· Advances in Medical Sciences · Vol. 54(2) · 2009 · pp 269-276 · DOI: 10.2478/v10039-009-0037-0

Effect of the class I metabotropic glutamate receptor antagonist AIDA on certain behaviours in rats with experimental chronic hyperammonemia

Grzęda E, Wiśniewska RJ*

Department of Pharmacology, Medical University of Białystok, Białystok, Poland

* CORRESPONDING AUTHOR:
Department of Pharmacology,
Medical University of Bialystok,
Mickiewicza 2c,
15-222 Białystok, Poland
Tel. +48 085 7485 570; Fax: +48 085 7485 575;
e-mail: zfarm@amb.edu.pl (Róża J. Wiśniewska)

Received 30.06.2009
Accepted 08.09.2009
Advances in Medical Sciences
Vol. 54(2) · 2009 · pp 269-276
DOI: 10.2478/v10039-009-0037-0

ABSTRACT

Purpose: This study examines possible interactions between behavioral effects and mGluR1(class I metabotropic glutamate receptor) by injecting AIDA [(RS)-1-aminoindan-1,5-dicarboxylic acid] in rats with experimental chronic hyperammonemia (chHA).

Material/Methods: The effects of mGluR1 antagonist on some behaviors were tested in control groups of rats and in rats with chHA. Experimental chHA was induced by intraperitoneal injection of ammonium acetate (12 mmol/kg) for five consecutive days. We used the following behavioural tests: the open field test, the passive avoidance test and the elevated "plus" maze.

Results: In control rats AIDA administered intracerebroventricularly (i.c.v.) at the dose 100 nmol decreased the number of crossings and bar approaches in the open field test and impaired acquisition and recall in the passive avoidance situation. ChHA significantly inhibited locomotor and exploratory activity and profoundly impaired acquisition and recall processes in the passive avoidance test and significantly increased acute stress responses. AIDA increased locomotor activity in chHA rats (especially number of crossed fields and rearings) and produced anxiety enhancement in rats with chHA. AIDA used in rats with chHA significantly improved acquisition and retrieval processes.

Conclusions: The obtained results suggest that AIDA, the antagonist of mGluR1, had beneficial effects on learning and memory in rats with experimental chronic hyperammonemia.

Key words: behaviour, experimental chronic hyperammonemia, AIDA, rat

INTRODUCTION

Ammonia is produced by proteins metabolism and degradation. High ammonia levels cause functional disturbances of the central nervous system [1]. To avoid these toxic effects, ammonia is usually detoxified in the liver by incorporation into urea. In the central nervous system (CNS) the urea cycle does not take place, thus the only manner to detoxify ammonia is its incorporation in glutamine by the astrocytic enzyme glutamine synthetase [2].

In chronic liver disease (e.g. cirrhosis, hepatitis) ammonia detoxification is impaired, thus leading to high concentrations of ammonia in the blood, which in turn causes an increased accumulation of ammonia and glutamine in the brain [2]. It is well known that ammonia at elevated concentrations is toxic to the brain [3]. Ammonia can exert diverse effects on nervous tissue, including membrane depolarization [4], alternation of intracellular pH, increased conversion of glutamate to glutamine and changes of respiratory enzymes [5]. Impairment of long-term potentiation (LTP) in hyperammonemia may be involved in the impairment of the cognitive function in patients with hepatic encephalopathy [6]. LTP is an activity-dependent form of increased transmission efficacy at synapses and is considered to by involved in some forms of learning and memory [7]. Hyperammonemia is considered the main contributing factor to the neurological alternations found in hepatic encephalopathy [3]. The signs of hepatic encephalopathy in patients with chronic liver disease range from alterations in the sleep-waking cycle and motor coordination to changes in personality and gradually developing intellectual impairment [6, 8].

Animal models of chronic hyperammonemia can make an important contribution to the understanding of the

pathophysiology of the effects of hyperammonemia on the brain. As in humans with chronic hyperammonemia, a variety of neurophysiologic parameters are impaired in rats and mice [2,5, 9].

The molecular mechanism by which hyperammonemia leads to impaired cerebral function have not been clarified. There are therefore a large number of possible sites of molecular targets for interference by high ammonia levels of glutamatergic neurotransmission [3]. Alterations at the level of glutamate receptors have been described in hyperammonemic rats [2]. Glutamate receptors, which are the major excitatory receptors within the central nervous system, are objects of particular attention since their regulation appears to be crucial for controlling synaptic operation during learning and memory [10]. It is known that excitatory transmission in the brain is largely mediated by glutamate acting through different classes of receptors: ionotropic (AMPA-α-amino-3-hydroxyl-5-methyl-4-isoxazole-propionate, NMDA- N-methyl-D-aspartate acid) and metabotropic (mGluRs) [11].

Although the effects of hyperammonemia and liver failure on ionotropic glutamate receptors have been studied by different groups, only very few studies have addressed the effects on mGluRs. Eight metabotropic glutamate receptors have been identified. mGluRs are classified into three groups (I-III) on the basis of the similarities in coupling mechanisms and the pharmacology of the receptors. mGluRs are also implicated in long term potentiation and in memory formation. [12]. There is a specific role of mGluR1 in certain types of synaptic plasticity and learning [13]. Group I receptors (mGluR1 and mGluR5) are positively linked to phospholipase C and therefore direct activation of mGluR 1/5 results in increased phosphoinositide turnover [2,14].

