.ntt.2010.07.006>.
- Day, N. L., Richardson, G. A., Geva, D., & Robles, N. (1994). Alcohol, marijuana, and tobacco: Effects of prenatal exposure on offspring growth and morphology at age six. *Alcoholism: Clinical and Experimental Research*, 18(4), 786–794. <http://dx.doi.org/10.1111/j.1530-0277.1994.tb00041.x>.
- De Chiara, V., Angelucci, F., Rossi, S., Musella, A., Cavasinni, F., Cantarella, C., ... Centonze, D. (2010). Brain-derived neurotrophic factor controls cannabinoid CB1 receptor function in the striatum. *Journal of Neuroscience*, 30(24), 8127–8137. <http://dx.doi.org/10.1523/JNEUROSCI.1683-10.2010>.
- De March, Z., Zuccato, C., Giampia, C., Patassini, S., Bari, M., Gasperi, V., ... Fusco, F. R. (2008). Cortical expression of brain derived neurotrophic factor and type-1 cannabinoid receptor after striatal excitotoxic lesions. *Neuroscience*, 152(3), 734–740. <http://dx.doi.org/10.1016/j.neuroscience.2007.11.044>.
- de Salas-Quiroga, A., Díaz-Alonso, J., García-Rincón, D., Remmers, F., Vega, D., Gómez-Cañas, M., ... Galve-Roperh, I. (2015). Prenatal exposure to cannabinoids evokes long-lasting functional alterations by targeting CB 1 receptors on developing cortical neurons. *Proceedings of the National Academy of Sciences of the United States of America*, 112(44), 1–6. <http://dx.doi.org/10.1073/pnas.1514962112>.
- Devane, W. A., Dysarz, F. A., III, Johnson, M. R., Melvin, L. S., & Howlett, A. C. (1988). Determination and characterization of a cannabinoid receptor in rat brain. *Molecular Pharmacology*, 34(5), 605–613.
- Di Marzo, V., Fontana, A., Cadas, H., Schinelli, S., Cimino, G., Schwartz, J. C., & Piomelli, D. (1994). Formation and inactivation of endogenous cannabinoid anandamide in central neurons. *Nature*, 372(6507), 686–691. <http://dx.doi.org/10.1038/372686a0>.
- Diamond, A., & Lee, K. (2011). Interventions shown to aid executive function development in children 4 to 12 years old. *Science*, 333(6045), 959–964. <http://dx.doi.org/10.1126/science.1204529>.
- Díaz-Alonso, J., Aguado, T., Wu, C.-S., Palazuelos, J., Hofmann, C., Garcez, P., ... Galve-Roperh, I. (2012). The CB(1) cannabinoid receptor drives corticospinal motor neuron differentiation through the Ctip2/Satb2 transcriptional regulation axis. *The Journal of Neuroscience*, 32(47), 16651–16665. <http://dx.doi.org/10.1523/JNEUROSCI.0681-12.2012>.
- Dinh, T. P., Carpenter, D., Leslie, F. M., Freund, T. F., Katona, I., Sensi, S. L., ... Piomelli, D. (2002). Brain monoglyceride lipase participating in endocannabinoid inactivation. *Proceedings of the National Academy of Sciences of the United States of America*, 99(16), 10819–10824. <http://dx.doi.org/10.1073/pnas.152334899>.
- Dinh, T. P., Kathuria, S., & Piomelli, D. (2004). RNA interference suggests a primary role for monoacylglycerol lipase in the degradation of the endocannabinoid 2-arachidonylglycerol. *Molecular Pharmacology*, 66(5), 1260–1264. <http://dx.doi.org/10.1124/mol.104.002071>.
- Dinis-Oliveira, R. J. (2016). Metabolomics of  $\Delta^9$ -tetrahydrocannabinol: Implications in toxicity. *Drug Metabolism Reviews*, 2532(1), 80–87. <http://dx.doi.org/10.3109/03602532.2015.1137307>.
- du Plessis, S. S., Agarwal, A., & Syriac, A. (2015). Marijuana, phytocannabinoids, the endocannabinoid system, and male fertility. *J. Assist. Reprod. Genet.* 32(11), 1575–1588. <http://dx.doi.org/10.1007/s10815-015-0553-8> (Epub 2015 Aug 16. Review. PMID: 26277482).
- El Marroun, H., Hudziak, J. J., Tiemeier, H., Creemers, H., Steegers, E. A., Jaddoe, V. W., ... Huizink, A. C. (2011). Intrauterine cannabis exposure leads to more aggressive behavior and attention problems in 18-month-old girls. *Drug and Alcohol Dependence*, 118(2–3), 470–474. <http://dx.doi.org/10.1016/j.drugalcdep.2011.03.004>.
- El Marroun, H., Tiemeier, H., Franken, I. H. A., Jaddoe, V. W. V., Van Der Lugt, A., Verhulst, F. C., ... White, T. (2015). Prenatal cannabis and tobacco exposure in relation to brain morphology: A prospective neuroimaging study in young children. *Biological Psychiatry*, 79(12), 1–9. <http://dx.doi.org/10.1016/j.biopsych.2015.08.024>.
- El Marroun, H., Tiemeier, H., Steegers, E. A., Jaddoe, V. W., Hofman, A., Verhulst, F. C., ... Huizink, A. C. (2009). Intrauterine cannabis exposure affects fetal growth trajectories: The Generation R Study. *Journal of the American Academy of Child and Adolescent Psychiatry*, 48(12), 1173–1181. [http://dx.doi.org/10.1016/S1090-798X\(10\)79394-2](http://dx.doi.org/10.1016/S1090-798X(10)79394-2).
- ElSohly, M. A., Mehmedic, Z., Foster, S., Gon, C., Chandra, S., & Church, J. C. (2016). Changes in cannabis potency over the last 2 decades (1995–2014): Analysis of current data in the United States. *Biological Psychiatry*, 79(7), 613–619. <http://dx.doi.org/10.1016/j.biopsych.2016.01.004>.
- Falcon, M., Pichini, S., Joya, J., Pujadas, M., Sanchez, A., Vall, O., ... Pellegrini, M. (2012). Maternal hair testing for the assessment of fetal exposure to drug of abuse during early pregnancy: Comparison with testing in placental and fetal remains. *Forensic Science International*, 218(1–3), 92–96. <http://dx.doi.org/10.1016/j.forsciint.2011.10.022>.
- Fergusson, D. M., Horwood, L. J., & Northstone, K. (2002). Maternal use of cannabis and pregnancy outcome. *BJOG: An International Journal of Obstetrics and Gynaecology*, 109(1), 21–27.
- Frank, D. A., Kuranz, S., Appugliese, D., Cabral, H., Chen, C., Crooks, D., ... Rose-Jacobs, R. (2014). Problematic substance use in urban adolescents: Role of intrauterine exposures to cocaine and marijuana and post-natal environment. *Drug and Alcohol Dependence*, 142, 181–190. <http://dx.doi.org/10.1016/j.drugalcdep.2014.06.014>.
