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European Journal of Pharmacology 431 (2001) 43–46

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Short communication

Effect of glucose on antipsychotic drug-induced changes in dopamine neuronal activity

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Received 25 June 2001; received in revised form 7 September 2001; accepted 20 September 2001

Abstract

The effect of glucose on antipsychotic drug-induced changes in the spontaneous activity of rat midbrain dopaminergic neurons was tested with the cells-per-track extracellular electrophysiology method. After daily s.c. treatment with vehicle, haloperidol (0.5 mg/kg), or clozapine (20 mg/kg) for 21 days, rats were anesthetized and recordings performed on one side of the brain. Then, glucose (250 mg/kg, i.v.) was administered and recordings were made from the contralateral midbrain. Glucose significantly reduced the number of spontaneously active A9 and A10 dopaminergic cells per track in control rats, but significantly attenuated the chronic haloperidol- and clozapine-induced reductions in dopaminergic cells per track. These results suggest that caloric intake may influence antipsychotic drug-induced changes in the population activity of midbrain dopaminergic neurons.

Keywords: Antipsychotic; Clozapine; Haloperidol; Dopamine electrophysiology; Obesity

1. Introduction

Patients treated chronically with antipsychotic drugs usually experience weight gain due to overeating, which often progresses to obesity (Ganguli, 1999). The underlying mechanism for antipsychotic drug-induced weight gain is not clearly understood (Baptista, 1999), but this phenomenon may be related to the finding that the levels of striatal dopamine D_2 receptors are low in drug-free obese patients (Wang et al., 2001). In this light, the possible role of the electrophysiological effects of antipsychotic drugs are of interest. In rats, chronic administration of the typical antipsychotic drug haloperidol results in a reduction in the number of spontaneously active dopaminergic neurons in the substantia nigra pars compacta (A9) and ventral tegmental area (A10) of the rat midbrain, whereas the atypical antipsychotic drug clozapine selectively reduces the number of active A10 neurons (Chiodo and Bunney, 1983; White and Wang, 1983a; Skarsfeldt, 1992). In these studies, rats were treated with antipsychotic drugs for 3 weeks, a duration of treatment shown to be sufficient to achieve maximal decreases in the numbers of active dopaminergic neurons (White and Wang, 1983b). These reductions in population activity have been attributed to the induction of depolarization block of dopaminergic neurons, and appear to be associated with a large decrease in terminal dopamine release (Lane and Blaha, 1987). This effect, combined with concurrent blockade of postsynaptic dopamine receptors by antipsychotic drugs, suggests that dopaminergic neurotransmission is profoundly diminished during chronic treatment (Grace et al., 1997).

In control rats, low autoreceptor-selective doses of dopamine receptor agonists hyperpolarize dopaminergic neurons and inhibit their activity. This hyperpolarizing action, however, reverses the depolarization inactivation of dopaminergic neurons induced by chronic antipsychotic drug administration (Bunney and Grace, 1978; Grace and Bunney, 1986; Skarsfeldt, 1992). Repeated treatment of patients with low doses of dopamine receptor agonists reverses antipsychotic drug-induced weight gain (Baptista et al., 1987).

Acute i.v. administration of glucose inhibits the firing rate of individual midbrain dopaminergic neurons in rats

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0014-2999/01/$ - see front matter
PII: S0014-2999(01)01408-X

(Saller and Chiodo, 1980). This raises the possibility that caloric intake, like dopamine receptor agonist administration, may influence the degree of depolarization block in the dopaminergic neuronal population during chronic antipsychotic drug administration. We carried out experiments to test the hypothesis that glucose can attenuate the chronic antipsychotic drug-induced reduction in the number of spontaneously active rat midbrain dopaminergic neurons.

