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Anxiolytic properties of the antipsychotic alkaloid alstonine

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Abstract

Anxiolytic properties may be a crucial feature of newer antipsychotics associated with the improvement of negative symptoms in schizophrenic patients. The indole alkaloid alstonine acts as an atypical antipsychotic in behavioral models, but differs in its dopamine and serotonin binding profile. The purpose of this study was to verify if alstonine possesses anxiolytic properties in mice. The hole-board and light/dark models were used; moreover, the participation of D_1, 5-HT_2, NMDA and γ-aminobutyric acid (GABA) receptors was likewise investigated. Alstonine clearly behaves as anxiolytic in both hole-board and light/dark situations. Pretreatment with the 5-$HT_{2A/2C}$ serotonin receptor antagonist ritanserin reverted the effects of alstonine in both the hole-board and light/dark models, suggesting the involvement of these receptors in the alstonine mechanism of action. The involvement of glutamate NMDA receptors should also be considered, given that alstonine partially reversed the increase in locomotion induced by MK-801 in the hole board, as well as MK-801-induced hyperlocomotion in motor activity apparatus.

Keywords: Alstonine; Antipsychotics; Anxiety; Serotonin; Glutamate; Hole-board; Light/dark; Mice

1. Introduction

A satisfactory understanding of the pathophysiology underlying schizophrenia is still to be unveiled (Andreasen, 2000; Crow, 2000; Terenius, 2000; Lara and Souza, 2000). While treatment of schizophrenia has been significantly ameliorated with the introduction of newer atypical antipsychotics, the contribution of each component of these new drugs' manifold mechanism of action to the overall patients improvement is not clear (Lindenmayer, 2000). Besides clinical contribution, the study of newer antipsychotics, such as clozapine and sulpiride, highlighted the role of receptors other than dopamine D_2 (namely, dopamine D_1, 5-HT_2 and NMDA) in schizophrenia.

Anxiety is a syndrome common to many nervous disorders and in schizophrenia, antipsychotic therapy reduces anxiety concomitant with the alleviation of the psychosis (Bourin et al., 2001). Recently, a significant amount of attention has been given to the links between anxiety and schizophrenia (Turnbull and Bebbington, 2001). Schizophrenic patients may exhibit depression, anxiety and fear, often hard to distinguish from primary negative symptoms of schizophrenia. It has been suggested that the anxiolytic property of certain antipsychotics is a crucial feature for ameliorating the so-called negative symptoms, which affect the quality life in some schizophrenics (Cao and Rodgers, 1997; Sakamoto et al., 1998; Huppert et al., 2001). It is known that the relative efficacy on positive and negative symptoms, cognition, psychotic anxiety and depression, suicidality and quality of life varies among different antipsychotic compounds (Blin, 1999); it is also recognized that differences in the diverse behavioral experimental models may be indicative of diverse clinical profile (Moore, 1999).

The indole alkaloid alstonine is the major component of a plant-based remedy, traditionally used in Nigeria to treat mental illnesses (Costa-Campos et al., 1999). While decoctions prepared with alstonine-containing plants are used orally in African traditional medicine (Iwu, 1993), intra-

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peritonial administration to mice shows significant effects within 30 min. In mice models (haloperidol catalepsy, amphetamine lethality, amphetamine and apomorphine stereotypes), alstonine shows antipsychotic properties (0.5–1.0 mg/kg ip); the profile is closer to atypical antipsychotics (clozapine and sulpiride) than to older neuroleptics (haloperidol and chlorpromazine). Nevertheless, unlike most atypical antipsychotics, alstonine does not affect [^{3}H]SCH23390 or [^{3}H]Spiperone binding to cortex membranes, indicating the lack of direct interaction with D_1, D_2 and 5-HT_{2A} receptors (Costa-Campos et al., 1998). While the mechanism(s) of action of alstonine remains to be elucidated, a consistent bell-shaped dose–effect relationship (activity in the range of 0.5–2.0 mg/kg) in animal models suggests that alstonine interacts with more than one neurotransmitter system (Costa-Campos et al., 1998).

Considering that the claimed therapeutic effects of alstonine-based formulations in humans include sedation in mentally ill patients, the similarity of alstonine experimental behavioral profile with that of clozapine, and the possibility of an innovative mechanism of action, the purpose of this study was to verify if alstonine possess anxiolytic properties in the hole-board and light/dark mice models. The participation of D_1, 5-HT_2, NMDA and γ-aminobutyric acid (GABA) receptors in the effects of alstonine was also examined in these models by pretreatment with appropriate antagonists. The effects of alstonine were further studied on spontaneous locomotion, MK-801-induced hyperlocomotion and rota rod performance.

