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Review

The endocannabinoid-CB_1 receptor system in pre- and postnatal life

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Abstract

Recent research suggests that the endogenous cannabinoids ("endocannabinoids") and their cannabinoid receptors have a major influence during pre- and postnatal development. First, high levels of the endocannaboid anandamide and cannabinoid receptors are present in the preimplantation embryo and in the uterus, while a temporary reduction of anandamide levels is essential for embryonal implantation. In women accordingly, an inverse association has been reported between fatty acid amide hydrolase (the anandamide degrading enzyme) in human lymphocytes and miscarriage. Second, CB_1 receptors display a transient presence in white matter areas of the pre- and postnatal nervous system, suggesting a role for CB_1 receptors in brain development. Third, endocannabinoids have been detected in maternal milk and activation of CB_1 receptors appears to be critical for milk sucking by newborn mice, apparently activating oral–motor musculature. Fourth, anandamide has neuroprotectant properties in the developing postnatal brain. Finally, prenatal exposure to the active constituent of marihuana (Δ^9-tetrahydrocannabinol) or to anandamide affects prefrontal cortical functions, memory and motor and addictive behaviors, suggesting a role for the endocannabinoid CB_1 receptor system in the brain structures which control these functions. Further observations suggest that children may be less prone to psychoactive side effects of Δ^9-tetrahydrocannabinol or endocannabinoids than adults. The medical implications of these novel developments are far reaching and suggest a promising future for cannabinoids in pediatric medicine for conditions including "non-organic failure-to-thrive" and cystic fibrosis.

Keywords: Cannabinoid; CB_1 receptor; Endocannabinoid-CB receptor system; Development; Failure-to-thrive; Implantation

Contents

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1. Introduction

Cannabinoid CB_1 receptors in the mature organism are widely and densely distributed in neural as well as non-neural tissue including brain, reproductive, immune, digestive systems as well as in peripheral neurons (Fride, 2002c; Maccarrone et al., 2002; Parolaro et al., 2002; Pertwee, 1997; Pinto et al., 2002). CB_2 receptors are mainly found in non-neural tissue (Lutz, 2002; Pertwee, 1997) although their presence on peripheral nerves is possible (Ibrahim et al., 2003). In the developing organism, CB_1 receptors have been investigated more thoroughly than that of CB_2 receptors.

Endogenous ligands for the cannabinoid receptors, denoted as "endocannabinoids", include thus far anandamide (arachidonyl ethanol amide (Devane et al., 1992)), 2-arachidonoyl glycerol (2-AG (Mechoulam et al., 1995)), noladin (arachidonyl glyceryl ether (Hanus et al., 2001)), the antagonist/partial agonist virodhamine (Porter et al., 2002) and NADA (*N*-arachidonoyl-dopamine (Walker et al., 2002)). This newly discovered physiological system will be denoted the 'endocannabinoid CB receptor' system.

2. Pre- and postnatal development of the endocannabinoid CB receptor system

2.1. Cannabinoid (CB_1 and CB_2) receptors

Cannabinoid CB_1 and CB_2 receptor mRNA has been detected as early as the pre-implantation period in the embryonal mouse (Paria and Dey, 2000) and has also been described around day 11 of gestation (Buckley et al., 1998). Postnatally, a gradual increase in CB_1 receptor mRNA (McLaughlin and Abood, 1993) and in the density of CB_1 receptors has been measured (Belue et al., 1995; Rodriguez de Fonseca et al., 1993) in whole brain.

Similar developmental patterns of CB_1 receptors were found during human pre- and postnatal development. Thus CB_1 receptors were detected at week 14 of gestation in the human embryo (Biegon and Kerman, 2001). In the 20th week of gestation, a selective expression of CB_1 receptors was recorded in the limbic area CA of the hippocampus and the basal nuclear group of the amygdala, as compared to a much wider CB_1 receptor expression of CB_1 mRNA in the adult human brain (Wang et al., 2003). A progressive increase in the concentrations of CB_1 receptors was found in the frontal cortex, hippocampus basal ganglia and cerebellum between the fetal period and adulthood (Mato et al., 2003). In contrast, high CB_1 receptor concentrations were present on several white matter neuronal tracts of the fetus, but had disappeared by infancy (Mato et al., 2003). In the same study, it was shown that at all stages of development, the CB_1 receptors were functionally active, since the agonist WIN55,212-2 ((*R*)-(+)-[2,3-dihydro-5-methyl-3-[4-morpholinylmethyl]-pyrrolo[1,2,3-de]1,4-benzoxazin-6-yl](1-naphthyl) methanone mesylate) stimulated [^{35}S]GTPγS binding.

