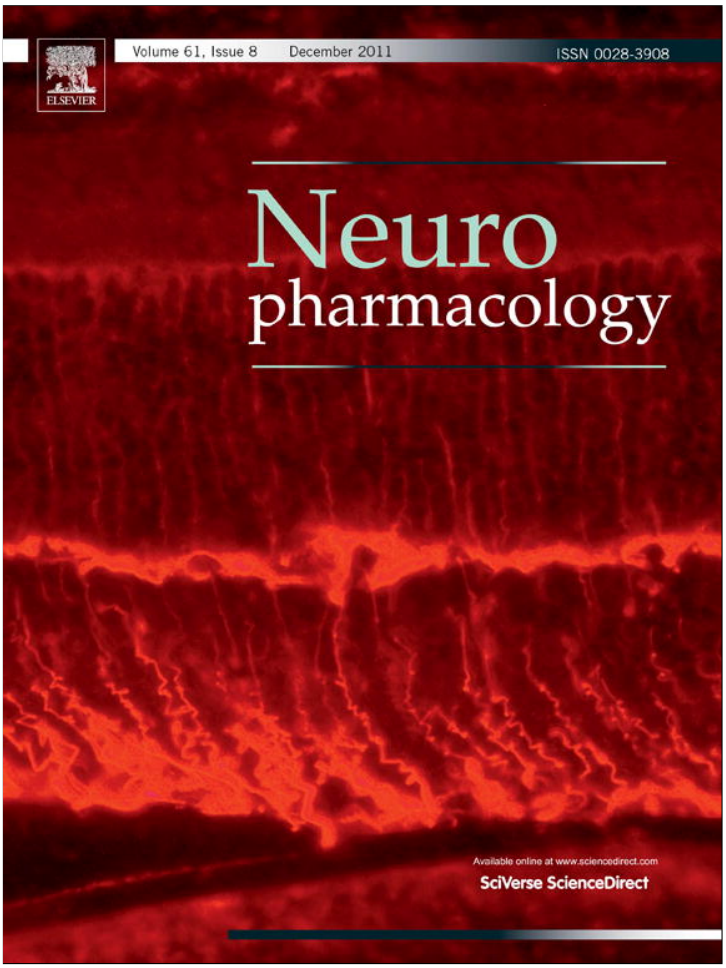

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On the role of brain 5-HT$_7$ receptor in the mechanism of hypothermia: Comparison with hypothermia mediated via 5-HT$_{1A}$ and 5-HT$_3$ receptor

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ABSTRACT

Intracerebroventricular administration of selective agonist of serotonin 5-HT$_7$ receptor LP44 (4-[2-(methylthio)phenyl]-N-(1,2,3,4-tetrahydro-1-naphthalenyl)-1-pyperasinehexanamide hydrochloride; 10.3, 20.5 or 41.0 nmol) produced considerable hypothermic response in CBA/Lac mice. LP44-induced (20.5 nmol) hypothermia was significantly attenuated by the selective 5-HT$_7$ receptor antagonist SB 269970 (16.1 fmol, i.c.v.) pretreatment. At the same time, intraperitoneal administration of LP44 in a wide range of doses 1.0, 2.0 or 10.0 mg/kg (2.0, 4.0, 20.0 μmol/kg) did not cause considerable hypothermic response. These findings indicate the implication of central, rather than peripheral 5-HT$_7$ receptors in the regulation of hypothermia. The comparison of LP44-induced (20.5 nmol) hypothermic reaction in eight inbred mouse strains (DBA/2J, CBA/Lac, C57BL/6, BALB/c, ICR, AKR/J, C3H and Asn) was performed and a significant effect of genotype was found.

In the same eight mouse strains, functional activity of 5-HT$_{1A}$ and 5-HT$_3$ receptors was studied. The comparison of hypothermic responses produced by 5-HT$_7$ receptor agonist LP44 (20.5 nmol, i.c.v.) and 5-HT$_{1A}$ receptor agonist 8-OH-DPAT 1.0 mg/kg, i.p. (3.0 μmol/kg), 5-HT$_3$ receptor agonist m-CPBG (40.0 nmol, i.c.v.) did not reveal considerable interstrain correlations between 5-HT$_7$ and 5-HT$_{1A}$ or 5-HT$_3$ receptor-induced hypothermia. The selective 5-HT$_7$ receptor antagonist SB 269970 (16.1 fmol, i.c.v.) failed to attenuate the hypothermic effect of 8-OH-DPAT 1.0 mg/kg, i.p. (3.0 μmol/kg) and m-CPBG (40.0 nmol, i.c.v.) indicating that the brain 5-HT$_7$ receptor is not involved in the hypothermic effects of 8-OH-DPAT or m-CPBG. The obtained results suggest that the central 5-HT$_7$ receptor plays an essential role in the mediation of thermoregulation independent of 5-HT$_{1A}$ and 5-HT$_3$ receptors.

