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5-HT_3 Receptor-independent Inhibition of the Depolarization-induced ^{86}Rb Efflux from Human Neuroblastoma Cells, TE671, by Ondansetron

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Abstract

The 5-HT_3-receptor antagonist, ondansetron, has been shown to have positive effects in selected in-vivo models of memory impairment and anxiety. The exact mechanisms underlying such bioactivities are unknown. In the present work, an ^{86}Rb efflux bioassay was used to show that ondansetron has a unique ability to block voltage-gated potassium channels in TE671 human neuroblastoma cells.

This intrinsic potassium-channel-blocking (KCB) property is relatively weak (IC50 20 μM), but is not shared by other 5-HT_3-receptor ligands including zatosetron, MDL 72222, LY 278, 584, zacopride, 1-phenylbiguanide, and ICS 205-930 (tropisetron). Pre-incubation of the target neuroblastoma cells with several 5-HT-receptor ligands including 5-hydroxytryptamine, 8-OH-DPAT, ketanserin, 2-methyl-5-HT, as well as a number of potent 5-HT_3 agonists and antagonists and two selective neurotoxins, failed to abolish the KCB action of ondansetron.

A preliminary structure-activity relationship analysis indicates that the KCB activity of ondansetron is almost entirely attributable to its structural nucleus, 2,3-dihyro-9-methyl-4(1H)-carbazolone. It is hypothesized that the KCB action of ondansetron is mediated through receptors other than 5-HT_3 receptors. The KCB activity of ondansetron may be a significant factor in the in-vivo cognition-enhancing activities of this compound, conceivably due to depolarization of the hippocampal synaptic membranes and a consequent augmentation of neurotransmission.

Ondansetron, 1,2,3,9-tetrahydro-9-methyl-3-[(2-methyl-1H-imidazol-1-yl)methyl]-4H-carbazol-4-one (Fig. 1), is a selective antagonist of the 5-HT_3-receptor subtype (reviewed by Kilpatrick et al 1990; Hoyer 1991). This compound has been reported to have positive effects in models of anxiety, schizophrenia, alcohol withdrawal and age-associated memory impairment, and it is currently in clinical trials for anxiety and cognition (Barnes et al 1990; Costall et al 1990; Costall & Naylor 1992; Oxford et al 1992; Tyers et al 1993).

The exact mechanism for the cognition-enhancing properties of ondansetron is not well understood. However, the 5-HT_3 receptor has been shown to mediate the inhibitory effects of 5-HT on acetylcholine release in the entorhinal cortical tissue in-vitro (Barnes et al 1989, 1990). Blandina et al (1988) have shown that 5-HT and 2-methyl-5-HT could stimulate dopamine release from striatal slices and this effect could be inhibited by low concentrations of MDL 72222 and ICS 205-930 (tropisetron) (Fig. 1), potent and selective antagonists of the 5-HT_3-receptor subtype. Blockade of the 5-HT_3 receptor by its specific antagonists such as ondansetron is expected to facilitate cholinergic function, thus enhancing cognition (Barnes et al 1989, 1990; Costall & Naylor 1992). There are also several lines of neuroanatomical evidence implicating the 5-HT_3 receptor in central functions such as cognition, anxiety and depression. These include the observation that the 5-HT_3 receptors are generally found presynaptically on nerve terminals or fibres on non-monoaminergic neurons. These receptors are notably present in the hippocampus, amygdala and entorhinal cortex (Kidd et al 1993)

Whereas 5-HT_1-, 5-HT_2- and 5-HT_4-receptor subtypes are G-protein coupled and modulate potassium (K) channels, the 5-HT_3 receptor does not appear to be involved in G-protein, signal transduction mediated events (Hoyer 1991). Instead, the 5-HT_3 receptor has been suggested to be a member of the ligand-gated ion channels, forming a monovalent-selective cation channel permeable to both Na^+ and K^+ but not Ca^{2+}, analogous to the kainate receptor (Henderson 1990; Kilpatrick et al 1990). This has also been confirmed by the functional expression of a cloned 5-HT_3 receptor cDNA in *Xenopus* oocytes (Maricq et al 1991).