2.2. Endocannabinoids

Anandamide has been detected from the early stages of the embryo in very high a amounts (Paria and Dey, 2000). During the fetal period, anandamide is present at much lower (1000-fold) concentrations than 2-arachidonoyl glycerol (Fernandez-Ruiz et al., 2000). Moreover, the fetal–postnatal developmental pattern differs between the two endocannabinoids. Thus, whereas concentrations of anandamide gradually increase throughout development until adult levels are reached (Berrendero et al., 1999), fetal levels of 2-arachidonoyl glycerol are similar to those in young and in adult brains with a remarkably distinct peak on the first day after birth in rats (Berrendero et al., 1999).

In the postnatal hypothalamus, anandamide displays a hardly perceptible rise from day 5 through adulthood, with peak levels immediately before puberty (Wenger et al., 2002).

Thus, the endocannabinoids and their receptors are abundantly present from the early developmental stages and are therefore likely to be important in the maturation of the nervous system and its functions.

3. The role of the endocannabinoid CB receptor system in gestation

CB_1 and CB_2 receptors are already present in the pre-implantation mouse embryo (Paria and Dey, 2000), the CB_1 receptor at higher concentrations than those in the brain (Yang et al., 1996). These observations led to the discovery that cannabinoids and endocannabinoids arrest the development of 2-cell embryos into blastocytes. Subsequent studies with CB_1 and CB_2 receptor antagonists indicated that the cannabinoid-induced embryonal growth arrest is mediated by CB_1 and not by CB_2 receptors (Paria and Dey, 2000; Schmid et al., 1997; Yang et al., 1996).

Further, anandamide levels in the uterus are very high; in order for implantation to take place, uterine anandamide levels have to be lowered on the day and on the site of implantation. Failing to do so prevents implantation of the embryo (Schmid et al., 1997). Thus the reduction in anandamide is a critical condition for the implantation and, hence, survival of the embryo.

Anandamide levels appear to be fine-tuned by varying levels of fatty acid amine hydrolase (FAAH), the enzyme which degrades anandamide. FAAH mRNA is present in preimplantation and implanting embryos as well as in the implantation site of uterus (Paria and Dey, 2000; Paria et al., 1999) where it inversely correlates with anandamide levels. Maccarrone et al. (2000a) reported down-regulation of FAAH in the uterus of pregnant and pseudopregnant mice during the implantation period. The same study also

suggested that FAAH levels are regulated by sex hormones. Moreover, when FAAH concentrations in the lymphocytes of women who spontaneously aborted were compared to FAAH concentrations in lymphocytes of women who gave birth, lower levels of the enzyme were measured in lymphocytes of women who subsequently aborted (Maccarrone et al., 2000b).

4. Role of cannabinoid receptors in neuronal development

Studies on the expression and functionality of the human CB_1 receptor in the developing brain have demonstrated that fetal brain CB_1 receptors are functionally active not only in regions which contain cannabinoid CB_1 receptors throughout life, such the cerebral cortex and hippocampus, but also in white matter such as the capsula interna and pyramidal tract and in proliferative zones such as the subventricular zone (Mato et al., 2003). These observations are consistent with investigations on the developing rat brain, in which "atypical distribution patterns" of CB_1 receptors (i.e., a transient presence during development in regions where none are found at adulthood) were detected in white matter regions including the corpus callosum and anterior commissure between gestational day 21 and postnatal day 5. These findings suggest a role for endocannabinoids in neuronal development (Fernandez-Ruiz et al., 2000; Romero et al., 1997).

Therefore, the varying developmental patterns of cannabinoid CB_1 receptors in different brain regions (Berrendero et al., 1999; McLaughlin and Abood, 1993) are probably due to the transient presence of cannabinoid CB_1 receptors in "atypical" regions during brain development (Fernandez-Ruiz et al., 2000).