1. Introduction

The involvement of brain serotonin (5-HT) system in thermoregulation is generally known. The hypothermic effect of peripheral (Lagerspetz et al., 1974; Popova and Konusova, 1985) as well as central (Lin et al., 1983; Popova and Kudryavtseva, 1985) administration of 5-HT or its precursor 5-hydroxytryptophan was shown. It was concluded that central and peripheral 5-HT systems are independently participating in the thermoregulation (Popova and Konusova, 1985).

The identification of 14 types and subtypes of receptors mediating 5-HT action has opened a new area to explore 5-HT-related neurophysiology (Barnes and Sharp, 1999). Although the role of different 5-HT receptor types in 5-HT-induced hypothermia attracted considerable attention, the problem can not be considered as solved.

Among a large variety of 5-HT receptors the implication of 5-HT$_{1A}$ receptor in thermoregulation has been the subject of much experimentation (Goodwin et al., 1987; Hjorth, 1985). Selective agonists of 5-HT$_{1A}$ receptor produced considerable hypothermic response, and this effect has been used as a test for 5-HT$_{1A}$ receptor functional activity (Hjorth, 1985; Overstreet et al., 1996; Popova et al., 2005). Previously, we reported the changes in the expression of 5-HT$_{1A}$ receptor gene linked with natural hibernation and associated hypothermia (Naumenko et al., 2008). Recently the implication of central 5-HT$_3$ receptor in thermoregulation and its interaction with 5-HT$_{1A}$ receptor was reported (Naumenko et al., 2009).

5-HT$_7$ receptors are also known to be involved in thermoregulation (Sjogren and Svenningsson, 2007). It was shown that the hypothermic effect of 5-carboxamidotryptamine – the agonist of 5-HT$_{1A}$, 5-HT$_{1B}$, 5-HT$_{1D}$, 5-HT$_5$ and 5-HT$_7$ serotonin receptors was

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attenuated by a selective antagonist of 5-HT$_7$ receptors SB 269970 (Guscott et al., 2003).

At the same time, it has been shown that there was no hypothermic response to intraperitoneally administered 5-HT in 5-HT$_7$ knockout mice (Hedlund et al., 2003). The data of Hedlund and coworkers suggested that 5-HT$_7$ receptor mediates the action of 5-HT$_{1A}$ and 5-HT$_3$ receptors in the brain serotonin system, although this fact is questionable because of considerably later phylogenetic differentiation of 5-HT$_7$ receptors in comparison with the primordial 5-HT$_1$ receptors (Peroutka and Howell, 1994). Anyway, the both Guscott and Hedlung research groups administered 5-CT and 5-HT peripherally (intraperitoneally). This fact together with the poor ability of peripheral 5-HT to cross the blood-brain barrier (Carley and Radulovacki, 1999; Yoshioka et al., 1992) and low selectiveness and peripheral effects of 5-CT (Villalon et al., 2000) indicate that the role of the central 5-HT$_7$ receptor in the mechanism of thermoregulation remains unclear.

The data on the involvement of the 5-HT$_{1A}$ and 5-HT$_7$ receptors in the 5-HT$_{1A/7}$ receptor agonist 8-OH-DPAT-induced hypothermia are also very contradictory. On the one hand it has been shown that pretreatment of the mice with a 5-HT$_{1A}$ receptor selective antagonist (WAY-100135) completely blocked the hypothermic response to 8-OH-DPAT in all doses (0.3–1.0 mg/kg; Hedlund et al., 2004). On the other hand, a selective antagonist of 5-HT$_7$ receptors SB 269970, reduced hypothermic response produced by 0.3 mg/kg (0.9 μmol/kg) of 8-OH-DPAT but not 1.0 mg/kg (3.0 μmol/kg) (Faure et al., 2006; Hedlund et al., 2004). It has been also found that at lower doses 0.3–0.6 mg/kg, i.p. (0.9–1.8 μmol/kg) 8-OH-DPAT decreased body temperature in 5-HT$_7$(+/+) mice but not in 5-HT$_7$(−/−) mice, whereas at a higher dose 1.0 mg/kg, i.p. (3.0 μmol/kg) 8-OH-DPAT-induced hypothermia in both 5-HT$_7$(−/−) and 5-HT$_7$(+/+) mice (Hedlund et al., 2004).

In fact, the 8-OH-DPAT pK$_i$ values (pK$_i$ is 8.5 at human 5-HT$_{1A}$ receptor) (Watson et al., 2000) with a minor affinity to 5-HT$_7$ receptor (pK$_i$ is 6.6 at the human 5-HT$_7$ receptor expressed in HEK 293 cells, according Tocris Bioscience) allow to consider it as a preferential agonist of 5-HT$_{1A}$ receptor. But at the same time there are a lot of studies where researchers use 8-OH-DPAT as 5-HT$_{1A}$ selective (Gronier, 2008; Inam et al., 2009; Jabeen and Haleem, 2008), 5-HT$_{1A/7}$ mixed (Gogos et al., 2010; Knoch et al., 2006; Perez-Garcia and Meneses, 2009) or 5-HT$_{1A}$ principal (Eriksson et al., 2008) agonist.