AIDA is a selective mGluR1 antagonist, and possesses a potential role in the hippocampus-dependent forms of learning [13].

The purpose of our study was to investigate an influence of AIDA on behaviour in rats with chronic hyperammonemia.

MATERIALS AND METHODS

Animals

The study was conducted on male Wistar rats (Central of Experimental Medicine, Medical University of Bialystok) weighing 160–180 g. They were housed in cages (55 × 40 × 20cm), six animals per cage, in an air-conditioned (humidity 50–60%) and temperature-controlled (22°C) room under 12 h light/12 h dark cycle beginning at 7 a.m. The animals were fed a standard diet, food and water were freely available. The experiments were carried out between 8 a.m. and 12 p.m. Each animal was used only once and the same rat was not used in a different test. We have used 174 rats for these experiments (6-8 weeks old).

The experimental procedures applied in this study were in compliance with the Board for Ethical Affairs and Supervisions over Research on Animals and Individuals, Medical Academy of Bialystok (No 2006/36). Every effort was made to minimize the number of animals used and their suffering. All experiments were in accordance with the EU Directive 86/609/EEC and international guidelines on the ethical use of animals.

Drugs

(RS)-1-Aminoindan-1,5-dicarboxylic acid (AIDA, Tocris Cookson, UK) was dissolved in 0,9% NaCl (pH 7,4) and administered only once, 15 min. before experiments into the lateral ventricle of the brain (i.c.v.) through the implanted cannulas at a dose of 100 nmol per single rat in the volume of 5 µl [15].

Control rats received saline (0.9 % NaCl) i.c.v. in the volume of 5µl.

Surgery

A week before the experiments the rats were anesthetized with Vetbutal (pentobarbitalum, Biowet Puławy) at a dose of 50 mg/kg *ip*. and the polyethylene cannulas were implanted into the lateral brain ventricle (i.c.v.) [16] at the following coordinates: depth: 4 mm from the surface of the skull, 2.5 mm to the right from the sagittal suture and 1 mm behind the coronary suture. Cannulas were fixed to the skull bones with acrylic Deltamed PM 16 glue (Chemical Factory, Oświęcim, Poland). The injections were made using a Hamilton microliter syringe with a 0.3 mm external diameter. After implantation, rats were housed individually. After termination of each experiment, all animals were sacrificed, their brains were removed, and the sites of implantation of cannulas and injection were verified macroscopically after brain sectioning. Animals with inappropriate injection sites were not used for analysis.

Hyperammonemia induced by ammonium acetate

Experimental chHA was induced by intraperitoneal injection of ammonium acetate (POCH Gliwice) (12 mmol/kg) for five consecutive days [17]. All experiments have been started 24 hours after last application of ammonium acetate. The blood ammonia level was measured by Blood Ammonia test. In rats treated with ammonium acetate, a marked increase in blood ammonia was observed. Mean increases were 612,7% (one day after last injection of ammonium acetate), 556,7 % (three days after last application), and 469,3 % (fifteen days after last application) compared to control rats. Blood ammonia levels (µmol/l) in rats treated with ammonium acetate were 459±45,9 –s.e.m. (n = 8), 417±43,7–s.e.m. (n = 6) and 352±26,9–s.e.m. (n = 7) respectively while in control rats it was 75± 6,5 (n = 8).

Behavioural testing

All experiments were carried out in a quiet, dimly lit room between 8 a.m. and 12 a.m. with each group equally represented at the times of testing. Each group comprised 8–10 rats. Rats were randomly allocated to treatment groups and used only once. Passive avoidance responses were selected to estimate acquisition and recall memory. Moreover, the putative influence of the treatment on motor and exploratory activity was tested in open field, respectively. After the experiments, rats were anesthetized with Vetbutal at the dose of 20 mg/kg per rat (i.p.) and then they were killed by decapitation.

Passive avoidance

The response was induced using the one-trial learning method of Ader et al. (1972) [18]. The apparatus consisted of a 6 x 25 cm platform illuminated with a 25 W electric bulb connected through a 6 x 6 cm opening with a dark compartment (40 x 40 x 40 cm). The floor of the cage was made of metal rods 3 mm in diameter, spaced by 1 cm. The investigation took advantage of the natural preference of rats to stay in dark compartments. The test lasted 3 days. On the first day, after 2 min of habituation in the dark compartment, rats were immediately removed. Two similar trials, at an interval of 2 min, were carried out on the second day. After the first trial, rats were allowed to stay in the dark compartment for 10–15 s. In the second trial, when a rat entered the dark compartment, it received a foot shock (0.25 mA, 3 s) delivered through the metal rods. The presence of the passive avoidance was checked 24 h later. Rats were placed on the illuminated platform once more and latency to enter the dark compartment was measured, with the cut- off time at 300 s. To determine a possible effect of drug treatment on retrieval, according to the protocol proposed by Matthies [19], AIDA was administrated on the third day 15 min before the retention test. To determine a possible AIDA effect on acquisition, the drug at the dose of 15 mg/kg per rat was given immediately before induction of passive avoidance.