5. Neuroprotection in the developing organism

Similarly to the neuroprotective effects of the endocannabinoid CB_1 receptor system in adults (Fride and Shohami, 2002), activation of CB_1 receptors in postnatal rats (7 days old) with WIN55,212 prevented neuronal loss (in a model of acute asphyxia), both immediate and delayed cell death. However, only delayed neurotoxicity was inhibited by the CB_1 receptor antagonist *N*-(piperidiny-1-yl)-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1*H*-pyrazole-3-carboxamide (SR141716A) (Martinez-Orgado et al., 2003). Moreover, exogenously applied anandamide reduced ouabain-induced neuronal damage in 1- and 7-day-old rat pups. The CB_1 receptor antagonist SR141716A reversed this neuroprotective effect of anandamide only in the 7-day-old pups (Van der Stelt et al., 2001). This study also suggested, however, the absence of a tonic neuroprotective activity of anandamide, since neither anandamide nor 2-archidonoyl glycerol was elevated after ouabain administration. In addition, SR141716A, when injected alone, did not enhance the ouabain-induced neuronal damage (Van der Stelt et al., 2001). In contrast, Hansen et al. (2001) observed a dramatic increase of anandamide precursors in the infant rat brain after head trauma, which is consistent with a role for anandamide as an endogenous neuroprotectant in the adult brain (Fride and Shohami, 2002; Panikashvili et al., 2001).

6. Prenatal manipulation of the endocannabinoid CB receptor system and the developing brain

Since the 1960s, a multitude of studies have attempted to assess potential adverse effects of marihuana use during pregnancy, on the offspring. Although description of the teratogenicity of the cannabis plant and its major psychoactive constituent Δ^9-tetrahydrocannabinol is beyond the scope of this article, the outcome of such studies has implications for the importance of the endocannabinoid CB receptor system during development. Thus functions which are not affected by prenatal Δ^9-tetrahydrocannabinol are unlikely to be regulated by the developing endocannabinoid CB receptor system. Conversely, the development of structures or functions which are significantly altered in the prenatally exposed offspring is probably regulated, at least in part, by the endocannabinoid CB receptor system. Therefore, major trends in the teratogenicity research of cannabinoids will be reviewed briefly.

In animal studies, a number of behavioral, hormonal and neurochemical sequelae of prenatal Δ^9-tetrahydrocannabinol exposure have been reported, mainly between the 1960s and 1980s. Taken together, the majority of these investigations have reported subtle somatic as well as functional impairments, immediately after birth and/or later in life (see Fride and Mechoulam, 1996a; Fried, 1996; Fried et al., 2001). It seems, however, that confounding variables such as impaired feeding patterns of the cannabinoid-exposed dams may have been responsible for many of these prenatal Δ^9-tetrahydrocannabinol effects (Hutchings et al., 1989, 1991).

Recent studies have focused on more specific deficits in the prenatally exposed offspring. For example, pre- and postnatal exposure to Δ^9-tetrahydrocannabinol interfered with normal dopamine-dependent motor functions and the hypothalamic–pituitary–adrenal stress axis (Bonnin et al., 1995; Navarro et al., 1994; Ramos et al., 2002; Rodriguez de Fonseca et al., 1991). Further, prenatal Δ^9-tetrahydrocannabinol facilitated morphine self-administration in the adult female rat offspring, while a number of brain areas including the prefrontal cortex, amygdala and hippocampus displayed altered concentrations of mu-opioid receptors (Vela et al., 1998). Memory retention in the adult offspring in a passive avoidance task was disrupted by prenatal exposure to the synthetic cannabinoid WIN55,212. The memory impairment was correlated with a shortening of long-term potentiation and a reduction in extracellular

glutamate in the hippocampus (Mereu et al., 2003). In an elegantly designed prospective study of the children of marihuana smoking mothers, Fried and colleagues have specifically pointed at a subtle but significant impairment of higher cognitive ('executive') functioning, which is ascribed to the prefrontal cortex. This deficiency only becomes apparent from the age of 4, since at this age executive functioning starts to be expressed (Fried, 1996; Fried et al., 1998, 2003).

Since the identification of the endocannabinoids (see Fride, 2002c), the effects of prenatally manipulating the endogenous system have been investigated, often comparing the results to the administration of exogenous cannabinoids. These experiments comprise an important extension of the pre- and perinatal studies, because they represent a more physiological manipulation of the endocannabinoid CB receptor system. Thus, when anandamide was administered daily at low doses (0.02 mg/kg) to rats during the last week of pregnancy, only transient, mainly inhibitory, effects on a number of reproductively relevant hormones were detected. The effects had disappeared within days after birth (Wenger et al., 1997). Interestingly, Fried et al. (2001) did not record changes in pubertal milestones in the children of marijuana smoking mothers.