- Freudenthal, R. I., Martin, J., & Wall, M. E. (1972). Distribution of  $\Delta^9$ -tetrahydrocannabinol in the mouse. *British Journal of Pharmacology*, 44(2), 244–249. <http://dx.doi.org/10.1111/j.1476-5381.1972.tb07260.x>.
- Fried, P. A. (1976). Short and long-term effects of pre-natal Cannabis inhalation upon rat offspring. *Psychopharmacology*, 50(3), 285–291. <http://dx.doi.org/10.1007/BF00426846>.
- Fried, P. A. (1980). Marijuana use by pregnant women: Neurobehavioral effects in neonates. *Drug and Alcohol Dependence*, 6(6), 415–424. [http://dx.doi.org/10.1016/0376-8716\(80\)90023-X](http://dx.doi.org/10.1016/0376-8716(80)90023-X).
- Fried, P. A. (2002). The consequences of marijuana use during pregnancy: A review of the human literature. *Women and Cannabis: Medicine, Science, and Sociology*, 2(3), 85–104.
- Fried, P. A., Buckingham, M., & Von Kulmiz, P. (1983). Marijuana use during pregnancy and perinatal risk factors. *American Journal of Obstetrics and Gynecology*, 146(8), 992–994. [http://dx.doi.org/10.1016/0002-9378\(83\)90989-4](http://dx.doi.org/10.1016/0002-9378(83)90989-4).
- Fried, P. A., James, D. S., & Watkinson, B. (2001). Growth and pubertal milestones during

- adolescence in offspring prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 23(5), 431–436. [http://dx.doi.org/10.1016/S0892-0362\(01\)00161-1](http://dx.doi.org/10.1016/S0892-0362(01)00161-1).
- Fried, P. A., & O'Connell, C. M. (1987). A comparison of the effects of prenatal exposure to tobacco, alcohol, cannabis and caffeine on birth size and subsequent growth. *Neurotoxicology and Teratology*, 9(2), 79–85. [http://dx.doi.org/10.1016/0892-0362\(87\)90082-1](http://dx.doi.org/10.1016/0892-0362(87)90082-1).
- Fried, P. A., O'Connell, C. M., & Watkinson, B. (1992). 60- and 72-month follow-up of children prenatally exposed to marijuana, cigarettes, and alcohol: Cognitive and language assessment. *Journal of Developmental & Behavioral Pediatrics*, 13(6), 383–391.
- Fried, P. A., & Smith, A. M. (2001). A literature review of the consequences of prenatal marijuana exposure - An emerging theme of a deficiency in aspects of executive function. *Neurotoxicology and Teratology*, 23(1), 1–11. [http://dx.doi.org/10.1016/S0892-0362\(00\)00119-7](http://dx.doi.org/10.1016/S0892-0362(00)00119-7).
- Fried, P. A., & Watkinson, B. (1988). 12- and 24-month neurobehavioral follow-up of children prenatally exposed to marijuana, cigarettes and alcohol. *Neurotoxicology and Teratology*, 10(4), 305–313. [http://dx.doi.org/10.1016/0892-0362\(88\)90032-3](http://dx.doi.org/10.1016/0892-0362(88)90032-3).
- Fried, P. A., & Watkinson, B. (1990). 36- and 48-month neurobehavioral follow-up of children prenatally exposed to marijuana, cigarettes, and alcohol. *Journal of Developmental & Behavioral Pediatrics*, 11(2), 49–58.
- Fried, P. A., & Watkinson, B. (2001). Differential effects on facets of attention in adolescents prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 23(5), 421–430. [http://dx.doi.org/10.1016/S0892-0362\(01\)00160-X](http://dx.doi.org/10.1016/S0892-0362(01)00160-X).
- Fried, P. A., Watkinson, B., Dillon, R. F., & Dulberg, C. S. (1987). Neonatal neurological status in a low-risk population after prenatal exposure to cigarettes, marijuana, and alcohol. *Journal of Developmental & Behavioral Pediatrics*, 8(6), 318–326 ([https://doi.org/10.1016/S0892-0362\(87\)0806-0318502.00.00](https://doi.org/10.1016/S0892-0362(87)0806-0318502.00.00)).
- Fried, P. A., Watkinson, B., & Gray, R. (1992). A follow-up study of attentional behavior in 6-year-old children exposed prenatally to marijuana, cigarettes, and alcohol. *Neurotoxicology and Teratology*, 14(5), 299–311.
- Fried, P. A., Watkinson, B., & Gray, R. (1998). Differential effects on cognitive functioning in 9- to 12-year olds prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 20(3), 293–306. [http://dx.doi.org/10.1016/S0892-0362\(97\)00091-3](http://dx.doi.org/10.1016/S0892-0362(97)00091-3).
- Fried, P. A., Watkinson, B., & Gray, R. (1999). Growth from birth to early adolescence in offspring prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 21(5), 513–525. [http://dx.doi.org/10.1016/S0892-0362\(99\)00009-4](http://dx.doi.org/10.1016/S0892-0362(99)00009-4).
- Fried, P. A., Watkinson, B., & Gray, R. (2003). Differential effects on cognitive functioning in 13- to 16-year-olds prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 25(4), 427–436. [http://dx.doi.org/10.1016/S0892-0362\(03\)00029-1](http://dx.doi.org/10.1016/S0892-0362(03)00029-1).
- Fried, P. A., Watkinson, B., & Siegel, L. S. (1997). Reading and language in 9- to 12-year olds prenatally exposed to cigarettes and marijuana. *Neurotoxicology and Teratology*, 19(3), 171–183. [http://dx.doi.org/10.1016/S0892-0362\(97\)00015-9](http://dx.doi.org/10.1016/S0892-0362(97)00015-9).
- Fu, J., Bottegoni, G., Sasso, O., Bertorelli, R., Rocchia, W., Masetti, M., ... Piomelli, D. (2012). A catalytically silent FAAH-1 variant drives anandamide transport in neurons. *Nature Neuroscience*, 15(1), 64–69. <http://dx.doi.org/10.1038/nn.2986>.
- Galve-Roperh, I., Chirchiu, V., Diaz-Alonso, J., Bari, M., Guzmán, M., & Maccarrone, M. (2013). Cannabinoid receptor signaling in progenitor/stem cell proliferation and differentiation. *Progress in Lipid Research*, 52(4), 633–650. <http://dx.doi.org/10.1016/j.plipres.2013.05.004>.
- Galve-Roperh, I., Palazuelos, J., Aguado, T., & Guzmán, M. (2009). The endocannabinoid system and the regulation of neural development: Potential implications in psychiatric disorders. *European Archives of Psychiatry and Clinical Neuroscience*, 259(7), 371–382. <http://dx.doi.org/10.1007/s00406-009-0028-y>.