Responses to 5-HT that are mediated through 5-HT_3 receptors include depolarization and noradrenaline release in postganglionic autonomic neurons, depolarization and neurotransmitter (e.g. acetylcholine) release in parasympathetic and sensory neurons, as well as in the enteric nervous system (Richardson & Engel 1986).

At high doses, ondansetron and zatosetron prolong the action potential duration in the canine heart and increase the Q-Tc interval. This effect has been postulated to be due to the interaction of these compounds with K^+ channels in cardiac muscle that is similar to, but not identical with, the 5-HT_3 receptor (Williams et al 1991). A potent and selective

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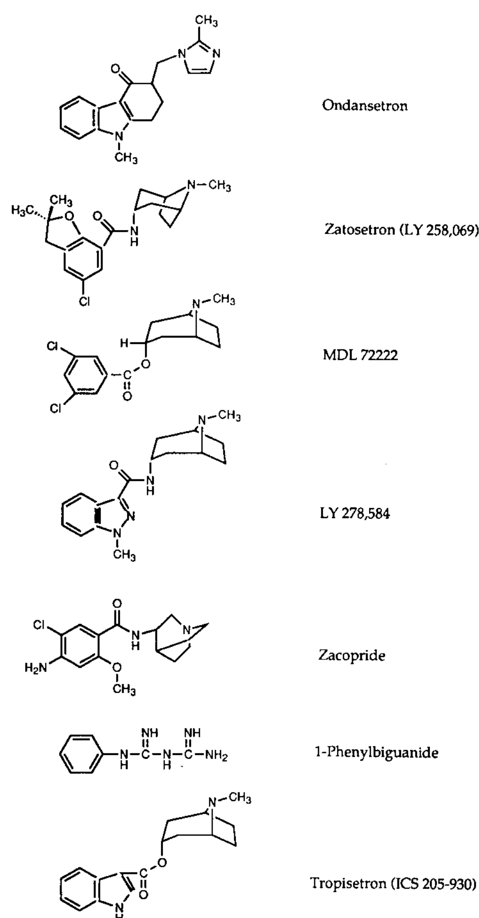

FIG. 1. Structure of 5-HT_3-receptor ligands.

5-HT_3 antagonist, tropisetron, has been shown to block cardiac potassium, sodium and calcium currents and possess strong antiarrhythmic activities (Williams et al 1985; Scholtysik 1987; Scholtysik et al 1988; Hof et al 1993).

In an effort to further investigate a possible association of the 5-HT_3 receptor with the voltage-gated K^+ channels, we used an ^{86}Rb-efflux assay to test the effect of a number of the 5-HT-receptor agonists and antagonists on the voltage-gated K^+ channels in a human neuroblastoma cell line depolarized with high concentrations of KCl. Since ^{86}Rb is a surrogate ion for K^+, modulation of its efflux from pre-loaded viable target cells represents an accurate assessment of the activity of various K^+ channels (Arner & Stallcup 1981; Daniel et al 1991; Toral et al 1994).

The functional significance of the potassium channel blocking (KCB) action of ondansetron can be connected to the hypothesis that KCBs have potential in enhancing cognition, presumably through the modulation of long-term potentiation and augmentation of neurotransmission (Ben Ari et al 1989; Lavertsky & Jarvik 1992; Harvey 1993). Such an activity may indeed contribute to the observed in-vivo memory-improving action of ondansetron which, by and large, has remained unexplained.

Materials and Methods

Buffers and reagents
Chemicals and drugs were purchased from Sigma Chemical Co. (St Louis, MO), Aldrich Chemical Co. (Milwaukee, WI), MTM Research Chemicals Inc. (Windham, NH), and Research Biochemicals Inc. (Natick, MA).

