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Differential effects of potassium channel blockers on extracellular concentrations of dopamine and 5-HT in the striatum of conscious rats

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1 The selective Ca^{2+}-activated K^+ channel blocker apamin increased extracellular 5-hydroxytryptamine (5-HT) concentrations in the striatum when administered through the microdialysis probe at doses of 0.1 mM and 1 mM. Extracellular dopamine concentrations increased only at the highest dose administered (1 mM).

2 Mast cell degranulating peptide (MCDP), which blocks the dendrotoxin sensitive delayed rectifier (DR) current, increased extracellular concentrations of dopamine at doses of 10 μM - 100 μM but had no effect on 5-HT.

3 The non selective K^+ channel blocker tetraethylammonium (TEA) induced a dose-dependent (1 mM - 10 mM) increase in extracellular dopamine concentrations and an increase in 5-HT which showed little or no dose-dependency.

4 4-Aminopyridine (4-AP), a blocker with some similar characteristics to MCDP, increased extracellular dopamine concentrations at doses of 10 μM - 1 mM, but had no effect on 5-HT.

5 These findings suggest that dopamine release may be modulated by DR-like current and/or A-current K^+ channels. However, in view of the similar effects of MCDP and 4-AP at the concentrations used it is more likely that the dendrotoxin-sensitive DR-like current is involved. In contrast, 5-HT release appears to be modulated by Ca^{2+}-activated K^+ channels.

Keywords: Potassium channel blocker; dopamine; 5-HT and microdialysis

Introduction

The role of potassium channels (K^+ channels) in controlling the membrane potential (and hence the excitability) of neurones is well established (Hille, 1984). There is a variety of different subtypes of K^+ channels which have been classified on the basis of their voltage gating properties, second messenger regulation and sensitivity to calcium (Rudy, 1988). Neuronal membranes contain many, if not all, K^+ channel subtypes, with different neurone types possessing different proportions of these channel subtypes. This heterogeneity of the distribution of K^+ channel subtypes probably contributes to the different electrical conductance properties of individual neurones (Rogawski, 1985). K^+ channel activity is postulated to participate in the control of neurotransmitter release from nerve terminals by dictating the time course of repolarization. For example, blockade of K^+ channels can prolong the duration of action potentials, which leads to delayed closure of calcium channels, increased calcium influx and thus increased release of transmitter. There are many examples of these K^+ channel blockers which can be divided into two categories: organic molecules such as 4-aminopyridine (4-AP), tetraethylammonium (TEA) and quinine; and the natural toxins such as the dendrotoxins, charybdotoxin and apamin (Castle *et al.*, 1989; Strong, 1990; Colatsky, 1992). A number of these K^+ channel blockers have been shown to potentiate action potentials in various nerve tissues (Alkadhi & Hogan, 1989; Castle *et al.*, 1989; Haj-Dahmane *et al.*, 1991; Hu & Fredholm, 1991) and to increase the *in vitro* release of acetylcholine (Harvey & Karlsson, 1980; Anderson & Harvey, 1988), γ-aminobutyric acid (GABA, de Belleroche & Gardiner, 1988; Schweitz *et al.*, 1990), noradrenaline (Hu *et al.*, 1991; Clemens *et al.*, 1993; Ennis & Minchin, 1993) and dopamine (Jin & Fredholm, 1994). *In vivo* microdialysis studies from freely moving animals have further shown 4-AP-evoked increases in extracellular dopamine, acetylcholine and 5-hydroxytryptamine (5-HT) (Damsma *et al.*, 1988; Pei *et al.*, 1993; 1995).