2. Materials and methods

2.1. Animals

Experiments were performed with male adult mice, CF1 strain, received from Fundação Estadual de Experimentação e Produção da Saúde (FEEPS), immediately after weaning (21 days). Animals were maintained in our own animal facilities under controlled environment (22 ± 1 °C, 12-h light/dark cycle, free access to food [Nuvilab CR1] and water) until they were 10 weeks old (30–40 g). All procedures were carried out according to institutional policies on experimental animal handling.

2.2. Drugs

Clozapine, sulpiride, chlorpromazine, amphetamine and diazepam were obtained from Sigma (USA) and haloperidol was used from Haldol (Janssen Farmacêutica do Brasil). SCH23390, ritanserin and MK-801 were acquired from RBI (USA). Drugs and vehicles were administered intraperitoneally, except for clozapine, which was given subcutaneously, always as 0.1 ml/10 g of body weight. Alstonine was isolated according to Iwu and Court (1977) (see also Iwu and Klayman, 1992). Alstonine, picrotoxine and MK-801 were dissolved in distilled water. Clozapine and sulpiride were dissolved in a minimal volume of 0.1 N HCl and due volume completed with distilled water. The pH was adjusted to 5–6 by adding 1 N NaOH. Diazepam was diluted in propylene glycol (20% PPG). Ritanserin and SCH23390 were diluted in DMSO (10%).

2.3. Hole board

The hole-board apparatus (Ugo Basile, Italy) consisted of a gray Perspex panel (40 × 40 cm, 2.2-cm thick) with 16 equidistant holes (3-cm diameter) in the floor. Photocells below the surface of the holes provided the measures of the number of head-dips. The board was positioned 15 cm above the table and was divided (with black water-resistant marker) into nine squares of 10 × 10 cm. The method was

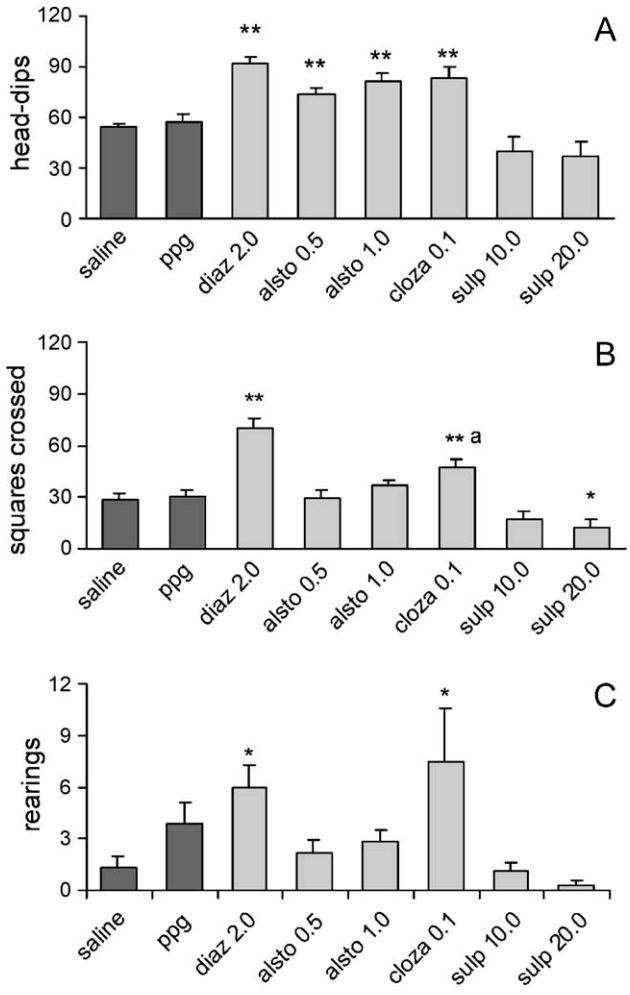

Fig. 1. Effect of alstonine, diazepam, clozapine and sulpiride in the mice hole-board model. Alstonine (alsto, 0.5 and 1.0 mg/kg ip), diazepam (diaz, 2.0 mg/kg ip), clozapine (cloza, 0.1 mg/kg sc) and sulpiride (sulp, 10.0 and 20.0 mg/kg ip), or vehicles (saline or PPG) were injected 30 min prior to the behavior measurements: (A) head-dips; (B) squares crossed; (C) rearing. Each column represents the mean ± S.E.M. ($n = 10–14$). $^{**}P < .01$, $^{*}P < .05$ vs. vehicle-treated group. a = $P < .05$ vs. diazepam. ANOVA was followed by Student–Newmann–Keuls analysis.

adapted from Takeda et al. (1998). Mice were transported to the dimly lit laboratory at least 1 h before testing. Each animal was individually placed in the center of the board (facing away from the observer) and the following parameters were noted for 5 min: the latency to the first head-dip, the number of rearings and spontaneous movements (number of squares crossed with all four paws). Groups ($n = 10–15$) were treated with diazepam (2.0 mg/kg), clozapine (0.1 mg/kg), sulpiride (10.0 and 20.0 mg/kg), alstonine (0.5 and 1.0 mg/kg) or vehicles (0.9% saline or 20% PPG) 30 min prior to the testing. In a second experiment, the animals were treated with saline or antagonists intraperitoneally (0.1 mg/kg SCH23390, 2.0 mg/kg ritanserin, 0.1 mg/kg MK-801 and 1.0 mg/kg picrotoxine) 30 min before saline or alstonine (1.0 mg/kg). The test was performed 30 min after the last drug administration.