Ondansetron (GR38032F) was kindly provided by Glaxo Group Research, Greenford (UK). Other commercially unavailable compounds were provided by their respective manufacturers.

Drugs were dissolved at a stock concentration of 10 mM, either in MOPS-PSS (4-morpholine propanesulphonic acid in physiological saline) pH 7·4 (NaCl, 120 mM; KCl, 7·0 mM; $CaCl_2$, 2·0 mM; $MgCl_2$, 1.0 mM; MOPS, 20 mM and oubain, 10 μM) or dimethylsulphoxide (DMSO), and were subsequently diluted in the appropriate buffers to the desired concentration.

Depolarizing solution (DS-100) consisted of MOPS-PSS, containing KCl (100 mM), replacing the equivalent concentrations of NaCl.

Cell culture and ^{86}Rb loading
Human neuroblastoma cells TE671 were obtained from American Type Culture Collection (ATCC HBT 139) and were maintained at 37°C in Dulbecco's modified Eagle medium (DMEM) supplemented with 10% foetal bovine serum, 4·5 g L^{-1} glucose and 2·0 mM L-glutamine. Cells were plated, loaded with ^{86}Rb, and allowed to adhere to the bottom of the wells in 96-well microtitre plates overnight (Daniel et al 1991).

^{86}Rb-efflux assay
This assay was performed as previously described by Daniel et al (1991), and subsequently modified (Toral et al 1994). Briefly, the growth medium in the microtitre plate was discarded by a sharp flicking of the plate. The adherent cell layer was washed three times with 200 μL MOPS-PSS. The cells were incubated for 30 min at room temperature (21°C) with 200 μL DS-100, in the presence, or the absence of a test drug. Supernatant (150 μL) from each well was removed and counted. Cell layer was solubilized in 200 μL 0·1% Tween 20 in water and 150 μL was also counted in a Packard 2200 CA liquid scintillation counter using a spectral window (0–1700). All supernatants were counted in 7·0 mL distilled water.

The % total efflux (i.e. under depolarizing condition induced by DS-100 buffer) was calculated as: (counts min^{-1} in supernatant)/(counts min^{-1} in supernatant + counts min^{-1} in cell extract) × 100. The % net efflux was calculated as: % total efflux in DS-100 − % basal efflux in MOPS-PSS.

For the competition studies, ^{86}Rb-loaded TE671 cells were first incubated at room temperature (21°C) for 10 min with 400 μM (4 × concentration) of the competing ligand in 50 μL MOPS-PSS. Following that period, 150 μL DS-100 buffer with various doses of ondansetron was added into each well. Cells were further incubated for 20 min at room temperature, supernatants collected and counted and percent efflux calculated as described above.

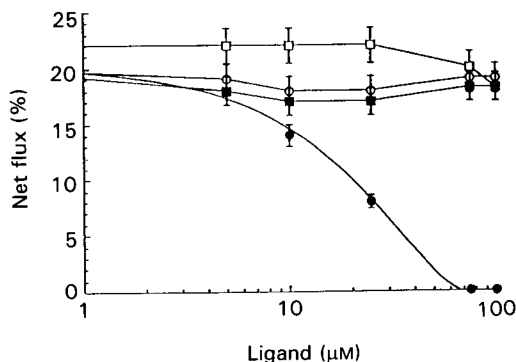

FIG. 2. Inhibition of K^+-stimulated ^{86}Rb efflux from TE671 cells by ondansetron. Target TE671 cells pre-loaded with ^{86}Rb were incubated with DS-100 solution containing various doses of either ondansetron (●), or MDL 72222 (□), LY 278, 584 (■), or 1-phenylbiguanide (○). Percent net efflux was calculated by a subtraction of the basal efflux (in MOPS-PSS) from the total efflux (in DS-100). Bars represent ± one standard deviation of four replicate wells in a representative experiment.

