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Research Article

The value of extended amygdala structures in emotive effects of narcogenic with diverse chemical structure

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Abstract

Introduction: Studies on the mechanisms of the reinforcing action of opioid and non-opioid narcotics confirmed the existence in the brain of a specialized system named the extended amygdala.

Materials and methods: To clarify the value of the extended amygdala structures (bed nucleus, central nucleus of the amygdala and nucleus accumbens shell) in the mechanisms of unconditioned and conditioned reinforcement activated by various narcogenic, this paper carried out a neuropharmacological analysis of these effects, using blockade of dopamine receptors, GABA, opioids and CRF receptors within these brain structures, as well as an analysis of behavioral responses by self-stimulation (unconditioned reinforcement) and conditioned place preference (CPP) (conditioned reinforcement).

Results and discussion: The central amygdala and the bed nucleus have a controlling influence on the hypothalamus, which is predominantly of CRF-, GABA- and dopaminergic nature. Through D1 dopamine receptors,, a direct positive (activating) effect on the lateral hypothalamus is made. The D2 receptor blockade of the nucleus accumbens prevents narcogenic from exerting the reinforcing properties, which are primarily stimulating. The blockade of the D1 receptors of the nucleus accumbens by SCH-23390 prevents the expression of unconditioned and conditioned reinforcing properties of predominantly opiates and opioids. The blockade of $GABA_A$ receptors in the nucleus accumbens with bicuculline prevents the manifestation of the primary and secondary reinforcing properties (CPP) of psychostimulant drugs (amphetamine), without affecting the effects of opiates and opioids (fentanyl and leu-enkephalin).

Conclusion: The pharmacological analysis proves that CRF, dopamine and GABA receptors are most important for the correction of reinforcement activated by various narcogenic.

Keywords

reinforcement, extended amygdala, narcogenic, amphetamine, fentanyl, leu-enkephalin, dopamine, corticoliberin.

Introduction

The evolution of views on the mechanisms of the reinforcing action of opioid and non-opioid narcotics confirmed the existence in the brain of a specialized system of emotiogenic structures, first of all, structures of the extended amygdala. It includes the bed nucleus of the stria terminalis (BNST), the central nucleus of the amygdala (CeA), the medial part (shell) of the nucleus accumbens (NA) and the substantia innominata receiving the innervation

from the dopaminergic neurons of the ventral forebrain bundle (Koob 2009a, Koob 2009b, Shabanov and Lebedev 2008). It is the structures of the extended amygdala complex, originally identified on the basis of their neuroanatomical similarity (Alheid and Heimer 1996) that make up the main structural and functional system for delivering emotional and motivational effects of various narcogenic mediating their action on effector organs (Giardino et al. 2018, Shabanov and Lebedev 2008).

In order to clarify the value of the extended amygdala structures (the bed nucleus, the central nucleus of the amygdala and the nucleus accumbens shell) in the mechanisms of unconditioned and conditioned reinforcement activated by various narcogenic (psychotropic substances), a neuropharmacological analysis of these effects was carried out in this paper, using blockade of dopamine, GABA, opioids and CRF receptors within these brain structures, and an analysis of behavioral responses by self-stimulation (unconditioned reinforcement) and conditioned place preference (conditioned reinforcement). Thus, a number of fundamental questions of the experimental and clinical pharmacology of narcogenic were addresses to, namely: 1) to find out the value of the structures of the extended amygdala complex in the emotive effects of the narcogenic (psychotropic drugs) of various chemical structures; 2) to analyze the mechanisms of conjugation of unconditioned and conditioned reinforcing properties of narcogenic during emotive reactions, and finally, 3) to identify the main targets for influencing the reinforcing mechanisms of the brain in order to reduce the addictive effect of the studied opioid and non-opioid psychotropic drugs.

Materials and methods

Animals

The experiments were performed on 869 Wistar males rats weighing 200–220 g, contained in a group of 5 individuals (before the implantation of electrodes into the brain) in standard plastic boxes in vivarium conditions of the Neuropharmacology Department of the Institute of Experimental Medicine. The air temperature was maintained within 20–22 °C, relative humidity – 50–70%. The animals were kept with free access to water and food in the inverted light conditions of 8.00–20.00. All the experiments were conducted in the autumn-winter period. The study was approved by the local ethical committee of the Institute of Experimental Medicine.