2.4. Light/dark

The light/dark apparatus consisted of a rectangular box (46 × 27 × 30 cm), divided into one small (18 × 27 cm) and one large (27 × 27) area, with an opening door (7.5 × 7.5 cm) located in the center of the partition at floor level. The small compartment was painted in black, whereas the large compartment was painted in white and was brightly illuminated with a 60-W cold light source. The method was adapted from Li and Quock (2001). Each animal was individually placed in the center of the light compartment (facing away from the door) and the following parameters were noted for 5 min: latency of the first crossing from one compartment to the other, time spent in the light zone and the number of crossings between the light and dark compartments. The test was performed in a quiet, darkened room. The mice were kept in this room for at least 1 h before the test. Groups ($n = 14–25$) were treated with diazepam (1.0 mg/kg), clozapine (0.1 mg/kg), sulpiride (10.0 and 20.0 mg/kg), alstonine (0.5 and 1.0 mg/kg) or vehicle (0.9% saline or 20% PPG) 30 min prior to the testing. In a second experiment, the animals were treated with saline or ritanserin (2.0 mg/kg) 30 min before saline or alstonine (1.0 mg/kg). The test was performed 30 min after the last drug administration.

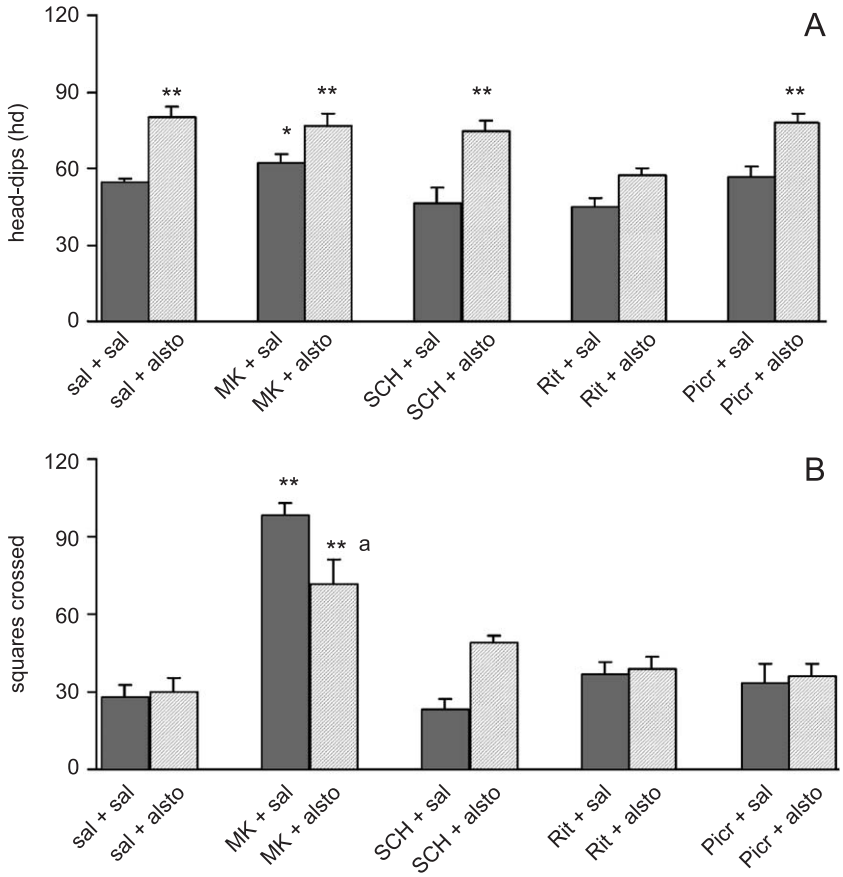

Fig. 2. Effect of the interaction of alstonine and antagonists in the mice hole-board model. Animals were treated with saline or the antagonists SCH23390 (SCH 0.1 mg/kg ip), ritanserin (Rit, 2.0 mg/kg), MK-801 (MK, 0.1 mg/kg) and picrotoxine (Picr, 1.0 mg/kg) 30 min prior to vehicle (solution saline, 0.9%) or alstonine (alsto, 1.0 mg/kg). (A) Head-dips and (B) squares crossed. The test was performed 30 min after the last drug administration. Each column represents the mean ± S.E.M. ($n = 10–14$). ** $P < .01$, * $P < .05$ vs. vehicle-treated group. a = $P < .01$, vs. MK-801-saline. ANOVA was followed by Student–Newmann–Keuls analysis.