2. Materials and methods

2.1. Animal treatment and electrophysiology procedure

Male Sprague-Dawley rats (225–300 g; Charles River, Wilmington, MA) were group-housed and maintained on a regular 12:12-h light/dark cycle with food and water available ad libitum. Animals were injected s.c. once daily for 21 days with either water (1 ml/kg), haloperidol (0.5 mg/kg), or clozapine (20 mg/kg). Two hours after the last injection, rats were anesthetized with chloral hydrate (400 mg/kg, i.p.) and placed in a stereotaxic instrument. A lateral tail vein was cannulated and body temperature maintained at 36–37 °C with a heating pad. A burr hole was drilled in the skull unilaterally over the substantia nigra (A9) and ventral tegmental area (A10) (anterior 3.2–4.2 mm, lateral 0.2–2.4 mm; Paxinos and Watson, 1986). Single-barrel micropipettes filled with 1 M NaCl were used to record extracellular action potentials. Spontaneously active dopaminergic neurons were identified as previously described (Bunney et al., 1973).

In the A9 and A10 regions, the recording pipette was lowered 12 times through a stereotaxically defined block of tissue as described previously (Chiodo and Bunney, 1983). The number of spontaneously active dopaminergic neurons encountered per electrode track was determined in vehicle- and drug-treated rats. Next, a burr hole was drilled in the skull over the contralateral midbrain, the rat was injected with saline or with glucose (250 mg/kg) via a tail vein (0.5 ml/kg), and determination of the number of dopaminergic cells per track was begun immediately on that side.

2.2. Data analysis

The effects of chronic treatments on dopaminergic cell population activity were compared with two-way analysis of variance (ANOVA) with region (A9/A10) and treatment as independent variables, followed by the Newman–Keuls test for group comparisons. The within-subject effects (before/after) of acute glucose treatment on dopaminergic cell population activity were evaluated with the paired *t*-test.

3. Results

Mean group firing rates ranged from 3.8 to 4.8 spikes/s, and there were no significant treatment effects on this measure. The cells per track results from electrophysiological recordings in A9 and A10 are summarized in Figs. 1 and 2, respectively. In control rats treated daily with water, the mean ($\pm$S.E.M.) number of spontaneously active A9 dopaminergic neurons was 1.12 ± 0.08 ($N = 9$). In A10, the mean value was 1.11 ± 0.06 ($N = 9$). In comparison to water-treated controls, 21-day treatment with 0.5 mg/kg haloperidol significantly reduced the number of spontaneously active A9 (0.52 ± 0.05) and A10 (0.48 ± 0.07) neurons ($P < 0.05$, Newman–Keuls, $N = 8$). Chronic clozapine resulted in selective reduction in A10 dopaminergic cells per track ($P < 0.05$, $N = 5$). Immediately after these recordings were performed, each rat was injected i.v. with saline or glucose, and then the numbers of A9 and A10 cells per track were determined in the contralateral midbrain.

Glucose significantly reduced the number of spontaneously active A9 and A10 neurons in chronic water-treated rats (post-glucose: A9, 0.65 ± 0.07, $P < 0.05$; A10, 0.67 ± 0.04, $P < 0.05$; compared to pre-glucose, paired *t*-test, $N = 9$). In contrast, acute glucose significantly attenuated the reduction in the number of spontaneously active A9 and A10 dopaminergic neurons in haloperidol-treated rats ($P < 0.05$, $N = 8$), and significantly attenuated the reduc-