The aim of the current research was to study the effect of central and peripheral administration of a selective 5-HT$_7$ receptor agonist on the body temperature, to compare this effect with hypothermia mediated by 5-HT$_{1A}$ and 5-HT$_3$ receptors and to clarify the 5-HT$_7$ receptor role in 8-OH-DPAT-induced hypothermia in mice.

2. Materials and methods

2.1. Animals

Experiments were carried out on adult male mice of DBA/2J, CBA/Lac, AKR/J, C57BL/6, BALB/c, ICR, C3H and Asn inbred strains. The mice were housed under standard laboratory conditions in a natural light–dark cycle (16 h light and 8 h dark) with free access to water and food. Three days before the experiment the mice were weighed (about 25 g) and were isolated into individual cages to remove the group effect. All experimental procedures were in compliance with Guidelines for the Use of Animals in Neuroscience Research, 1992.

2.2. Drugs

To investigate the role of the 5-HT$_7$ receptor in the 5-HT-related hypothermia we used the highly selective 5-HT$_7$ agonist LP44 (4-[2-(Methylthio)phenyl]-N-(1,2,3,4-tetrahydro-1-naphthalenyl)-1-piperazinehexanamide hydrochloride, Sigma Aldrich, USA), which demonstrated a high affinity to 5-HT$_7$ receptor (Ki = 0.22 nM) and selectivity over 5-HT$_{1A}$ and 5-HT$_{2A}$ receptors (200- and >1000-fold, respectively, according Sigma Aldrich); the highly selective 5-HT$_7$ antagonist SB 269970 hydrochloride ((2R)-1-[(3-Hydroxyphenyl)sulfonyl]-2-[2-(4-methyl-1-piperidinyl) ethyl]pyrrolidine hydrochloride, Tocris Bioscience, USA), showed high affinity to 5-HT$_7$ receptor (pKi = 8.9) and selectivity over 5-HT$_{5A}$ and 5-HT$_{1B}$ receptors (pKi values are 7.2 and 6.0 respectively, according Tocris Bioscience).

2.3. Central and peripheral 5-HT$_7$ receptor in thermoregulation

To discriminate peripheral and central effects of the 5-HT$_7$ receptor, the agonist of 5-HT$_7$ receptor – LP44, was dissolved in sterile water and administered intraperitoneally 1.0, 2.0 or 10.0 mg/kg (2.0, 4.0, or 20.0 μmol/kg) or intracerebrally (5.1, 10.3, 20.5 or 41.0 nmol) into the left lateral ventricle, AP: −0.5, L: −1.6 mm, DV: 2 mm (Slotnick and Leonard, 1975). Before central drug administration the animals were anesthetized during 20–30 s with diethyl ether. The control groups received sterile water. The volume of the centrally administered solutions was 5 μl.

In the separate series, LP44 was administered (20.5 nmol, i.c.v.) to 5-HT$_7$ receptor antagonist SB 269970-pretreated mice. SB 269970 was dissolved in sterile water and administered intracerebrally (16.1 fmol) 15 min prior to LP44 injection. The control group received sterile water 15 min before LP44. The optimal dose of the SB 269970 (16.1 fmol) that was able to attenuate LP44-induced hypothermic response was chosen in the series of preliminary experiments.

2.4. Central 5-HT$_7$ receptor in 5-HT$_{1A}$, 5-HT$_3$ receptor and 5-HT-induced hypothermia

To investigate the role of central 5-HT$_7$ receptor in the mechanism of 5-HT-induced hypothermia, in the separate experimental series serotonin creatinine sulfate (5-HT, Roanal, Budapest, Hungary) was administered (61.7 nmol, i.c.v.) to 5-HT$_7$ receptor antagonist SB 269970-pretreated CBA mice. 5-HT was dissolved in sterile water and administered intracerbroventricularly. SB 269970 was dissolved in sterile water and administered intracerebrally (16.1 fmol) 15 min prior to 5-HT injection. The control groups received sterile water 15 min before administration of water or serotonin. The optimal dose of the 5-HT (61.7 nmol) that was able to induce a significant hypothermic response was chosen in the series of preliminary experiments.

To study the genetically defined variations in 5-HT$_7$ receptor functional activity the hypothermic reaction produced by i.c.v. administration of LP44 (20.5 nmol) was evaluated in mice of eight inbred strains (DBA/2J, CBA/Lac, AKR/J, C57BL/6, BALB/c, ICR, C3H and Asn).

To study 5-HT$_{1A}$ receptor functional activity (sensitivity), the hypothermic reaction produced by agonist of 5-HT$_{1A}$ receptor 8-OH-DPAT (8-hydroxy-2-(di-n-propylamino)tetralin, Research Biochemicals Inc. USA) was evaluated (Overstreet et al., 1996) in a separate experimental series. Since the 5-HT$_{1A}$ receptor is mainly localized centrally (Barnes and Sharp, 1999; Saudou and Hen, 1994) and 8-OH-DPAT is known to cross the blood–brain barrier, 8-OH-DPAT was dissolved in saline and administered intraperitoneally 1.0 mg/kg (3.0 μmol/kg).