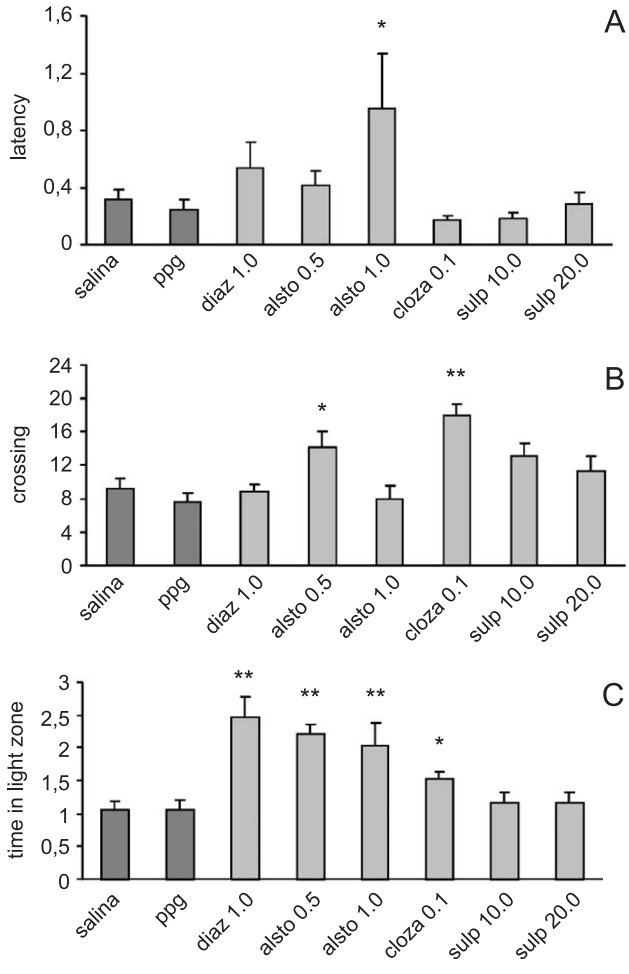

Fig. 3. Effect of alstonine, diazepam, clozapine, and sulpiride in the mice light/dark model. Alstonine (alsto, 0.5 and 1.0 mg/kg ip), diazepam (diaz, 1.0 mg/kg ip), clozapine (cloza 0.1 mg/kg sc), sulpiride (sulp 10.0 and 20.0 mg/kg ip) or saline (sal) were injected 30 min prior to the measurements of exploratory behavior: (A) latency for the first crossing; (B) number of crossings; (C) time spent in the light zone. Each column represents the mean ± S.E.M. ($n = 15–20$). $^{**}P < .01$, $^{*}P < .05$ vs. vehicle-treated group. ANOVA was followed by Student–Newmann–Keuls analysis.

2.5. Spontaneous locomotor activity

The method for the spontaneous locomotor activity was adapted from Creese et al. (1976). Activity cages (45 × 25 × 20 cm, Albarsch Electronic Equipment), equipped with three parallel photocells, automatically record the number of crossings. Animals were individually habituated to an activity cage for 10 min before receiving the following treatments ($n = 10–16$): saline, clozapine (0.1 and 0.5 mg/kg), sulpiride (10.0 and 20.0 mg/kg) and alstonine (0.1, 0.5 and 1.0 mg/kg). The animals returned to the activity cages 30 min after treatments, and the crossings were recorded for 15 min.

2.6. Hyperlocomotion induced by MK-801

The method was adapted from Ninan and Kulkarni (1998). Locomotion activity was measured using the activity cages described above. The mice ($n = 10–25$) were treated with saline, alstonine (0.5 and 1.0 mg/kg), clozapine (0.1, 0.2, 0.3 and 0.5 mg/kg) and sulpiride (10.0 and 20.0 mg/kg), and 30 min later received MK-801 (0.25 mg/kg). Control animals received saline only. The mice were individually placed in the activity cages, 30 min after MK-801 administration, and motor activity was recorded for 5 min, starting 2 min after the mice was placed in the cage (considered as exploration of the new environment).

2.7. Rota rod

The method was adapted from Bristow et al. (1996). Mice were initially trained to remain on the rota rod apparatus (18 rpm) for 120 s; those that did not remain on the bar for at least two out of three consecutive trials were discarded. Clozapine (0.1 mg/kg), sulpiride (10.0 and 20.0 mg/kg), alstonine (0.5 and 1.0 mg/kg) or vehicles were administered 24 h after this initial training ($n = 12–19$). The latency to fall from the rota rod (one 60-s trial) was determined 30, 60, 90 and 120 min after drug administration.