Implantation of electrodes and cannulas into brain structures

Implantation of electrodes and cannulas into the brain of rats was performed under Nembutal anesthesia (50 mg/kg), using a stereotactic apparatus manufactured by Medicor, Hungary. Nickel-chromium monopolar electrodes in glass insulation (electrode diameter 0.25 mm, length of the exposed tip 0.25–0.30 mm, its thickness 0.12 mm) were implanted bilaterally into the lateral hypothalamic nucleus according to the following coordinates: AP = 2.5 mm posterior to bregma, SD = 2.0 mm lateral to sagittal suture, H = 8.4 mm below the skull surface (König and Klippel 1963). An indifferent nickel-chromium electrode was attached to the skull of an animal. All electrodes were termintaed in a micro-connector, which was fixed on the skull with self-hardening plastic.

Metal guide cannulas made of stainless steel with a diameter of 0.2 mm were implanted unipolarly into the right central nucleus of the amygdala according to the following coordinates: AP = 2.8 mm posterior to bregma, SD = 3.9 mm lateral to sagittal suture, H = 8.2 mm below the skull surface, or into the right bed nucleus of the stria terminalis according to the coordinates: AP = 0.5 mm posterior to bregma, SD = 1.5 mm lateral to the sagittal suture, H = 6.7 mm below the skull surface (Fig. 1), or into the right nucleus accumbens shell (Fig. 2): AP = 2.2 anterior to bregma, SD = 1.2 mm lateral to sagittal suture, H = 6.5 mm below the skull surface, according to the atlas by K.P.König and A.A.Klippel (1963). The cannulas were fixed on the skull of the animal with self-hardening plastic, and, after the operation, were closed with a special obturator, which was temporarily removed to introduce substances into the brain structure. With the intrastructural introduction of substances, metal micro-cannulas (diameter 100 μm), the tips of which were 0.2 mm longer than the guides, were inserted into the guides.

Behavioral experiments were not started until 10 days after surgery. At the end of all behavioral experiments, the morphological control of the localization of the tips of the electrodes was performed on a series of frontal sections of the brain, which were stained according to the Nissl method and previously coagulated through implanted electrodes with a current of 1 mA for 30 s.

Methods of self-stimulation of the brain in rats

To reproduce the brain self-stimulation in rats, the classic version of brain self-stimulation in the form of pedal self-stimulation in the Skinner's box was used. The training procedure for rats to press the pedal in the Skinner's box to produce electrical stimulation of the brain (rectangular pulses of negative polarity, 1 ms duration, 100 Hz frequency, for 0.4 s, current threshold values in the "fixed ratio" mode – FR1) started no earlier than 10 days after the implantation of electrodes into the brain.

To repeat stimulation, an animal was forced to re-press the pedal. The frequency, time of each pedal pressing and the thresholds of self-stimulation reactions were analyzed. Pharmacological agents were injected on the 3rd day of the experiment, after the stabilization of the reaction using a fixed value of current. The number of pedal pressings was recorded for 10 minutes of the experiment, and the "mismatch" coefficient was calculated; then the intrastructural micro-injection of the drug was made, and, 15–20 minutes

