Dopamine, but not noradrenaline, contributes to opiate withdrawal-induced anxiety in the VTA

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Background

• Drug addiction is maintained by negative reinforcement – alleviation of the negative withdrawal state.¹
• Acute (one time) administration of morphine, an opiate drug, elicits negative emotional symptoms of withdrawal such as anxiety in rats.²
• The withdrawal potentiated startle (WPS) paradigm is used to assess the negative withdrawal state in rats. It relies on the acoustic startle reflex, which has been shown to be elevated during acute morphine withdrawal, and represents the anxiety-like symptoms of withdrawal.³
• Research in the Gewirtz laboratory has shown that one of the brain structures involved in mediating withdrawal from opiate drugs such as morphine is the ventral tegmental area (VTA). Morphine causes the VTA to release dopamine and excite downstream targets, which may be related to the onset of withdrawal.

• In addition, increased levels of noradrenaline are also present during opiate withdrawal. 3-4
• We aim to test whether both the dopaminergic and adrenergic systems engage to produce WPS when morphine is supplied to the VTA. Thus, we administer propranolol, an adrenergic receptor antagonist, to determine whether noradrenaline is recruited by the VTA during withdrawal. Apomorphine, a dopamine agonist, is given to determine the actions of dopamine during withdrawal.

Methods

• Acoustic Startle Reflex: Rats were exposed to 20 95 and 105-dB white noise bursts presented in a pseudorandom order. Two habituation sessions were given prior to morphine exposure. On test days animals received startle tests before and after morphine exposure (pretreat and posttest, respectively).
• Cannulation Surgery: Bilateral guide cannulae were implanted into the VTA (AP:+3.3, ML:+1.0, DV:+.72 from Bregma). Correct placement was confirmed by histological analysis.
• Microinfusion: Morphine sulfate (2 mg) or saline was infused in a volume of 0.5 μl over a period of 2 min.
• Injection: Morphine sulfate (10 mg/kg), apomorphine (50 and 100 μg/kg), propranolol (10mg/kg) and saline controls injected s.c.
• Expt 1: Time course of WPS following intracranial morphine. Rats were infused with morphine or saline (0 hrs) in or dorsally into the VTA (control). Startle was tested 2, 4, 6, and 8 hrs after infusion.
• Expt 2: Effects of systemic propranolol on intra-VTA morphine. Rats were infused with intra-VTA morphine or sterile saline (0 hrs) and injected with propranolol 3:50 hrs later. Startle was tested at 4:00 hrs.
• Expt 3: Effects of systemic apomorphine on systemic morphine. Rats were injected with morphine or saline (0 hrs) followed by injections of apomorphine 3:50 hrs later. Startle was tested at 4:00 hrs.

Results

Fig 1 Time course of spontaneous withdrawal potentiated startle following intra-VTA morphine. Experimental time line (A). The effect of intra-VTA infusion shows significant startle potentiation four hours after administration as compared to the saline-treated control group (B). Morphine infused dorsally into the VTA does not differ from the saline group (C).

Fig 2 Systemic propranolol does not attenuate withdrawal-potentiated startle. Anxiolytic drug propranolol was delivered prior to the four-hour morphine withdrawal peak (A), and did not startle dose-dependently (B), suggesting that the agonistic action prevents withdrawal caused by morphine.

Fig 3 Systemic apomorphine attenuates startle potentiation during withdrawal. Apomorphine, a dopamine agonist, was used before the posttest (A). It decreases the startle response and significantly reduce the startle response due to withdrawal as compared to the saline group (B).

Conclusions

• Acute exposure to morphine in rats leads to peak withdrawal at four hours in the WPS paradigm.
• Systemic administration of propranolol, a noradrenergic antagonist, does not attenuate withdrawal potentiation startle caused by intracranial delivery of morphine into the VTA.
• This effect could be due to noradrenaline getting activated by a different brain structure. An experiment in which morphine is administered to other local structures, again followed by propranolol, could help pinpoint where noradrenaline gets activated during withdrawal.
• The dopamine agonist apomorphine administered systemically prevents startle potentiation at the peak time of morphine withdrawal. This dose-dependent attenuation indicates that increased levels of dopamine act to oppose the manifestation of morphine withdrawal.
• A proposed mechanism might be that withdrawal is caused by declining levels of morphine at four hours after the initial transient rise in dopaminergic activity in VTA targets. Apomorphine mimics dopamine, and thus sufficient dopaminergic activity is recovered to prevent the onset of withdrawal.
• Attenuation by systemic apomorphine leads us to speculate its target structures in the brain; further study must include intracranial administration of the agonist into a specific location.

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References