We have found decreased inflammatory responsiveness in mice which had been prenatally exposed to anandamide (Fride and Mechoulam, 1996a). We have performed experiments on the adult offspring of mice injected daily with anandamide (or Δ^9-tetrahydrocannabinol) during the last week of gestation, with the aim to detect changes specific to the endocannabinoid CB receptor system per se (Fride and Mechoulam, 1996a). Thus when these offspring were observed in the "tetrad" (a series of four in vivo assays for cannabinoid-mediated effects), they performed similarly to naïve animals which had been injected with 5–10 mg/kg Δ^9-tetrahydrocannabinol (Fride et al., 1996). In other words, the prenatally exposed offspring displayed a permanent, moderate "high". Consistent with these data, we found a higher concentration of CB_1 receptors in the forebrain of anandamide and Δ^9-tetrahydrocannabinol-exposed offspring (Fride et al., 1996). These data are consistent with an overactive endogenous cannabinoid CB_1 receptor system resulting, perhaps, in a greater vulnerability to the addictive potential of cannabis or other drugs (Vela et al., 1998).

In a recent study, Viggiano et al. (2003) induced an elevation of endogenous anandamide levels by administering the reuptake inhibitor AM-404 (*N*-(4-hydroxyphenyl) arachidonoyl amide) to pregnant "NHE" rats, an animal model for "attention deficit hyperactivity disorder" (ADHD). Thus they reduced certain aspects of hyperactivity and the hypertrophicity of the mesocorticolimbic dopamine system in the offspring.

Taken together, prenatal manipulation of the endocannabinoid CB_1 receptor system, whether by exogenous or endogenous agents, produces long-term effects in the offspring. These include disruptions of memory, addictive and motor behaviors and higher cognitive 'executive' functioning of the prefrontal cortex, suggesting that the endocannabinoid CB receptor system plays a role in the prenatal development of cognitive function and addictive behaviors. This is consistent with reports that CB_1 receptors are involved in neural development in the fetus (see Role of cannabinoid receptors in neuronal development). Further investigations of the role of the endocannabinoid CB receptor system in the development of these functions and their neurochemical/anatomical underpinnings seem therefore warranted.

7. Milk suckling and survival during the neonatal period

The involvement of marihuana in feeding and appetite was demonstrated decades ago (Abel, 1971; Fride, 2002b); endocannabinoids appear to fulfill a similar role (Fride, 2002c; Williams and Kirkham, 1999). Endocannabinoids have been detected in bovine as well as human milk, 2-arachidonoyl glycerol (2-AG) in at least 100- to 1000-fold higher concentrations than anandamide (Di Marzo et al., 1998; Fride et al., 2001).

Is it possible that the high levels of CB_1 receptor mRNA and 2-AG which have been observed on the first day of life in structures including the hypothalamic ventromedial nucleus (Berrendero et al., 1999) (which is associated with feeding behavior) comprise a major stimulus for the newborn to initiate milk intake?

In a series of studies performed in neonatal mice, we have demonstrated that CB_1 receptors are critically important for the initiation of the suckling response. Thus a single injection of the CB_1 receptor antagonist SR141716A to newborn mice completely inhibited milk ingestion and subsequent growth in most pups (75–100%) and death followed within days after antagonist administration (Fride et al., 2001). The antagonist must be administered within 24 h after birth in order to obtain the full effect: Injections on day 2 result in a 50% death rate; SR141716A administration on day 5 has no effect at all on pup growth and survival (Fride, 2002b; Fride et al., 2003b).

Subsequent studies indicated that the catastrophic effect of CB_1 receptor blockade in neonates is dose-dependent and specifically mediated by CB_1 receptors. Thus CB_2 antagonists did not affect pup suckling and growth, and Δ^9-tetrahydrocannabinol co-application almost completely reversed the SR141716A-induced growth failure (Fride et al., 2001). We have replicated this phenomenon now in three different strains of mice (Sabra, C57BL/6 and ICR).