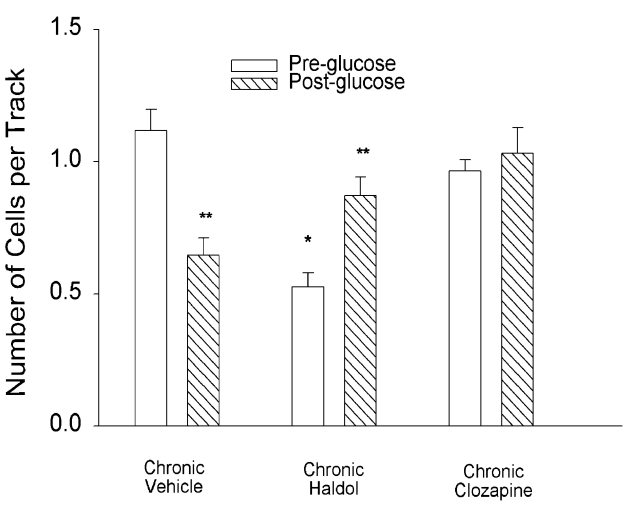

Fig. 1. Acute glucose administration (250 mg/kg, i.v.) reduced the number of spontaneously active A9 dopaminergic cells per track in 21-day vehicle-treated rats ($N = 9$). Repeated (21-day) haloperidol treatment (Haldol, 0.5 mg/kg/day, s.c.) significantly reduced A9 dopaminergic cells per track ($N = 8$ rats), whereas 21-day clozapine treatment (20 mg/kg/day, s.c., $N = 5$) had no effect compared to controls. Acute glucose injection significantly attenuated the effects of chronic haloperidol. Data are mean $\pm$ S.E.M. $^{*}P < 0.01$, chronic treatment vs. chronic vehicle (ANOVA, post hoc Newman–Keuls test) and $^{**}P < 0.05$ paired *t*-test (pre- and post-glucose).

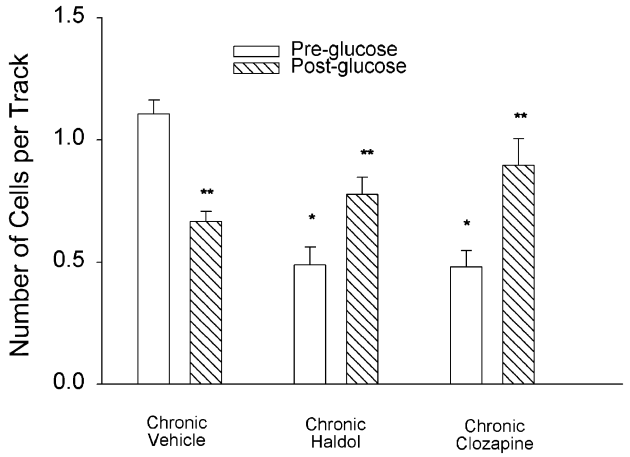

Fig. 2. Acute glucose administration (250 mg/kg, i.v.) reduced the number of spontaneously active A10 dopaminergic cells per track in 21-day vehicle-treated rats ($N = 9$). Repeated (21-day) haloperidol treatment (Haldol, 0.5 mg/kg/day, s.c.) significantly reduced A9 and A10 dopaminergic cells per track ($N = 8$ rats), whereas 21-day clozapine treatment (20 mg/kg/day, s.c.) selectively reduced A10 dopaminergic cells per track. Acute glucose injection significantly attenuated the inhibitory effects of chronic haloperidol and clozapine. Data are mean ± S.E.M. $^{*}P < 0.05$, chronic treatment vs. chronic vehicle (ANOVA, post hoc Newman–Keuls test) and $^{**}P < 0.05$ paired *t*-test (pre- and post-glucose).

tion in the number of spontaneously active A10 dopaminergic neurons in clozapine-treated rats ($P < 0.05$, $N = 5$).

4. Discussion

Chronic administration of typical antipsychotic drugs reduces the number of spontaneously active A9 and A10 dopaminergic neurons in anesthetized rats (Bunney and Grace, 1978; Chiodo and Bunney, 1983; White and Wang, 1983a; Skarsfeldt, 1992). These reductions have been attributed to induction of depolarization inactivation of the dopaminergic neuron plasma membrane, which may develop in response to reduced long-loop feedback inhibition emanating from terminal fields of dopaminergic neurons (Grace and Bunney, 1986). In this condition of maintained depolarization, the membrane is unable to repolarize, the spike-generating mechanism of the cell is unable to operate, and the cell is unable to spontaneously fire. It has been suggested by one group that antipsychotic drug-induced depolarization block in this model, although of preclinical utility, is an artifact of anesthesia (Melis et al., 1998). Grace et al. (1997) have presented a comprehensive review of the attributes of this model.