To investigate the effect of 5-HT$_7$ receptor in the 5-HT$_{1A}$ receptor-induced hypothermia selective agonist of 5-HT$_{1A}$ receptor 8-OH-DPAT was administered 1.0 mg/kg, i.p. (3.0 μmol/kg) to selective 5-HT$_7$ receptor antagonist SB 269970-pretreated CBA mice. SB 269970 was dissolved in sterile water and administered intracerebrally (16.1 fmol) 15 min prior to 8-OH-DPAT injection.

To study 5-HT$_7$ and 5-HT$_3$ receptor interrelation in hypothermia regulation, selective agonist of 5-HT$_3$ receptor – 1-(3-Chlorophenyl)biguanide hydrochloride (m-CPBG; Tocris Bioscience, UK) that is known to be very selective 5-HT$_3$ receptor agonist (pKi = 8.8 for 5-HT$_3$ receptor and less than 5.0 for 5-HT$_{2A}$ and 5-HT$_{1A}$ receptors; Jorgensen, 2007) was dissolved in sterile water and administered (40.0 nmol, i.c.v.) to selective 5-HT$_7$ receptor antagonist SB 269970-pretreated CBA mice. SB 269970 was dissolved in sterile water and administered intracerebrally (16.1 fmol) 15 min prior to m-CPBG injection.

To study possible effect of 5-HT$_{1A}$ receptor on the 5-HT$_7$ receptor-induced hypothermia, LP44 (20.5 nmol) was administered i.c.v. to 5-HT$_{1A}$ receptor selective antagonist WAY-100635-pretreated CBA mice. WAY-100635 (N-[2-[4-(2-Methoxyphenyl)-1-piperazinyl]ethyl]-N-2-pyridinylcyclohexanecarboxamide maleate salt, Sigma Aldrich, USA) is known to be a highly potent and selective (pKi at human serotonin 5-HT$_{1A}$ receptors = 9.5 and only 7.4 at dopamine D(4.4) receptors; Martel et al., 2007). WAY-100635 was dissolved in saline and administered intraperitoneally at the dose of 1.0 mg/kg (1.9 μmol/kg) 10 min prior to LP44 administration. The control groups received saline 10 min before LP44 injection.

To prove the selectiveness of 5-HT$_{1A}$ agonist 8-OH-DPAT it was administered to 5-HT$_{1A}$ receptor selective antagonist WAY-100635-pretreated mice. WAY-100635 was dissolved in saline and administered intraperitoneally at the dose of 1.0 mg/kg (1.9 μmol/kg) 10 min prior to 8-OH-DPAT administration. The control groups received saline 10 min before 8-OH-DPAT injection.

2.5. Body temperature measurement

The body temperature was measured by KJT thermocouple thermometer (Hanna Instruments, Singapore) with a copper-constantan rectal probe for mice (Physitemp Instruments, USA) before and every 15 min after drug administration. The expression of hypothermic reaction was calculated as the difference (delta t °C)

between the initial body temperature and the temperature 30 min (for m-CPBG) or 20 min (for 8-OH-DPAT, LP44 and 5-HT) after drug administration.

2.6. Statistical analysis

The results were presented as m ± SEM and compared by means of repeated measures ANOVA, two-way or one-way ANOVA followed by post-hoc Fisher's test. The interstrain correlation study was performed by Pearson correlation using strain means.

3. Results

3.1. Central and peripheral 5-HT$_7$ receptor in thermoregulation

Central administration of LP44 (5.1, 10.3, 20.5 or 41.0 nmol) produced considerable dose-dependent hypothermic effect in CBA mice (Fig. 1A). Repeated measures analysis revealed effect of dose ($F_{4,30} = 20.7$; $p < 0.001$), time of the measurement ($F_{3,90} = 23.9$; $p < 0.001$) and interaction of these factors ($F_{12,90} = 5.98$; $p < 0.001$). When administered at the dose 41.0 nmol, LP44 led to dramatic and long-lasting reduction of body temperature for more then 3 °C at 15 and 30 min after drug administration. The body temperature recovered to baseline within 105 min (Fig. 1A). At dose 20.5 nmol, LP44 produced about two-fold weaker response compared with 41.0 nmol (Fig. 1A). At doses, 10.3 and 5.1 nmol LP44 led to a weak but significant hypothermic response in 15 min after drug administration compared with the control group, however the body temperature recovered to baseline within 30 min (Fig. 1A). At the same time, the hypothermic effect of LP44 administered at dose 20.5 nmol lasted significantly longer, allowing it to be detected accurately in 20 min after drug administration ($F_{1,27} = 40.7$; $p < 0.001$).

In contrast to i.c.v administration, intraperitoneal injection of the selective agonist of 5-HT$_7$ receptor LP44 in a wide range of doses 1.0, 2.0 and 10.0 mg/kg (2.0, 4.0 and 20.0 μmol/kg) failed to affect the body temperature (Fig. 1B).