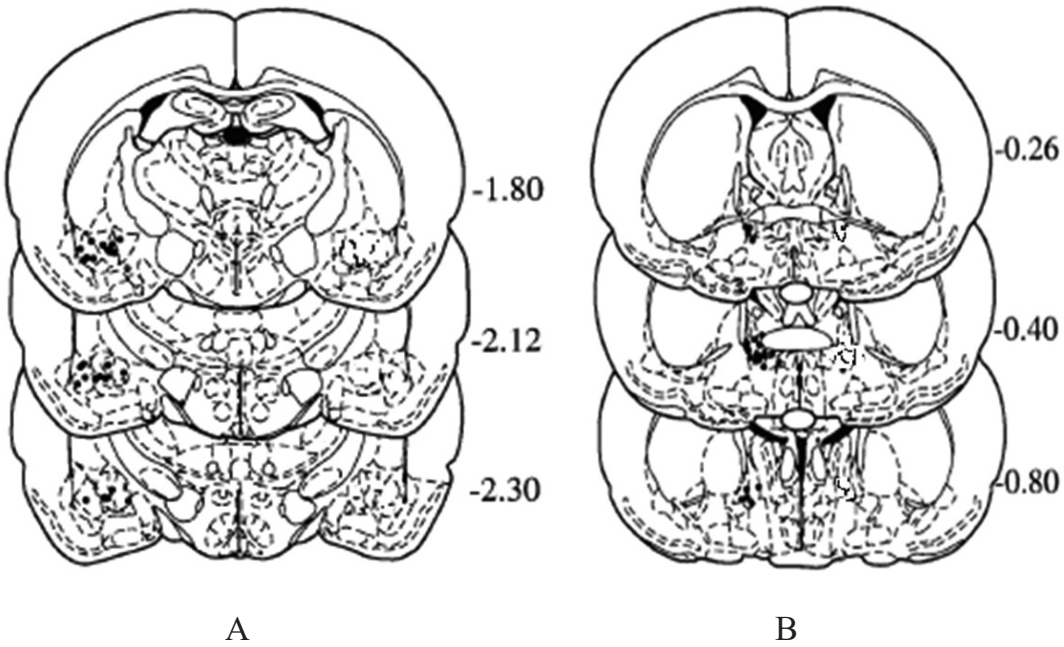

Figure 1. Morphological picture of zones of microinjection of substances into the central nucleus of the amygdala (A) and the bed nucleus of the stria terminalis (B) The coordinates are according to the atlas by K.P. König and A.A. Klippel (1963). Frontal sections are shown in mm relative to bregma.

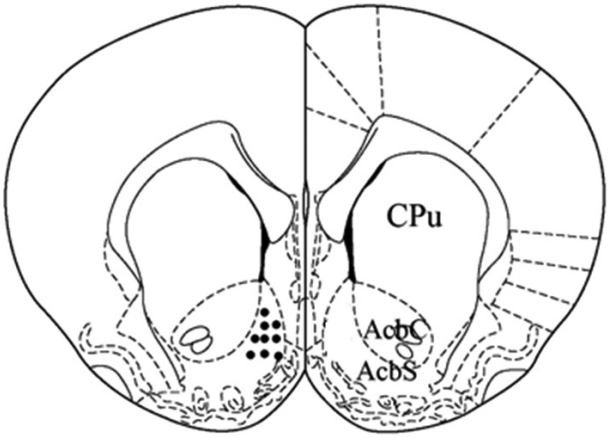

Figure 2. Projections (locations) of injections of pharmacological agents into the nucleus accumbens shell 2.2 mm anterior to bregma of the rat's skull (indicated by dark circles). **Note:** AcbS – n. accumbens shell, AcbC – n. accumbens core, CPu –n. caudatum and putamen.

later, both the number of pedal pressings and the "mismatch" coefficient were recorded over a 10-minute interval (Shabanov and Lebedev 2013). The "mismatch" coefficient ranges from -1 to +1 and shows a share of activation of the positive and negative reinforcing phase of self-stimulation.

Conditioned place preference

The conditioned place preference (CPP) was produced in a 60×30×30 cm apparatus consisting of two square chambers (compartments) of the same size, connected by a 10×10 cm door. The inner surface of one compartment was painted white and the other – dark. The texture of the floor was different: in one chamber it was a fine grate, in the other – a smooth dark brown floor. The acquisition of CPP was carried out for 8 days (Rosas et al. 2017). On the 1st day, the rat was placed for 10 minutes in the apparatus with the door open to familiarize and determine the initial preference of one of the apparatus compartments. Starting from the 2nd day of the experiment, each rat was injected with either one of the pharmacological preparations (on the 2nd, 4th and 6th days), or saline (on the 3rd, 5th and 7th days) and immediately placed for 60 minutes in the apparatus: in the non-preferable compartment in case of the administration of narcogenic and in the preferable compartment in case of the injection of saline. The door between the compartments of the apparatus in this case was closed. On the 8th day of the experiment, the door was opened, and the animal was placed for 10 minutes in the non-preferable compartment without the administration of the drug. Time spent in each of the compartments, as well as the number of shuttles between the compartments. An increase in time spent in the originally non-preferable compartment of the chamber was interpreted as a conditioned place preference (the main criterion is a 50% increase in the time spent in the non-preferable compartment of the total exposure). An additional criterion of preference was a general increase in the number of shuttles between the compartments.