Peripheral administration of low doses of dopamine receptor agonists normally inhibits the firing rate of dopaminergic neurons. This effect is due to stimulation of impulse-regulating somatodendritic dopamine autoreceptors, which are linked to the opening of potassium channels. Thus, the inhibitory effect of autoreceptor stimulation is associated with hyperpolarization of the dopaminergic cell membrane. Low doses of dopamine receptor agonists, however, reverse antipsychotic drug-induced depolarization inactivation of dopaminergic neurons (Bunney and Grace, 1978). This paradoxical effect of dopamine autoreceptor stimulation is due to repolarization of the cell membrane with resultant reactivation of the spike-generating mechanism (Grace and Bunney, 1986).

Glucose potentiates acute haloperidol-induced catalepsy and inhibits the firing rate of rat midbrain dopaminergic neurons (Saller and Chiodo, 1980; Saller and Kopin, 1981). The dose-dependent duration of action of glucose on firing rate is related to the duration of blood glucose elevation: 250 mg/kg glucose profoundly inhibits firing rate for at least 30 min, and was therefore suitable for use in the current study (Saller and Chiodo, 1980). In preliminary i.v. dose–response experiments, we confirmed that this dose of glucose inhibits the firing rate of dopaminergic neurons (data not shown). This inhibitory effect may be related to the reduction in the number of dopaminergic cells per track observed in control (chronic water-treated) rats after acute glucose injection in the current experiments. Acute administration of glucose, however, significantly attenuated chronic haloperidol- and clozapine-induced reductions in the numbers of dopaminergic cells per track. Analogous to the effects of dopamine receptor agonists, the acute inhibitory effects of glucose on firing rate may underlie its ability to reverse the effects of chronic haloperidol and clozapine on the numbers of dopaminergic cells per track. The mechanism of glucose effects on rat dopaminergic neuron electrophysiology remains to be elucidated, but includes effects on an ATP-sensitive potassium channel (Levin, 2000; Marinelli et al., 2000). In contrast, cat nigral dopaminergic neurons are not responsive to glucose administration (Strecker et al., 1983).

Chronic treatment of patients with antipsychotic drugs is associated with significant weight gain (Baptista, 1999; Ganguli, 1999). Obesity in drug-free patients is associated with a reduction in striatal dopamine D_2 receptors compared to control subjects (Wang et al., 2001). A reduction in dopaminergic neurotransmission may occur in both of these situations. The current results suggest that food intake may increase the number of spontaneously firing dopaminergic neurons during chronic antipsychotic drug treatment. Because fluctuations in glucose availability may influence the activity of dopaminergic neurons (Saller and Chiodo, 1980), perhaps excessive caloric intake is in part a behavioral response to antipsychotic drug-induced reduction in the number of spontaneously active dopaminergic neurons. Following acute pharmacological reversal of antipsychotic drug-induced depolarization block in rats, dopaminergic neurons do not always return immediately to the depolarized, inactive state (Grace and Bunney, 1986). This raises the possibility that the frequency and quantity of caloric intake is linked to changes in population activity of dopaminergic neurons. Additional studies are required

to understand the time-course and dose-responsiveness of the interaction between caloric intake and antipsychotic drug effects on dopaminergic neurons.

Acknowledgements

The authors thank Dr. L.A. Chiodo for helpful comments during this study. This work was supported in part by Texas Tech University Health Sciences Center.

References

Baptista, T., 1999. Body weight gain induced by antipsychotic drugs: mechanisms and management. Acta Psychiatr. Scand. 100, 3–16.

