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Increased palatability of ethanol after prenatal ethanol exposure is mediated by the opioid system

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Abstract

Previous studies have shown that prenatal exposure to a moderate dose of ethanol (2 g/kg) during the last days of gestation of the rat (17–20) not only increases postnatal intake of the drug but also enhances the palatability of ethanol's taste when measured with a taste reactivity test. Prenatal administration of the opioid antagonist naloxone, together with ethanol, reduces ethanol intake. The aim of the present study was to analyze whether this decreased intake of ethanol after the administration of naloxone is accompanied by a reduction in ethanol's palatability. Results show that preweanling rats exposed prenatally to ethanol alone displayed more ethanol intake and more ingestive responses in reaction to its taste than non-exposed pups. Simultaneous prenatal administration of naloxone with ethanol prevented both the increased intake of ethanol and the higher amount of appetitive responses to its taste. These results indicate that the opioid system plays an important role in the effect of enhanced palatability of ethanol's taste after its prenatal exposure. Results also support the hypothesis of a conditioned response established in utero as a consequence of the association between ethanol's chemosensory and reinforcing aspects, the latter mediated by the opioid system.

Keywords: Rat; Ethanol; Prenatal exposure; Infant; Taste reactivity test; Naloxone; Intake; Conditioned response

The rat fetus has the capacity for perceiving the chemosensory properties of the amniotic fluid and other substances present in their prenatal milieu (Pedersen et al., 1986, 1983; Smotherman and Robinson, 1990). This fetal capacity has a direct relation to postnatal responses towards those substances (Teicher and Blass, 1977). For instance, it has been demonstrated that olfactory cues guiding rat neonates in their first nipple attachment are substances contained in the amniotic fluid. Contamination of the amniotic fluid with a flavored substance has been found to modify that early suckling behavior (Blass and Pedersen, 1980), and also can increase intake of that substance later in life (Smotherman, 1982a). It also has been shown that the rat fetus has the capacity for acquiring conditioned responses to chemosensory stimuli and that this prenatal associative memory can be expressed during infancy and adolescence (Abate et al., 2002; Chotro and Arias, 2003; Molina and Chotro, 1991; Stickrod et al., 1982).

The opioid system seems to be implicated in learning processes modulating the acquisition of taste or odor preferences during early infancy (Kehoe and Blass, 1986). Some authors suggest that this neurochemical system has a distinctive role in neonatal rat learning, temporally limited to a sensitive period that ends on postnatal day 9, in which pups learn easily odor preferences (Roth and Sullivan, 2001, 2003). Although it is not specified by those authors, it is conceivable that this sensitive period may also include the last prenatal period. In fact during the last days of gestation it has been observed that, as previously mentioned, chemosensory preferences can be acquired (Abate et al., 2001; Chotro and Arias, 2003; Molina and Chotro, 1991; Smotherman, 1982b; Stickrod et al., 1982) and also that the opioid system is involved in prenatal learning processes (Arnold et al., 1993; Chotro and Arias, 2003; Robinson et al., 1993a). It has been demonstrated that in the rat fetus, the opioid receptors subtypes μ and κ are functional and are capable of modulating fetal behavior during the last days of gestation (Smotherman et al., 1993). It also has been shown that the activity of the opioid system (specifically, μ opioid receptors) can be conditioned prenatally after pairing a

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chemosensory CS with a US that promotes the release of endogenous opioids, and that the rat fetus is capable of exhibiting a conditioned opioid response when the CS is again presented (Arnold et al., 1993). The administration of opioid antagonists, either nonselective or selective for each receptor subtype, has been found to be effective in modifying those fetal responses known to be regulated by the opioid system (Robinson et al., 1993b; Smotherman and Robinson, 1994).

Prenatal exposure to ethanol has been shown to affect subsequent recognition of the drug's odor and/or taste (Chotro and Molina, 1990; Chotro and Spear, 1997). Several studies have demonstrated that the rat fetus not only can perceive ethanol's chemosensory properties but also can form an association involving the taste and odor of this drug (Chotro et al., 1991; Chotro and Molina, 1990; Molina and Chotro, 1991). The fetus may also learn about the toxic aspects of the drug. When ethanol is administered to the pregnant rat, fetuses are exposed to the chemosensory as well as the toxic aspects of ethanol. The drug is rapidly distributed to fetal tissues reaching levels in fetal blood equal to those in maternal circulation (Szeto, 1989). Alcohol also accumulates in the amniotic fluid, and previous data have demonstrated that 60 min after the administration of a relatively low ethanol dose to the pregnant dam, the concentration of the drug in the amniotic fluid is enough to be perceived by the rat fetus (Chotro and Molina, 1990; Szeto, 1989). This prenatal experience with ethanol chemosensory aspects together with its toxic properties has been found to promote subsequent changes in responsiveness to the drug. For example, repeated administrations of low to moderate ethanol doses (1 or 2 g/kg) to the pregnant rat during gestational days (GD) 17–20 have been found to increase ethanol consumption in infant rats (Arias and Chotro, 2005; Chotro and Arias, 2003) (Dominguez et al., 1996, 1998; Molina et al., 1995).