Results

Inhibition by the 5-HT_3 receptor selective ligands of the K^+-stimulated ^{86}Rb efflux from TE671 cells was measured. To this end, ^{86}Rb-loaded viable target cells were depolarized with a buffer containing 100 mM KCl (DS-100) and in the presence, or the absence, of a 5-HT_3 receptor-selective ligand. The value of the basal efflux (in MOPS-PSS) was subtracted from the total ^{86}Rb efflux induced by DS-100, and the net percent efflux was calculated. As shown in Fig. 2, ondansetron inhibited the net ^{86}Rb efflux in a dose-dependent manner with an IC50 value of approximately $20 \pm 1{\cdot}4\,\mu M$. This value remained in the 20–40 μM range in all subsequent assays. Ondansetron did not have any significant effect on the rate of basal efflux. Two other 5-HT_3 selective antagonists, MDL 72222 and LY 278, 584, and an agonist, 1-phenylbiguanide, at concentrations up to 200 μM did not significantly alter the K^+-stimulated ^{86}Rb efflux from TE671 cells. In addition, other 5-HT_3-receptor antagonists, (−)-zacopride, zatosetron, tropisetron, or the potent 5-HT_3-receptor agonist, 1-(*m*-chlorophenyl)-biguanide, used at concentrations up to 200 μM did not significantly inhibit the K^+-stimulated ^{86}Rb efflux (not shown). At concentrations higher than 200 μM, tropisetron showed some KCB activity, but due to its poor solubility at high concentrations, the IC50 value remained undetermined.

We next examined the effect of a number of other 5-HT-receptor ligands on the inhibitory (KCB) action of ondansetron. TE671 cells loaded with ^{86}Rb were pre-incubated with the individual 5-HT receptor selective ligands, used at a final concentration of 100 μM in the physiological buffer, MOPS-PSS. Target cells were then incubated with the increasing doses (0–200 μM) of ondansetron in the depolarization buffer, DS-100. The percent net efflux of ^{86}Rb and the IC50 values for ondansetron were calculated in each experiment. Neither 5-HT, nor any of the 5-HT_3 receptor-selective ligands, MDL 72222, LY 278, 584, 1-phenylbiguanide, or 2-methyl 5-HT significantly altered the inhibitory activity of ondansetron. The 5-HT_{1A} agonist 8-OH-DPAT ((±)-8-hydroxydipropylaminotetralin hydrobromide), and the 5-$HT_{2/1C}$ antagonist ketanserin were also ineffective in blocking the action of ondansetron on ^{86}Rb efflux. Furthermore, two other 5-HT_3-directed ligands metoclopramide (an antagonist) and quipazine *N*-methyl dimaleate (an agonist), used at 100 μM also failed to abolish the KCB activity of ondansetron through competition for the common binding sites on the 5-HT_3 receptor. Moreover, two 5-HT receptor-selective neurotoxins, 5,7- and 6,7-dihydroxytryptamine creatinine sulphate, used at a final concentration of 100 μM, did not significantly shift the IC50 value of ondansetron (results not shown).

In an effort to obtain some correlations between the chemical structure of ondansetron and its KCB activity, a number of compounds comprising the core structure of ondansetron were tested for KCB activity. The main nucleus of ondansetron, 2,3-dihydro-9-methyl-4(1H)-carbazolone, inhibited the efflux of ^{86}Rb from TE671 cells, with IC50 value of 50 μM, while 1,2,3,4-tetrahydrocabazole showed an IC50 value of 130 μM (Table 1). Other constituents of ondansetron; 1-methylindole, indole, cyclohexanone and 2-methylimidazole were found either extremely weak, or inactive as KCBs (Table 1).

Table 1. Potassium-channel blocking activity of compounds comprising the core structure of ondansetron.