In this study we use the technique of *in vivo* microdialysis to determine the effect of K^+ channel blockers with varying channel selectivities on the extracellular concentrations of dopamine and 5-HT in the striatum of freely moving rats. The blockers used were: (1) apamin, a 2 kDa toxin found in the venom of the European honey bee (Habermann, 1984) which blocks the small conductance Ca^{2+}-activated K^+ channels (SK channels) (Banks *et al.*, 1979); (2) mast cell degranulating peptide (MCDP), a basic 2.6 kDa peptide (also isolated from the venom of the European honey bee) which acts at a dendrotoxin-sensitive delayed rectifier (DR) K^+ channels (Strong, 1990); (3) tetraethylammonium (TEA), a quaternary ammonium with blocking effects at the voltage-gated DR (I_{DR}) and large conductance Ca^{2+}-activated K^+ channels (BK channels) (Rudy, 1988) and (4) 4-aminopyridine (4-AP), a tertiary amine which has been shown to block a subset of voltage-gated delayed rectifier currents (I_k) and transient A currents (I_A) (Rudy, 1988).

Methods

Male Sprague-Dawley rats (280-350g Charles River) were used in all experiments. Animals were group housed in cages with food and water available *ad libitum*. Following surgery the animals were singly housed in Plexiglass cages (30 cm sq.) with food and water available *ad libitum*.

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Surgical procedure

Following anaesthesia with ketamine/xylazine (66.6:6.67 mg kg^{-1}i.m, 1 ml kg^{-1}) the animals were secured in a stereotaxic frame with ear and incisor bars. A microdialysis guide cannula (CMA/Microdialysis, Sweden) was implanted and cemented to the skull with dental acrylic. Co-ordinates for the striatum were taken according to Paxinos and Watson (1986): RC −0.4; L −3.7; V −2.8 mm from the dura. The wound was sutured and the animals left to recover for 24 h in their home cages with free access to food and water.

Microdialysis

The dialysis probe (o.d. 0.5 mm, membrane length 4 mm; CMA/Microdialysis, Sweden) was implanted, via the guide cannula, into the striatum of the unrestrained rat 24 h post surgery. The probe was perfused with artificial cerebrospinal fluid (aCSF) (composition, mM: NaCl 125, KCl 3.0, $MgSO_4$ 0.75, and $CaCl_2$ 1.2 pH 7.4) at a flow rate of 1 μl min^{-1}. A three hour stabilization period was allowed following probe implanatation after which time dialysis sampling was carried out by a modification of the method of Routledge *et al.* (1993). Three control samples were taken before drug infusion to achieve a steady baseline. Test compounds or aCSF were then infused for 30 min followed by a further 30 min wash out period. This was repeated for increasing concentration of compound. The compounds tested were apamin, MCDP, TEA and 4-AP; all test compounds were made up in aCSF. For vehicle-treated animals the same procedure was used with aCSF replacing test compound. A 20 min sampling regime was used throughout the experimental period. At the end of the experiment the probe placement was verified histologically and data from animals with incorrect probe placement were discarded.

Analysis of dialysates

Dopamine and 5-hydroxytryptamine (5-HT) were separated by reverse phase high performance liquid chromatography (h.p.l.c.) (C18 ODS2 column, 100 × 4.6 mm, Jones Chromatography, Mid Glamorgan, Wales) and detected by an ANTEC electrochemical detector (ANTEC, Netherlands) set at a potential of 0.65V vs a Ag/AgCl reference electrode. Mobile phase was delivered by a LKB 2150 h.p.l.c. pump (LKB/Pharmacia, Milton Keynes, U.K) at 1 ml min^{-1} and contained 0.1 M NaH_2PO_4 buffer at pH 3.8, 0.1 mM EDTA, 1 mM 1-octane sodium sulphonic acid and 17.5% methanol.

Data analysis

The fmol perfusate values of transmitters for the first three baseline samples were averaged and this value denoted as 100%. Subsequent sample values were expressed as a percentage of this preinfusion control value. Results were analysed by 2 way analysis of variance followed by *post hoc* comparison of means by use of the Super ANOVA software application (Abacus Concepts Inc., Berkeley, CA 1989) on an Apple Macintosh computer.

Materials

All chemicals used were analytical grade and were purchased from BDH chemicals (Poole, Dorset). 4-AP, MCDP, TEA and apamin were purchased from Sigma chemicals (Poole, Dorset).