While these experiments were in progress, CB_1 receptor-deficient mice were generated in two different laboratories (Ledent et al., 1999; Zimmer et al., 1999). In order to resolve the paradox of the existence of mice completely lacking CB_1 receptors, with our finding that blockade of CB_1 receptors after birth is incompatible with survival, at least for most neonates, we studied early development and

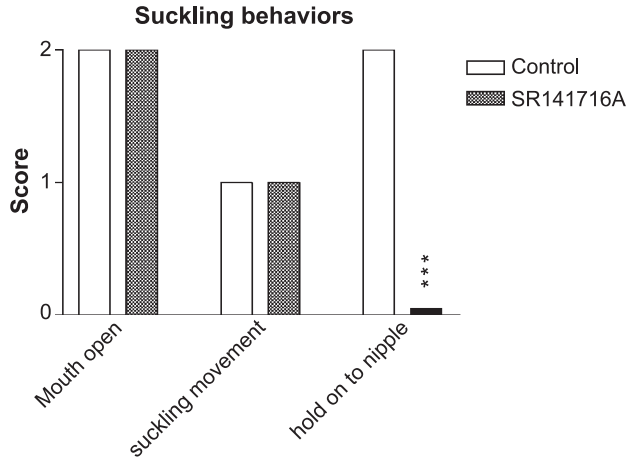

Fig. 1. Suckling behaviors in ICR mouse pups. Half the pups of each litter was injected with SR141716A (20 mg/kg in Ethanol:emulphor:saline=1:1:18, see (Fride et al., 2001)) within 24 h after birth. The next day they were observed for suckling parameters with an anesthetized dam (opening of the mouth, score 0–2, suckling movements, score 0–1, the strength by which the pup holds onto the nipple, score 0–2). ***P<0.001, SR141716A-injected pups, cf. control, Mann–Whitney test.

milk intake of neonatal $CB_1^{-/-}$ pups, provided by Dr. Zimmer's laboratory (Fride et al., 2003b). Interestingly, CB_1 knockout pups did not nurse on the first day of life. However, by day 3 of life they had developed normal suckling behavior. Their weight gain, though, remained significantly reduced compared to the C57BL/6 background strain. Further, as expected, the growth curve of CB_1 receptor knockout mice was not affected by neonatal injections of the cannabinoid CB1 receptor antagonist. On the other hand, survival rate and the initiation of the suckling response were significantly inhibited by the CB_1 receptor blocker, suggesting the existence of an additional "CB_3" receptor, possibly up-regulated in the $CB_1^{-/-}$ knockout mice (Fride et al., 2003b).

Recent experiments in our laboratory were designed to further analyze potential physiological/behavioral mediators by which the neonatally administered CB_1 receptor antagonist prevents the development of milk ingestion. Thus 2- to 11-day-old pups which had been injected with SR141716A, or with vehicle, within 24 h after birth, were allowed to nurse from an anesthetized nursing dam. While vehicle-injected pups all located the nipples and nursed from the dam on every testing day, none of the SR141716A-injected pups did so on the day after injection. Only the pups which survived the SR14171A injection gradually developed the suckling response and suckled like controls by the end of the first week (Ezra and Fride, unpublished observations). In a further experiment, we manually brought the pups in proximity of the nipple and scored them for components of behavior which are required for successful milk ingestion (opening of the mouth, score 0–2, suckling movements, score 0–1, the strength by which

