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Histamine H_3 receptor-mediated inhibition of depolarization-induced, dopamine D_1 receptor-dependent release of [^{3}H]-γ-aminobutyric acid from rat striatal slices

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1 A study was made of the regulation of [^{3}H]-γ-aminobutyric acid ([^{3}H]-GABA) release from slices of rat striatum by endogenous dopamine and exogenous histamine and a histamine H_3-agonist. Depolarization-induced release of [^{3}H]-GABA was Ca^{2+}-dependent and was increased in the presence of the dopamine D_2 receptor family antagonist, sulpiride (10 μM). The sulpiride-potentiated release of [^{3}H]-GABA was strongly inhibited by the dopamine D_1 receptor family antagonist, SCH 23390 (1 μM). Neither antagonist altered basal release.

2 The 15 mM K^+-induced release of [^{3}H]-GABA in the presence of sulpiride was inhibited by 100 μM histamine (mean inhibition $78 \pm 3\%$) and by the histamine H_3 receptor-selective agonist, immepip, 1 μM (mean inhibition $81 \pm 5\%$). The IC_{50} values for histamine and immepip were 1.3 ± 0.2 μM and 16 ± 2 nM, respectively. The inhibitory effects of histamine and immepip were reversed by the H_3 receptor antagonist, thioperamide, 1 μM.

3 The inhibition of 15 mM K^+-induced [^{3}H]-GABA release by immepip was reversed by the H_3 receptor antagonist, clobenpropit, K_d 0.11 ± 0.04 nM. Clobenpropit alone had no effect on basal or stimulated release of [^{3}H]-GABA.

4 Elevated K^+ caused little release of [^{3}H]-GABA from striatal slices from reserpinized rats, unless the D_1 partial agonist, R(+)-SKF 38393, 1 μM, was also present. The stimulated release in the presence of SKF 38393 was reduced by 1 μM immepip to the level obtained in the absence of SKF 38393.

5 These observations demonstrate that histamine H_3 receptor activation strongly inhibits the dopamine D_1 receptor-dependent release of [^{3}H]-GABA from rat striatum; primarily through an interaction at the terminals of GABA neurones.

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Keywords: GABA release; rat striatum; dopamine D_1 receptors; dopamine D_2 receptors; histamine H_3 receptors; immepip; clobenpropit; thioperamide; SKF 38393; reserpine

Abbreviations: [^{3}H]-GABA, [^{3}H]-γ-aminobutyric acid; SNr, substantia nigra pars reticulata

Introduction

We have reported previously that activation of histamine H_3 receptors located on the terminals of striatonigral projection neurones in rat substantia nigra pars reticulata (SNr) selectively inhibits the component of depolarization-induced release of [^{3}H]-γ-aminobutyric acid ([^{3}H]-GABA) which is dependent on concomitant dopamine D_1 receptor stimulation (Garcia *et al*., 1997). The striatonigral projection neurones have axon collaterals which remain within the striatum (Kawaguchi *et al*., 1990; reviewed in Gerfen & Wilson, 1996) and the release of striatal GABA is subject to the same interplay between D_1 and D_2 receptors as in SNr; D_1 agonists and D_2 antagonists both causing an increase in GABA release (Girault *et al*., 1986; Floran *et al*., 1990; Harsing & Zigmond, 1997). The striatum is also rich in histamine H_3 receptors (Arrang *et al*., 1987a; Cumming *et al*., 1991b; Pollard *et al*., 1993; Ligneau *et al*., 1994; Jansen *et al*., 1994) and striatal quinolinic acid lesions result in a parallel decrease in the numbers of ipsilateral dopamine D_1 and histamine H_3 receptors, both in SNr and striatum (Ryu *et al*., 1994). These observations suggest that D_1 and H_3 receptors are colocalized on the same terminals in the striatum, as in SNr (Garcia *et al*., 1997), and, hence, that depolarization-induced, D_1 receptor-dependent release of [^{3}H]-GABA in striatum may be regulated by H_3 receptor activation in the same way as in SNr. We report here a study of the effects of ligands acting at dopamine D_1 and D_2 and histamine H_3 receptors on depolarization-induced release of [^{3}H]-GABA from slices of rat striatum. A preliminary account of some of these results has been presented to the British Pharmacological Society (Arias-Montaño *et al*., 2000).

Methods

Measurement of [^{3}H]-GABA release from slices of rat striatum

The striatum was dissected from vibratome-cut slices (300 μm) of rat brain (Wistar strain, males, 250–300 g,

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bred in the Centro de Investigacion), cut into smaller pieces, and incubated for 30 min at 37°C in 4.5 ml of a modified Krebs-Henseleit solution (composition in mM: NaCl 134, KCl 4.75, $MgSO_4$ 1, KH_2PO_4 1.25, $NaHCO_3$ 25, $CaCl_2$ 2 and D-glucose 10) gassed continuously with O_2/CO_2 (95 : 5, v v^{-1}). The slices were then incubated for 30 min with 80 nM [^{3}H]-GABA in 4.5 ml Krebs-Henseleit solution containing 10 μM aminooxyacetic acid. At the end of this period, excess radiolabel was removed by washing twice with Krebs-Henseleit solution containing 10 μM aminooxyacetic acid and 10 μM nipecotic acid, which were present in the superfusion solution for the rest of the experiment. The slices were then apportioned randomly between the chambers of a superfusion apparatus (volume of each chamber 80 μl; 20 chambers in parallel) and superfused with the medium at a rate of 0.5 ml min^{-1} for 30 min. The design of the superfusion chambers was essentially as described by Aceves & Cuello (1981), except that the electrodes for electrical stimulation were omitted. Basal release of [^{3}H]-GABA was measured by collecting four or five fractions of the superfusate at 4 min intervals (each fraction 2 ml) before release was stimulated by changing to a solution containing 15 mM K^+ (composition in mM: NaCl 55.6, Na_2SO_4 39.2; K_2SO_4 6.87, $MgSO_4$ 1, KH_2PO_4 1.25, $NaHCO_3$ 25, $CaCl_2$ 2 and D-glucose 10: Floran *et al*., 1988) and a further six fractions collected. In experiments in which sulpiride (10 μM) or SCH 23390 (1 μM) was present in every incubation, they were added 4 min before the first basal fraction was collected.