3.2. Central 5-HT$_7$ receptor in 5-HT$_{1A}$, 5-HT$_3$ receptor and 5-HT-induced hypothermia

It was shown that 5-HT$_7$ receptor selective antagonist SB 269970 pretreatment (16.1 fmol, i.c.v.) significantly attenuated LP44-induced (20.5 nmol, i.c.v.) hypothermia ($F_{1,16} = 14.8$; $p = 0.014$). LP44 reduced body temperature for 3.4 ± 0.42 °C, whereas SB 269970 pretreatment diminished this temperature reduction to 1.4 ± 0.33 °C (Fig. 2A). At the same time, SB 269970 pretreatment (16.1 fmol, i.c.v.) did not produce any essential effect

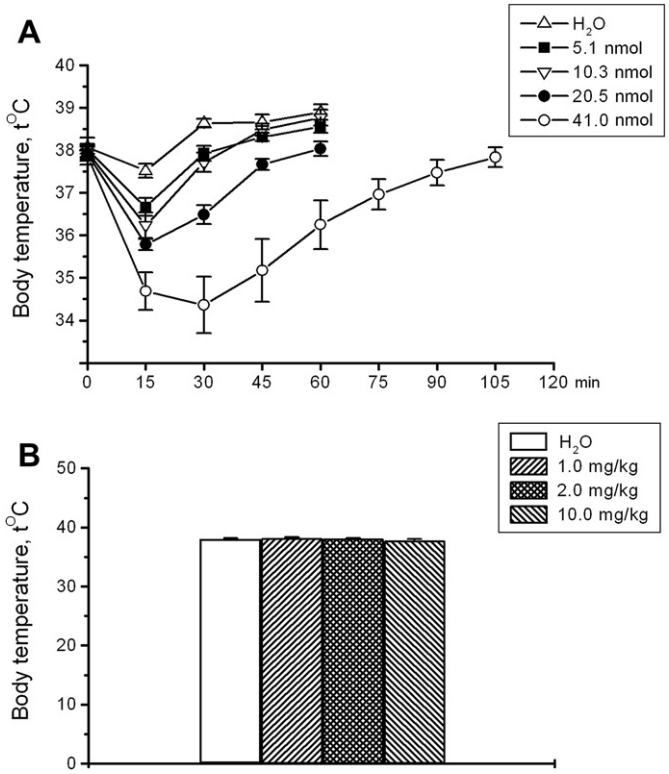

Fig. 1. Effect of intracerebroventricular administration of 5-HT$_7$ receptor agonist LP44 on the body temperature in CBA mice (A). The lack of temperature response to intraperitoneally administered LP44 in CBA mice (B). Body temperature was measured 30 min after intraperitoneal administration of LP44. The data are presented as mean ± SEM of no less than 8 mice.

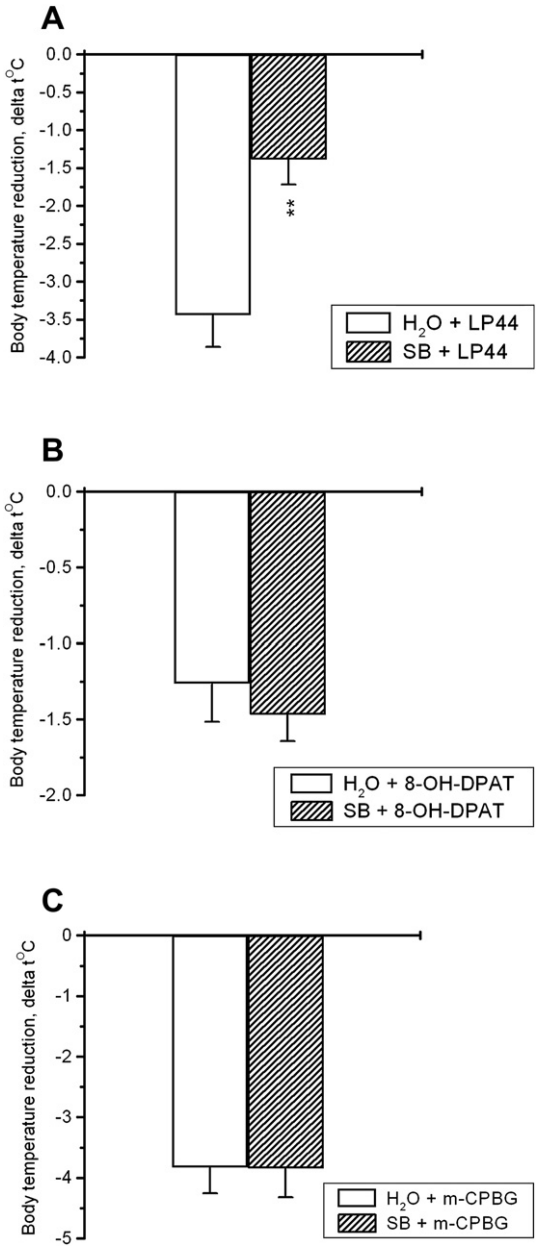

Fig. 2. The body temperature changes in pretreated with SB 269970 (i.c.v., 16.1 fmol) or water CBA mice in (A) 20 min after LP44 administration (i.c.v., 20.5 nmol); (B) 20 min after 8-OH-DPAT administration i.p., 1.0 mg/kg (3.0 μmol/kg); (C) 30 min after m-CPBG administration (i.c.v., 40.0 nmol). The data presented as mean ± SEM of no less than 8 animals. $^{**}p < 0.01$ vs. control group.

on the 8-OH-DPAT-induced 1.0 mg/kg, i.p. (3.0 μmol/kg) hypothermic response ($F_{1,16} = 0.4$; $p > 0.05$; Fig. 2B) as well as on the m-CPBG-induced (40.0 nmol, i.c.v.) hypothermic response ($F_{1,16} = 0.0003$; $p > 0.05$; Fig. 2C).