- García-Gil, L., Romero, J., Ramos, J. A., & Fernández-Ruiz, J. J. (1999). Cannabinoid receptor binding and mRNA levels in several brain regions of adult male and female rats perinatally exposed to delta-9-tetrahydrocannabinol. *Drug and Alcohol Dependence*, 55(1–2), 127–136.
- Garg, M., Garrison, L., Leeman, L., Hamidovic, A., Borrego, M., Rayburn, W. F., & Bakhireva, L. (2016). Validity of self-reported drug use information among pregnant women. *Maternal and Child Health Journal*, 20(1), 41–47. <http://dx.doi.org/10.1007/s10995-015-1799-6>.
- Garrett, E. R., & Hunt, A. C. (1977). Pharmacokinetics of  $\Delta^9$ -tetrahydrocannabinol in dogs. *Journal of Pharmaceutical Sciences*, 66(3), 395–407. <http://dx.doi.org/10.1002/jps.2600660322>.
- Gianutos, G., & Abbatiello, E. R. (1972). The effect of pre-natal *Cannabis sativa* on maze learning ability in the rat. *Psychopharmacologia*, 27(2), 117–122. <http://dx.doi.org/10.1007/BF00439370>.
- Ginsburg, B. C., Hrubá, L., Zaki, A., Javors, M. A., & McMahon, L. R. (2014). Blood levels do not predict behavioral or physiological effects of  $\Delta^9$ -tetrahydrocannabinol in rhesus monkeys with different patterns of exposure. *Drug and Alcohol Dependence*, 139, 1–8. <http://dx.doi.org/10.1016/j.drugalcdep.2014.02.696>.
- Glaz-Sandberg, A., Dietz, L., Nguyen, H., Oberwittler, H., Aderjan, R., & Mikus, G. (2007). Pharmacokinetics of 11-nor-9-carboxy- $\Delta^9$ -tetrahydrocannabinol (CTHC) after intravenous administration of THC in healthy human subjects. *Clinical Pharmacology and Therapeutics*, 82(1), 63–69. <http://dx.doi.org/10.1038/sj.cpt.6100199>.
- Goldschmidt, L., Day, N. L., & Richardson, G. A. (2000). Effects of prenatal marijuana exposure on child behavior problems at age 10. *Neurotoxicology and Teratology*, 22(3), 325–336. [http://dx.doi.org/10.1016/S0892-0362\(00\)00066-0](http://dx.doi.org/10.1016/S0892-0362(00)00066-0).
- Goldschmidt, L., Richardson, G. A., Cornelius, M. D., & Day, N. L. (2004). Prenatal marijuana and alcohol exposure and academic achievement at age 10. *Neurotoxicology and Teratology*, 26(4), 521–532. <http://dx.doi.org/10.1016/j.ntt.2004.04.003>.
- Goldschmidt, L., Richardson, G. A., Larkby, C., & Day, N. L. (2016). Early marijuana initiation: The link between prenatal marijuana exposure, early childhood behavior, and negative adult roles. *Neurotoxicology and Teratology*, 58, 40–45. <http://dx.doi.org/10.1016/j.ntt.2016.05.011>.
- Goldschmidt, L., Richardson, G. A., Willford, J., & Day, N. L. (2008). Prenatal marijuana exposure and intelligence test performance at age 6. *Journal of the American Academy of Child and Adolescent Psychiatry*, 47(3), 254–263. <http://dx.doi.org/10.1097/CHI.0b013e318160b3f0>.
- Grant, K. S., & Rice, D. C. (2008). Exposure to environmental chemicals and developmental risk. In T. M. Burbacher, G. P. Sackett, & K. S. Grant (Eds.), *Primate models of children's health and developmental disabilities* (pp. 377–419). Elsevier.
- Gray, K. A., Day, N. L., Leech, S., & Richardson, G. A. (2005). Prenatal marijuana exposure: Effect on child depressive symptoms at ten years of age. *Neurotoxicology and Teratology*, 27, 439–448. <http://dx.doi.org/10.1016/j.ntt.2005.03.010>.
- Grotenhermen, F. (2003). Pharmacokinetics and pharmacodynamics of cannabinoids. *Clinical Pharmacokinetics*, 42(4), 327–360. <http://dx.doi.org/10.2165/00003088-200342040-00003>.
- Grotenhermen, F., Russo, E., & Zuardi, A. W. (2017). Even high doses of oral cannabidiol do not cause THC-like effects in humans: Comment on Merrick et al. cannabis and cannabinoid research 2016;1(1):102–112; DOI: 10.1089/can.2015.0004. *Cannabis and Cannabinoid Research*, 2(1), 1–4. <http://dx.doi.org/10.1089/can.2015.0004>.
- Harkany, T., Keimpema, E., Barabás, K., & Mulder, J. (2008). Endocannabinoid functions controlling neuronal specification during brain development. *Molecular and Cellular Endocrinology*, 286(1–2), S84–S90. <http://dx.doi.org/10.1016/j.mce.2008.02.011>.
- Harkany, T., Mackie, K., & Doherty, P. (2008). Wiring and firing neuronal networks: Endocannabinoids take center stage. *Current Opinion in Neurobiology*, 18(3), 338–345. <http://dx.doi.org/10.1016/j.conb.2008.08.007>.
- Huestis, M. A. (2007). Human cannabinoid pharmacokinetics. *Chemical Biodiversity*, 4(8), 1770–1804. <http://dx.doi.org/10.1002/cbdv.200790152.Human>.
- Hunault, C. C., Mensinga, T. T., de Vries, I., Kelholt-Dijkman, H. H., Hoek, J., Kruidenier, M., ... Meulenbelt, J. (2008). Delta-9-tetrahydrocannabinol (THC) serum concentrations and pharmacological effects in males after smoking a combination of tobacco and cannabis containing up to 69 mg THC. *Psychopharmacology*, 201(2), 171–181.
- Hunt, C., & Jones, R. (1980). Tolerance and disposition of tetrahydrocannabinol in man. *Journal of Pharmacology and Experimental Therapeutics*, 215(1), 35–44. <http://dx.doi.org/10.1017/CBO9781107415324.004>.
- Hurd, Y. L., Wang, X., Anderson, V., Beck, O., Minkoff, H., & Dow-Edwards, D. (2005). Marijuana impairs growth in mid-gestation fetuses. *Neurotoxicology and Teratology*, 27(2), 221–229. <http://dx.doi.org/10.1016/j.ntt.2004.11.002>.
- Hutchings, D. E., Brake, S. C., Banks, A. N., Nero, T. J., Dick, L. S., & Zmitrovich, A. C. (1991). Prenatal delta-9-tetrahydrocannabinol in the rat: Effects on auditory startle in adulthood. *Neurotoxicology and Teratology*, 13(4), 413–416. [http://dx.doi.org/10.1016/0892-0362\(91\)90090-J](http://dx.doi.org/10.1016/0892-0362(91)90090-J).