Locomotor and exploratory activity

The open field test was used to estimate locomotor activity in all groups of rats. The apparatus consisted of a square with 100 x l00 cm white floor, which was divided by 8 lines into 25 equal squares, and surrounded by white walls, 47 cm high. Four plastic bars, 20 cm high, were located at four different line crossings in the central area of the floor. A single rat was placed inside the apparatus for 1 min adaptation. Subsequently, crossings, rearings and bar approaches were counted for 5 min. AIDA (100 nmol) was given 15 min before the test.

Elevated "plus" maze

The maze (constructed of grey coloured wooden planks) consisted of two open arms, 50 cm (length) × 10 cm (width), and two closed arms, 50 cm (length) × 10 cm (width) × 40 cm (height), covered with a removable lid, such that the open or closed arms were opposite to each other. The maze was elevated to a height of 50 cm from the floor. Ten minutes after the injection, a naive rat was placed for 5 min in a pretest arena (60 × 60 × 35 cm, constructed from the same material) prior to exposure to the maze. This step allows the facilitation of exploratory behaviour. The experimental procedure was similar to that described by Pellow et al. (1985) [20]. Immediately after the pretest exposure, rats were placed in the centre of the elevated "plus" maze facing one of the open arms. During the 5 min test period the following measurements were taken: the number of entries into the open and closed arms and the time spent in the open and closed arms. An entry was defined as entering with all four feet into one arm. An increase in open arm entries and increase in time spent in the open arms is indicative of potential anxiolytic activity, as rats naturally prefer the closed arms. AIDA was given 15 min before test.

Statistical analysis

The statistical significance of the results was computed by two-way analysis of variance (ANOVA II) followed by Newman-Keuls test, except for passive avoidance behaviour which was assessed with Mann-Whitney ranking test. F-ratios, degrees of freedom and p-values are reported only for significant differences. For all comparisons, differences between particular groups with P equal to or lower than 0.05 were considered as significant. Statistical analyses were carried out using Statistica 6 software.

RESULTS

The effect of AIDA on acquisition and recall of passive avoidance in control and hyperammonemic rats (Tab. 1).

The latency was shortened in hyperammonemic rats vs. control group of rats in acquisition and recall of a passive avoidance situation. Experimental chHA decreased acquisition of a passive avoidance responses (at about 30 sec.). AIDA significantly shortened re-entry time in control rats but prolonged time to entrance in the dark compartment in hyperammonemic rats.
Chronic hyperammonemia significantly decreased recall in a passive avoidance test (by about 25 sec). The AIDA attenuated retrieval deficits induced by chHA, but significantly shortened re-entry time in control rats.

The influence of AIDA on locomotor and exploratory activity of control and hyperammonemic rats in the open field test (Fig. 1).

We observed that AIDA administered i.c.v. at a dose of 100 nmol/rat decreased the number of crossed fields and bar approaches, but it had no effect on the number of the rearings in control rats.

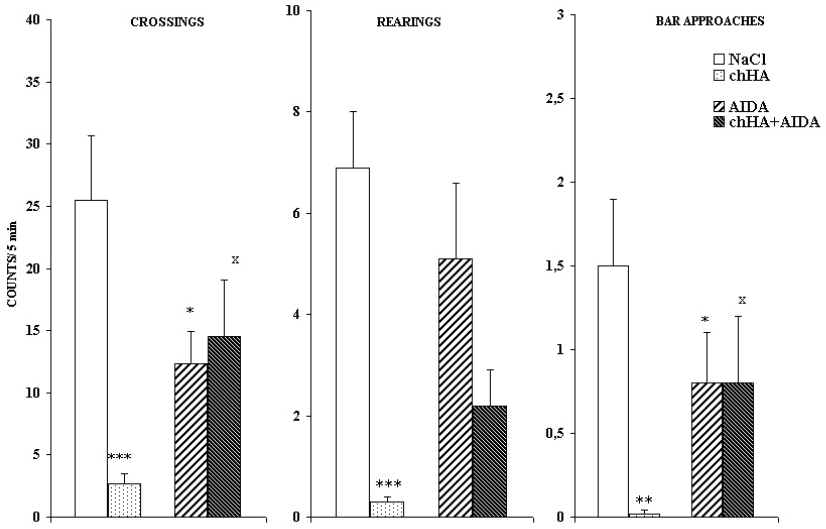
Experimental-induced chronic hyperammonemia significantly decreased locomotor and exploratory activity (number of crossed fields, rearings and bar approaches) while

Table 1. **Influence of (RS)-1-Aminoindan-1,5-dicarboxylic acid (AIDA) on the acquisition and recall of passive avoidance responses.**