Central administration of 5-HT (61.7 nmol) produced a significant hypothermic response in CBA mice ($F_{2,20} = 5.4$, $p < 0.01$; Fig. 3). Serotonin reduced temperature for 1.6 ± 0.43 °C that considerably differ from control group. At the same time, SB 269970 pretreatment (16.1 fmol, i.c.v.) did not produce any essential effect on the 5-HT-induced (61.7 nmol, i.c.v.) hypothermic response ($p > 0.05$ compared with water plus serotonin group). 5-HT reduced the body temperature of the mice pretreated with SB 269970 for 1.5 ± 0.35 °C.

It was shown that selective 5-HT_{1A} receptor antagonist WAY-100635 pretreatment 1.0 mg/kg, i.p. (1.9 μmol/kg) abolished ($F_{1,19} = 19.9$; $p < 0.001$) the hypothermic effect of 5-HT_{1A} agonist 8-OH-DPAT 1.0 mg/kg, i.p. (3.0 μmol/kg). The body temperature reduced for 2.3 ± 0.21 °C in response to 8-OH-DPAT administration in saline-pretreated mice, whereas WAY-100635 pretreatment diminished this temperature reduction to 0.3 ± 0.26 °C (Fig. 4A). At the same time, WAY-100635 pretreatment 1.0 mg/kg, i.p. (1.9 μmol/kg) did not produce any essential effect on the LP44-induced (20.5 nmol, i.c.v.) hypothermia ($F_{1,22} = 0.03$; $p > 0.05$; Fig. 4B).

In order to study the role of genotype in hypothermic effect of the 5-HT_7 receptor agonist, LP44 was administered i.c.v. at the dose of 20.5 nmol to mice of eight inbred strains. LP44 produced significant strain-specific hypothermic response ($F_{7,87} = 5.9$, $p < 0.001$; Fig. 5A). The most significant body temperature decrease was revealed in C57BL/6 mice. The delta t °C was 3.6 ± 0.37, whereas in the rest investigated inbred strains the decrease of body temperature varied from 2.6 ± 0.29 °C in C3H mice to 1.0 ± 0.38 °C in AKR mouse strain.

The administration of the selective agonist of 5-HT_{1A} receptor 8-OH-DPAT to mice of the same inbred strains, produced strain-specific hypothermic reaction as well ($F_{7,72} = 8.98$, $p < 0.001$; Fig. 5B). The most considerable body temperature decline was revealed in Asn and BALB mice (Fig. 5B). The body temperature in Asn mice decreased for 2.6 ± 0.33 °C and for 2.1 ± 0.26 °C in BALB mice, whereas in the rest investigated strains the decrease of body temperature varied from 1.2 ± 0.18 °C in CBA mice to 0.5 ± 0.22 °C in AKR mouse strain.

I.c.v. administered m-CPBG (40.0 nmol) also produced significant strain-specific hypothermic response ($F_{7,67} = 4.3$, $p < 0.001$; Fig. 5C) in mice of investigated inbred strains. The most significant body temperature decrease was revealed in Asn (5.9 ± 0.39 °C) and CBA (5.3 ± 0.78 °C) mouse strains. The body temperature decrease in Asn and in CBA mice was more than two-fold higher than in AKR mouse strain (2.6 ± 0.55 °C).

Two-way ANOVA demonstrated significant effect of drugs (m-CPBG, 8-OH-DPAT, LP44) on the body temperature ($F_{2,225} = 128.6$, $p < 0.001$), the role of genotype ($F_{7,225} = 9.9$, $p < 0.001$) and their interaction ($F_{14,225} = 128.6$, $p < 0.001$).

The comparison of interstrain differences in 5-HT_7-, 5-HT_3- and 5-HT_{1A}-receptor-induced hypothermia using strain means did not reveal correlation for all studied mouse strains.

4. Discussion

Here we have shown for the first time that activation of central 5-HT_7 receptors with selective agonist LP44 produced considerable dose-dependent hypothermic reaction in mice. This response was significantly attenuated with 5-HT_7 receptor antagonist SB 269970 indicating 5-HT_7 receptor-mediation of LP44-induced hypothermia. At the same time, intraperitoneal administration of LP44 in a wide range of doses failed to produce distinct effect on body temperature in mice. Taken together, these data suggest: 1) the involvement of central rather than peripheral 5-HT_7 receptors in the mechanisms of thermoregulation, and 2) inability of 5-HT_7 receptor agonist LP44 to cross blood–brain barrier.