- Hutchings, D. E., & Dow-Edwards, D. (1991). Animal models of opiate, cocaine, and cannabis use. *Clinics in Perinatology*, 18(1), 1–22.
- Hutchings, D. E., Fico, T. A., Banks, A. N., Dick, L. S., & Brake, S. C. (1991). Prenatal delta-9-tetrahydrocannabinol in the rat: Effects on postweaning growth. *Neurotoxicology and Teratology*, 13(2), 245–248. [http://dx.doi.org/10.1016/0892-0362\(91\)90018-R](http://dx.doi.org/10.1016/0892-0362(91)90018-R).
- Hutchings, D. E., Gamagaris, Z., Miller, N., & Fico, T. A. (1989). The effects of prenatal exposure to delta-9-tetrahydrocannabinol on the rest-activity cycle of the pre-weaning rat. *Neurotoxicology and Teratology*, 11, 353–356.
- Hutchings, D. E., Martin, B. R., Gannagaris, Z., Miller, N., & Fico, T. (1989). Plasma concentrations of delta-9-tetrahydrocannabinol in dams and fetuses following acute or multiple dosing in rats. *Life Sciences*, 44, 697–701.
- Hutchings, D. E., Morgan, B., Brake, S. C., Shi, T., & Lasalle, E. (1987). Delta-9-tetrahydrocannabinol during pregnancy in the rat: I. Differential effects on maternal nutrition, embryotoxicity, and growth in the offspring. *Neurotoxicology and Teratology*, 9(1), 39–43. [http://dx.doi.org/10.1016/0892-0362\(87\)90068-7](http://dx.doi.org/10.1016/0892-0362(87)90068-7).
- Isoherranen, N., & Thummel, K. E. (2013). Drug metabolism and transport during pregnancy: How does drug disposition change during pregnancy and what are the mechanisms that cause such changes? *Drug Metabolism and Disposition*, 41(2), 256–262. <http://dx.doi.org/10.1124/dmd.112.050245>.
- Jaddoe, V. W. V., Van Duijn, C. M., Van Der Heijden, A. J., MacKenbach, J. P., Moll, H. A., Steegers, E. A. P., ... Hofman, A. (2008). The Generation R Study: Design and cohort update until the age of 4 years. *European Journal of Epidemiology*, 23(12), 801–811. <http://dx.doi.org/10.1007/s10654-008-9309-4>.
- Jaddoe, V. W. V., Van Duijn, C. M., Van Der Heijden, A. J., MacKenbach, J. P., Moll, H. A., Steegers, E. A. P., ... Hofman, A. (2010). The Generation R Study: Design and cohort update 2010. *European Journal of Epidemiology*, 25(11), 823–841. <http://dx.doi.org/10.1007/s10654-010-9516-7>.
- Karlsson, M., Contreras, J. A., Hellman, U., Tornqvist, H., & Holm, C. (1997). cDNA cloning, tissue distribution, and identification of the catalytic triad of monoglyceride lipase. Evolutionary relationship to esterases, lysophospholipases, and haloperoxidases. *Journal of Biological Chemistry*, 272(43), 27218–27223. <http://dx.doi.org/10.1074/jbc.272.43.27218>.
- Katona, I., & Freund, T. F. (2008). Endocannabinoid signaling as a synaptic circuit breaker in neurological disease. *Nature Medicine*, 14(9), 923–930. <http://dx.doi.org/10.1038/nm.f1869>.
- Katona, I., & Freund, T. F. (2012). Multiple functions of endocannabinoid signaling in the brain. *Annual Review of Neuroscience*, 35(1), 529–558. <http://dx.doi.org/10.1146/annurev-neuro-062111-150420>.
- Kauert, G. F., Ramaekers, J. G., Schneider, E., Moeller, M. R., & Toennes, S. W. (2007). Pharmacokinetic properties of  $\Delta^9$ -tetrahydrocannabinol in serum and oral fluid. *Journal of Analytical Toxicology*, 31, 288–293. <http://dx.doi.org/10.1093/jat/31.5.288>.
- Keimpema, E., Barabás, K., Morozov, Y. M., Tortoriello, G., Torii, M., Cameron, G., ... Harkany, T. (2010). Differential subcellular recruitment of monoacylglycerol lipase generates spatial specificity of 2-arachidonoyl glycerol signaling during axonal

- pathfinding. *The Journal of Neuroscience*, 30(42), 13992–14007. <http://dx.doi.org/10.1523/JNEUROSCI.2126-10.2010>.
- Keimpema, E., Mackie, K., & Harkany, T. (2011). Molecular model of cannabis sensitivity in developing neuronal circuits. *Trends in Pharmacological Sciences*, 32(9), 551–561. <http://dx.doi.org/10.1016/j.tips.2011.05.004>.
- Klausner, H. A., & Dingell, J. V. (1971). The metabolism and excretion of  $\Delta^9$ -tetrahydrocannabinol in the rat. *Life Sciences*, 10, 49–59. [http://dx.doi.org/10.1016/0024-3205\(71\)90245-1](http://dx.doi.org/10.1016/0024-3205(71)90245-1).
- Klumbers, L. E., Beumer, T. L., van Hasselt, J. G., Lipplaa, A., Karger, L. B., Kleinloog, H. D., ... van Gerven, J. M. (2012). Novel  $\Delta^9$ -tetrahydrocannabinol formulation Namisol® has beneficial pharmacokinetics and promising pharmacodynamic effects. *British Journal of Clinical Pharmacology*, 74(1), 42–53.
- Lapoint, J., James, L. P., Moran, C. L., Nelson, L. S., Hoffman, R. S., & Moran, J. H. (2011). Severe toxicity following synthetic cannabinoid ingestion. *Clinical Toxicology*, 49(8), 760–764. <http://dx.doi.org/10.3109/15563650.2011.609822>.
- Laprairie, R. B., Bagher, A. M., Kelly, M. E. M., & Denovan-Wright, E. M. (2015). Cannabidiol is a negative allosteric modulator of the cannabinoid CB1 receptor. *British Journal of Pharmacology*, 172(20), 4790–4805. <http://dx.doi.org/10.1111/bph.13250>.
- Lee, D., Vandrey, R., Mendu, D. R., Murray, J. A., Barnes, A. J., & Huestis, M. A. (2015). Oral fluid cannabinoids in chronic frequent cannabis smokers during ad libitum cannabis smoking. *Drug Testing and Analysis*, 7(6), 494–501.
- Leech, S. L., Larkby, C. A., Day, R., & Day, N. L. (2006). Predictors and correlates of high levels of depression and anxiety symptoms among children at age 10. *Journal of the American Academy of Child and Adolescent Psychiatry*, 45(2), 223–230. <http://dx.doi.org/10.1097/01.chi.0000184930.18552.4d>.