Pharmacological substances used for analysis

For pharmacological analysis, a psychomotor stimulant amphetamine (β-phenylisopropylamine hydrochloride;

Sigma, USA; 1 mg/kg), synthetic opiate analgesic fentanyl citrate (Vector, Russia; 0.1 mg/kg), barbiturate pentobarbital sodium (ethaminal sodium; Vector, Russia; 5 mg/kg), opioid leu-enkephalin (H-Tyr-Gly-Gly-Phe-Leu-OH; Sigma, USA; 1 mg/kg), an opioid receptor antagonist naloxone (Narcanti; Du Pont de Nemur, Germany; 1 mg/kg) were administered intraperitoneally 30 minutes before the study of self-stimulation (after determining its background values). Bicuculline (Sigma, USA), a $GABA_A$ receptor antagonist, lidocaine (Egis, Hungary), an influx Na^+ channel blocker (local anesthetic), SCH-23390 (halobenzazepine; Sigma, USA), a D_1 dopamine receptor antagonist, sulpiride (Sanofi-Aventis France, France), a D_2 dopamine receptor antagonist, and astressin (DS Laboratories, USA), a non-selective CRF antagonist; all at a dose of 1 μg were injected intrastructurally into one of the structures of the extended amygdala (n. accumbens shell, central nucleus of amygdala, bed nucleus of the stria terminalis) through a cannula implanted into the brain structure (Shabanov and Lebedev 2013). The substances (1 μg per injection) were dissolved in distilled water and injected in a volume of 1 μl using a CMA-100 microinjector (Sweden) for 30 s 15–20 min before the testing after determining the initial (background) values of the lateral hypothalamus self-stimulation. Considering the chronic nature of the experiment (it lasted on average 30–40 days for each rat), the pharmacological agents were re-administered to each animal via cannula with an interval of at least 5 days between injections so that one operated rat received the same pharmacological substance 3–4 times. Each time before the administration of the substance, the background values of the self-stimulation reaction were determined, which were qualified as the control values for this experiment. Overall, the total number of the experiments (10–12 for each substance), rather than just the number of animals studied, was taken into account. This principle of substance injection is common for this kind of studies (Koob 2017, Shabanov and Lebedev 2013).

Results and discussion

Involvement of the nucleus accumbens shell in the reinforcement

A study of the involvement of the nucleus accumbens shell in the realization of the reinforcing effects of psycho-stimulant (amphetamine) and hypno-sedative (pentobarbital, fentanyl citrate and leu-enkephalin) narcogenic in rats in models of unconditioned (self-stimulation of the brain) and conditioned (acquisition and expression of CPP) reinforcement showed that intrastructural administration of blockers, as a rule, leads to a decrease in the reinforcing effect of narcogenic (Table 1).

An exception was the enhancement of the reinforcing properties of leu-enkephalin by lidocaine in the test of acquisition and expression of CPP, of bicuculline and SCH-23390 on the acquisition and expression of CPP by fentanyl citrate and pentobarbital and astressin on self-stimulation of the brain, stimulated by leu-enkephalin. That is, opiates (fentanyl citrate) and opioids (leu-enkephalin) responded most often by enhancing the reinforcing properties after intrastructural administration of pharmacological blockers.

In all other cases, intra-accumbental administration of the studied pharmacological agents, as a rule, led to a decrease in the reinforcing properties of the narcogenic. At the same time, in almost all combinations, the used pharmacological blockers reduced the unconditioned and conditioned reinforcing properties of a psycho-stimulant amphetamine. This, of course, is of practical interest from the point of view of the biological prevention of abusing psycho-active substances of the amphetamine type. On the other hand, the activation of the reinforcing properties of the brain by pentobarbital was often resistant to the action of dopamine receptor antagonists (sulpiride and SCH-23390), which emphasized the inexpediency of using neuroleptics to treat behavioral dependence on barbiturates.

Overall, a sufficiently high anti-narcogenic activity of the used pharmacological blockers was demonstrated when they were intrastructurally administered into the n. accumbens shell, which should be taken into account when developing issues of biological prevention of dependence on psychotropic drugs.