Baptista, T., Parada, M., Hernandez, L., 1987. Long term administration of some antipsychotic drugs increases body weight and feeding in rats. Are D2 dopamine receptors involved? Pharmacol. Biochem. Behav. 27, 399–405.

Bunney, B.S., Grace, A.A., 1978. Acute and chronic haloperidol treatment: comparison of effects on nigral dopaminergic cell activity. Life Sci. 23, 1715–1728.

Bunney, B.S., Walters, J.R., Roth, R.H., Aghajanian, G.K., 1973. Dopaminergic neurons: effect of antipsychotic drugs and amphetamine on single cell activity. J. Pharmacol. Exp. Ther. 195, 560–571.

Chiodo, L.A., Bunney, B.S., 1983. Typical and atypical neuroleptics: differential effects of chronic administration on the activity of A9 and A10 midbrain dopaminergic neurons. J. Neurosci. 3, 1607–1619.

Ganguli, R., 1999. Weight gain associated with antipsychotic drugs. J. Clin. Psychiatry 60 (Suppl. 21), 20–24.

Grace, A.A., Bunney, B.S., 1986. Induction of depolarization block in midbrain dopamine neurons by repeated administration of haloperidol: analysis using in vivo intracellular recording. J. Pharmacol. Exp. Ther. 238, 1092–1100.

Grace, A.A., Bunney, B.S., Moore, H., Todd, C.L., 1997. Dopamine-cell depolarization block as a model for the therapeutic actions of antipsychotic drugs. Trends Neurosci. 20, 31–37.

Lane, R.F., Blaha, C.D., 1987. Chronic haloperidol decreases dopamine release in striatum and nucleus accumbens in vivo: depolarization block as a possible mechanism of action. Brain Res. Bull. 18, 135–138.

Levin, B.E., 2000. Glucose-regulated dopamine release from substantia nigra neurons. Brain Res. 874, 158–164.

Marinelli, S., Bernardi, G., Giacomini, P., Mercuri, N.B., 2000. Pharmacological identification of the K(+) currents mediating the hypoglycemic hyperpolarization of rat midbrain dopaminergic neurones. Neuropharmacology 39, 1021–1028.

Melis, M., Mereu, G., Lilliu, V., Quartu, M., Diana, M., Gessa, G.L., 1998. Haloperidol does not produce dopamine cell depolarization-block in paralyzed, unanesthetized rats. Brain Res. 783, 127–132.

Paxinos, G., Watson, C., 1986. The Rat Brain in Stereotaxic Coordinates. 2nd edn. Academic Press, New York, NY.

Saller, C.F., Chiodo, L.A., 1980. Glucose suppresses basal firing and haloperidol-induced increases in the firing rate of central dopaminergic neurons. Science 210, 1269–1271.

Saller, C.F., Kopin, I.J., 1981. Glucose potentiates haloperidol-induced catalepsy. Life Sci. 29, 2337–2341.

Skarsfeldt, T., 1992. Electrophysiological profile of the new atypical neuroleptic, sertindole, on midbrain dopamine neurones in rats: acute and repeated treatment. Synapse 10, 25–33.

Strecker, R.E., Steinfels, G.F., Jacobs, B.L., 1983. Dopaminergic unit activity in freely moving cats: lack of relationship to feeding, satiety, and glucose injections. Brain Res. 260, 317–321.

Wang, G.-J., Volkow, N.D., Logan, J., Pappas, N.R., Wong, C.T., Zhu, W., Netusil, N., Fowler, J.S., 2001. Brain dopamine and obesity. Lancet 357, 354–357.

White, F.J., Wang, R.Y., 1983a. Differential effects of classical and atypical antipsychotic drugs on A9 and A10 dopamine neurons. Science 221, 1054–1057.

White, F.J., Wang, R.Y., 1983b. Comparison of the effects of chronic haloperidol treatment on A9 and A10 dopamine neurons in the rat. Life Sci. 32, 983–993.