Histamine, immepip, clobenpropit and thioperamide were present from 12 min before the change to the medium containing 15 mM K^+. The superfusate fractions were mixed with 10 ml scintillator (4 g 2,5-diphenyloxazole + 0.2 g, 1,4-bis-2-(5-phenyloxazolyl)-benzene in 1 l toluene/Triton X-100, 2 : 1, v v^{-1}) and the tritium content determined by scintillation counting. It has been shown that >90% of the tritium released by a depolarizing stimulus from rat striatum is [^{3}H]-GABA (Kuriyama *et al*., 1984; Harsing & Zigmond, 1997). To determine the total amount of tritium remaining in the tissue, the contents of each chamber were collected, treated with 1 ml 1 M HCl and allowed to stand for 1 h before addition of scintillator.

Pretreatment of animals with reserpine

Rats were pretreated with reserpine (5 mg kg^{-1}, i.p.) 24 h before preparation of striatal slices. Control animals were treated with the same volume (1 ml kg^{-1}) of vehicle (7% w v^{-1} lactic acid). The reserpine treatment has previously been shown to reduce striatal dopamine levels by 95% (Garcia *et al*., 1997).

Analysis of data

[^{3}H]-GABA release was expressed initially as a fraction of the total amount of tritium remaining in the tissue. Basal fractional release per 2 ml superfusate fraction varied quite widely between chambers, but was normally in the range 0.003 – 0.020 (released tritium usually 1000 – 7000 d.p.m.), although occasional values were outside this range. The within-treatments variability in an experiment was greatly reduced by expressing the amount of tritium in each fraction as a percentage of the amount of tritium present in the fraction collected immediately before the change to the medium containing 15 mM K^+ (i.e. the release in fraction 4 or 5 was set to 100%). In most experiments, 5 – 6 replicate determinations were made at each drug concentration or drug combination tested.

The effect of drugs on the basal release of [^{3}H]-GABA was assessed by comparing the fractional release in fraction 2 or 3 (immediately before exposure of the tissue to drug, e.g. sulpiride) and fraction 4 or 5 (immediately prior to exposure to 15 mM K^+), using the paired *t*-test.

A measure of the degree of inhibition of the release of [^{3}H]-GABA was obtained by comparing the areas under the appropriate release curves between the first and last fractions collected after the change to high K^+, making the assumption that the basal release of [^{3}H]-GABA would have remained unchanged at the level measured in the fraction immediately preceding K^+ stimulation (set to unity in the normalization procedure above). In three experiments in which this was tested the basal release in fraction 10 was 90 ± 4% of that in fraction 5.

To obtain an unbiased estimate of IC_{50} values, concentration-response data for the inhibition of [^{3}H]-GABA release by histamine and immepip, and for the reversal by clobenpropit of the inhibitory action of immepip, were fitted by non-linear regression to an hyperbola. The actual equation fitted was:

$$\text{Response} = \text{Resp}_{max}C/(C + IC_{50}) \quad (1)$$

where $Resp_{max}$ is the maximum response (maximum per cent inhibition or per cent of control [^{3}H]-GABA release), C is the concentration of histamine, immepip or clobenpropit and IC_{50} is the concentration giving the half maximal response (inhibition or reversal of inhibition). The K_d for clobenpropit in reversing the inhibition by immepip was calculated from the curves for immepip and clobenpropit using the method of Lazareno & Roberts (1987); Dickenson & Hill (1993).

To test for statistical differences between treatments, the area under the release curve in the presence of elevated K^+ was calculated for each individual chamber and the data then analysed as described previously (Garcia *et al*., 1997).

Chemicals

[2,3-^{3}H]-γ-Aminobutyric acid ([^{3}H]-GABA), specific activity 82 Ci.$mmol^{-1}$, was obtained from Amersham Pharmacia Biotech. Aminooxyacetic acid hemihydrochloride, 2,5-diphenyloxazole and (±)-nipecotic acid were purchased from Sigma; histamine dihydrochloride, R(+)-SCH 23390 hydrochloride (R(+)-7-chloro-8-hydroxy-3-methyl-1-phenyl-2,3,4,5-tetrahydro-1H-3-benzazepine hydrochloride), R(+)-SKF 38393 hydrochloride ((±)-1-phenyl-2,3,4,5-tetrahydro-(1H)-3-benzazepine-7,8-diol hydrochloride), (±)-sulpiride and thioperamide maleate from Research Biochemicals International; and 1,4-bis-2-(5-phenyloxazolyl)-benzene from Packard. Clobenpropit dihydrobromide and immepip dihydrobromide were kind gifts from Prof H. Timmerman, Vrije Universiteit, Amsterdam.

Stock solutions of sulpiride (1 mM) were made in 100 μM ascorbic acid.