Compound	IC50[a] (μM)
Ondansetron	25 ± 1·8
2,3-Dihydro-9-methyl-4(1H)-carbazolone	50 ± 3·5
1,2,3,4-Tetrahydrocarbazole	130 ± 9·1
1-Methylindole	500 ± 35
Indole	> 800
Cyclohexanone	> 800
2-Methylimidazole	> 800

[a] Arithmetic mean of quadruplicate assay wells ± standard deviation.

To test whether ondansetron could possibly act on a slow inactivating (delayed rectifier) K^+ channel, an ^{86}Rb-efflux assay using cultured Chinese Hamster Ovary (CHO) cells transfected with a cloned cDNA encoding a human hippocampus slow inactivating K^+ channel, HHiKI (Kv 1·5), was employed. Ondansetron (400 μM) did not show a significant inhibition of the K^+-stimulated ^{86}Rb efflux from these cells, when the latter were depolarized with 100 mM KCl in the presence of ondansetron (results not shown).

In addition, we also tested the KCB activity of ondansetron (up to 200 μM) on the rate of ^{86}Rb efflux from TE671 cells treated with the calcium ionophore A23187. No significant inhibition by ondansetron or any of its chemical constituents (shown in Table 1) of the ^{86}Rb efflux from the target TE671 cells was observed.

Discussion

Data obtained in this work points to a strong possibility that ondansetron possesses a distinct voltage-gated KCB activity in the human neuroblastoma cells, TE671. While this activity can only be detected at high concentrations of ondansetron, it appears to be a property unique to that 5-HT_3 compound. Several other 5-HT_3 receptor antagonists, as well as four specific 5-HT subtype selective agonists,

and two 5-HT receptor selective neurotoxins either showed no intrinsic KCB activity, or failed to affect the KCB activity of ondansetron. This is despite the fact that in several bioassays, compounds such as tropisetron, MDL 72222, and (−)-zacopride show much higher binding affinities for the 5-HT_3 receptor than ondansetron (Hoyer 1991; Oxford et al 1992).

Our present observations could have at least one significant implication. The fact that none of the 5-HT_3 receptor-selective ligands, or those specific for other 5-HT receptor subtypes showed intrinsic KCB activities, or could competitively inhibit the KCB activity of ondansetron, may indicate that the observed KCB action in the neuroblastoma cells, TE671, is unlikely to be mediated via the 5-HT_3 receptor.

From a mechanistic point of view, three possibilities can be envisioned. First, because of its chemical structure, ondansetron may directly interact with one or more voltage-gated K^+-channels in the neuroblastoma TE671 cells and cause their inactivation or blockade. This may be analogous to the observation that the potent and selective 5-HT_3-receptor antagonist tropisetron, used at micromolar concentrations, blocks cardiac potassium, sodium and calcium currents, and prolongs the cardiac action potential duration (Williams et al 1985; Scholtysik 1987; Scholtysik et al 1988). Intrinsic K^+ channel-blocking activity of ondansetron and zatosetron, used at high doses, has also been implicated in the prolongation of the action potential duration in the canine heart (Williams et al 1991).

The fact that ondansetron, but not tropisetron or MDL 72222 shows a KCB activity in the ^{86}Rb efflux assay, indicates that the carbazolone and not the tropanyl pharmacophore may be responsible for conferring the KCB activity on ondansetron, as indicated by our observation (Table 1), that 2,3-dihydro-9-methyl-4(1H)-carbazolone moiety of ondansetron has a KCB activity (IC50 50 μM), close to that shown by ondansetron (IC50 25 μM). The presence of a protonated imidazole ring in ondansetron (Oxford et al 1992) may enhance the affinity of that compound for its receptor site, presumably by providing a positive charge. The imidazolyl moiety may also help to increase the lipophilicity of ondansetron, improving its interaction with the 5-HT_3 receptor (Oxford et al 1992), or with the voltage-gated K^+ channels. The potency of the carbazolone structure, as a KCB, decreases when the carbonyl and 9-methyl groups are removed to form a tetrahydrocarbazole pharmacophore (IC50 130 μM). This reduction in the KCB activity may parallel the report by Oxford et al (1992) that tetrahydrocarbazole is approximately one-hundredth as potent as a 5-HT_3 receptor antagonist than tetrahydrocarbazolone. Removal of the saturated 6-membered carbonyl ring from 2,3-dihydro-9-methyl-4(1H)-carbazolone (Table 1) yields 1-methylindole which proved to be much weaker KCB (IC50 500 μM). This may be partly attributed to a reduced lipophilicity in the resulting compounds. Evidently, on their own, the indole, cyclohexanone or 2-methylimidazole substructures of ondansetron have no detectable KCB activity up to 800 μM (Table 1).