Involvement of the central nucleus of the amygdala in reinforcement

The studies showed that the central nucleus of the amygdala plays an important role in the realization of the unconditioned and conditioned reinforcing effects of the psycho-stimulant (amphetamine) and hypno-sedative (pentobarbital, fentanyl citrate and leu-enkephalin) narcogenic in rats. The first issue which was faced in the experiment was the neurohormonal component of the central nucleus of the amygdala. The injection of astressin into it, a non-selective antagonist of corticoliberin receptors (CRF), inhibited the self-stimulation of the lateral hypothalamus by 55%. This emphasizes the importance of the central amygdala nucleus as a stres-inducing source of reinforcement regulation.

After intra-amygdalar administration of the CRF antagonist, all the studied narcogenic, with the exception of leu-enkephalin, showed high reinforcing properties. Moreover, against the background of other receptor blockers (lidocaine, SCH-23390, sulpiride), narcogens, as a rule, activated the reaction of the lateral hypothalamus self-stimulation. Quite different patterns emerged in the study of the acquisition and expression of CPP in rats, when no psycho-activating effect of narcogenic drugs was observed, with rare exceptions (bicuculline moderately increased the expression of CPP of fentanyl citrate, and naloxone – the expression of CPP of pentobarbital). In all other cases, the effects of pharmacological blockers, as a rule, led to a decrease in the acquisition and expression of CPP of all the studied narcogenic. There was also another

Table 1. Comparison of the effects of pharmacological blockers administered into the nucleus accumbens shell on unconditioned and conditioned reinforcing properties of psycho-stimulant and hypno-sedative narcogenic in rats.

Drugs	Amphetamine	Fentanyl citrate	Pentobarbital	Leu-enkephalin
Self-stimulation of the lateral hypothalamus				
Bicuculline	-	0	--	0
Lidocaine	-	--	-	+
SCH-23390	-	--	0	0
Sulpiride	--	0	0	+
Astressin	0	--	0	-
Acquisition of CPP				
Bicuculline	--	-	-	+
Lidocaine	-	-	-	-
SCH-23390	-	-	+	--
Sulpiride	--	-	-	0
Naloxone	-	--	-	--
Expression of CPP				
Bicuculline	--	+	-	0
Lidocaine	--	-	-	++
SCH-23390	-	+	-	--
Sulpiride	--	-	0	-
Naloxone	--	--	+	-

Note: "0" – no effect; "+" – a weak activating effect; "++" – a moderate activating effect; "+++" – a strong activating effect; "-" – a weak inhibitory effect; "--" – a moderate inhibitory effect.

Table 2. Comparison of the effects of pharmacological blockers administered into the amygdala on unconditioned and conditioned reinforcing properties of psycho-stimulant and hypno-sedative narcogenic in rats.

Drugs	Amphetamine	Fentanyl citrate	Pentobarbital	Leu-enkephalin
Self-stimulation of the lateral hypothalamus				
Astressin	+++	+	++	--
Lidocaine	++	0	+	+
SCH-23390	0	+	+	+
Sulpiride	+	0	+	+
Acquisition of CPP				
Bicuculline	-	-	--	0
Lidocaine	-	--	0	-
SCH-23390	-	-	0	--
Sulpiride	--	-	0	0
Naloxone	-	--	0	--
Expression of CPP				
Bicuculline	--	+	-	0
Lidocaine	-	--	-	-
SCH-23390	--	---	--	---
Sulpiride	--	-	0	-
Naloxone	-	--	+	+

Note: "0" – no effect; "+"– a weak activating effect; "++"– a moderate activating effect; "+++" – a strong activating effect; "-" – a weak inhibitory effect; "--" – a moderate inhibitory effect; "---" – a strong inhibitory effect.

exception: the reinforcing properties of pentobarbital remained unchanged towards the acquisition of CPP after administration of most pharmacological blockers in the central nucleus of the amygdala (lidocaine, SCH-23390, sulpiride, naloxone). At the same time, antagonists of dopamine receptors (SCH-23390 and sulpiride) and naloxone showed maximum activity suppressing the reinforcing properties (Table 2).