Results

Effects of apamin on extracellular concentrations of dopamine and 5-HT

Application of apamin (0.1 mM − 1 mM) through the microdialysis probe caused a significant concentration-dependent increase in striatal dialysate concentrations of 5-HT ($P<0.05$) (Figure 1b) from basal preinfusion levels (91.6 ± 34.6 fmol per 20 μl) to a maximum of $1 \times 10^5 \pm 2.7 \times 10^4$% of preinfusion values at the highest infusion dose of 1 mM ($t=360$ min). Apamin (1 mM) infusions also produced a significant increase in extracellular dopamine concentrations (Figure 1a) from basal (199.6 ± 75.4 fmol) to a maximum of 1069 ± 917% of preinfusion control levels. Animals treated with 100 μM apamin exhibited forepaw treading, head twitches and flat body posture which is characteristic of the 5-HT behavioural syndrome (Jacobs, 1976). However administration of higher doses of apamin (i.e. 1 mM) induced convulsive behaviour in all animals.

Effect of MCDP on extracellular concentrations of dopamine and 5-HT

MCDP administration increased striatal extracellular concentrations of dopamine ($P<0.05$) at concentrations of 10 and 100 μM (Figure 2a) to a maximum of 218 ± 84% of preinfusion control values after 200 min. No significant changes in 5-HT levels were observed at any of the MCDP concentrations used in this study (Figure 2b).

Effects of TEA on extracellular concentrations of dopamine and 5-HT

Application of TEA (1 μM − 10 mM) evoked a concentration-dependent increase in extracellular dopamine concentrations

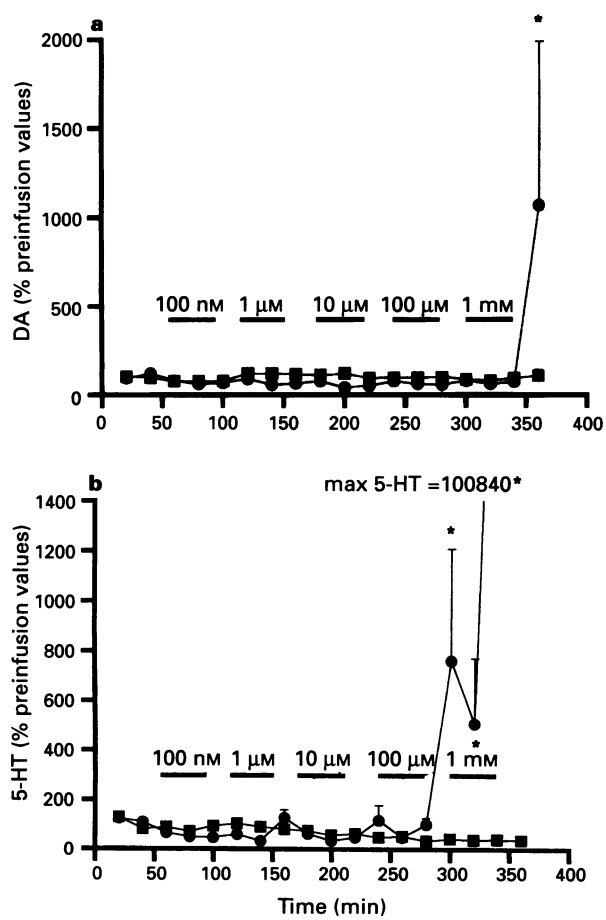

Figure 1 Effects of cumulative infusion of apamin on extracellular concentrations of (a) dopamine (DA) and (b) 5-hydroxytryptamine (5-HT) in rat striatal dialysates. Apamin treated animals (●) vehicle treated animals (■). Data are shown as mean ± s.e.mean for 6 animals per treatment group. Bars denote infusion of apamin. *Demonstrates statistical significance ($P<0.05$) compared to vehicle controls.

($P<0.05$) reaching a maximum of $500\pm193\%$ of preinfusion control levels following 1 mM infusions ($t=250$ min) (Figure 3a). A smaller but still significant ($P<0.05$) increase in extracellular concentrations of 5-HT was also observed with a maximum increase of $187\pm34\%$ ($t=260$ min) after administration of 1 mM TEA (Figure 3b).