Results

Effect of D_2 and D_1 dopamine receptor blockade and of omission of Ca^{2+} from the medium on [^{3}H]-GABA release

In the absence of the dopamine D_2 receptor antagonist, sulpiride, increasing the K^+ concentration in the superfusion medium from 6–15 mM caused only a small increase in the release of [^{3}H]-GABA from striatal slices (Figure 1). However, in the presence of 10 μM sulpiride the release was markedly enhanced (mean stimulation 2.9±0.2 fold of basal, $n=3$; Figure 1) and was well maintained over the period for which samples were collected. In contrast, sulpiride had no significant effect on basal [^{3}H]-GABA release (Figure 1). Sulpiride (10 μM) was therefore included in all subsequent experiments, except where specifically indicated.

The depolarization-induced release of [^{3}H]-GABA in the presence of sulpiride was highly dependent on concomitant D_1 receptor activation, since it was strongly inhibited by the dopamine D_1 receptor antagonist, SCH 23390 (1 μM) (Figure 1); mean inhibition 84±6%, $n=3$. SCH 23390 had no significant effect on basal [^{3}H]-GABA release (Figure 1).

The depolarization-induced release of [^{3}H]-GABA was strongly Ca^{2+}-dependent, since omission of Ca^{2+} from the superfusion medium and increasing Mg^{2+} from 1–3 mM reduced depolarization-induced [^{3}H]-GABA release to a low level; mean inhibition 84±6%, $n=3$ (data not shown).

Effect of histamine and immepip on [^{3}H]-GABA release

Depolarization-induced [^{3}H]-GABA release was strongly inhibited by 100 μM histamine (Figure 2A) and by the selective histamine H_3 receptor agonist, immepip (1 μM) (Figure 2B). The mean inhibitions in this series of experiments were 78±3% and 81±5% for histamine and immepip, respectively, both $n=3$. Neither 100 μM histamine nor 1 μM immepip had any significant effect on basal [^{3}H]-GABA release.

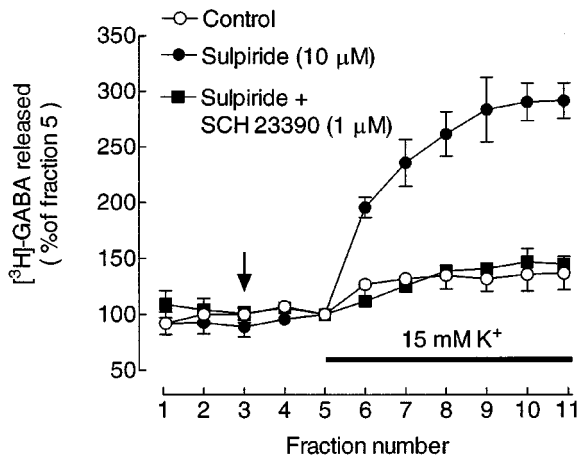

Figure 1 Modulation of depolarization-induced [^{3}H]-GABA release by dopamine receptor antagonists. Values are expressed as a percentage of the fractional release of [^{3}H]-GABA in fraction 5 and represent the means±s.e.mean from three experiments. Drugs were added at the vertical arrow and the K^+ in the medium increased for the period indicated by the horizontal bar.

The inhibitory effect of the agonists on depolarization-induced [^{3}H]-GABA release was blocked by the H_3 receptor antagonist, thioperamide (1 μM) (Figure 2A,B). In the presence of histamine/immepip + thioperamide, the extent of the release was not significantly different from control.

The inhibitory actions of histamine and immepip on depolarisation-induced [^{3}H]-GABA release were concentration-dependent (Figure 3). The value for 300 μM histamine is from a single experiment, since in two other experiments this concentration of histamine caused a statistically significant release of [^{3}H]-GABA in normal K^+ medium. The best-fit values of log IC_{50}±estimated s.e.mean for histamine and immepip were 3.10±0.12 and 1.19±0.15, respectively, (concentrations expressed as nM) (IC_{50} 1.3±0.2 μM for histamine and 15.5±2.3 nM for immepip).