A second mechanism which might underlie the KCB activity of ondansetron is that the compound could interact with a hitherto undescribed 5-HT_3 (or 5-HT_x)-receptor subtype in TE671 cells, with little or no affinity for other 5-HT_3 ligands tested in this work. This notion may be supported by several other reports. There are significantly different affinity constants for the same competitive 5-HT_3 antagonist such as MDL 72222 or tropisetron in different bioassay systems (Richardson & Engel 1986). The 5-HT_3-receptor antagonists tropisetron, renzapride and zacopride have significant affinities for the recently characterized 5-HT_4 receptor (Oxford et al 1992). Furthermore, in the neuroblastoma cells NG108-15, a chemical modification by *N*-bromosuccinimide of tryptophyl residues in the 5-HT_3 receptor molecule leads to a significant loss of binding affinity or the value of B_{max} displayed by zacopride and 2-methyl-5-HT. However, no significant changes in these parameters were observed for ondansetron or granisetron (Miquell et al 1991). These findings, and others (Wong et al 1993), may render support to the hypothesis that at least a sub-population of ondansetron binding sites in neuroblastoma cells may be distinct from the known 5-HT_3-receptor sites recognized by ligands such as zacopride, MDL 72222, or LY 278, 584. It might be suggested that there exists a heterogeneous population of 5-HT_x (including 5-HT_3) receptors having dual functions, namely as 5-HT_x receptors, as well as the voltage-gated K^+ channels.

Thirdly, ondansetron may interact with a receptor in TE671 cells, entirely unrelated to any 5-HT receptors or K^+ channels. This receptor may mediate a cascade of events leading to the inactivation of a voltage-gated K^+ channel. This possibility has also been raised for other 5-HT receptor ligands (Feuerstein & Hertting 1986; Kilpatrick et al 1990; Nogradi 1993).

In the present investigation, we used two 5-HT receptor-selective neurotoxins, 5,7- and 6,7-dihydroxytryptamine creatinine sulphate to deplete the target TE671 cells of 5-HT-binding sites. These reagents failed to affect the KCB activity of ondansetron, rendering further support to the hypothesis that the 5-HT receptor may not be involved in this process.

Although the association of the 5-HT_3 receptor with the ligand-gated potassium conductance has been demonstrated in several neuronal systems (Bobker & Williams 1990), the present work is the first reported observation of the association of that receptor and its selective antagonist, ondansetron, with the voltage-gated K^+ channels. Notwithstanding the observation that the carbazolone moiety appears to be mostly responsible for the KCB activity of ondansetron, it is possible that several mechanisms, including those outlined above, may play a role. An interplay of these mechanisms may have a net inhibitory effect on the voltage-gated K^+ channels in the neuroblastoma TE671 cells, as reflected in the rate of ^{86}Rb efflux.