Thus, when comparing the effects of pharmacological blockers of different receptors ($GABA_A$, D_1, D_2 of dopamine, opioids, CRF) and influx sodium ion channels acting at different points of application or targets (the n. accumbens shell and the central nucleus of the amygdala), they significantly differ. The n. accumbens shell, regarded as the target of exposure to pharmacological agents, behaves predictably, providing in most cases negative responses to unconditioned and conditioned reinforcing stimuli.

The central nucleus of the amygdala appears to be a convergence point of both reinforcing signals and non-specific stress-inducing inputs, due to which the response to reinforcement is determined not only by the expected positive effect of the narcogen, but also by the non-specific motivational component related to a stress response.

Involvement of the bed nucleus of the stria terminalis in the reinforcement

Summarizing the results, it is important to emphasize that the bed nucleus, related to the structures of the extended amygdala, is directly involed in the formation and reali-

Table 3. Comparison of the effects of pharmacological blockers administered into the bed nucleus of stria terminalis on unconditioned and conditioned reinforcing properties of psycho-stimulant and hypno-sedative narcogens in rats.

Drugs	Amphetamine	Fentanyl citrate	Pentobarbital	Leu-enkephalin
Self-stimulation of the lateral hypothalamus				
Bicuculline	-	-	+	0
Lidocaine	-	-	0	-
SCH-23390	--	--	--	---
Sulpiride	--	---	0	-
Naloxone	-	--	+	--
Acquisition of CPP				
Bicuculline	--	++	+++	++
Lidocaine	-	++	-	+++
SCH-23390	--	---	--	---
Sulpiride	--	--	0	-
Naloxone	--	---	-	--
Expression of CPP				
Bicuculline	--	++	+++	++
Lidocaine	-	++	-	+++
SCH-23390	--	---	---	---
Sulpiride	--	--	0	-
Naloxone	--	--	-	--

Note: "0" – no effect; "+"– a weak activating effect; "++"– a moderate activating effect; "+++" – a strong activating effect; "-" – a weak inhibitory effect; "--" – a moderate inhibitory effect; "---" – a strong inhibitory effect.

zation of the unconditioned and conditioned reinforcing properties of the psycho-stimulant and hypno-sedative (pentobarbital, fentanyl citrate and leu-enkephalin) narcogenic in rats (Table 3).

The pharmacological analysis made by using receptor blocking agents ($GABA_A$, D_1, D_2 dopamine, opioids) or influx sodium ions antagonist (lidocaine) showed that the reinforcing effects of amphetamine were the most labile in the models of self-stimulation of the lateral hypothalamus and conditioned place preference (acquisition and expression of CPP), which decreased under the influence of all the investigated substances. The effects of synthetic opioid fentanyl citrate may be enhanced under the influence of intra-structurally administered bicuculline or lidocaine, if using the techniques to study conditioned reinforcement (CPP, both options of acquisition and expression), and, on the contrary, may be suppressed in unconditioned reinforcement methods (self-stimulation). Similar, but more variable responses, were obtained for leu-enkephalin.

The most problematic results relate to pentobarbital, the reinforcing effects of which can both be enhanced (by bicuculline in the self-stimulation of the brain and CPP) and suppressed (primarily by SCH-23390). This indicates a lack of knowledge of D_1 dopamine receptors in the implementation of the reinforcing properties of narcogenic and emphasizes the prospect of their possible use as anti-narcotic drugs. The use of D_2 dopamine receptor antagonists (typical and atypical neuroleptics) for this purpose is not always effective.

The present work aimed to clarify the significance of the CRF, GABA, dopamine and opioids systems in the main structures of the extended amygdala (the bed nucleus of the stria terminalis, the central nucleus of the amygdala and the nucleus accumbens shell) for the reinforcing effects of a number of psycho-active substances (opiates, opioids, psycho-stimulants, barbiturates) on the self-stimulation of the lateral hypothalamus, as well as the acquisition and expression of the conditional place preference (CPP) in rats. Intrastructural administration of various blockers of receptors localized in these brain structures was selected as the main methodological research tool, with an assessment of the implementation of the self-stimulation of the lateral hypothalamus as an indicator of unconditioned reinforcement, and CPP as conditioned reinforcement (Schreiber et al. 2019, Waraczynski et al. 2015). That is, the similarities and differences in the mechanisms of unconditioned and conditioned reinforcement were compared in various behavioral models that are most common for studying the described phenomena.