To investigate the role of genotype in the mechanism of 5-HT_7 receptor-mediated hypothermia, the LP44-induced hypothermic

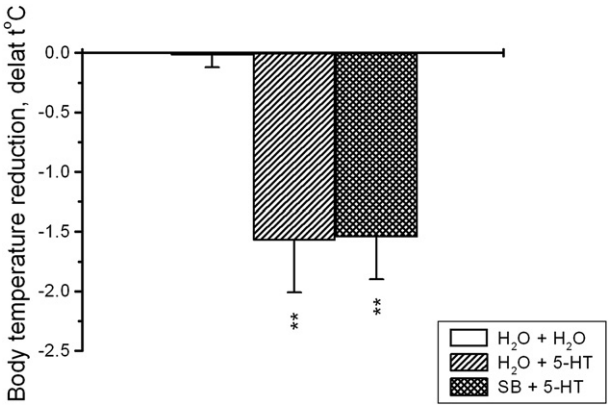

Fig. 3. The body temperature changes in pretreated with SB 269970 (i.c.v., 16.1 fmol) or water CBA mice in 20 min after 5-HT (i.c.v., 61.7 nmol). The data presented as mean ± SEM of no less than 7 animals. **$p < 0.01$ vs. control (water plus water) group.

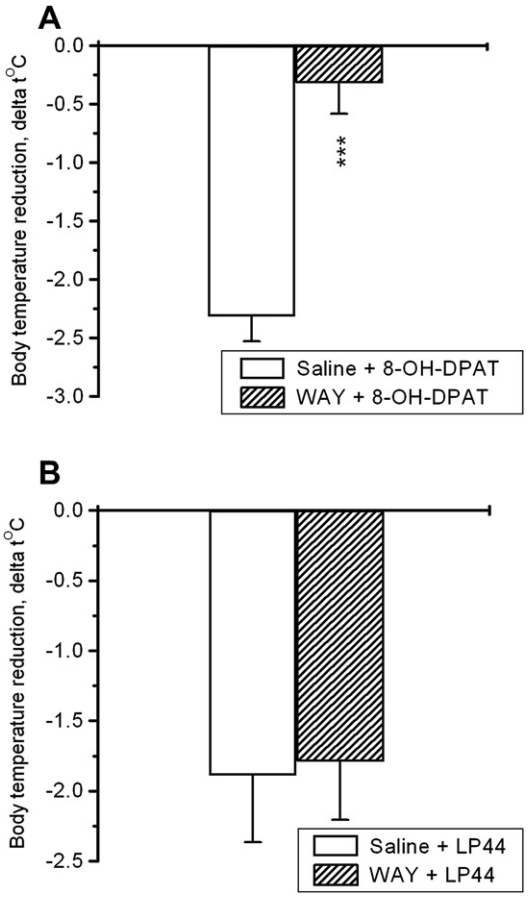

Fig. 4. The body temperature changes in pretreated with WAY-100635 i.p., 1.0 mg/kg (1.9 μmol/kg) or saline CBA mice in (A) 20 min after 8-OH-DPAT administration i.p., 1.0 mg/kg (3.0 μmol/kg); (B) 20 min after LP44 administration (i.c.v., 20.5 nmol). The data presented as mean ± SEM of no less than 9 animals. ***$p < 0.001$ vs. control group.