Groups	Acquisition (sec) M ± SEM (Q1; Q2)	N	Recall (sec) M ± SEM (Q1; Q2)	N
NaCl	46,5 ± 5,95 (38,0; 64)	12	45,5 ± 5,33 (28,0; 64,0)	10
AIDA	17,5 ± 4,37(9,5; 32,5)**	12	22,5 ± 2,92 (17,5; 35,0)**	10
chHA	15,5 ± 3,3 (8,0; 20,0)*	12	19,0 ± 3,07 (15,0; 29,0)**	11
chHA+AIDA	186,5 ± 70,8 (50,0; 200,0)xx, ##	13	53,0 ± 7,83 (41,0; 60,0)xx, #	10

Rats received AIDA at the dose of 100 nmol / kg i.c.v. Control group received 0,9 % NaCl. The retention latencies were expressed as the median (M) and interquartile range (Q1, Q3). The data were analyzed by two-tailed Mann-Whitney U-test.
*p<0.05, **p<0,01, vs NaCl; xx p<0,01, vs chHA; # p<0,05, ## p<0.01 vs AIDA.

Figure 1. **The effect of AIDA on the number of crossings, rearings and bar approaches in the open field test in control and hyperammonemic rats.**



Columns represent means ± SEM of the values obtained from 9-12 rats. Control group received 5 µl 0.9 % NaCl icv.
*p < 0.05, **p<0.01,***p < 0.001 vsNaCl; x p<0,05 vs HA (ANOVA, Newman-Keuls tests).

AIDA increased this activity in chHA rats (especially number of crossed fields and bar approaches).

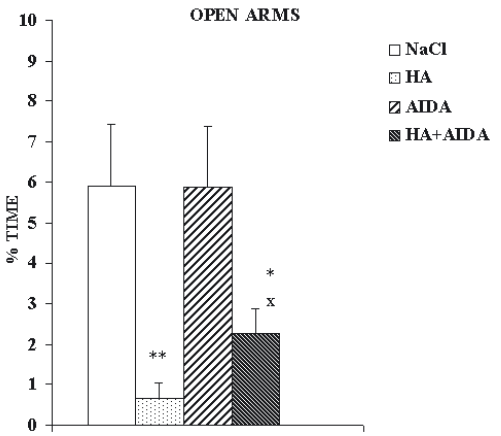
The analysis of the number of crossed fields revealed significant treatments x chHA interaction (F(3,40)=5,84; p<0,05). In the number of rearings we observed significant interaction between treatment x chHA (F(3,40)=5,09; p<0,05).

The effect of AIDA on the activity of control and HA rats in the elevated "plus" maze (Fig. 2).

ChHA decreased the time spent in the open arms (%). AIDA prolonged the time spent in the open arms (%) in experimental hyperammonemic rats.

The analysis of the correct choices in "plus" maze revealed a significant main effect of treatments x hyperammonemia interaction (F(3,36)= 5,78; p<0,05).

Figure 2. **The effect of HA+AIDA in rats on the % time spend in open arms in elevated plus-maze test.**



Columns represent means ± SEM of the values obtained from 10-12 rats.
Control group received 5 µl 0.9 % NaCl icv.
*p<0.05, **p<0.01 vs NaCl; x p<0.05 vs HA (ANOVA, Newman-Keuls test).

DISCUSSION

In our experiments, we observed that AIDA, a selective mGluR1 antagonist administered i.c.v. at the dose of 100 nmol/rat significantly impaired acquisition and recall of memory processes in passive avoidance situation in control rats. Experimental chronic hyperammonemia (chHA) impaired memory in rats in passive avoidance test. AIDA attenuated acquisition and recall deficits induced by chHA in this experiment.

Like other metabolic disorders, hyperammonemia is associated with dysfunction on multiple neurotransmitters systems in the brain [21]. The impairment of the glutamate-NO-cGMP pathway in chronic hyperammonemia would contribute to the neurotransmission alteration and the cerebral function impairment . The inhibition of the NO-induced formation of cGMP in hyperammonemic rats could also result in impaired long-term potentation, which in turn could impair memory and learning [22].

It is known that chronic HA impairs glutamatergic neurotransmission in several brain areas [23]. Although the effects of hyperammonemia and liver failure on ionotropic glutamate receptors have been studied by different groups, only very few studies have addressed the effects on mGluRs [2].

Lombardini et al. [24] reported that ammonia potentiated the effects of mGluRs coupled to G proteins that inhibit adenylate cyclase, indicating that high ammonia concentrations differentially affect the effects induced by activation of different types of mGluRs in cerebral cortex.

Saez et al. [25] showed that hyperammonemia alters the function of signal transduction pathways associated with mGluRs that modulate phosphorylation of the microtubule-associated protein MAP-2.

These in vitro studies suggest that the processes modulated by mGluRs may be altered in brain of hyperammonemic rats in vivo. Group I metabotropic glutamate receptors, are widely expressed throughout the brain including nucleus accumbens neurons [2]. The nucleus accumbens, located in the ventral part of the striatum plays a central role in motor and cognitive processes [26].

AIDA in our experiments impaired acquisition and recall in control rats, but in groups of rats with chHA significantly facilitated memory in passive avoidance test.