Effects of 4-AP on extracellular concentrations of dopamine and 5-HT

4-AP infusions induced significant, concentration-related increases in striatal dopamine concentrations. ($P<0.05$) but had no effect on 5-HT (Figure 4a,b, respectively). 4-AP (10 μM) increased dopamine $148\pm28\%$ of preinfusion control levels. This level was maintained throughout subsequent infusions with a small increase after an infusion of 1 mM, reaching a maximum level of $172\pm39\%$; this increase coincided with the animals exhibiting seizure like behaviour. All animals exhibited myoclonic like forepaw jerks and contralateral head twitches which progressively worsened during infusion.

A summary of the differential effect of apamin, MCDP, TEA and 4-AP on the release of dopamine and 5-HT and each blocker's selectivity for K^+ channel subtypes can be seen in Table 1.

Discussion

These data demonstrate that various K^+ channel blockers differentially modulate extracellular concentrations of dopamine and 5-HT in rat striatum, suggesting that release of these transmitters may be modulated by different K^+ channels.

High concentrations of apamin caused a marked increase in the extracellular concentrations of 5-HT and a more modest and delayed increase in extracellular dopamine concentrations. Apamin is one of the most selective K^+ channel blockers available, showing specificity for low conductance Ca^{2+} activated K^+ channels (SK). However its specificity for these particular channels is over a concentration range of 1 – 100 nM *in vitro* (Strong, 1990). It is therefore possible that the effect seen here following application of 100 μM and 1 mM apamin through the dialysis probe is mediated by a mechanism other than blockade of Ca^{2+}-activated K^+ channels. This is more likely for dopamine which only shows increases in response to application of 1 mM apamin infusions. This contasts with the work of Steketee and Kalivas (1990) who showed that microinjection of apamin into the dopaminergic nuclei A10, produced a dose-dependent increase in motor activity which was inhibited by the dopamine D_2 receptor antagonist haloperidol. However, these effects may reflect differences in the innervation of the two different regions. This discrepancy may result from different sites and methods of administration of the peptide since the *in vivo* recovery across the probe is difficult to measure. Since apamin was administered to terminal region, an alternative explanation is that the Ca^{2+}-activated K^+ channels are located on the cell bodies rather than the terminals of dopaminergic neurones. However, the most straight forward interpretation is that apamin-sensitive Ca^{2+}-activated K^+ channels (SK channels) modulate presynaptic 5-HT but not dopamine release in the striatum.

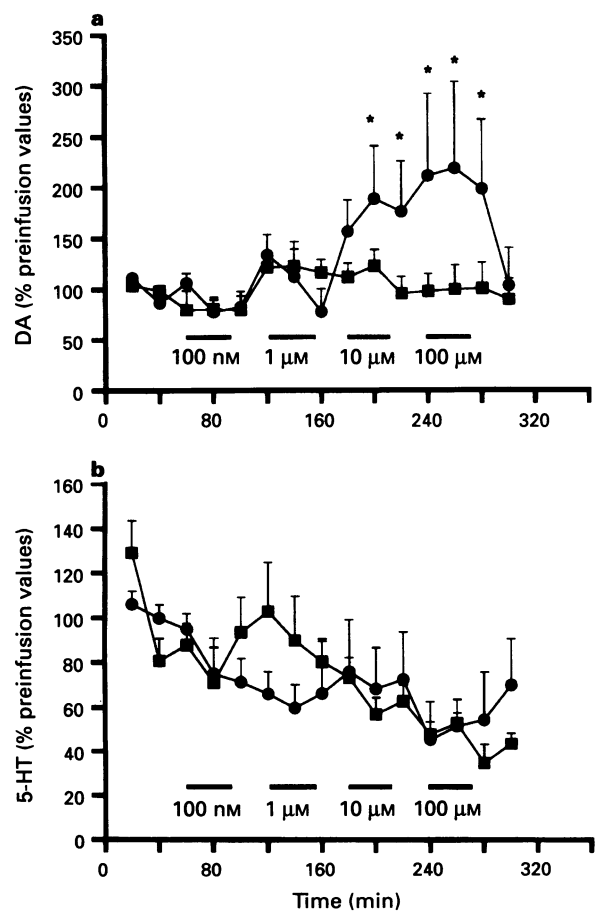

Figure 2 Effects of cumulative infusion of mast cell degranulating peptide (MCDP) on extracellular concentrations of (a) dopamine (DA) and (b) 5-hydroxytryptamine (5-HT) in rat striatal dialysates. MCDP treated animals (●) vehicle treated animals (■). Data are shown as mean±s.e.mean for 6 animals per treatment group. Bars denote infusion of MCDP. *Demonstrates statistical significance ($P<0.05$) compared to vehicle controls.