- Lemberger, L., Axelrod, J., & Kopin, I. J. (1971). Metabolism and disposition of tetrahydrocannabinols in naive subjects and chronic marijuana users. *Annals of the New York Academy of Sciences*, 191(1), 142–154. <http://dx.doi.org/10.1111/j.1749-6632.1971.tb13994.x>.
- Leuschner, J. T. A., Harvey, D. J., Bullingham, R. E. S., & Paton, W. D. M. (1986). Pharmacokinetics of delta-9-tetrahydrocannabinol in rabbits following single or multiple intravenous doses. *Drug Metabolism and Disposition*, 14(2), 230–238.
- Linn, S., Schoenbaum, S. C., Monson, R. R., Rosner, R., Stubblefield, P. C., & Ryan, K. J. (1983). The association of marijuana use with outcome of pregnancy. *Am. J. Public Health*, 73(10), 1161–1164 (PMID: 6604464).
- Lipari, R., International, R., Kroutil, L. A., & Pemberton, M. R. (2015). Risk and protective factors and initiation of substance use: Results from the 2014 National Survey on Drug Use and Health. Retrieved from <https://www.samhsa.gov/data/sites/default/files/NSDUH-DR-FRR4-2014rev/NSDUH-DR-FRR4-2014.pdf>.
- López-Gallardo, M., López-Rodríguez, A. B., Llorente-Berzal, Á., Rotllant, D., Mackie, K., Armario, A., ... Viveros, M. P. (2012). Maternal deprivation and adolescent cannabinoid exposure impact hippocampal astrocytes, CB1 receptors and brain-derived neurotrophic factor in a sexually dimorphic fashion. *Neuroscience*, 204, 90–103. <http://dx.doi.org/10.1016/j.neuroscience.2011.09.063>.
- Maccarrone, M., Guzmán, M., Mackie, K., Doherty, P., & Harkany, T. (2014). Programming of neural cells by (endo)cannabinoids: From physiological rules to emerging therapies. *Nature Reviews Neuroscience*, 15(12), 786–801. <http://dx.doi.org/10.1038/nrn3846>.
- Mark, K., Gryczynski, J., Axenfeld, E., Schwartz, R. P., & Terplan, M. (2017). Pregnant women's current and intended cannabis use in relation to their views toward legalization and knowledge of potential harm. *Journal of Addiction Medicine*, 11(3), 211–216. <http://dx.doi.org/10.1097/ADM.0000000000000299> (PMID: 28252456).
- Marrs, W., & Stella, N. (2007). 2-AG + 2 new players = forecast for therapeutic advances. *Chemistry & Biology*, 14(12), 1309–1311. <http://dx.doi.org/10.1016/j.chembiol.2007.12.004>.
- Marsicano, G., Goodenough, S., Monory, K., Hermann, H., Eder, M., Cannich, A., ... Lutz, B. (2003). CB1 cannabinoid receptors and on-demand defense against excitotoxicity. *Science*, 302(5642), 84–88. <http://dx.doi.org/10.1126/science.1088208>.
- Marsot, A., Audebert, C., Attolini, L., Lacarelle, B., Micallef, J., & Blin, O. (2016). Comparison of cannabinoid concentrations in plasma, oral fluid and urine in occasional cannabis smokers after smoking cannabis cigarette. *Journal of Pharmacy & Pharmaceutical Sciences*, 19(3), 411. <http://dx.doi.org/10.18433/J3F31D>.
- Martin, B. R., Dewey, W. L., Harris, L. S., & Beckner, J. S. (1977). 3H- $\Delta^9$ -tetrahydrocannabinol distribution in pregnant dogs and their fetuses. *Research Communications in Chemical Pathology and Pharmacology*, 17(3), 457–470 Retrieved from <http://www.ncbi.nlm.nih.gov/pubmed/897339>.
- Mathews, C. A., Scharf, J. M., Miller, L. L., Macdonald-Wallis, C., Lawlor, D. A., & Ben-Shlomo, Y. (2014). Association between pre- and perinatal exposures and Tourette syndrome or chronic tic disorder in the ALSPAC cohort. *British Journal of Psychiatry*, 204(1), 40–45. <http://dx.doi.org/10.1192/bjp.bp.112.125468>.
- Mazur, A., Lichti, C. F., Prather, P. L., Zielinska, A. K., Bratton, S. M., Gallus-Zawada, A., ... Moran, J. H. (2009). Characterization of human hepatic and extrahepatic UDP-glucuronosyltransferase enzymes involved in the metabolism of classic cannabinoids. *Drug Metabolism and Disposition: The Biological Fate of Chemicals*, 37(7), 1496–1504. <http://dx.doi.org/10.1124/dmd.109.026898>.
- Mckinney, M. K., & Cravatt, B. F. (2005). Structure and function of fatty acid amide hydrolase. *Annual Review of Biochemistry*, 74(1), 411–432. <http://dx.doi.org/10.1146/annurev.biochem.74.082803.133450>.
- McLemore, G. L., & Richardson, K. A. (2016). Data from three prospective longitudinal human cohorts of prenatal marijuana exposure and offspring outcomes from the fetal period through young adulthood. *Data in Brief*, 9, 753–757. <http://dx.doi.org/10.1016/j.dib.2016.10.005>.
- Mereu, G., Fà, M., Ferraro, L., Cagiano, R., Antonelli, T., Tattoli, M., ... Cuomo, V. (2003). Prenatal exposure to a cannabinoid agonist produces memory deficits linked to dysfunction in hippocampal long-term potentiation and glutamate release. *Proceedings of the National Academy of Sciences of the United States of America*, 100(8), 4915–4920. <http://dx.doi.org/10.1073/pnas.0537849100>.
- Metna-Laurent, M., & Marsicano, G. (2015). Rising stars: Modulation of brain functions by astroglial type-1 cannabinoid receptors. *Glia*, 63(3), 353–364. <http://dx.doi.org/10.1002/glia.22773>.
- Miller, A. M., & Stella, N. (2008). CB2 receptor-mediated migration of immune cells: It can go either way. *British Journal of Pharmacology*, 153(2), 299–308. <http://dx.doi.org/10.1038/sj.bjp.0707523>.
- Moreno, M., Trigo, J. M., Escuredo, L., Rodriguez de Fonseca, F., & Navarro, M. (2003). Perinatal exposure to  $\Delta^9$ -tetrahydrocannabinol increases presynaptic dopamine D2 receptor sensitivity: A behavioral study in rats. *Pharmacology Biochemistry and Behavior*, 75(3), 565–575. [http://dx.doi.org/10.1016/S0091-3057\(03\)00117-5](http://dx.doi.org/10.1016/S0091-3057(03)00117-5).
- Mulder, J., Aguado, T., Keimpema, E., Barabás, K., Ballester Rosado, C. J., Nguyen, L., ... Harkany, T. (2008). Endocannabinoid signaling controls pyramidal cell specification and long-range axon patterning. *Proceedings of the National Academy of Sciences of the United States of America*, 105(25), 8760–8765. <http://dx.doi.org/10.1073/pnas.0803545105>.