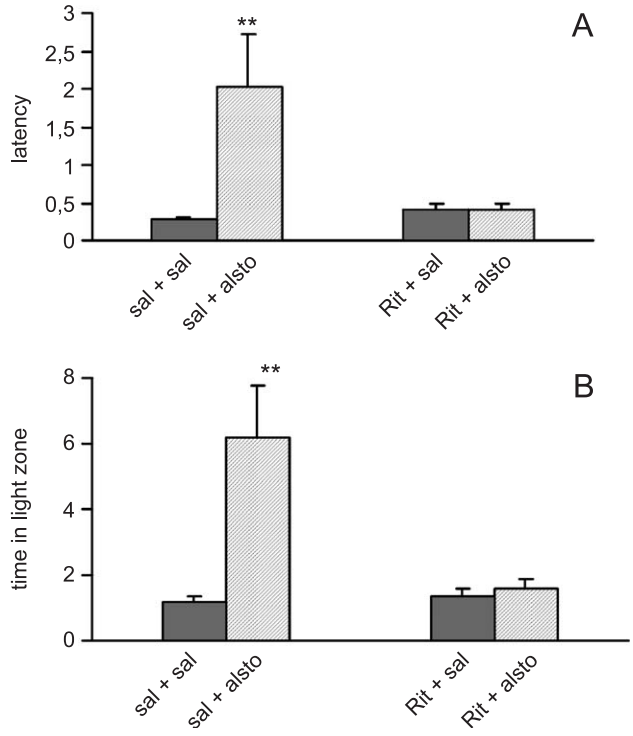

Fig. 4. Effect of the interaction of alstonine and ritanserin in the light/dark model: (A) latency and (B) time spent in the light zone. Animals were treated with saline (sal) or the antagonist ritanserin (Rit, 2.0 mg/kg) prior to saline or alstonine (alsto, 1.0 mg/kg). The test was performed 30 min after the last drug administration. Each column represents the mean ± S.E.M. ($n = 10–12$). $^{**}P < .01$ vehicle-treated group. ANOVA was followed by Student–Newmann–Keuls analysis.

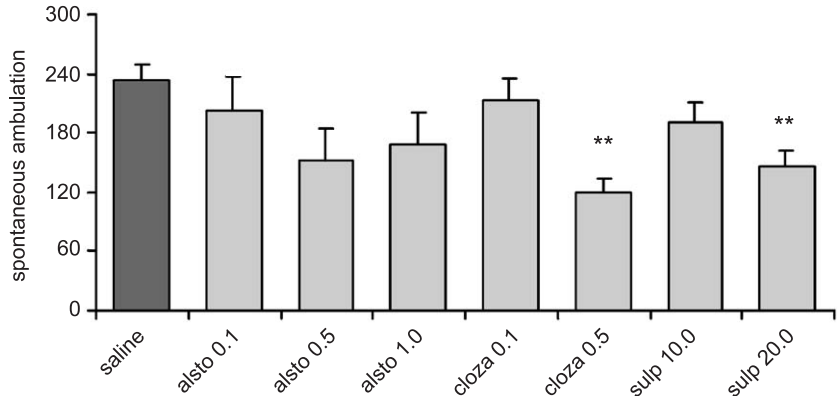

Fig. 5. Effect of alstonine (alsto, 0.1, 0.5 and 1.0 mg/kg), clozapine (cloza, 0.1 and 0.5 mg/kg), sulpiride (sulp, 10.0 and 20.0 mg/kg) or saline on spontaneous locomotor activity. Each column represents the mean ± S.E.M. ($n = 10$). * * $P < .01$ vs. vehicle-treated group. ANOVA was followed by Student–Newmann–Keuls analysis.

2.8. Statistical analysis

Results are expressed as mean ± S.E.M. The data were analyzed by one-way ANOVA followed by Student–Newman–Keuls post hoc analysis. The comparison between the data in rota rod performance at different time points were analyzed using a general linear model (GLM) with repeated measures (drug treatment vs. time), with time as the repeated measure. Student–Newman–Keuls post hoc analysis was used to determine the differences among specific groups.

3. Results

Fig 1 shows the results obtained with the hole board. Alstonine [0.5 and 1.0 mg/kg; $F(2,34) = 14.89$, $P < .01$] and clozapine [0.1 mg/kg; $F(1,22) = 26.84$, $P < .01$] significantly increased the number of head-dips, an effect comparable to diazepam [2.0 mg/kg; $F(2,38) = 42.17$, $P < .01$; Fig. 1A]. Locomotion (squares crossed) was increased by clozapine [0.1 mg/kg; $F(1,22) = 8.07$, $P < .01$] and even more so by diazepam [2.0 mg/kg; $F(2,38) = 27.29$, $P < .01$]. Alstonine was devoid of effect, and locomotion was significantly decreased by sulpiride [20.0 mg/kg; $F(2,29) = 3.19$, $P < .05$; Fig. 1B]. Clozapine [0.1 mg/kg; $F(1,22) = 5.39$, $P < .05$] and diazepam, to a lesser extent [$F(2,38) = 5.31$, $P < .05$], significantly increased rearing (Fig. 1C). None of the drugs affected the latency for the first head-dip (data not shown).