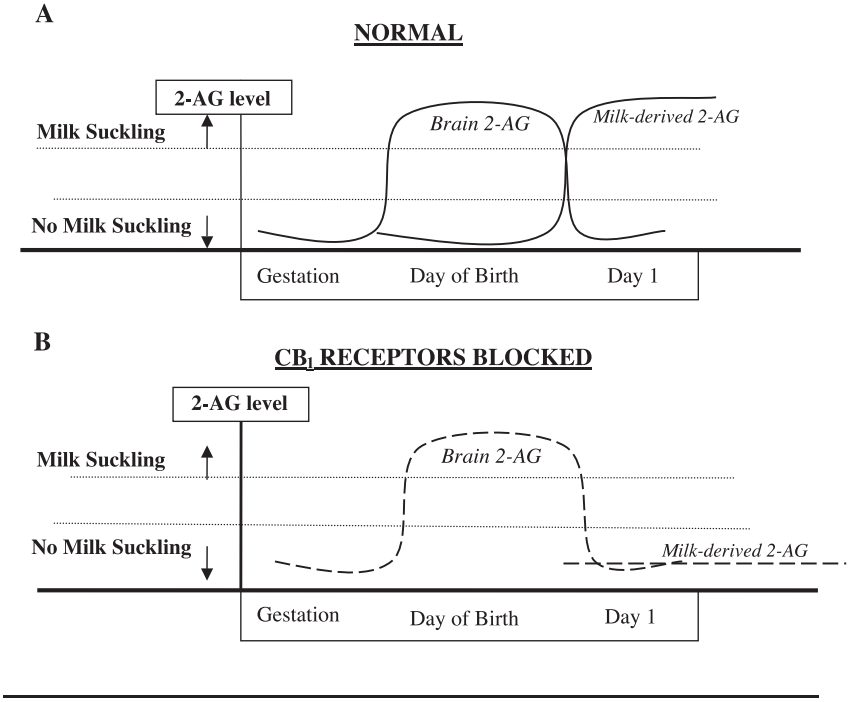

Fig. 2. A schematic model of the first days of life in a rodent pup. (A) On the day of birth, brain levels of 2-AG are sufficiently high to stimulate suckling, through activation of cannabinoid CB_1 receptors. Cannabinoid CB_1 receptor activation enables sucking and holding on to the nipple (see Fig. 1 and Milk suckling and survival during the neonatal period). Subsequently, the ingested maternal milk supplies new 2-AG which stimulates the CB_1 receptor in pup brains from day 2 and further on. (B) When CB_1 receptors are blocked on the day of birth, the first sucking episodes induced by brain 2-AG are not translated into milk ingestion, due to the putative oral–motor impairment. Hence no 2-AG from milk is supplied and the endogenous 2-AG is apparently not present at sufficiently high levels to stimulate milk sucking from day 2 onward.

the pup holds onto the nipple, score 0–2). We observed that SR141716A-treated pups open their mouth and made suckling movements, but they were not able to hold on to the nipple (Fig. 1, Ezra and Fride, unpublished observations). These data suggest that CB1 receptor blockade does not interfere with the motivation to suckle milk ('appetite'), but interferes with the efficiency of the oral musculature.

Based on data gathered thus far (Berrendero et al., 1999; Fride et al., 2001a,b, 2003b), we now propose the following model for the initiation of the milk suckling process during the first days postnatally in the mouse (see Fig. 2): At birth, the 2-AG content in the brain ('brain 2-AG') is sufficiently high to stimulate the suckling response. Normally then, the pups start sucking soon after birth. Upon milk intake, 2-AG from the maternal milk ('milk-derived 2-AG') elevates the levels of 2-AG in the pup's brain, so that by the second day of life and further on, the milk-derived 2-AG stimulates suckling. If brain 2-AG is prevented from effecting milk sucking (as in the Cannabinoid CB1 receptor antagonist-treated pups), milk is not ingested, therefore, milk-derived 2-AG is not present in the brain to stimulate milk sucking on day 2 of life, and the 'window' to develop a pattern of suckling behavior has closed.

Based on the complex relationship between thermoregulation, ultrasonic vocalization (Blumberg and Sokoloff, 2001; Kraebel et al., 2002; McGregor et al., 1996), suckling (Stern and Azzara, 2002; Stern and Lonstein, 1996) and maternal behavior (Branchi et al., 1998; Brunelli et al., 1994), we decided to study body temperature and ultrasonic vocalizations, in SR141716A-treated pups throughout postnatal development. Thus we have observed that the SR141716A-treated pups are hypothermic, while their ultrasonic vocalizations are inhibited (a preliminary report of these data was reported in Fride et al., 2003a). Present experiments are aimed at delineating the sequence of events induced by the blockade of the CB_1 receptor immediately after birth: is a hypothermic pup unable to call his mother to stimulate the suckling response? Or perhaps, does the pup who fails to call his mother become hypothermic and thus does not have the motor capability to suckle? Alternatively, is CB_1 receptor activation necessary for ultrasonic vocalizations as shown for 11- to 12-day-old rat pups (McGregor et al., 1996), thereby interfering with the pup's ability to attract maternal attention and nursing?