- Musshoff, F., & Madea, B. (2006). Review of biologic matrices (urine, blood, hair) as indicators of recent or ongoing cannabis use. *Therapeutic Drug Monitoring*, 28(2), 155–163. <http://dx.doi.org/10.1097/01.ftd.0000197091.07807.22>.
- Naef, M., Russmann, S., Petersen-Felix, S., & Brenneisen, R. (2004). Development and pharmacokinetic characterization of pulmonary and intravenous delta-9-tetrahydrocannabinol (THC) in humans. *Journal of Pharmaceutical Sciences*, 93(5), 1176–1184.
- Narimatsu, S., Matsubara, K., Shimonishi, T., Watanabe, K., Yamamoto, I., & Yoshimura, H. (1988). Enzymatic oxidation of 7-hydroxylated delta 8-tetrahydrocannabinol to 7-oxo-delta 8-tetrahydrocannabinol by hepatic microsomes of the guinea pig. *Drug Metabolism and Disposition: The Biological Fate of Chemicals*, 16(1), 156–161.
- Narimatsu, S., Watanabe, K., Matsunaga, T., Yamamoto, I., Imaoka, S., Funae, Y., & Yoshimura, H. (1990). Cytochrome P-450 isozymes in metabolic activation of delta 9-tetrahydrocannabinol by rat liver microsomes. *Drug Metabolism and Disposition*, 18(6).
- Narimatsu, S., Watanabe, K., Matsunaga, T., Yamamoto, I., Imaoka, S., Funae, Y., & Yoshimura, H. (1992). Cytochrome P-450 isozymes involved in the oxidative metabolism of delta 9-tetrahydrocannabinol by liver microsomes of adult female rats. *Drug Metabolism and Disposition*, 20(1), 79–83.
- National Academies Press (2017). *The health effects of cannabis and cannabinoids: The current state of evidence and recommendations for research*. Washington, D.C.: National Academies of Sciences Engineering and Medicine <http://dx.doi.org/10.17226/24625>.
- Navarrete, M., & Araque, A. (2010). Endocannabinoids potentiate synaptic transmission through stimulation of astrocytes. *Neuron*, 68(1), 113–126. <http://dx.doi.org/10.1016/j.neuron.2010.08.043>.
- Navarro, M., Rubio, P., & Rodriguez de Fonseca, F. (1995). Behavioral consequences of maternal exposure to natural cannabinoids in rats. *Psychopharmacology*, 122(1), 1–14.
- Newsom, R. J., & Kelly, S. J. (2008). Perinatal delta-9-tetrahydrocannabinol exposure disrupts social and open field behavior in adult male rats. *Neurotoxicology and Teratology*, 30(3), 213–219. <http://dx.doi.org/10.1016/j.jnt.2007.12.007>.
- Noland, J. S., Singer, L. T., Short, E. J., Minnes, S., Arendt, R. E., Kirchner, H. L., & Bearer, C. (2005). Prenatal drug exposure and selective attention in preschoolers. *Neurotoxicology and Teratology*, 27, 429–438. <http://dx.doi.org/10.1016/j.jnt.2005.02.001>.
- Nomura, D. K., Morrison, B. E., Blankman, J. L., Long, J. Z., Kinsey, S. G., Marcondes, M. C. G., ... Cravatt, B. F. (2011). Endocannabinoid hydrolysis generates brain prostanoid ligands that promote neuroinflammation. *Science*, 334(6057), 809–813. <http://dx.doi.org/10.1126/science.1209200>.
- O'Shea, M., & Mallet, P. E. (2005). Impaired learning in adulthood following neonatal  $\Delta^9$ -THC exposure. *Behavioural Pharmacology*, 16(5–6), 455–461 (<https://doi.org/00008877-200509000-00019> [pii]).
- Ohlsson, A., Lindgren, J. E., Wahlen, A., Agurell, S., Hollister, L. E., & Gillespie, H. K. (1980). Plasma delta-9 tetrahydrocannabinol concentrations and clinical effects after oral and intravenous administration and smoking. *Clinical Pharmacology and Therapeutics*, 28(3), 409–416. <http://dx.doi.org/10.1038/clpt.1980.181>.
- Oliveira da Cruz, J. F., Robin, L. M., Drago, F., Marsicano, G., & Metna-Laurent, M. (2016). Astroglial type-1 cannabinoid receptor (CB1): A new player in the tripartite synapse. *Neuroscience*, 323, 35–42. <http://dx.doi.org/10.1016/j.neuroscience.2015.05.002>.
- Parikh, N., Kramer, W. G., Khurana, V., Smith, C. C., & Veticaden, S. (2016). Bioavailability study of dronabinol oral solution versus dronabinol capsules in healthy volunteers. *Clinical Pharmacology: Advances and Applications*, 8, 155–162.
- Pertwee, R. G. (2008). The diverse CB1 and CB2 receptor pharmacology of three plant cannabinoids: delta9-tetrahydrocannabinol, cannabidiol and delta9-tetrahydrocannabinol. *British Journal of Pharmacology*, 153(2), 199–215. <http://dx.doi.org/10.1038/sj.bjp.0707442>.
- Pertwee, R. G., Howlett, A. C., Abood, M. E., Alexander, S. P. H., Di Marzo, V., Elphick, M. R., ... Ross, R. A. (2010). International Union of Basic and Clinical Pharmacology. LXXIX. Cannabinoid receptors and their ligands: Beyond CB1 and CB2. *Pharmacological Reviews*, 62(4), 588–631. <http://dx.doi.org/10.1124/pr.110.003004>.
- Piomelli, D. (2003). The molecular logic of endocannabinoid signalling. *Nature Reviews Neuroscience*, 4(11), 873–884. <http://dx.doi.org/10.1038/nrn1247>.
- Renard, J., Norris, C., Rushlow, W., & Laviolette, S. R. (2017). Neuronal and molecular effects of cannabidiol on the mesolimbic dopamine system: Implications for novel schizophrenia treatments. *Neuroscience & Biobehavioral Reviews*, 75, 157–165. <http://dx.doi.org/10.1016/j.neubiorev.2017.02.006>.
- Renard, J., Rosen, L. G., Loureiro, M., De Oliveira, C., Schmid, S., Rushlow, W. J., & Laviolette, S. R. (2017). Adolescent cannabinoid exposure induces a persistent subcortical hyper-dopaminergic state and associated molecular adaptations in the

- prefrontal cortex. *Cerebral Cortex*, 27(2), 1297–1310. <http://dx.doi.org/10.1093/cercor/bhv335>.
- Richardson, G. A., Day, N. L., & Goldschmidt, L. (1995). Prenatal alcohol, marijuana, and tobacco use: Infant mental and motor development. *Neurotoxicology and Teratology*, 17(4), 479–487. [http://dx.doi.org/10.1016/0892-0362\(95\)00006-D](http://dx.doi.org/10.1016/0892-0362(95)00006-D).