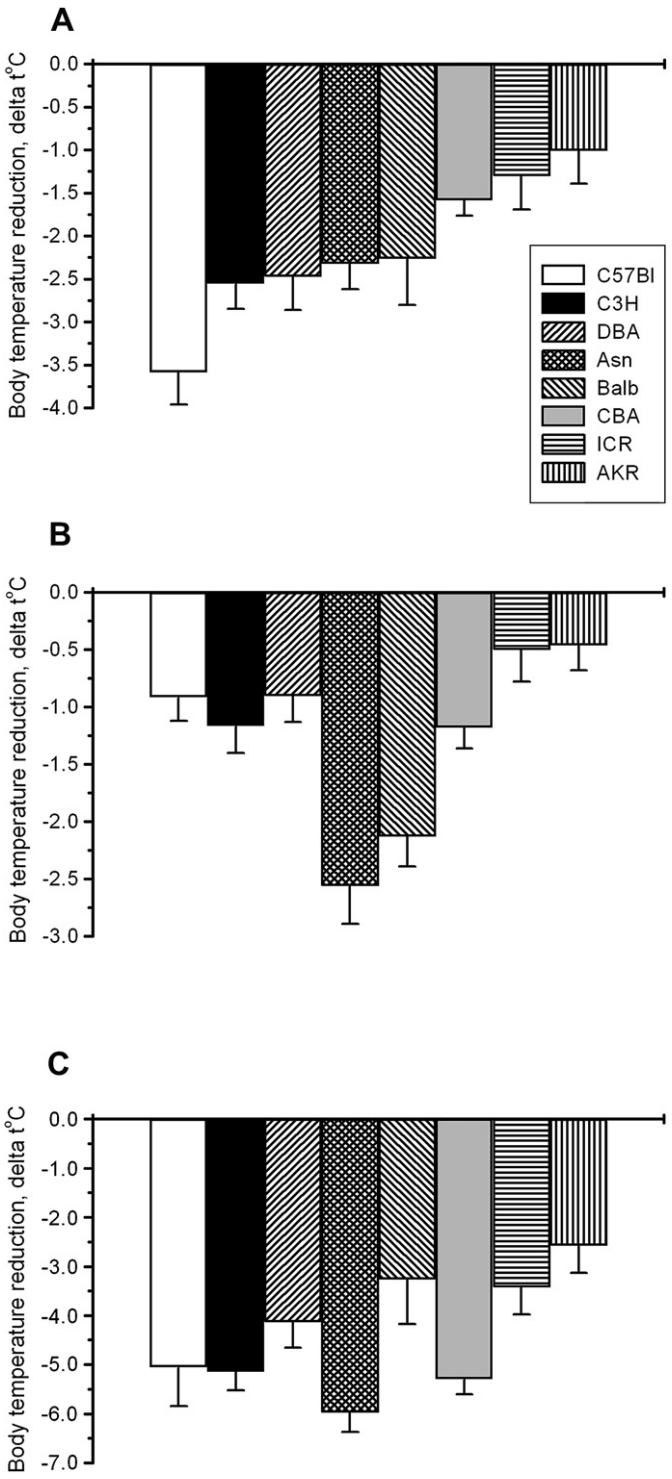

Fig. 5. The interstrain differences in hypothermic response produced by i.c.v. administration of 5-HT_7 receptor agonist LP44 (A), i.p. administration of 5-HT_{1A} receptor agonist 8-OH-DPAT (B) and i.c.v. administration of 5-HT_3 receptor agonist m-CPBG (C). The difference in the body temperature measured before and 20 min (A, B) or 30 min (C) after drug administration (delta t °C) is presented.

response in eight inbred mouse strains was studied. Significant interstrain differences in the hypothermic response to LP44 administration in eight investigated inbred mouse strains were shown. The most significant body temperature decrease was revealed in C57BL/6 mice, suggesting the highest functional activity of 5-HT_7 receptor in this strain.

The comparison of interstrain differences in 5-HT_7 and 5-HT_{1A} or 5-HT_3 receptor-induced hypothermia using strain means did not reveal any correlation for all studied mouse strains indicating the lack of interaction between 5-HT_7 and 5-HT_{1A} or 5-HT_3 receptors in the control of the body temperature.

The 5-HT_7 receptor selective antagonist SB 269970 attenuated the effect of 5-HT_7 agonist, but produced no effect on the 5-HT-induced (61.7 nmol, i.c.v.) hypothermia. These results are not in agreement with the data on absence of hypothermic response to 5-HT in 5-HT_7 knockout mice (Hedlund et al., 2003) that is likely attributed to the peripheral administration of 5-HT in this study and compensatory reorganization of 5-HT receptor system in 5-HT_7 knockout mice.

At the same time, blockade of 5-HT_7 receptors with the selective antagonist SB 269970 produced no effect on the 8-OH-DPAT-induced 1.0 mg/kg, i.p. (3.0 μmol/kg) hypothermia, that coincided with the data of Hedlungs' research group (Hedlund et al., 2004). These results taken together with the data on 8-OH-DPAT pK_i values (Watson et al., 2000) indicate that 8-OH-DPAT specifically binds with 5-HT_{1A} receptors.

Blockade of 5-HT_{1A} receptor with the selective antagonist WAY-100635 1.0 mg/kg, i.p. (1.9 μmol/kg) completely prevented 8-OH-DPAT-induced 1.0 mg/kg, i.p. (3.0 μmol/kg) hypothermic response confirming the notion that 8-OH-DPAT specifically binds with 5-HT_{1A} receptors.

It is important that the same dose of WAY-100635 did not attenuate 5-HT_7 receptor-induced hypothermia that together with the data on the lack of any correlation between 5-HT_7- and 5-HT_{1A} receptor-induced hypothermia allow us to reject the implication of 5-HT_{1A} receptor in the 5-HT_7 receptor-induced hypothermia. Blockade of 5-HT_7 receptor with SB 269970 produced no effect on the 5-HT_3 receptor-induced hypothermic response either confirming the absence of 5-HT_7 receptor role in the 5-HT_3 receptor-induced hypothermia.

In summary, we demonstrated that intracerebroventricular but not intraperitoneal administration of selective 5-HT_7 receptor agonist LP44 produced a considerable and dose-dependent hypothermic response in mice indicating the involvement of central rather then peripheral 5-HT_7 receptors in thermoregulation. Considerable difference in genetically defined functional activity of 5-HT_7 receptor was revealed. The lack of any interstrain correlation between 5-HT_7 and 5-HT_{1A}- or 5-HT_3-receptor-induced hypothermia and the lack of 5-HT_7 receptor selective antagonist effect on the 8-OH-DPAT-, m-CPBG-induced hypothermia indicate that: 1) 8-OH-DPAT specifically binds with 5-HT_{1A} receptor; 2) central 5-HT_7 receptor plays essential role in the mechanism of thermoregulation independent of 5-HT_{1A} and 5-HT_3 receptors.

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