At a first glance, our finding that ondansetron is a relatively weak KCB (IC50 25 μM), precludes such an activity from contributing to the in-vivo cognition enhancing properties of that compound due, presumably, to the enhancement of long-term potentiation of synaptic transmission (Lee et al 1986; Kondo et al 1992; Teyler & Discenna 1987). This apparently irreconcilable discrepancy between the in-vitro and the in-vivo potency of ondansetron is likely to be due to the relative insensitivity of the ^{86}Rb efflux assay, which rarely predicts the in-vivo potency of a

potassium-channel modulator. This notion is supported by several independent reports on the activity of the potassium-channel activator, cromakalim. In the rat and conscious renal hypertensive cat, this compound causes a rapid, significant and sustained fall in the blood pressure at 0·05–0·1 mg kg^{-1}, whereas it stimulates ^{86}Rb efflux from the rat portal vein or the rabbit mesenteric artery only when used at 10–50 μM (Weir & Weston 1986; Coldwell & Howlett 1987; Longman et al 1988; Buckingham et al 1989).

As a second possibility, although the metabolites of ondansetron, largely resulting from *N*-demethylation and hydroxylation at positions 6, 7 and 8, have little affinity for the 5-HT_3 receptor (Oxford et al 1992), these and other metabolites may have a markedly more significant KCB activity than ondansetron itself. Clearly, further investigation in this area is warranted.

Furthermore, the availability of radiolabelled ondansetron, together with the techniques of chemical cross linking, protein chemistry and molecular biology should clarify, at the molecular level, the identity of the receptor sites with which ondansetron interacts leading to the inhibition of the voltage-gated potassium channels.

References

Arner, L. S., Stallcup, W. B. (1981) Rubidium efflux from neural cell lines through voltage-dependent potassium channels. Dev. Biol. 83: 138–145

Barnes, J. M., Barnes, N. M., Costall, B., Naylor, R. H., Tyers, M. B. (1989) 5-HT_3 receptors mediate inhibition of acetylcholine release in cortical tissue. Nature 338: 762–763

Barnes, J. M., Costall, B., Coughlan, J., Domeney, A. M., Gerrard, P. A., Kelly, M. E., Naylor, R. J., Onaivi, E. S., Tomkins, D. M., Tyers, M. B. (1990) The effects of ondansetron, a 5-HT_3 receptor antagonist on cognition in rodents and primates. Pharmacol. Biochem. Behav. 35: 955–962

Ben Ari, Y., Cherubini, F., Aniksztejn, L., Roisin, M. P., Charriaut-Marlangue, C. (1989) Mechanism of induction of long term potentiation by the mast cell degranulating peptide. Pharmacopsychiatry 22: 107–110

Blandina, P., Goldfarb, J., Green, J. P. (1988) Activation of a 5-HT_3 receptor releases dopamine from rat striatal slice. Eur. J. Pharmacol. 155: 349–350

Bobker, D. H., Williams, J. T. (1990) Ion conductances affected by 5-HT receptors subtypes in mammalian neurons. Trends Neurosci. 13: 169–173

Buckingham, R. E., Hamilton, T. C., Howlett, D. R., Mootoo, S., Wilson, C. (1989) Inhibition by glibenclamide of the vasorelaxant action of cromakalim in the rat. Eur. J. Pharmacol. 97: 57–64

Coldwell, M. C., Howlett, D. R. (1987) Specificity of action of the novel antihypertensive agent, BRL34915, as a potent potassium channel activator. Biochem. Pharmacol. 36: 3663–3669

Costall, B., Naylor, R. J. (1992) The psychopharmacology of 5-HT_3 receptors. Pharmacol. Toxicol. 71: 401–415

Costall, B., Naylor, R. J., Tyers, M. B. (1990) The psychopharmacology of 5-HT_3 receptors. Pharmacol. Ther. 47: 181–202

Daniel, S., Malkowitz, L., Wang, H.-C., Beer, B., Blume, A. J., Ziai, M. R. (1991) Screening for potassium channel modulators by a high through-put 86-rubidium efflux assay in a 96-well microtiter plate. J. Pharmacol. Methods 25: 185–193

Feuerstein, T. J., Hertting, G. (1986) Serotonin (5-HT) enhances hippocampal noradrenaline (NA) release: evidence for facilitatory 5-HT receptors within the CNS. Naunyn Schmiedebergs Arch. Pharmacol. 333: 191–197