- Richardson, G. A., Day, N. L., & Taylor, P. M. (1989). The effect of prenatal alcohol, marijuana, and tobacco exposure on neonatal behavior. *Infant Behavior & Development*, 12(2), 199–209. [http://dx.doi.org/10.1016/0163-6383\(89\)90006-4](http://dx.doi.org/10.1016/0163-6383(89)90006-4).
- Richardson, G. A., Ryan, C., Willford, J., Day, N. L., & Goldschmidt, L. (2002). Prenatal alcohol and marijuana exposure: Effects on neuropsychological outcomes at 10 years. *Neurotoxicology and Teratology*, 24(3), 309–320. [http://dx.doi.org/10.1016/S0892-0362\(02\)00193-9](http://dx.doi.org/10.1016/S0892-0362(02)00193-9).
- Rosenthaler, S., Pöhn, B., Kolmanz, C., Nguyen Huu, C., Krewenka, C., Huber, A., ... Moldzio, R. (2014). Differences in receptor binding affinity of several phytocannabinoids do not explain their effects on neural cell cultures. *Neurotoxicology and Teratology*, 46, 49–56. <http://dx.doi.org/10.1016/j.ntt.2014.09.003>.
- Ross, R. A. (2009). The enigmatic pharmacology of GPR55. *Trends in Pharmacological Sciences*, 30(3), 156–163. <http://dx.doi.org/10.1016/j.tips.2008.12.004>.
- Rubio, P., Rodríguez de Fonseca, F., Muñoz, R. M., Ariznavarreta, C., Martín-Calderón, J. L., & Navarro, M. (1995). Long-term behavioral effects of perinatal exposure to delta 9-tetrahydrocannabinol in rats: possible role of pituitary-adrenal axis. *Life Sci*, 56(23–24), 2169–2176 (PMID: 7776846).
- Saurel-Cubizolles, M. J., Prunet, C., & Blondel, B. (2014). Cannabis use during pregnancy in France in 2010. *BJOG: An International Journal of Obstetrics and Gynaecology*, 121(8), 971–977. <http://dx.doi.org/10.1111/1471-0528.12626>.
- Scher, M. S., Richardson, G. A., Coble, P. A., Day, N. L., & Stoffer, D. S. (1988). The effects of prenatal alcohol and marijuana exposure: Disturbances in neonatal sleep cycling and arousal. *Pediatric Research*, 24(1), 101–105.
- Schwilke, E. W., Schwöpe, D. M., Karschner, E. L., Lowe, R. H., William, D., Kelly, D. L., ... Huestis, M. A. (2009).  $\Delta^9$ -tetrahydrocannabinol (THC), 11-hydroxy-THC, and 11-nor-9-carboxy-THC plasma pharmacokinetics during and after continuous high-dose oral THC. *Clinical Chemistry*, 55(12), 2180–2189. <http://dx.doi.org/10.1373/clinchem.2008.122119>.
- Sharir, H., & Abood, M. E. (2010). Pharmacological characterization of GPR55, a putative cannabinoid receptor. *Pharmacology and Therapeutics*, 126(3), 301–313. <http://dx.doi.org/10.1016/j.pharmthera.2010.02.004>.
- Silva, L., Zhao, N., Popp, S., & Dow-Edwards, D. (2012). Prenatal tetrahydrocannabinol (THC) alters cognitive function and amphetamine response from weaning to adulthood in the rat. *Neurotoxicology and Teratology*, 34(1), 63–71. <http://dx.doi.org/10.1016/j.ntt.2011.10.006>.
- Smith, A. M., Fried, P. A., Hogan, M. J., & Cameron, I. (2004). Effects of prenatal marijuana on response inhibition: An fMRI study of young adults. *Neurotoxicology and Teratology*, 26(4), 533–542. <http://dx.doi.org/10.1016/j.ntt.2004.04.004>.
- Smith, A. M., Fried, P. A., Hogan, M. J., & Cameron, I. (2006). Effects of prenatal marijuana on visuospatial working memory: An fMRI study in young adults. *Neurotoxicology and Teratology*, 28(2), 286–295. <http://dx.doi.org/10.1016/j.ntt.2005.12.008>.
- Smith, A. M., Mioduszewski, O., Hatchard, T., Byron-Alhassan, A., Fall, C., & Fried, P. A. (2016). Prenatal marijuana exposure impacts executive functioning into young adulthood: An fMRI study. *Neurotoxicology and Teratology*, 58, 53–59. <http://dx.doi.org/10.1016/j.ntt.2016.05.010>.
- Sonon, K. E., Richardson, G. A., Cornelius, J., Kim, K. H., & Day, N. L. (2015). Prenatal marijuana exposure predicts marijuana use in young adulthood. *Neurotoxicology and Teratology*, 47, 10–15. <http://dx.doi.org/10.1016/j.ntt.2014.11.003>.
- Stella, N. (2010). Cannabinoid and cannabinoid-like receptors in microglia, astrocytes, and astrocytomas. *Glia*, 58(9), 1017–1030. <http://dx.doi.org/10.1002/glia.20983>.
- Stella, N. (2012). Neuroscience. Inflammation to rebuild a brain. *Science*, 338(6112), 1303–1304. <http://dx.doi.org/10.1126/science.1232331>.
- Stella, N., & Piomelli, D. (2001). Receptor-dependent formation of endogenous cannabinoids in cortical neurons. *European Journal of Pharmacology*, 425(3), 189–196. [http://dx.doi.org/10.1016/S0014-2999\(01\)01182-7](http://dx.doi.org/10.1016/S0014-2999(01)01182-7).
- Stella, N., Schweitzer, P., & Piomelli, D. (1997). A second endogenous cannabinoid that modulates long-term potentiation. *Nature*, 388(6644), 773–778. <http://dx.doi.org/10.1038/42015>.
- Stott, C., White, L., Wright, S., Wilbraham, D., & Guy, G. (2013). A Phase I, open-label, randomized, crossover study in three parallel groups to evaluate the effect of Rifampicin, Ketoconazole, and Omeprazole on the pharmacokinetics of THC/CBD oromucosal spray in healthy volunteers. *SpringerPlus*, 2(1), 236. <http://dx.doi.org/10.1186/2193-1801-2-236>.
- Thompson, B. L., Levitt, P., & Stanwood, G. D. (2009). Prenatal exposure to drugs: Effects on brain development and implications for policy and education. *Nature Reviews Neuroscience*, 10(4), 303–312. <http://dx.doi.org/10.1038/nrn2598>.
- Todd, S. M., & Arnold, J. C. (2016). Neural correlates of interactions between cannabidiol and  $\Delta^9$ -tetrahydrocannabinol in mice: Implications for medical cannabis. *British Journal of Pharmacology*, 173(1), 53–65. <http://dx.doi.org/10.1111/bph.13333>.