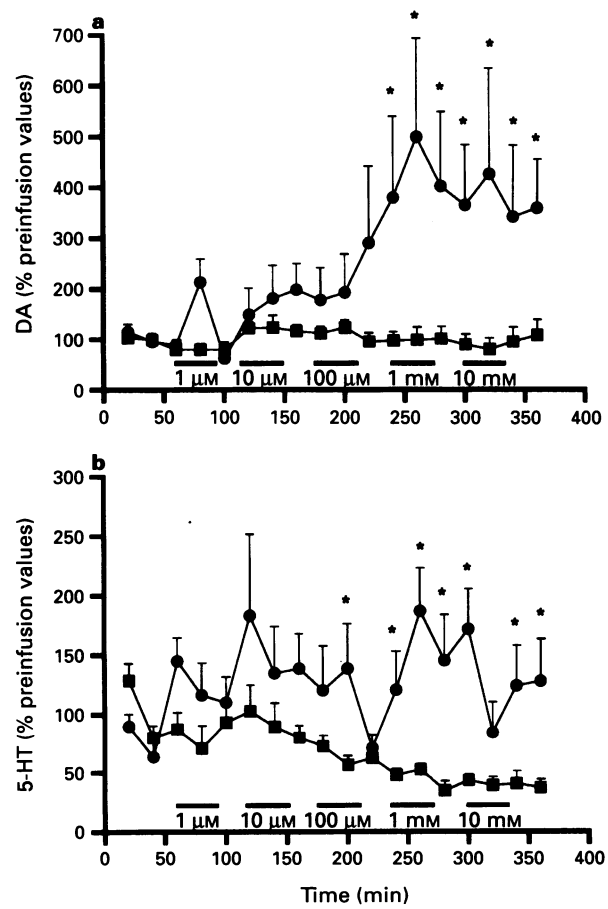

Figure 3 Effects of cumulative infusion of tetraethylammonium (TEA) on extracellular concentrations of (a) dopamine (DA) and (b) 5-hydroxytryptamine (5-HT) in rat striatal dialysates. TEA treated animals (●) vehicle treated animals (■). Data are shown as mean±s.e.mean for 6 animals per treatment group. Bars denote infusion of TEA. *Demonstrates statistical significance ($P<0.05$) compared to vehicle controls.

In contrast to apamin, MCDP had no significant effect on extracellular 5-HT concentrations but did produce a significant, concentration-related increase in dopamine. Although these two peptides exhibit some sequence homology, their channel specificities and hence sites of action are different. MCDP is selective for a class of fast activating dendrotoxin-sensitive DR channels (Strong, 1990) which contrasts to apamin's specificity for the SK channel. The differences in the *in vivo* observations may therefore be a function of the differing sites of action of the two toxins.