The increase in head-dips induced by alstonine was not modified by previous administration of SCH23390, MK-801 or picrotoxine, but was prevented by ritanserin (Fig. 2A). As expected, the number of squares crossed was increased by MK-801 [compared with saline only; $F(3,43) = 34.32$, $P < .01$], an increase attenuated by alstonine (Fig. 2B).

In the light/dark test, alstonine (1.0 mg/kg) significantly increased the latency for the first crossing from the light to the dark compartment [$F(2,58) = 4.61$, $P < .05$; Fig. 3A]. The number of crossings between compartments was increased by alstonine [0.5 mg/kg; $F(2,58) = 4.60$, $P < .05$]

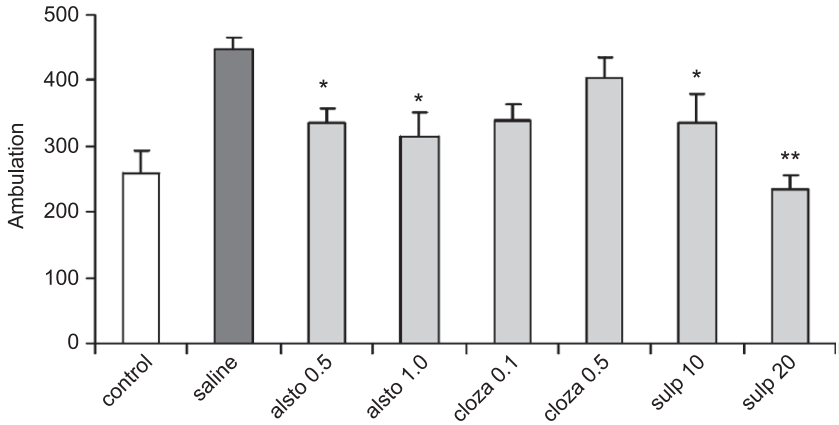

Fig. 6. Effect of alstonine (alsto, 0.1, 0.5 and 1.0 mg/kg), clozapine (cloza, 0.1 and 0.5 mg/kg), sulpiride (sulp, 10.0 and 20.0 mg/kg) or saline on hyperlocomotion induced by MK-801 (0.25 mg/kg). Control animals received saline only. Each column represents the mean ± S.E.M. ($n = 10$). * * $P < .01$, * $P < .05$ vs. vehicle-treated group. ANOVA was followed by Student–Newmann–Keuls analysis.

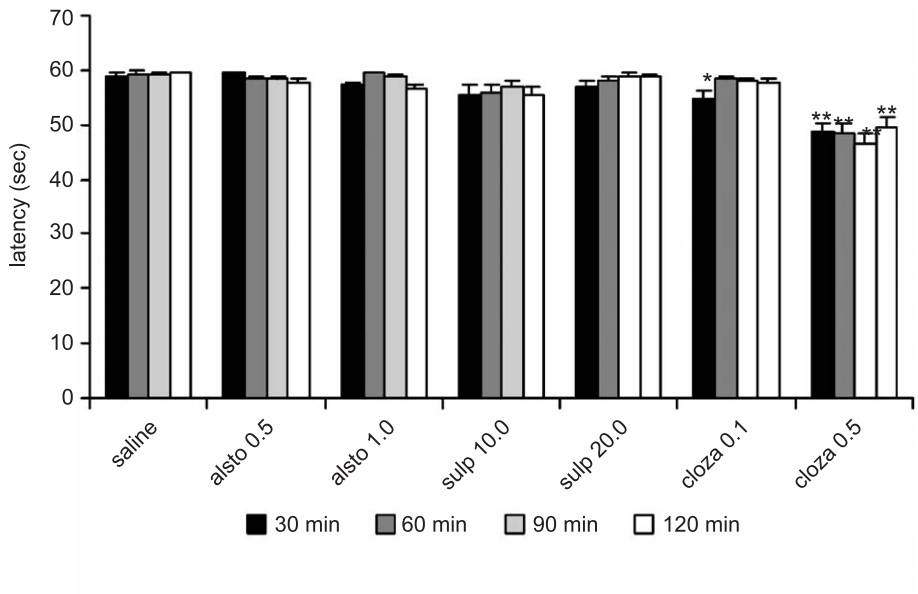

Fig. 7. Effects of saline, alstonine (alsto, 0.5 and 1.0 mg/kg), clozapine (cloza, 0.1 and 0.5 mg/kg) and sulpiride (sulp, 10.0 and 20.0 mg/kg) on the rota rod test. The time spent on the rota rod (maximum of 60 s) was determined at 30, 60, 90 and 120 min posttreatment. Each column represents the mean ± S.E.M. ($n = 10–12$). $^{**}P < .01$ vs. vehicle-treated group. ANOVA was followed by Student–Newmann–Keuls analysis.