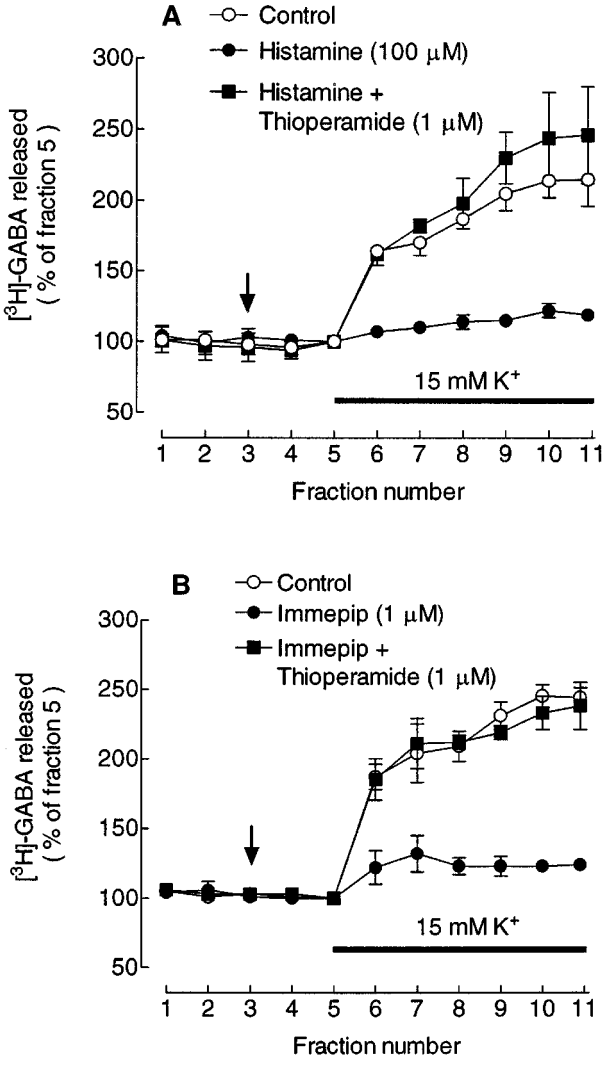

Figure 2 Inhibition of depolarisation-induced [^{3}H]-GABA release by histamine and immepip and reversal by thioperamide. (A) Effect of 100 μM histamine in the absence and presence of 1 μM thioperamide. (B) Effect of 1 μM immepip in the absence and presence of 1 μM thioperamide. In both panels values are expressed as a percentage of the fractional release of [^{3}H]-GABA in fraction 5 and represent the means±s.e.mean from three experiments. Drugs were added at the vertical arrow and the K^+ in the medium increased for the period indicated by the horizontal bar. Sulpiride (10 μM) was present throughout.

The high degree to which the release of [^{3}H]-GABA is inhibited by SCH 23390 makes it difficult to test whether the inhibitory action of H_3 agonists is selective for the D_1 receptor-dependent component of release. In three experiments in which comparison was made, the extent of the inhibition of depolarization-induced [^{3}H]-GABA release in the presence of 1 μM SCH 23390, 84$\pm$6%, was not significantly different from that in the presence of 1 μM SCH 23390 + 1 μM immepip, 88$\pm$9% inhibition.

Reversal of the inhibitory action of immepip by clobenpropit

The potent effect of immepip and its reversal by thioperamide strongly suggests that the inhibition is mediated by histamine H_3 receptors. To gain more quantitative evidence, we have investigated the concentration-dependence of the effect of the selective H_3 receptor antagonist, clobenpropit. Acting alone, 1 μM clobenpropit had no significant effect on either basal or depolarization-induced release of [^{3}H]-GABA (Figure 4A). However, clobenpropit reversed in a concentration-dependent manner the inhibition of depolarization-induced [^{3}H]-GABA release by 1 μM immepip (Figure 4B). There was a marked variability in some of the experiments in this series, which is reflected in the large estimated error associated with the best-fit value of the IC_{50}, 7.3$\pm$2.1 nM. The calculated K_d, for clobenpropit was 0.11$\pm$0.04 nM.

Effect of SKF 38393 and immepip on depolarization-induced [^{3}H]-GABA release in striatum from reserpinized rats

The evidence indicates that H_3 receptor activation inhibits dopamine-dependent release of [^{3}H]-GABA in rat striatum, but does not indicate whether the action is at the level of dopamine release or is a direct effect at GABA terminals. To establish whether there is an interaction at GABA terminals, measurements were made on depolarization-induced [^{3}H]-GABA released from striatal slices from animals treated with reserpine 24 h previously, which reduces striatal dopamine to very low levels (Garcia *et al*., 1997). Sulpiride was also omitted from the superfusion medium in these experiments.

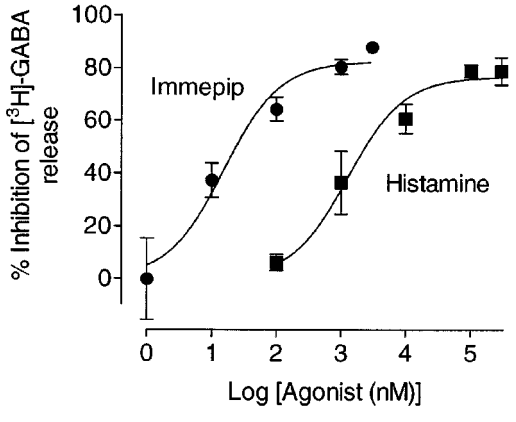

Figure 3 Concentration-dependence of the inhibition by histamine and immepip of depolarisation-induced [^{3}H]-GABA release. Values are means$\pm$s.e.mean from 3–6 independent determinations at each concentration, except for 300 μM histamine, which is from a single experiment (see text). The curves drawn are best-fit lines to an hyperbola (see Methods).