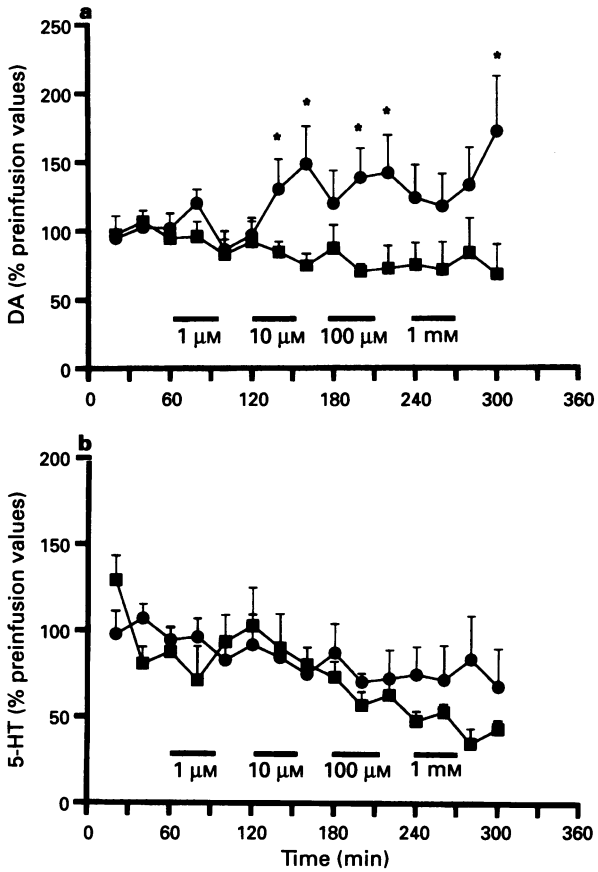

Figure 4 Effects of cumulative infusion of 4-aminopyridine (4-AP) on extracellular concentrations of (a) dopamine (DA) and (b) 5-hydroxytryptamine (5-HT) in rat striatal dialysates. 4-AP treated animals (●) vehicle treated animals (■). Data are shown as mean ± s.e.mean for 6 animals per treatment group. Bars denote infusion of 4-AP. *Demonstrates statistical significance ($P<0.05$) compared to vehicle controls.

TEA infusion produced significant increases in both dopamine and 5-HT extracellular concentrations. However, these changes though qualitatively similar were very different quantitatively. The increase in dopamine demonstrated a dose-dependency which was not observed with 5-HT, which showed little or no dose relationship. This may be explained by the differing selectivity of TEA for any one particular type of K^+ channel. The increases in dopamine concentrations were approximately equivalent (for equivalent infusion concentrations) for both MCDP and TEA. This could be attributed to the fact that both blockers show action at the same K^+ channels i.e. dendrotoxin-sensitive DR-like K^+ channel. TEA-induced increases in extracellular concentrations of 5-HT may be a function of the action of this blocker at the large conductance Ca^{2+}-activated K^+ channels (Noack, 1992). However, the difference in the quantitative effects between the two transmitters suggests a greater selectivity of TEA for the dendrotoxin-sensitive DR-like K^+ channel.

4-AP (10 μM – 1 mM) increased dialysate concentrations of dopamine but had no effect on 5-HT, suggesting that 4-AP-sensitive K^+ channels are present on dopaminergic terminals but absent from 5-hydroxytryptaminergic terminals. Similar increases in dopamine after intrastriatal administration have been previously obtained by Damsma *et al.* (1988). Although 4-AP is not totally selective it does possess a degree of specificity for A-currents (I_A) and some action at the dendrotoxin-sensitive DR (Rudy, 1988). The effects of 4-AP are comparable to those K^+ channel blockers with similar specificities, i.e. MCDP, in that dopamine but not 5-HT was increased. The effective concentrations of 4-AP suggest the involvement of the dendrotoxin-sensitive delayed-rectifier-like current as apposed to the A-current, which usually requires higher concentrations to block the channel (Storm, 1988). Infusion of 1 mM 4-AP induced seizures in all animals, the behavioural characteristics of which were somewhat different to those caused by apamin. 4-AP-induced seizures have been shown to be mediated by $GABA_B$ and excitatory amino acid receptors (Siniscalchi & Avoli, 1992) so it is possible that the seizures observed were mediated by modulation of one or both of these receptors.