- Toennes, S. W., Schneider, K., Kauert, G. F., Wunder, C., Moeller, M. R., Theuvsen, E. L., & Ramaekers, J. G. (2011). Influence of ethanol on cannabinoid pharmacokinetic parameters in chronic users. *Analytical and Bioanalytical Chemistry*, 400(1), 145–152.
- Tornqvist, H., & Belfrage, P. (1976). Purification and some properties of a monoacylglycerol-hydrolyzing enzyme of rat adipose tissue. *Journal of Biological Chemistry*, 251(3), 813–819.
- Tortoriello, G., Morris, C. V., Alpar, A., Fuzik, J., Shirran, S. L., Calvigioni, D., ... Harkany, T. (2014). Miswiring the brain:  $\Delta^9$ -tetrahydrocannabinol disrupts cortical development by inducing an SCG10/stathmin-2 degradation pathway. *The EMBO Journal*, 33(7), 668–685. <http://dx.doi.org/10.1002/emboj.201386035>.
- Trezza, V., Campolongo, P., Cassano, T., Macheda, T., Dipasquale, P., & Carratù, M. R. (2008). Effects of perinatal exposure to delta-9-tetrahydrocannabinol on the emotional reactivity of the offspring: A longitudinal behavioral study in Wistar rats. *Psychopharmacology*, 198, 529–537. <http://dx.doi.org/10.1007/s00213-008-1162-3>.
- Trezza, V., Campolongo, P., Manduca, A., Morena, M., Palmery, M., Vanderschuren, L. J. M. J., & Cuomo, V. (2012). Altering endocannabinoid neurotransmission at critical developmental ages: Impact on rodent emotionality and cognitive performance. *Frontiers in Behavioral Neuroscience*, 6, 1–12. <http://dx.doi.org/10.3389/fnbeh.2012.00002>.
- Turner, S. E., Williams, C. M., Iversen, L., & Whalley, B. J. (2017). Molecular pharmacology of phytocannabinoids. *Progress in the Chemistry of Organic Natural Products*, 103, 61–101. [http://dx.doi.org/10.1007/978-3-319-45541-9\\_3](http://dx.doi.org/10.1007/978-3-319-45541-9_3).
- van Gelder, M. M. H. J., Reefhuis, J., Caton, A. R., Werler, M. M., Druschel, C. M., & Roeleveld, N. (2010). Characteristics of pregnant illicit drug users and associations between cannabis use and perinatal outcome in a population-based study. *Drug and Alcohol Dependence*, 109(1–3), 243–247. <http://dx.doi.org/10.1016/j.drugaldep.2010.01.007>.
- Varner, M. W., Silver, R. M., Rowland Hogue, C. J., Willinger, M., Parker, C. B., Thorsten, V. R., Goldenberg, R. L., Saade, G. R., Dudley, D. J., Coustan, D., Stoll, B., Bukowski, R., Koch, M. A., Conway, D., Pinar, H., Reddy, U. M., & Eunice Kennedy Shriver National Institute of Child Health and Human Development stillbirth collaborative research network (2014). Association between stillbirth and illicit drug use and smoking during pregnancy. *Obstet. Gynecol.* 123(1), 113–125. <http://dx.doi.org/10.1097/AOG.0000000000000052>.
- Vitalis, T., Lainé, J., Simon, A., Roland, A., Letierrier, C., & Lenkei, Z. (2008). The type 1 cannabinoid receptor is highly expressed in embryonic cortical projection neurons and negatively regulates neurite growth in vitro. *European Journal of Neuroscience*, 28(9), 1705–1718. <http://dx.doi.org/10.1111/j.1460-9568.2008.06484.x>.
- Wall, M. E., Sadler, B. M., Brine, D., Taylor, H., & Perez-Reyes, M. (1983). Metabolism, disposition, and kinetics of delta-9-tetrahydrocannabinol in men and women. *Clinical Pharmacology and Therapeutics*, 34(3), 352–363. <http://dx.doi.org/10.1038/clpt.1983.179>.
- Warf, B. (2014). High points: An historical geography of cannabis. *Geographical Review*, 104(4), 414–438. <http://dx.doi.org/10.1111/j.1931-0846.2014.12038.x>.
- Warner, T. D., Roussos-Ross, D., & Behnke, M. (2014). It's not your mother's marijuana: Effects on maternal-fetal health and the developing child. *Clinics in Perinatology*, 41(4), 877–894. <http://dx.doi.org/10.1016/j.clp.2014.08.009>.
- Warshak, C., Regan, J., Moore, B., Magner, K., Kritzer, S., & Hook, J. V. (2015). Association between marijuana use and adverse obstetrical and neonatal outcomes. *Journal of Perinatology*, 35(12), 991–995. <http://dx.doi.org/10.1038/jp.2015.120>.
- Watanabe, K., Yamaori, S., Funahashi, T., Kimura, T., & Yamamoto, I. (2007). Cytochrome P450 enzymes involved in the metabolism of tetrahydrocannabinols and cannabidiol by human hepatic microsomes. *Life Sciences*, 80(15), 1415–1419. <http://dx.doi.org/10.1016/j.lfs.2006.12.032>.
- Westfall, R. E., Janssen, P. A., Lucas, P., & Capler, R. (2009). Survey of medicinal cannabis use among childbearing women: Patterns of its use in pregnancy and retrospective self-assessment of its efficacy against “morning sickness”. *Complementary Therapies in Clinical Practice*, 15(4), 242–246. <http://dx.doi.org/10.1016/j.ctcp.2009.07.001>.
- Wise, R. A. (2004). Dopamine, learning and motivation. *Nature Reviews Neuroscience*, 5(6), 483–494. <http://dx.doi.org/10.1038/nrn1406>.
- Wu, C.-S., Zhu, J., Wager-Miller, J., Wang, S., O'Leary, D., Monory, K., ... Lu, H. C. (2010). Requirement of cannabinoid CB1 receptors in cortical pyramidal neurons for appropriate development of corticothalamic and thalamocortical projections. *The European Journal of Neuroscience*, 32(5), 693–706. <http://dx.doi.org/10.1111/j.1460-9568.2010.07337.x>.
- Zammit, S., Thomas, K., Thompson, A., Horwood, J., Menezes, P., Gunnell, D., ... Harrison, G. (2009). Maternal tobacco, cannabis and alcohol use during pregnancy and risk of adolescent psychotic symptoms in offspring. *British Journal of Psychiatry*, 195(4), 294–300. <http://dx.doi.org/10.1192/bjp.bp.108.062471>.
- Zuckerman, B., Amaro, H., & Cabral, H. (1989). Validity of self-reporting of marijuana and cocaine use among pregnant adolescents. *The Journal of Pediatrics*, 115. [http://dx.doi.org/10.1016/S0022-3476\(89\)80668-7](http://dx.doi.org/10.1016/S0022-3476(89)80668-7).