Some studies have indicated that antagonism of classes I and II mGluRs inhibit memory. For example (R,S)-α-methyl-4-carboxyphenylglycine (MCPG) has been found to inhibit shock-reinforced spatial alternation learning, and to have an amnestic effect on step-down passive avoidance learning [27]. Nielsen et al. [28] have suggested that learning and memory deficits may be due to selective blockade of class I mGluRs in the hippocampal formation.

Christoffersen et al. [15] indicate that AIDA may exert a dual effect on learning: facilitation of short-term and inhibition of long-term memory in spatial task in rats.

mGluRs are implicated in learning and memory formation [27]. Group I mGluRs include mGluR1 and mGluR5, which are positively coupled to phosphoinositide hydrolysis/Ca^{2+} mobilization [29]. AIDA is a selective mGluR1 antagonist [29]. It has been suggested that targeting mGluRs sub-types may offer therapeutic potential in many central nervous system disorders [30]. Antagonist of group mGluR1 have most often been associated with neuroprotection, agonist have been found to either amplify of attenuate neurolnal cell death [29]. AIDA reduced traumatic neuronal injury in vivo and in vitro [31]. The blockade of class I mGluRs may enhance short term memory under certain learning conditions in control rats [15].

Some research has shown the existence of an interaction between group I mGluRs and other systems. Bordi et al. [32] found an indirect role of mGluRs in synaptic plasticity via regulation of GABA inhibition. mGluR1 and mGluR5 can be physically and functionally coupled to NMDA receptor. In the CA3 region of the hippocampus stimulation of both mGluR1 and mGluR5 enhances NMDA receptor function [29]. Has been suggested that interaction with NMDA receptor may be one of several mechanisms whereby inhibition of mGluR1 and mGluR5 receptors attenuate neurodegeneration in ischaemia condition [29].

On the other hand, many studies have indicated that NMDA receptors have been implicated in the appearance of LTP, and this receptor type seems to be very important for learning and memory [27, 33]. AIDA can disturb learning and memory because this mGluR1 antagonist leads to hypoactivation of NMDA receptor [29].

In chHA situations AIDA improves memory in rats in passive avoidance situations. Several reports indicate that the hyperammonemia showed increased concentrations of the excitatory Glu in the brain [1]. Moreover hyperammonemic rats show increased content of mGluR1 α in the nucleus accumbens, and this receptor mediates the altered responses. Changes in extracellular concentration of neurotransmitters induced by activation of group I mGluRs in the nucleus accumbens are strongly altered in chHA, resulting in enhanced glutamate and reduced dopamine concentrations in the extracellular fluid, when compared with control rats [1]. Canales et al reported [1] that major alterations induced by chHA in both neurotransmission and motor function are modulated by group I mGluRs in the nucleus accumbens. These alterations may contribute significantly to some of the cognitive dysfunctions characteristic of chHA in rats.

Considering that all cognitive effects had to be expressed by psychomotor activity, the influence of treatments on locomotor and exploratory behavior was checked in open field test. The changes in locomotor activity induced by AIDA may affect the data on passive avoidance test in our study. The number of entries, crossing and bar approaches has significantly decreased in group of rats with chHA, while AIDA increased locomotor activity in those rodents in the open field test (in two parameters: crossing and rearings). On the other hands we observed that AIDA decreased locomotor

activity in control rats. Some literature described that hepatic encephalopathy exhibited motor and coordination deficits in humans [34] and rats [2, 5]. Decreased locomotor activity also found in chHA rats was thought to be related to alterations affecting the neuronal circuits between basal ganglia end prefrontal cortex [35]. The basal ganglia produces signals that go to the thalamus which sends signals to the cortex to modulate movement execution [2]. The signals originated in the thalamus are modulated by substantia nigra pars reticulate (SNr), which sends inhibitory projections to the ventro-medial thalamus (VMT) [36]. The locomotor activity mediated by the basal ganglia-thalamus-cortex circuit is modulated by glutamate and GABA systems in SNr [37].

The motor activity mediated by the basal ganglia and thalamus-cortex circuit is modulated by glutamate and GABA in SNr. Activating glutamate receptors or blocking GABA receptors includes hypolocomotion in normal rats [36]. Glutamate receptor antagonists or GABA receptor agonists administered in SNr induce hyperlocomotion. Alternations in glutaminergic and/or GABA-ergic neurotransmission in SNr may therefore contribute to the psychomotor slowing in HA rats [36].

Car et al. [38] reported that AIDA administered icv at the dose of 100 nmol reduced crossing and rearings in the open field test in control rats. In our experiments we observed the same significant inhibition of rats' mobility.

According to some data, mGluR1 knock-out mice exhibited motor deficits and impaired motor function [39]. Also Moroni et al., have noticed that AIDA as the most potent and selective mGluR1 antagonist causes some difficulty in the initiation of movement [40].

mGluRs are involved in the generation of dopamine-dependent locomotor behaviours [39]. Caneles et al. [1] presented that chronic hyperammonemia reduces the increase in extracellular dopamine induced by activation of group I mGluRs.