and clozapine [0.1 mg/kg; $F(1,38) = 25.55$, $P < .01$; Fig. 3B]. Like diazepam [1.0 mg/kg; $F(2,60) = 17.09$, $P < .01$], alstonine [0.5 and 1.0 mg/kg; $F(2,58) = 8.11$, $P < .01$] increased the time spent exploring the light zone (Fig. 3C). This alstonine-induced increase in latency (Fig. 4A) and in the time spent in the light zone was prevented by previous administration of ritanserin (2.0 mg/kg; Fig. 4B). None of the drugs affected the number of crossings (data not shown).

As can be seen in Fig. 5, alstonine (0.5 and 1.0 mg/kg), sulpiride (10.0 mg/kg) and clozapine (0.1 mg/kg) did not interfere with the spontaneous locomotion, whereas clozapine [0.5 mg/kg; $F(2,49) = 8.27$, $P < .01$] and sulpiride [20.0 mg/kg; $F(2,53) = 5.69$, $P < .01$] reduced locomotion. Fig. 5 shows the alstonine (0.5 and 1.0 mg/kg), clozapine (0.2 and 0.3 mg/kg) and sulpiride (10.0 and 20.0 mg/kg) effects on MK-801-induced hyperlocomotion. Alstonine [$F(2,51) = 6.96$, $P < .05$] and the sulpiride [$F(2,51) = 13.25$, $P < .01$] prevented MK801-induced hyperlocomotion, whereas clozapine did not have significant effects (Fig. 6).

Alstonine per se did not induce deficits in rota rod performance at any of the doses or post treatment times studied. A significant deficit was observed with clozapine [0.5 mg/kg; $F(2,42) = 7.85$, $P < .01$] from 30 to 120 min after treatment and clozapine 0.1 mg/kg on 30 min (Fig. 7).

4. Discussion

Anxiety, a symptom accompanying various central nervous system disorders and a disorder by itself, is characterized in humans by a tense and physically exhaustive alertness (Treit, 1985). Other animal species display a variety of defensive reactions in response to predators, some understood as animal correlates of anxious states (Rodgers et al., 1995). Rodents demonstrate anxiety, fear and curiosity when placed in a new environment, and an overall assessment of behavior can be determined by observing freezing, grooming (fear) or rearing, head-dips (curiosity) and the number of fecal boluses (stress; Kennedy, 1978; File, 1987; Dalvi and Rodgers, 1999). The hole-board and light/dark models have been frequently used to detect and evaluate anxiolytic/anxiogenic properties of drugs (File and Wardill, 1975; Isogawa et al., 2003; Nic Dhonnchadha et al., 2003; Ohl et al., 2002; Takeda et al., 1998). Whereas the increase in the number of head-dips is the most reliable parameter in the hole-board model (Takeda et al., 1998), the time spent in the light area is the principal index of the anxiety in the light/dark model, accepting the fact that an intensely lighted area is extremely aversive to rodents. In both cases, the data must be analyzed in the light of possible sedation (Hascoët and Bourin, 1998), especially if it significantly affects locomotion.

Stress is used in the literature referring to a diversity of meanings including the stressor, the accompanying anxiety and the behavioral, physiological, endocrine or immune responses that are elicited by stressful episodes (Gispen-de Wied, 2000). Although it is generally accepted that stress plays a significant role in the development of psychiatric disorders, including depression, posttraumatic stress disorder and schizophrenia (Walker and Diffioro, 1997; Esch et al., 2002; Heim et al., 2000; Nemeroff, 1998), the mechanisms by which stress actually influences these disorders remain to be clarified.

The most significant findings of this study are that alstonine clearly behaves as anxiolytic in both animal models, and that its anxiolytic effects are antagonized by the previous administration of the 5-$HT_{2A/2C}$ receptor antagonist ritanserin. In the hole-board model, alstonine significantly increases the number of head-dips, without interfering with the rearing or the number of squares crossed, while diazepam induced significant increases in all three parameters. The effects of diazepam on rearing and locomotion are consistent with its effects on open field experiments (Ramanathan et al., 1998; Wieland et al., 1991). Different results obtained with alstonine and diazepam may be attributed to differential modulation of GABAergic transmission. In the light/dark test, alstonine significantly increases the time spent in light area exploration, as well as the latency for the first crossing from light to dark compartment (Bourin and Hascoët, 2003). An increase in the latency for the first crossing is considered a reliable parameter for measuring the disinhibitory behavior and decreased anxiety; in this case, a potential false positive due to sedation is ruled out by the lack of effects of alstonine in ambulation.