It was proved by the methods of the pharmacological analysis that the unconditioned and conditioned reinforcing effects of different narcogenic (psycho-stimulants, opiates, hypno-sedatives, and opioid neuropeptides) have their own characteristics of involving the emotive and mnestic components of the general integrative reinforcement mechanism and are mediated mainly by dopaminergic mechanisms of the mesocorticolimbic system and central mechanisms of stress associated with the participation of receptors for CRF, dopamine, GABA and opioids (Shabanov and Lebedev 2013). At the same time, blockade of CRF, dopamine, GABA and opioid receptors, as well as influx ionic Na^+ currents in the neurons of the central nucleus of the amygdala, the bed nucleus and the nucleus accumbens shell, eliminates or significantly reduces the reinforcing effects of narcogenic (amphetamine, fentanyl citrate, pentobarbital and leu-enkephalin). These neurochemical structures of the extended amygdala appear to be vital for influencing the reinforcement mechanisms (Klein et al. 2017). In particular, the central nucleus of the amygdala and the bed nucleus have a controlling influence on the hypothalamus, which is predominantly of CRF-, GABA- and dopaminergic nature (Pina and Cunningham 2017). Through D_1 dopamine receptors, a direct positive (activating) effect on the lateral hypothalamus is realized. The D_2

receptor blockade of the nucleus accumbens shell prevents the realization of the reinforcing properties (self-stimulation of the brain, CPP) of psychostimulant narcogenic drugs (Shabanov and Lebedev 2013). At the same time, the blockade of D_1 receptors of the nucleus accumbens shell by SCH-23390 prevents the acquisition of unconditioned and conditioned reinforcing properties of predominantly opiates and opioids. The blockade of the nucleus accumbens shell $GABA_A$ receptors with bicuculline prevents the manifestation of the primary and secondary reinforcing properties (CPP) of the psycho-stimulant narcogens (amphetamine), without influencing the effects of opiates and opioids (fentanyl citrate and leu-enkephalin).

Thus, using the pharmacological analysis, it was demonstrated that receptors of CRF, dopamine and GABA are the most important for the correction of reinforcement activated by various narcogenic (psycho-stimulants, opiate drugs, hypno-sedatives and opioid neuropeptides); therefore, they should be considered as promising targets for influencing the reinforcing mechanisms of the brain in order to reduce the narcotic effect of the studied psychotropic drugs. The data obtained open up the prospect of searching for agents with antagonistic activity against CRF, dopamine, GABA and opioid receptors to correct various forms of drug addiction.