In summary, the *in vivo* data presented here indicate that dopamine release may be modulated by delayed-rectifier-like currents or A-current K^+ channels or both. However in view of the low effective concentration of 4-AP, it is unlikely that classical I_A channels are involved. Rather, since MCDP has a similar release profile to 4-AP, it is more likely that the dendrotoxin-sensitive delayed-rectifier-like current mediates the

Table 1 Summary of the differential effects of K^+ channel blockers on extracellular dopamine (DA) and 5-hydroxytryptamine (5-HT)

K^+ channel blocker	*Effect on extracellular DA*	*Effect on extracellular 5-HT*	*K^+ channel selectivity*
Apamin	Large increase after 1 mM max. 1069 ± 917%	Large increase after 0.1 and 1 mM max. $1 \times 10^5 \pm 2.7 \times 10^4$%	Small conductance Ca^{2+}-activated channels.
MCDP	Increases after 10 – 100 μM max. 218 ± 84%	No effect	Dendrotoxin-sensitive DR channels.
TEA	Increases after 0.1 – 10 mM max. 500 ± 193%	Small increases after 1 – 10 mM max. 187 ± 34%	Dendrotoxin-sensitive DR channels, large conductance Ca^{2+}-activated channels.
4-AP	Increases after 10 μM – 1 mM max. 172 ± 39%	No effect	A-channels, dendrotoxin-sensitive DR channels.

Values given are means ± s.e.mean of treatment group ($n=6$) expressed as % of preinfusion control values. For key to abbreviations used see text.

release of dopamine. In contrast, 5-HT release appears to be solely under the influence of the Ca^{2+}-activated K^+ channels (SK and BK). This differential effect of K^+ blockers on the possible release of dopamine and 5-HT (Table 1) probably occurs because of the differential distribution of K^+ channel subtypes on chemically distinct nerve terminals. More studies are necessary to characterize further this heterogeneity.

References

ALKADHI, K.A. & HOGAN, Y.H. (1989). Effects of calcium on synaptic facilitation by potassium channel blockers in superior cervical ganglion of the rat. *Neuropharmacol.*, **28,** 75–81.

ANDERSON, A.J. & HARVEY, A.L. (1988). Effects of the potassium channel blocking dendrotoxins on acetylcholine release and motor nerve terminal activity. *Br. J. Pharmacol.*, **93,** 215–221.

BANKS, B.E.C., BURGESS, C.E., BURNSTOCK, C., CLARET, M., COCKS, T.M. & JENKINSON, D.H. (1979). Apamin blocks certain neurotransmitter-induced increases in potassium permeability. *Nature*, **282,** 415–417.

DE BELLEROCHE, J. & GARDINER, I.M. (1988). Inhibitory effect of 1,2,3,4-tetrahydro-9-aminoacridine on the depolarization-induced release of GABA from cerebral cortex. *Br. J. Pharmacol.*, **94,** 1017–1019.

CASTLE, N.A., HAYLETT, D.G. & JENKINSON, D.H. (1989). Toxins in the characterisation of potassium channels. *Trends Neurosci.*, **12,** 59–65.

CLEMENS, A., REPP, H. & HERTTING, G. (1993). Effect of K^+ channel blockers on the α_2-adrenoceptor-coupled regulation of electrically evoked noradrenaline release in hippocampus. *Arch. Pharmacol.*, **347,** 14–20.

COLATSKY, T.J. (1992). Potassium channel blockers: synthetic agents and their antiarrhythmic potential. In *Potassium Channel Modulators*, ed. Weston, A.H. & Hamilton, T.C. pp. 304–340, Oxford: Blackwell Scientific Publications.

DAMSMA, G., BIESSELS, P.T.M., WESTERINK, B.H.C., DE VRIES, J.B. & HORN, A.S. (1988). Differential effects of 4-aminopyridine and 2,4-diaminopyridine on the in vivo release of acetylcholine and dopamine in freely moving rats measured by intrastriatal dialysis. *Eur. J. Pharmacol.*, **145,** 15–20.

ENNIS, C. & MINCHIN, M.C.W. (1993). The effect of toxin I, a K^+ channel inhibitor, on [3H]noradrenaline release from rat cerebral cortex. *Eur. J. Pharmacol.*, **248,** 85–88.

HABERMANN, E. (1984). Apamin. *Pharmacol. Ther.*, **25,** 255–270.

HAJ-DAHMANE, S., HAMON, M. & LANFUMEY, L. (1991). K^+ channel and $5HT_{1A}$ autoreceptor interactions in the rat dorsal raphe nucleus: an *in vitro* electrophysiological study. *Neurosci.*, **41,** 495–505.