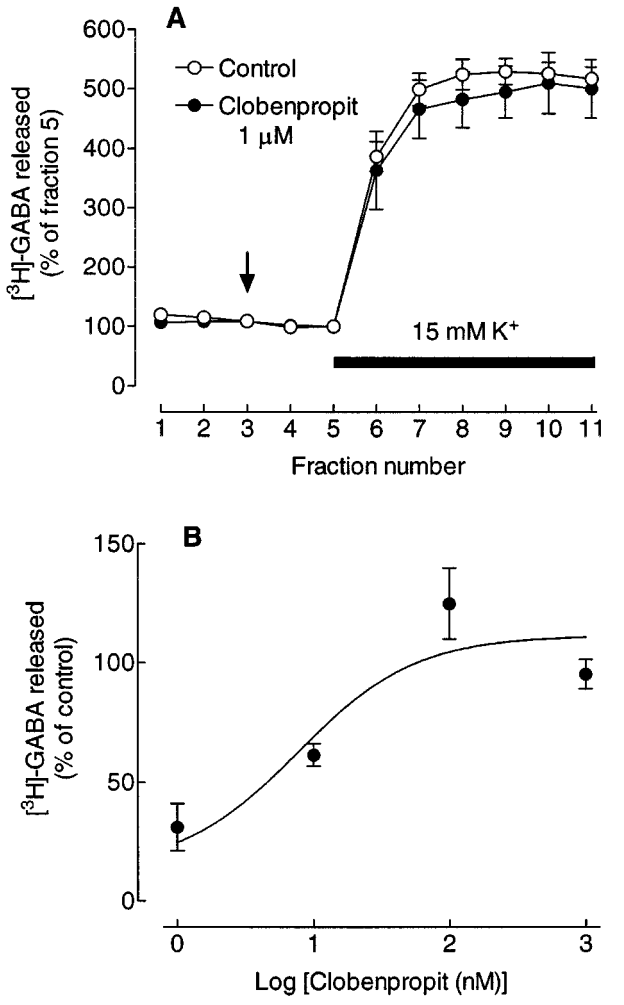

Figure 4 Effect of clobenpropit on [^{3}H]-GABA release. (A) Action of clobenpropit alone. Values are expressed as a percentage of the fractional release of [^{3}H]-GABA in fraction 5 and represent the means$\pm$s.e.mean from three experiments. Clobenpropit was added at the vertical arrow and the K^+ in the medium increased for the period indicated by the horizontal bar. (B) Concentration-dependence of the reversal by clobenpropit of the inhibition of [^{3}H]-GABA release by immepip. Immepip (1 μM) was present in all incubations with clobenpropit. Values are the per cent of control release of [^{3}H]-GABA, calculated from the relative areas under the curves (see Methods) and are the means$\pm$s.e.mean from 3–5 determinations. The curve drawn is the best-fit line to an hyperbola (see Methods). The foot of the curves has been fixed at 13% (mean inhibition by 1 μM immepip in this series of experiments 87.0$\pm$2.5%). Sulpiride (10 μM) was present throughout in (A) and (B).

In the absence of added drugs, depolarization with raised K^+ had a minimal effect on the release of [^{3}H]-GABA (Figure 5), consistent with the absence of significant dopamine release from the reserpinized slices. However, in the presence of the dopamine D_1 receptor agonist, R(+)-SKF 38393, 1 μM, the depolarization-induced release was markedly stimulated (Figure 5). Addition of 1 μM immepip reduced the depolarization-induced release in the presence of SKF 38393 to control levels (Figure 5), indicating that histamine H_3 and dopamine D_1 receptors are probably

colocalized on the GABA terminals. Immepip alone was without effect on release (Figure 5).

Discussion

It is clear that histamine inhibits dopamine-dependent [^{3}H]-GABA release from rat striatal slices in a manner similar to that reported using slices of rat SNr (Garcia *et al*., 1997). However, the larger amount of striatal tissue has made it possible to provide much stronger evidence that the inhibition is mediated by histamine H_3 receptors. The estimated K_d of 0.11 ± 0.04 nM for the H_3 receptor antagonist clobenpropit is in accord with literature values of 0.13 nM (Leurs *et al*., 1995a), 0.03 nM (Harper *et al*., 1999) and 0.08 nM (Valentine *et al*., 1999) and the potency of immepip as an inhibitor of dopamine-dependent [^{3}H]-GABA release (IC_{50} 16 ± 2 nM) is similar to that reported for inhibition of electrically-evoked twitches of guinea-pig jejunum (IC_{50} 10 nM; Leurs *et al*., 1995a). In addition, the effect of histamine and immepip is fully reversed by 1 μM thioperamide, although thioperamide may be a less selective H_3 antagonist than clobenpropit (Leurs *et al*., 1995b). There is no evidence for the involvement of either histamine H_1 or H_2 receptors in the inhibition of [^{3}H]-GABA release, since the inhibition is completely reversed by clobenpropit and thioperamide and the extent of the inhibition by histamine is the same, within error, as that produced by immepip.

Rat striatum has a low to moderate density of histaminergic fibres (Inagaki *et al*., 1988; Panula *et al*., 1989), but there is no indication of any release of endogenous histamine in our slices, since clobenpropit, in the absence of immepip, had no significant effect on either basal or depolarization-stimulated release of [^{3}H]-GABA. The lack of effect on basal release is consistent with the estimate of 50 nM for the lower limit of the extracellular concentration of histamine in rat striatum (Cumming *et al*., 1991a), which is well below the IC_{50} for histamine for inhibition of [^{3}H]-GABA release, 1.3 ± 0.2 μM. Histamine is released from histaminergic fibres on increasing extracellular K^+ (Arrang *et al*., 1983), but apparently not in sufficient amounts from striatal fibres to produce any inhibition of [^{3}H]-GABA release, as indicated by the lack of effect of clobenpropit. This is consistent with a report that no change was detected in the release of endogenous histamine from rat striatum, measured by microdialysis, when the K^+ concentration in the probe was increased to 156 mM (Russell *et al*., 1990).

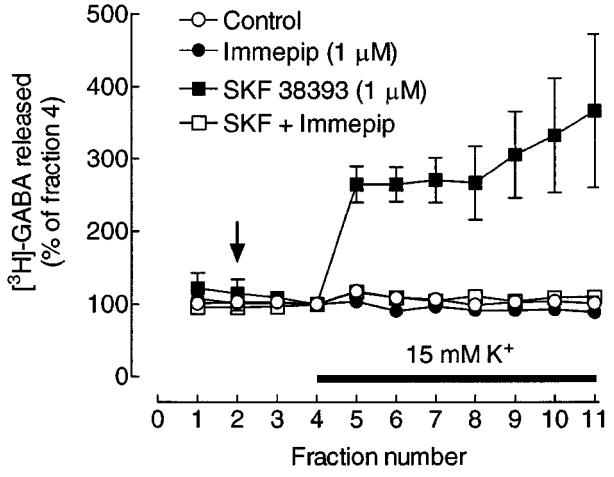

Figure 5 Effect of immepip on depolarization-induced, dopamine D_1 receptor-dependent, release of [^{3}H]-GABA from striatal slices from reserpinised rats. Values are the means $\pm$ s.e.mean from four (control and immepip) or five replicate determinations within a single experiment. Drugs were added at the vertical arrow and and K^+ in the medium increased for the period indicated by the horizontal bar. Similar results were obtained in two further experiments, except that the magnitudes of the release in the presence of SKF 38393 differed (max. 1.9 and 7.3 fold of basal release).