Cauli et al. [41] reported that activation of metabotropic glutamatereceptorsbyinjecting(S)3,5-dihydroxyphenylglycine (DHPG) in nucleus accumbens (Nacc) increases motor activity by different mechanisms in control rats and in rats with chronic liver failure due to portacaval shunt (PCS). In control rats DHPG increases extracellular dopamine in Nacc and induces locomotion by activation the normal circuit: Nacc, ventral pallidum, medial-dorsal thalamus, prefrontal cortex, which is not activated in PCS rats [41]. In this rats, DHPG activates an alternative circuit: Nacc, substantia nigra pars reticulata, ventro-medial thalamus, prefrontal cortex, which is not activated in control rats. The reasons by which liver failure leads to activation of an alternative circuit remain unclear [42]. In hyperammonaemia, activation of the alternative circuit and increased motor response following metabotropic glutamate receptors activation in Nacc seem due to an increase in extracellular glutamate which activates AMPA receptors [41]. The mechanisms by which liver failure alters motor function remain unclear.

HA plays an important role and that altered neurotransmission mediates the different neurological alterations. Different neurotransmitter systems are altered in HA. The functional consequences at these alterations depend on the role of each neurotransmitter in the brain areas involved in the control of different cerebral processes. The different types of cognitive, motor and other neurological alterations in chHA may be due to different alterations in different brain areas [41,43].

Anxiety could also be an important factor in the experiments, so it was evaluated in the plus maze. We have observed that chHA increased acute stress responses. AIDA produced anxiety reduction in rats with chHA manifested by an increase of time spent in open arms (%), which might suggest an anxiolytic-like activity. Our data are in agreement with most studies which have postulated effects of group I mGlu receptor antagonism ranging from neuroprotection and drug abuse to depression and anxiety [30]. It has been reported that AIDA, a noncompetitive antagonist with modest selectivity towards mGluR1 [40] does produce anxiolytic activity in conflict drinking test and plus maze but not in four-plate test in rats [44].

On the other hand Pietraszek et al., demonstrated an influence of EMQMCM (3-ethyl-2-methyl-quinolin-6-yl-4-methoxy-cyclohexyl-methanone methanesulfonate), a selective mGluR1 antagonist, on the behaviour of rats in the Geller-Seiffert test [45]. EMQMCM has axiogenic effect.

The noncompetitive mGluR1 receptor antagonist BAY 36-7620 had no effect in ultrasonic vocalization in healthy rats [30]. The variation in the results of these behavioural studies may by partially explained by differences in task complexity, animal models used and doses used. Anxiolytic effect of AIDA in group of rats with chHA may have been connected with the fact that mGlu1 receptors can physically and functionally influence N-methyl-D-aspartate (NMDA) receptors [45].

Our data seem to confirm an anxiolytic potential of mGluR1 antagonism in chHA state in rats.

It is very likely that the different effects of AIDA reported are due to blocking different mGluRs (very strongly 1 and a little 5) in different brain areas [13, 27]. mGluR1 and mGluR5 receptors play different roles in hippocampus. First of all mGluR1 and mGluR5 show distinct localization in this structure. mGluR5 is expressed in all principal cells but especially high in CA1 neurons [13]. mGlu R1 receptor's immunoreactivity is strongest in principal cells of the CA3 field and dentate granule cells but absent in CA1 [13].

It is known that mGluR1 receptors are implicated in long-term potentiation and in learning and memory formation [29]. Literature reports that chronic hyperammonemia imitates complex neuropsychiatric syndrome associated with chronic hepatocellular failure [19]. In our study we have shown that AIDA - a selective mGluR1 antagonist may have an influence on cognitive functions in rats with experimental, chronic hyperammonemia.

CONCLUSIONS

Chronic hyperammonemia profoundly impaired memory in passive avoidance, increased stress responses in elevated plus maze and locomotor activity in open field test. AIDA a selective mGluR1 antagonist improved memory deficits, locomotor activity and produced anxiety reduction in rats with hyperammonemia.

ACKNOWLEDGEMENTS

This work was supported by grant No. 3-10586 L from the State Committee for Scientific Research, Warszawa, Poland.

REFERENCES

1. Canales JJ, Elayadi A, Errami M, Llansola M, Cauli O, Felipo V. Chronic hyperammonemia alters motor and neurochemical responses to activation of group I metabotropic glutamate receptors in the nucleus accumbens in rats in vivo. Neurobiol Dis. 2003 Dec;14(3):380-90.

2. Cauli O, Llansola M, Rodrigo R, El Mlili N, Errami M, Felipo V. Altered modulation of motor activity by group I metabotropic glutamate receptors in the nucleus accumbens in hyperammonemic rats. Metab Brain Dis. 2005 Dec;20(4):347-58.

3. Corbalán R, Hernández-Viadel M, Llansola M, Montoliu C, Felipo V. Chronic hyperammonemia alters protein phosphorylation and glutamate receptor-associated signal transduction in brain. Neurochem Int. 2002 Aug-Sep;41(2-3):103-8.

4. Fan P, Szerb JC. Effects of ammonium ions on synaptic transmission and on responses to quisqualate and N-methyl-D-aspartate in hippocampal CA1 pyramidal neurons in vitro. Brain Res. 1993 Dec 31;632(1-2):225-31.