HARVEY, A.L. & KARLSSON, E. (1980). Dendrotoxin from the venom of the green mamba, *Dendroapsis augusticeps*. A neurotoxin that enhances the release of acetylcholine at neuromuscular junction. *Naunyn Schmied. Arch. Pharmacol.*, **312,** 1–7.

HILLE, B. (1984). In *Ionic Channels of Excitable Membranes*. ed. Hille, B. pp. 99–116 Sunderland, M.A.: Sinauer Assoc.

HU, P-S., BENISHIN, C. & FREDHOLM, B.B. (1991). Comparison of the effects of four dendrotoxin peptides, 4-aminopyridine and triethylammonium on the electrically evoked [^{3}H] noradrenaline release from rat hippocampus. *Eur. J. Pharmacol.*, **209,** 87–93.

HU, P-S. & FREDHOLM, B.B. (1991). 4-Aminopyridine induced increase in basal and stimulation evoked [^{3}H]-NA release in slices from rat hippocampus: Ca^{2+} sensitivity and presynaptic control. *Br. J. Pharmacol.*, **102,** 764–768.

JACOBS, B.L. (1976). An animal behaviour model for studying central serotonergic synapses. *Life Sci.*, **19,** 777–786.

JIN, S. & FREDHOLM, B.B. (1994). Role of NMDA, AMPA and kainate receptors in mediating glutamate and 4-aminopyridine induced dopamine and acetylcholine release from rat striatal slices. *Neuropharmacol.*, **33,** 1039–1048.

NOACK, T.H. (1992). Potassium channels in excitable cells: a synopsis. In *Potassium Channel Modulators*, ed. Weston, A.H. & Hamilton, T.C. pp. 1–14, Oxford: Blackwell Scientific Publications.

PAXINOS, G. & WATSON, C. (1986). *The Rat Brain in Stereotaxic Coodinates*. New York: Academic Press.

PEI, Q., ELLIOTT, M., GRAHAME-SMITH, D.G. & ZETTERSTROM, T. (1993). Quinine and 4-aminopyridine inhibit the stimulatory output of dopamine in nucleus accumbens and the behavioural activity produced by morphine. *Eur. J. Pharmacol.*, **249,** 243–246.

PEI, Q., LESLIE, R.A., GRAHAME-SMITH, D.G. & ZETTERSTROM, T.S. (1995). 5-HT efflux from rat hippocampus *in vivo* produced by 4-aminopyridine is increased by chronic lithium administration. *Neuroreport*, **6,** 716–720.

ROGAWSKI, M.A. (1985). The A current: how ubiquitous a feature of excitable cells is it? *Trends Neurosci.*, **8,** 214–219.

ROUTLEDGE, C., GURLING, J., WRIGHT, I.K. & DOURISH, C.T. (1993). Neurochemical profile of the selective and silent $5\text{-}HT_{1A}$ receptor antagonist WAY100135: an *in vivo* microdialysis study. *Eur. J. Pharmacol.*, **239,** 195–202.

RUDY, B. (1988). Diversity and ubiquity of K channels. *Neurosci.*, **25,** 729–749.

SCHWEITZ, H., BIDARD, J.N. & LAZDUNSKI, M. (1990). Purification and pharmacological characterisation of peptide toxins from black mamba (*Dendroapsis polyepsis*) venom. *Toxicon*, **28,** 847–856.

SINISCALCHI, A. & AVOLI, M. (1992). Modulation by $GABA_B$ receptors of spontaneous synchronous activities induced by 4-aminopyridine in the rat hippocampus. *Neurosci. Letts.*, **148,** 159–163.

STEKETEE, J.D. & KALIVAS, P.W. (1990). Effect of microinjection of apamin into the A10-dopamine region of rat. A behavioural and neurochemical analysis. *J. Pharmacol. Exp. Ther.*, **254,** 711–719.

STORM, J.F. (1988). Temporal integration by a slowly inactivating K current in hippocampal neurones. *Nature*, **336,** 379–381.

STRONG, P.N. (1990). Potassium channel toxins. *Pharmacol. Ther.*, **46,** 137–162.

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