The IC_{50} for histamine for inhibition of [^{3}H]-GABA release is in close agreement with the recently reported IC_{50} for histamine inhibition of corticostriatal transmission, 1.6 μM (Doreulee *et al*., 2001), but is much higher than that reported for histamine-induced inhibition of depolarization-induced release of [^{3}H]-histamine from rat brain slices, 40 nM (Arrang *et al*., 1983; Leurs *et al*., 1995a). However, the IC_{50} for inhibition of [^{3}H]-GABA release is nearer to the IC_{50} values for histamine-induced inhibition of depolarization-induced histamine synthesis in rat cerebral cortex, 0.34 μM (Arrang *et al*., 1987b) and for inhibition by histamine of the electrically-evoked release of [^{3}H]-noradrenaline (Schlicker *et al*., 1992) and [^{3}H]-serotonin (Smits & Mulder, 1991), *circa* 0.1 μM, from brain slices. The IC_{50} for histamine-induced inhibition of [^{3}H]-dopamine release in mouse striatum is not well defined (Schlicker *et al*., 1993), but appears to be of the same order as that we observe for inhibition of [^{3}H]-GABA release. The variation in IC_{50} values between tissues could reflect differences in receptor reserve or a difference in G protein coupling, possibly involving subtypes of the H_3 receptor (reviewed in Hill *et al*., 1997).

It should be noted that the overall inhibitory action of H_3-agonists on [^{3}H]-GABA release in striatum could involve some inhibition of striatal dopamine release, since in mouse striatum histamine and the H_3-agonist R(-)-α-methylhistamine are reported to cause *circa* 30% inhibition of the electrically-stimulated release of [^{3}H]-dopamine (Schlicker *et al*., 1993). H_3 receptors also appear to be present on dopaminergic terminals in rat striatum, since immepip produces a marked inhibition of depolarization-induced dopamine synthesis (Molina-Hernandez *et al*., 2000). However, the almost complete inhibition of dopamine-dependent [^{3}H]-GABA release in the reserpinized animals indicates that the major site of action of H_3 agonists is almost certainly on the terminals of GABA neurones. This is consistent with the reports that striatal quinolinic acid lesions result in a parallel decrease in the numbers of ipsilateral D_1 and H_3 receptors in striatum, as in SNr, (Ryu *et al*., 1994) and that H_3 receptor expression in the striatum is regulated, at least in part, by dopamine D_1 receptors (Ryu *et al*., 1996). The collaterals of the projection neurones would thus seem to be the most likely site of the D_1/H_3 interaction. The effects of H_3 agonists and dopamine on acetylcholine release in the ventral striatum (Prast *et al*., 1999) can also be explained by an interaction on GABA collaterals.

The GABA projection neurones make up over 90% of all the neurones in the striatum (Kawaguchi *et al*., 1995). However, the striatum also possesses at least two classes of GABA interneurone, at least one of which possesses D_1 receptors (Kawaguchi *et al*., 1995), and the possibility must be considered that some of the [^{3}H]-GABA release measured

might be from these interneurones. It may be noted that the pattern of depolarization-induced [^{3}H]-GABA release from the striatal slices (sustained or increasing with time) differs from the pattern observed in SNr (initial peak, then declining release) (Garcia *et al*., 1997). There is a report that [^{3}H]-GABA microinjected into the striatum of anaesthetized rats is taken up preferentially by one type of interneurone (Bolam *et al*., 1983), presumably reflecting a highly active GABA uptake system. This interneurone constitutes only 3–5% of striatal neurones, but has a dense arborization of local axon collaterals, stains strongly for GABA and glutamic acid decarboxylase, and has different electrophysiological properties from those of the projection neurones (Kawaguchi, 1993; Kawaguchi *et al*., 1995). However, the labelling conditions used in the present study, in which slices were exposed to an excess of [^{3}H]-GABA over an extended time, differ considerably from those employing a single microinjection of [^{3}H]-GABA. It should also be noted that there is currently no evidence that any of the classes of GABA interneurones express both D_1 and H_3 receptors and, consequently, that they might be a locus for the H_3/D_1 receptor interaction.

There is at present only limited evidence for the involvement of H_3 receptors in locomotor activity (Clapham & Kilpatrick, 1994), whereas the importance of the permissive role of D_1 receptors in the so-called 'direct' pathway through the basal ganglia is well documented (Gerfen & Wilson, 1996). However, the extent to which the D_1 receptor-dependent release of [^{3}H]-GABA in striatum and SNr is sensitive to inhibition by H_3 agonists is striking and could be important in circumstances in which there is a high local release of histamine, as may occur secondary to ischaemia (Adachi *et al*., 1991).

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References

ACEVES, J. & CUELLO, A.C. (1981). Dopamine release induced by electrical stimulation of microdissected caudate-putamen and substantia nigra. *Neuroscience,* **6,** 2069–2075.