5. Fedosiewicz-Wasiluk M, Hoły ZZ, Wiśniewska RJ, Wiśniewski K. The influence of NMDA, a potent agonist of glutamate receptor, on behavioral activity of rats with experimental hyperammonemia evoked by liver failure. Amino Acids. 2005 Feb;28(1):111-7.

6. Monfort P, Muñoz MD, Felipo V. Hyperammonemia impairs long-term potentiation in hippocampus by altering the modulation of cGMP-degrading phosphodiesterase by protein kinase G. Neurobiol Dis. 2004 Feb;15(1):1-10.

7. Bliss TV, Collingridge GL. A synaptic model of memory: long-term potentiation in the hippocampus. Nature. 1993 Jan 7;361(6407):31-9.

8. Clemmesen JO, Larsen FS, Kondrup J, Hansen BA, Ott P. Cerebral herniation in patients with acute liver failure is correlated with arterial ammonia concentration. Hepatology. 1999 Mar;29(3):648-53.

9. Muñoz MD, Monfort P, Gaztelu JM, Felipo V. Hyperammonemia impairs NMDA receptor-dependent long-term potentiation in the CA1 of rat hippocampus in vitro. Neurochem Res. 2000 Apr;25(4):437-41.

10. Trudeau F, Gagnon S, Massicotte G. Hippocampal synaptic plasticity and glutamate receptor regulation: influences of diabetes mellitus. Eur J Pharmacol. 2004 Apr 19;490(1-3):177-86.

11. Antzoulatos EG, Byrne JH. Learning insights transmitted by glutamate. Trends Neurosci. 2004 Sep;27(9):555-60.

12. Bortolotto ZA, Fitzjohn SM, Collingridge GL. Roles of metabotropic glutamate receptors in LTP and LTD in the hippocampus. Curr Opin Neurobiol. 1999 Jun;9(3):299-304.

13. Simonyi A, Serfozo P, Shelat PB, Dopheide MM, Coulibaly AP, Schachtman TR. Differential roles of hippocampal metabotropic glutamate receptors 1 and 5 in inhibitory avoidance learning. Neurobiol Learn Mem. 2007 Oct;88(3):305-11.

14. De Blasi A, Conn PJ, Pin J, Nicoletti F. Molecular determinants of metabotropic glutamate receptor signaling. Trends Pharmacol Sci. 2001 Mar;22(3):114-20.

15. Christoffersen GR, Christensen LH, Hammer P, Vang M. The class I metabotropic glutamate receptor antagonist, AIDA, improves short-term and impairs long-term memory in a spatial task for rats. Neuropharmacology. 1999 Jun;38(6):817-23.

16. Plech A, Rykaczewska-Czerwińska M, Bartosz-Bechowski H, Lombarska-Sliwińska D, Małota M, Szewczyk M, Brus R, Konopińska D. Insect neuropeptide leucopyrokinin analogues--synthesis and antinociceptive effect in rats. Pol J Pharmacol. 1997 Mar-Jun;49(2-3):119-26.

17. Jayakumar AR, Sujatha R, Paul V. Effect of ammonia on motor function in adult rats. Brain Res Bull. 1997;43(3):275-8.

18. Matthies H. Pharmacology of learning and memory. Trends Biochem Sci. 1980 Feb;1(1):333–7.

19. Ader R, Weijnen JAWM, Maleman P. Retention of a passive avoidance responses as a function of the intensity and duration of the electric shock. Psychonomic Sci. 1972 Mar; 26(3):125-32.

20. Pellow S, Chopin P, File SE, Briley M. Validation of open:closed arm entries in an elevated plus-maze as a measure of anxiety in the rat. J Neurosci Methods. 1985 Aug;14(3):149-67.

21. Chan H, Butterworth RF. Cell-selective effects of ammonia on glutamate transporter and receptor function in the mammalian brain. Neurochem Int. 2003 Sep-Oct;43(4-5):525-32.

22. Hermenegildo C, Montoliu C, Llansola M, Muñoz MD, Gaztelu JM, Miñana MD, Felipo V. Chronic hyperammonemia impairs the glutamate-nitric oxide-cyclic GMP pathway in cerebellar neurons in culture and in the rat in vivo. Eur J Neurosci. 1998 Oct;10(10):3201-9.

23. Monfort P, Muñoz MD, ElAyadi A, Kosenko E, Felipo V. Effects of hyperammonemia and liver failure on glutamatergic neurotransmission. Metab Brain Dis. 2002 Dec;17(4):237-50.

24. Lombardi G, Mannaioni G, Leonardi P, Cherici G, Carlà V, Moroni F. Ammonium acetate inhibits ionotropic receptors and differentially affects metabotropic receptors for glutamate. J Neural Transm Gen Sect. 1994;97(3):187-96.

25. Sáez R, Llansola M, Felipo V. Chronic exposure to ammonia alters pathways modulating phosphorylation of microtubule-associated protein 2 in cerebellar neurons in culture. J Neurochem. 1999 Dec;73(6):2555-62.

26. Heimer L. A new anatomical framework for neuropsychiatric disorders and drug abuse. Am J Psychiatry. 2003 Oct;160(10):1726-39.