The participation of the opioid system in the reinforcing properties of ethanol has been well demonstrated in humans and in animals. In fact, low blood ethanol levels have been found to stimulate the activity of the opioid system (Acquas et al., 1993), and the administration of μ-receptor agonists has been found to increase ethanol intake (Stromberg et al., 1997). The administration of nonselective opioid antagonists such as naloxone or naltrexone has been shown to reduce ethanol intake in adult rats and also to modify reactivity to its taste (Coonfield et al., 2002; Critcher et al., 1983; Hill and Kiefer, 1997; Stromberg et al., 1998a,b, 2002). It has been suggested that opioid antagonists reduce ethanol intake not only by decreasing the level of reward after ethanol ingestion but also by changing its palatability in a negative manner (Coonfield et al., 2002). However in all those studies in which a reduction of ethanol intake was observed under naltrexone, ethanol consumption returned to control levels when the naltrexone treatments were stopped (Coonfield et al., 2004; Goodwin et al., 2001; Hill and Kiefer, 1997). This may indicate that, at least with the doses used in those studies, naltrexone did not induce by itself a conditioned taste aversion to ethanol. Yet, there are studies showing that naloxone and naltrexone induce place aversion and taste aversion at moderate to high doses (Lett, 1985; Mucha, 1989; Mucha and Herz, 1985; Parker and Rennie, 1992; Stolerman et al., 1978).

Considering the role of the opioid system on perinatal learning processes as well as on the reinforcing aspects of ethanol, it has been proposed that the above mentioned increase in ethanol consumption observed after prenatal exposure to the drug could be a conditioned preference mediated by the opioid system. That is, the fetus perceives ethanol chemosensory aspects which could act as a conditioned stimulus (CS) and also experiences the reinforcing properties of the drug (unconditioned stimulus, US), the latter mediated by the opioid system. This hypothesis is supported by recent data showing that the effect of augmented ethanol intake was not observed in pups whose mothers were administered naloxone together with ethanol (Chotro and Arias, 2003). This result can be explained by the blockage of the reinforcing properties of ethanol, what would prevent the occurrence of the prenatal association. In that same study it was observed that postnatal re-exposure to ethanol and naloxone decreased ethanol intake in pups prenatally exposed to the drug, possibly by extinguishing the conditioned response.

In a more recent study the hedonic nature of the prenatal ethanol experience has been further investigated using a taste reactivity test in 14 day old rats (Arias and Chotro, 2005). It was found that pups exposed to ethanol in utero, when compared to non-exposed controls, not only consumed more ethanol but also displayed more appetitive responses (mouthing and paw licking) and less aversive behaviors (defined as general activity and wall climbing) in reaction to ethanol taste. Pups prenatally exposed to ethanol generalized those responses to a sucrose+quinine compound (SQ), which shares palatability attributes with alcohol (Kiefer et al., 1988), and they also showed higher intake of this solution than controls. Those results suggest that in the infant rat the palatability of the taste of ethanol was enhanced after exposure to the drug during the last days of gestation. They also agree with results of previous studies indicating that prenatal ethanol exposure induces a preference for this drug (Chotro and Arias, 2003; Chotro et al., 1996; Dominguez et al., 1996, 1998; Molina et al., 1995).

Accordingly, the aim of the present study was to analyze whether the administration of an opioid antagonist together with ethanol during the last gestational days affects the palatability of this drug. The response to the SQ compound taste, a stimulus that shares orosensory but not toxic properties with ethanol, was also analyzed. The hypothesis guiding the present study was that infant rats exposed prenatally to ethanol acquire in utero a conditioned preference for the ethanol taste and that naloxone administered together with ethanol prenatally will block or reduce the conditioning and therefore will decrease not only postnatal ethanol intake but also the palatability of ethanol's taste.

1. Materials and methods

Subjects for this experiment were 168 preweanling rats, 88 males and 80 females, obtained from 19 Wistar female rats.