ADACHI, N., OISHI, R. & SAEKI, K. (1991). Changes in the metabolism of histamine and monoamines after occlusion of the middle cerebral artery in rats. *J. Neurochem.,* **57,** 61–66.

ARIAS-MONTAÑO, J.A., FLORAN, B., GARCIA, M., ACEVES, J. & YOUNG, J.M. (2000). Histamine inhibits depolarisation-induced dopamine-dependent release of GABA in rat striatum via an action on H_3-receptors. *Br. J. Pharmacol.,* **129,** 66P.

ARRANG, J.M., GARBARG, M., LANCELOT, J.-C., LECOMTE, J.-M., POLLARD, H., ROBBA, M., SCHUNACK, W. & SCHWARTZ, J.-C. (1987a). Highly potent selective and potent ligands for histamine H_3 receptors. *Nature,* **327,** 117–123.

ARRANG, J.M., GARBARG, M. & SCHWARTZ, J.-C. (1983). Auto-inhibition of brain histamine release mediated by a novel class (H_3) of histamine receptor. *Nature,* **302,** 832–837.

ARRANG, J.M., GARBARG, M. & SCHWARTZ, J.C. (1987b). Autoinhibition of histamine synthesis mediated by presynaptic H_3-receptors. *Neuroscience,* **23,** 149–157.

BOLAM, J.P., CLARKE, D.J., SMITH, A.D. & SOMOGYI, P. (1983). A type of aspiny neuron in the rat neostriatum accumulates [^{3}H]γ-aminobutyric acid: combination of golgi-staining, autoradiography, and electron microscopy. *J. Comp. Neurol.,* **213,** 121–134.

CLAPHAM, J. & KILPATRICK, G.J. (1994). Thioperamide, the selective histamine H_3 receptor antagonist, attenuates stimulant-induced locomotor activity in the mouse. *Eur. J. Pharmacol.,* **259,** 107–114.

CUMMING, P., DAMSMA, G., FIBIGER, H.C. & VINCENT, S.R. (1991a). Characterization of extracellular histamine in the striatum and bed nucleus of the stria terminalis of the rat: an in vivo microdialysis study. *J. Neurochem.,* **56,** 1797–1803.

CUMMING, P., SHAW, C. & VINCENT, S.R. (1991b). High affinity histamine binding site is the H_3 receptor: characterization and autoradiographic localization in rat brain. *Synapse,* **8,** 144–151.

DICKENSON, J.M. & HILL, S.J. (1993). Adenosine A_1-receptor stimulated increases in intracellular calcium in the smooth muscle cell line, DDT_1MF-2. *Br. J. Pharmacol.,* **108,** 85–92.

DOREULEE, N., YANOVSKY, Y., FLAGMEYER, I., STEVENS, D.R., HAAS, H.L. & BROWN, R.E. (2001). Histamine H_3 receptors depress synaptic transmission in the corticostriatal pathway. *Neuropharmacol.,* **40,** 106–113.

FLORAN, B., ACEVES, J., SIERRA, A. & MARTINEZ-FONG, D. (1990). Activation of D_1 dopamine receptors stimulates the release of GABA in the basal ganglia of the rat. *Neurosci. Lett.,* **116,** 136–140.

FLORAN, B., SILVA, I. & ACEVES, J. (1988). Presynaptic modulation of the release of GABA by $GABA_A$ receptors in pars compacta and by $GABA_B$ receptors in pars reticulata of the rat substantia nigra. *Eur. J. Pharmacol.,* **150,** 277–286.

GARCIA, M., FLORAN, B., ARIAS-MONTAÑO, J.A., YOUNG, J.M. & ACEVES, J. (1997). Histamine H_3 receptor activation selectively inhibits dopamine D_1 receptor-dependent [^{3}H]-γ-aminobutyric acid release from depolarisation-stimulated slices of rat substantia nigra pars reticulata. *Neuroscience,* **80,** 241–249.

GERFEN, C.R. & WILSON, C.J. (1996). The basal ganglia. In *Integrated Systems of the CNS, Part III (Handbook of Chemical Neuroanatomy, Vol. 12)*. ed. Björklund, A., Hökfelt, T. & Swanson, L. pp. 369–466. Amsterdam: Elsevier.

GIRAULT, J.A., SPAMPINATO, U., GLOWINSKI, J. & BESSON, M.J. (1986). *In vivo* release of [^{3}H]γ-aminobutyric acid in the rat neostriatum – II. Opposing effects of D_1 and D_2 dopamine receptor stimulation in the dorsal caudate putamen. *Neuroscience,* **19,** 1109–1117.

HARPER, E.A., SHANKLEY, N.P. & BLACK, J.W. (1999). Characterization of the binding of [^{3}H]-clobenpropit to histamine H_3-receptors in guinea-pig cerebral cortex membranes. *Br. J. Pharmacol.,* **128,** 881–890.

HARSING, JR., L.G. & ZIGMOND, M.J. (1997). Influence of dopamine on GABA release in striatum: evidence for D_1-D_2 interactions and non-synaptic influences. *Neuroscience,* **77,** 419–429.

HILL, S.J., GANELLIN, C.R., TIMMERMAN, H., SCHWARTZ, J.C., SHANKLEY, N.P., YOUNG, J.M., SCHUNACK, W., LEVI, R. & HAAS, H.L. (1997). International Union of Pharmacology. XIII. Classification of histamine receptors. *Pharmacol. Rev.,* **49,** 253–278.

INAGAKI, N., YAMATODANI, A., ANDO-YAMAMOTO, M., TOHYAMA, M., WATANABE, T. & WADA, H. (1988). Organization of histaminergic fibres in the rat brain. *J. Comp. Neurol.,* **273,** 283–300.