Conclusions

1. The neuropharmacological analysis of the central mechanisms of unconditioned and conditioned reinforcing properties of chemically diverse narcogenic drugs revealed their functional contingency in the implementation of emotional reactions, provided both by individual structures of the extended amygdala (central nucleus of the amygdala, bed nucleus of the stria terminalis, nucleus accumbens shell) and specialized neurochemical systems (CRF, dopamine, GABA), considered as targets for influencing the reinforcing brain mechanism in order to reduce the narcotic effect of the studied psychotropic drugs of opioid and non-opioid types.
2. The blockade of CRF, dopamine, and GABA receptors, as well as influx Na^+ ion currents in the neurons of the central nucleus of the amygdala, in the bed nucleus and in the nucleus accumbens shell reduces the reinforcing effects of narcogenic (amphetamine, fentanyl citrate, pentobarbital and leu-enkephalin), which indicates the importance of these structures of the extended amygdala in the regulation of the reinforcement system activated by the action of psycho-active substances (narcogenic).
3. The involvement of separate neurochemical mechanisms of the extended amygdala structures (dopamine, GABA and opioids), which realize the reinforcing effects of narcogenic, depends on their directed action on the processes of unconditioned reinforcement (self-stimulation of the brain), the acquisition and expression of conditioned reinforcement (CPP). The reinforcing effects of various narcogenic drugs (psychomotor stimulants, opiate drugs, hypno-sedatives, and neuropeptides) have their own characteristics of involving the emotive and mnestic components of a common integrative reinforcement mechanism and are mediated by central stress mechanisms involving the receptors of CRF, dopamine and GABA, and, to a lesser extent, opioid receptors.
4. The blockade of CRF (astressin) receptors, the influx Na^+ ionic currents (lidocaine) or D_1 (SCH-23390) and D_2 dopamine receptors (sulpiride) in the extended amygdala reduces the self-stimulation of the lateral hypothalamus. According to the degree of inhibition of self-stimulation, the substances can be arranged it in the following order: CRF antagonist (astressin)> lidocaine> sulpiride> SCH-23390 (substances are arranged in decreasing order of activity). A similar pattern is also observed when astressin is administered into the nucleus accumbens shell, where the following pattern is observed in the degree of inhibition of self-stimulation: bicuculline> astressin> sulpiride> SCH-23390 (the substances are also arranged in decreasing order of activity).
5. Elimination of the modulating effects of the extended amygdala on the hypothalamus blocks the reinforcing properties of opiates (fentanyl citrate) and opioids (leu-enkephalin), without affecting the psycho-activating effect of the psycho-motor stimulant amphetamine and barbiturate pentobarbital. In the case of shifting the modulating effects from the nucleus accumbens shell to the hypothalamus (introduction of lidocaine, SCH-23390, astressin), the positive reinforcing effect of opiates (fentanyl citrate) is inverted into negative. None of the blockers, with the exception of astressin, significantly change the inhibitory effects of leu-enkephalin on hypothalamus self-stimulation, and astressin in the latter case aggravates the negative effect of leu-enkephalin on the self-stimulation reaction.
6. The blockade of $GABA_A$ receptors (bicuculline), the influx Na^+ ionic currents (lidocaine) or D1 (SCH-23390) dopamine receptors in the bed nucleus of the stria terminalis reduces, whereas the blockade of D_2 dopamine receptors (sulpiride) moderately increases self-stimulation of the lateral hypothalamus. According to the degree of inhibition of self-stimulation, the substances can be arranged in the following order: lidocaine> SCH-23390 ≈ bicuculline (the substances are arranged in decreasing order of activity).
7. The bed nucleus of the stria terminalis has a controlling influence on the hypothalamus, which is predominantly of GABA- and dopaminergic nature. GABA generates a negative (inhibitory) action. Through D_1 dopamine receptors a direct positive (activating) effect on the lateral hypothalamus

is realized, whereas D_2 dopamine receptors of the bed nucleus limit the positive effects of narcogenic.

8. Functional shutdown of the nucleus accumbens shell with lidocaine prevents the acquisition of unconditioned (self-stimulation) and conditioned (place preference) reinforcing properties of narcogenic: in the case of self-stimulation – all the studied narcogenic of stimulating and hypno-sedative patterns, in the case of the acquisition of CPP – mainly opiates and opioids (fentanyl citrate, enkephalin), in the case of the expression of CPP – predominantly psycho-motor stimulants (amphetamine).
9. The blockade of the D_2 dopamine receptors of the nucleus accumbens with sulpiride prevents the manifestation of the reinforcing properties (self-stimulation, CPP) of predominantly stimulating narcogenic (amphetamine). The D_1 receptor blockade of the nucleus accumbens shell with SCH-23390 prevents the manifestation of the reinforcing properties (self-stimulation, CPP) of mainly opiates and opioids (fentanyl citrate, leu-enkephalin).
10. The blockade of the $GABA_A$ receptors of the nucleus accumbens shell with bicuculline prevents the manifestation of unconditioned reinforcing properties (self-stimulation) of all the studied stimulating and hypno-sedative narcogenic (amphetamine, fentanyl citrate, pentobarbital, leu-enkephalin); At the same time, in relation to conditioned reinforcement (CPP), the $GABA_A$ receptor blockade is not always effective: it reduces the positive reinforcing effect of amphetamine and pentobarbital, without influencing the effects of opiates and opioids (fentanyl citrate and leu-enkephalin).
11. Consequently, the priority targets for influencing the reinforcing mechanisms of the brain in order to reduce the narcotic effect of the studied psychotropic drugs are the CRF, dopamine and GABA receptors in the structures of the extended amygdala, the selective blockade of which makes it possible to eliminate or significantly reduce the reinforcing effects of opiates (fentanyl citrate), opioids (leu-enkephalin), psycho-motor stimulants (amphetamine) and barbiturates (pentobarbital).

Conflict of interest

The authors declare neither competing financial interests, nor conflict of interests.

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