27. Riedel G, Platt B, Micheau J. Glutamate receptor function in learning and memory. Behav Brain Res. 2003 Mar 18;140(1-2):1-47.

28. Nielsen KS, Macphail EM, Riedel G. Class I mGlu receptor antagonist 1-aminoindan-1,5-dicarboxylic acid blocks contextual but not cue conditioning in rats. Eur J Pharmacol. 1997 May 20;326(2-3):105-8.

29. Szydlowska K, Kaminska B, Baude A, Parsons CG, Danysz W. Neuroprotective activity of selective mGlu1 and mGlu5 antagonists in vitro and in vivo. Eur J Pharmacol. 2007 Jan 5;554(1):18-29.

30. Spooren W, Ballard T, Gasparini F, Amalric M, Mutel V, Schreiber R. Insight into the function of Group I and Group II metabotropic glutamate (mGlu) receptors: behavioural characterization and implications for the treatment of CNS disorders. Behav Pharmacol. 2003 Jul;14(4):257-77.

31. Faden AI, O'Leary DM, Fan L, Bao W, Mullins PG, Movsesyan VA. Selective blockade of the mGluR1 receptor reduces traumatic neuronal injury in vitro and improvesoOutcome after brain trauma. Exp Neurol. 2001 Feb;167(2):435-44.

32. Bordi F, Reggiani A, Conquet F. Regulation of synaptic plasticity by mGluR1 studied in vivo in mGluR1 mutant mice. Brain Res. 1997 Jun 27;761(1):121-6.

33. Grzeda E, Wiśniewska RJ. Differentiations of the effect of NMDA on the spatial learning of rats with 4 and 12 week diabetes mellitus. Acta Neurobiol Exp (Wars). 2008;68(3):398-406.

34. Jover R, Compañy L, Gutiérrez A, Lorente M, Zapater P, Poveda MJ, Such J, Pascual S, Palazón JM, Carnicer F, Ferrandis F, Pérez-Mateo M. Clinical significance of extrapyramidal signs in patients with cirrhosis. J Hepatol. 2005 May;42(5):659-65.

35. Amodio P, Montagnese S, Gatta A, Morgan MY. Characteristics of minimal hepatic encephalopathy. Metab Brain Dis. 2004 Dec;19(3-4):253-67.

36. Cauli O, Llansola M, Erceg S, Felipo V. Hypolocomotion in rats with chronic liver failure is due to increased glutamate and activation of metabotropic glutamate receptors in substantia nigra. J Hepatol. 2006 Nov;45(5):654-61.

37. Trevitt T, Carlson B, Correa M, Keene A, Morales M, Salamone JD. Interactions between dopamine D1 receptors and gamma-aminobutyric acid mechanisms in substantia nigra pars reticulata of the rat: neurochemical and behavioral studies. Psychopharmacology (Berl). 2002 Jan;159(3):229-37.

38. Car H, Nadlewska A, Oksztel R, Wiśniewski K. AIDA influences behavior in rats pretreated with baclofen. Pol J Pharmacol. 2001 May-Jun;53(3):245-52.

39. Conquet F, Bashir ZI, Davies CH, Daniel H, Ferraguti F, Bordi F, Franz-Bacon K, Reggiani A, Matarese V, Condé F, Collingridge GL, Crepel F. Motor deficit and impairment of synaptic plasticity in mice lacking mGluR1. Nature. 1994 Nov 17;372(6503):237-43.

40. Moroni F, Lombardi G, Thomsen C, Leonardi P, Attucci S, Peruginelli F, Torregrossa SA, Pellegrini-Giampietro DE, Luneia R, Pellicciari R. Pharmacological characterization of 1-aminoindan-1,5-dicarboxylic acid, a potent mGluR1 antagonist. J Pharmacol Exp Ther. 1997 May;281(2):721-9.

41. Cauli O, Mlili N, Rodrigo R, Felipo V. Hyperammonaemia alters the mechanisms by which metabotropic glutamate receptors in nucleus accumbens modulate motor function. J Neurochem. 2007 Oct;103(1):38-46.

42. Cauli O, Mlili N, Llansola M, Felipo V. Motor activity is modulated via different neuronal circuits in rats with chronic liver failure than in normal rats. Eur J Neurosci. 2007 Apr;25(7):2112-22.

43. Vezina P, Kim JH. Metabotropic glutamate receptors and the generation of locomotor activity: interactions with midbrain dopamine. Neurosci Biobehav Rev. 1999 Mar;23(4):577-89.

44. Kłodzińska A, Tatarczyńska E, Stachowicz K, Chojnacka-Wójcik E. The anxiolytic-like activity of AIDA (1-aminoindan-1,5-dicarboxylic acid), an mGLu 1 receptor antagonist. J Physiol Pharmacol. 2004 Mar;55(1 Pt 1):113-26.

45. Pietraszek M, Sukhanov I, Maciejak P, Szyndler J, Gravius A, Wisłowska A, Płaźnik A, Bespalov AY, Danysz W. Anxiolytic-like effects of mGlu1 and mGlu5 receptor antagonists in rats. Eur J Pharmacol. 2005 May 2;514(1):25-34.