JANSEN, F.P., WU, T.S., VOSS, H.-P., STEINBUSCH, H.W.M., VOLLINGA, R.C., RADEMAKER, B., BAST, A. & TIMMERMAN, H. (1994). Characterization of the binding of the first selective radiolabelled histamine H_3-receptor antagonist, [^{3}H]-iodophenpropit, to rat brain. *Br. J. Pharmacol.,* **113,** 355–362.

KAWAGUCHI, Y. (1993). Physiological, morphological, and histochemical characterization of three classes of interneurone in rat neostriatum. *J. Neurosci.,* **13,** 4908–4923.

KAWAGUCHI, Y., WILSON, C.J. & EMSON, P.C. (1990). Projection subtypes of rat neostriatal matrix cells revealed by intracellular injection of biocytin. *J. Neurosci.,* **10,** 3421–3428.

KAWAGUCHI, Y., WILSON, C.J., AUGOOD, S.J. & EMSON, P.C. (1995). Striatal interneurones: chemical, physiological and morphological characterization. *Trends Neurosci.,* **18,** 527 – 535.

KURIYAMA, K., KANMORI, K., TAGUCHI, J.-I. & YONEDA, Y. (1984). Stress-induced enhancement of suppression of [^{3}H]GABA release from striatal slices by presynaptic autoreceptor. *J. Neurochem.,* **42,** 943 – 950.

LAZARENO, S. & ROBERTS, F.F. (1987). Measuring muscarinic antagonist potency using stimulated phosphoinositide breakdown in rat cortex slices. *Br. J. Pharmacol.,* **92,** 677P.

LEURS, R., SMIT, M.J. & TIMMERMAN, H. (1995a). Molecular pharmacological aspects of histamine receptors. *Pharmacol. Ther.,* **66,** 413 – 463.

LEURS, R., TULP, M.T.M., MENGE, W.M.B.P., ADOLFS, M.J.P., ZUIDERVELD, O.P. & TIMMERMAN, H. (1995b). Evaluation of the receptor selectivity of the H_3 receptor antagonists, iodophenpropit and thioperamide: an interaction with the 5-HT_3 receptor revealed. *Br. J. Pharmacol.,* **116,** 2315 – 2321.

LIGNEAU, X., GARBARG, M., VIZUETE, M.L., DÍAZ, J., PURAND, K., STARK, H., SCHUNACK, W. & SCHWARTZ, J.-C. (1994). [^{125}I]Iodoproxyfan, a new antagonist to label and visualize cerebral histamine H_3 receptors. *J. Pharmacol Exp. Ther.,* **271,** 452 – 459.

MOLINA-HERNANDEZ, A., NUÑEZ, A. & ARIAS-MONTAÑO, J.-A. (2000). Histamine H_3-receptor activation inhibits dopamine synthesis in rat striatum. *NeuroReport,* **11,** 163 – 166.

PANULA, P., PIRVOLA, U., AUVINEN, S. & AIRAKSINEN, M.S. (1989). Histamine-immunoreactive nerve fibres in the rat brain. *Neuroscience,* **28,** 585 – 610.

POLLARD, H., MOREAU, J., ARRANG, J.M. & SCHWARTZ, J.-C. (1993). A Detailed autoradiographic mapping of histamine H_3 receptors in rat brain areas. *Neuroscience,* **52,** 169 – 189.

PRAST, H., TRAN, M.H., FISCHER, H., KRAUS, M., LAMBERTI, C., GRASS, K. & PHILIPPU, A. (1999). Histaminergic neurones modulate acetylcholine release in the ventral striatum: role of H_3 histamine receptors. *Naunyn-Schmiedeberg's Arch. Pharmacol.,* **360,** 558 – 564.

RUSSELL, W.L., HENRY, D.P., PHEBUS, L.A. & CLEMENS, J.A. (1990). Release of histamine in rat hypothalamus and corpus striatum in vivo. *Brain Res.,* **512,** 95 – 101.

RYU, J.H., YANAI, K., IWATA, R., IDO, T. & WATANABE, T. (1994). Heterogenous distribution of histamine H3, dopamine D1 and D2 receptors in rat brain. *NeuroReport,* **5,** 621 – 624.

RYU, J.H., YANAI, K., ZHAO, X.-L. & WATANABE, T. (1996). The effect of dopamine D_1 receptor stimulation on the up-regulation of histamine H_3-receptors following destruction of the ascending dopaminergic neurones. *Br. J. Pharmacol.,* **118,** 585 – 592.

SCHLICKER, E., BEHLING, A., LÜMMEN, G. & GÖTHERT, M. (1992). Histamine H_3 receptor-mediated inhibition of noradrenaline release in the mouse brain cortex. *Naunyn-Schmiedeberg's Arch. Pharmacol.,* **345,** 489 – 493.

SCHLICKER, E., FINK, K., DETZNER, M. & GÖTHERT, M. (1993). Histamine inhibits dopamine release in the mouse striatum via presynaptic H_3 receptors. *J. Neural. Transm.,* **93,** 1 – 10.

SMITS, R.P.J.M. & MULDER, A.H. (1991). Inhibitory effects of histamine on the release of serotonin and noradrenaline from rat brain slices. *Neurochem. Int.,* **18,** 215 – 220.

VALENTINE, A.F., RIZZO, C.A., RIVELLI, M.A. & HEY, J.A. (1999). Pharmacological characterization of histamine H_3 receptors in human saphenous vein and guinea pig ileum. *Eur. J. Pharmacol.,* **366,** 73 – 78.

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