Although data from binding assays (Costa-Campos et al., 1998) do not suggest direct interaction of alstonine with 5-HT_2 receptors in cortical membranes, the results with the hole-board and light/dark models are indicative of the involvement of 5-$HT_{2A/2C}$ receptors. Pretreatment with ritanserin prevents the alstonine-induced increase in head-dips in the hole-board, and in all parameters (latency for first crossing, number of crossings and time spent in light zone) in the light/dark model. Alstonine partially reversed MK-801-induced increase in locomotion, both at the hole-board and locomotor activity cages. Accordingly, the inhibition of 5-HT_{2A} receptor function by antipsychotics has been previously reported to reduce the responsiveness to the effects of NMDA antagonists (Maurel-Remy et al., 1995; Zhang and Bymaster, 1999). A reduced responsiveness to an NMDA antagonist is compatible with the power of 5-HT_2 antagonists to enhance NMDA-mediated transmission (Arvanov and Wang, 1998; Spurney et al., 1999). In view of the complex interactions between serotonin and glutamate receptor subtypes (Breese et al., 2002), a possible role of NMDA glutamate receptors in alstonine mechanism(s) of action should be considered.

It has been long accepted that the regulation of fear and anxiety is strongly associated with the central γ-aminobutyric acid (GABA) and serotonergic (5-HT) systems (File, 1987). Recently however, the significance of other neurotransmitters systems (such as cholinergic, dopaminergic and glutamatergic) in modulating emotional behavior has received attention (Nakamura and Kurasawa, 2001). It has been suggested that NMDA antagonists may have potential as nonclassical anxiolytics (Dunn et al., 1989). In fact, MK-801 not only shows an anticonflict effect, but also enhances the anticonflict effect of benzodiazepine anxiolytics in mice (Kuribara et al., 1990; Plaznik et al., 1994, Reddy and Kulkarni, 1997). It is therefore noteworthy that, in agreement with data reported with the elevated plus-maze (Dunn et al., 1989), we report here a significant increase in head-dips with MK-801. The interaction between the dopaminergic and glutamatergic transmission (Carlsson and Carlsson, 1990) are of relevance to MK-801-induced alterations in exploratory behavior, stereotypy and ataxia.

The anxiolytic behavior of clozapine, previously reported with the elevated plus-maze (Cao and Rodgers, 1997; Szewczak et al., 1995) and the separation-induced vocalization (SIV) models of anxiety (Kehne et al., 2000), has been confirmed by our results in the hole-board and the light/dark models. These results correlate well with the observed clinical efficacy of clozapine in schizophrenic patients that presents anxiety symptoms (Blin, 1999). Despite indications that in humans, sulpiride reduces anxiety in neuroses (Costall and Naylor, 1995), experimental data are controversial, with results of anxiolytic (Barry et al., 1987) and anxiogenic (Simon et al., 1992) in the light/dark model, or yet, as in our study, devoid of clear effects in other models (Belzung and Berton, 1997; Cavazzuti et al., 1999; Groner et al., 1992; Redolat et al., 1991; Rex et al., 1998; Rodgers et al., 1994; Shimada et al., 1995).

The findings that 5-HT_2 or 5-HT_3 receptors blockade by ritanserin, ondansetron and other agents has a disinhibitory profile in animal models of anxiety (Costall and Naylor, 1991) provided further support to the serotoninergic modulation of anxiety. In fact, ritanserin has been shown to improve generalized anxiety disorder as efficaciously as the benzodiazepine anxiolytics (Graeff et al., 1998). Despite proven clinical effects (Delle Chiaie et al.; 1995; Griebel et al., 1997; Pollak et al., 1997; Rodgers; 1997), ritanserin shows inconsistent results in different animal models (Haller, 2001; Costall and Naylor, 1995; Rex et al., 1998). In this study, ritanserin was devoid of anxiolytic effects in the doses studied. Moreover, 5-HT_{1A} agonist effects of buspirone (receptor agonist) and other agents at the 5-HT somatodendritic receptors disinhibit behavior in animals and are anxiolytic in man (Costall and Naylor, 1997; Pokk and Zharkovsky, 1998; Haller, 2001). A serotoninergic involvement with the anxiolytic profile of alstonine is, therefore, not entirely surprising, and correlates well with the previously reported disinhibitory effects of clozapine and thioridazine (Costall and Naylor, 1995).

It has been proposed that new agents that modulate 5-HT_2 family serotonin receptors could have a significant impact on mental disease management (Jones and Blackburn, 2002; Roth and Shapiro, 2001) including anxiety, depression, schizophrenia and other disorders (Van Oekelen et al., 2003). Complementing the previously reported antipsychotic properties of alstonine, we report here that alstonine possesses anxiolytic properties, an effect likely to be related to the serotoninergic system. This study, therefore, adds to the idea that the indole alkaloid alstonine deserves further investigation as a tool in psychopharmacology and, conceivably, as a prototypic antipsychotic drug.

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