Whichever sequence of events underlies the sequelae of effects induced by CB_1 receptor blockade in neonates, we propose our experimental model as an explanatory framework for a condition in infants denoted "non-organic failure-to-thrive": Failure-to-thrive is commonly defined as an abnormally low weight and/or height for age (Reilly et al., 1999; Skuse, 1985). Non-organic failure-to-thrive is defined as failure-to-thrive without a known organic cause. Traditionally, impaired mother–infant relations were blamed for this condition (Jolley, 2003). However, recent research points to non-organic failure-to-thrive as a mild neurodevelopmental disorder or pathophysiology (Ramsay et al., 2002) in which an oral–motor defect plays a central role and results in deficient sucking and/or milk ingestion by the infant (Mathisen et al., 1989; Reilly et al., 1999; Suss-Burghart, 2000).

We propose, based on our findings, that an impaired endocannabinoid-CB_1 receptor system may underlie non-organic failure-to-thrive. This hypothesis is currently under investigation.

8. Cannabinoids in pediatric medicine

The gradual postnatal increase of anandamide and its CB_1 receptors (see Pre- and postnatal development of the endocannabinoid CB receptor system) is accompanied by a gradual maturing response to the psychoactive potential of Δ^9-tetrahydrocannabinol and anandamide in postnatal mice between birth and weaning (Fride and Mechoulam, 1996b).

This observation has important implications for cannabinoid therapy in children, since psychoactive side effects may be expected to be minor when treated with cannabinoids at a young age. Indeed, very high doses of Δ^8-tetrahydrocannabinol (approximately 0.64 mg/kg/treatment) were given to children between the ages 3 and 13 years who were undergoing chemotherapy for the treatment of various hematologic cancers, over long periods of time (up to 114 treatments, based on 4 treatments/24 h during the days of chemotherapy). The anti-emetic effects were impressive, whereas the side effects were minimal (Abrahamov and Mechoulam, 1995). In a case report study (Lorenz, 2003), eight children (ages 3–14 years) with a variety of severe neurological diseases were treated with Δ^9-tetrahydrocannabinol (0.04–0.12 mg/kg/day). Significant improvements in behavioral parameters including reduced spasticity, improved dystonia, increased interest in the surroundings and anti-epileptic activity were reported without notable adverse effects.

It is not clear, how, in the first study, the anti-emetic effects were achieved (presumably via the *area postrema*) and in the second, positive neurological benefit was derived in the absence of adverse psychological effects. Is it possible that a differential CB_1 receptor distribution appears during development, or that differential maturation of brain pathways is responsible for the clinical success? Clearly, further animal experiments and clinical investigations of cannabinoid treatment in the developing organism are warranted.

In a previous publication (Fride, 2002a), we have suggested that a deficient endocannabinoid CB receptor system may underlie at least some of the symptoms of cystic fibrosis, such as malnutrition, gastrointestinal problems, inflammatory exacerbations, and fatty acid imbalance. Therefore, treatment with cannabinoids or drugs which

target the endocannabinoid CB receptor system, such as anandamide reuptake inhibitors or inhibitors of endocannabinoid breakdown, or Δ^9-tetrahydrocannabinol itself, may benefit children (and perhaps young adults) afflicted with cystic fibrosis.

9. Conclusions

The endocannabinoids and their receptors (CB_1, CB_2 and the putative CB_3 receptor) (Breivogel et al., 2001; Fride et al., 2003b) fulfill a multitude of physiological functions, including immunological, neurological, psychiatric and cardiovascular. Our knowledge of the various roles of the endocannabinoid CB receptor system in developmental processes is still sketchy. However, from the knowledge accumulated until now, it appears that while the endocannabinoid CB receptor system contributes to various physiological processes in the adult, in the developing organism, proper functioning of the endocannabinoid CB receptor system is acutely critical for survival, at least during two specific stages: implantation of the embryo and the initiation of suckling in the newborn.

Interestingly, opposite requirements are involved in each of these processes. A reduction of anandamide and CB_1 receptor activation is necessary for implantation to take place (Paria and Dey, 2000), while the initiation of suckling requires the activation of CB_1 receptors, presumably by the presence of high levels of 2-AG (Fride, 2002b; Fride et al., 2001, 2003b).

Psychoactive side effects of cannabinoid treatment seem to be absent or much reduced in children. Although the reasons for this phenomenon are largely unclear, further understanding of the underlying mechanisms will hopefully lead to the development of cannabinoid-based therapeutic strategies for the treatment of disorders including infant "non-organic failure-to-thrive" and cystic fibrosis.

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