Harvey, A. L. (1993) Neuropharmacology of potassium ion channels. Med. Res. Rev. 13: 81–104

Henderson, G. (1990) Complexity of 5-HT pharmacology compounded by electrophysiological data. Trends Pharmacol. Sci. 11: 265–266

Hof, R. P., Hof, A., Novosel, D., Ziehut, W. (1993) Antiarrhythmic and hemodynamic effects of tropisetron in anesthetized rabbits. J. Cardiovasc. Pharmacol. 22: 499–505

Hoyer, D. (1991) The 5-HT receptor family: ligands, distribution and receptor-effector coupling. In: Rodgers, R. J., Cooper, S. J. (eds), 5-HT_{1A} Agonists, 5-HT_3 Antagonists and Benzodiazepines: their Comparative Behavioural Pharmacology. John Wiley & Sons, New York, pp 31–49

Kidd, E. J., Laporte, A. M., Langlois, X., Fattaccini, C.-M., Doyen, C., Lombard, M. C., Gozlan, H., Hamon, M. (1993) 5-HT_3 receptors in the rat central nervous system are mainly located on nerve fibers and terminals. Brain Res. 612: 289–298

Kilpatrick, G. J., Bunce, K. T., Tyers, M. B. (1990) 5-HT_3 receptors. Med. Res. Rev. 10: 441–475

Kondo, T., Ikenaka, K., Fujimoto, I., Aimoto, S., Kato, H., Ito, K. I., Taguchi, T., Morita, T., Kasai, M., Mikoshiba, K. (1992) K-Channel involvement in induction of synaptic enhancement by mast cell degranulating (MCD) peptide. Neurosci. Res. 13: 207–216

Lavertsky, E. P., Jarvik, L. F. (1992) A group of potassium-channel blockers-acetylcholine releasers: new potentials for Alzheimer's disease? A review. J. Clin. Psychopharmacol. 12: 110–118

Lee, W. L., Anwyl, R., Rowan, M. (1986) 4-Aminopyridine-mediated increase in long term potentiation in CA1 of the rat hippocampus. Neurosci. Lett. 70: 106–109

Longman, S. D., Clapham, J. C., Wilson, C., Hamilton, T. C. (1988) Cromakalim, a potassium channel activator: a comparison of its cardiovascular haemodynamic profile and tissue specificity with those of pinacidil and nicorandil. J. Cardiovasc. Pharmacol. 12: 535–542

Maricq, A. V., Peterson, A. S., Brake, A. J., Myers, R. M., Julius, D. (1991) Primary structure and functional expression of the 5HT_3 receptor, serotonin-gated ion channel. Science 254: 432–437

Miquel, M. C., Emerit, M. B., Gozlan, H., Hamon, M. (1991) Involvement of tryptophan residues in the specific binding of agonists/antagonists to 5-HT3 receptors in NG108-15 clonal cells. Biochem. Pharmacol. 42: 1453–1461

Nogradi, M. (1993) GYKI-46903. Drugs of the Future 18: 421–423

Oxford, A. W., Bell, J. A., Kilpatrick, G. J., Ireland, S. J., Tyers, M. B. (1992) Ondansetron and related 5-HT_3 antagonists: recent advances. Prog. Med. Chem. 29: 239–270

Richardson, B. P., Engel, G. (1986) The pharmacology and function of 5-HT_3 receptors. Trends. Neurosci. 9: 424–428

Scholtysik, G. (1987) Evidence for inhibition by ICS 205–930 and stimulation by BRL 34915 of K^+ conductance in cardiac muscle. Naunyn Schmiedebergs Arch. Pharmacol. 335: 692–696

Scholtysik, G., Imoto, Y., Yatani, A., Brown, A. M. (1988) 5-Hydroxytryptamine antagonist ICS 205–930 blocks cardiac potassium, sodium and calcium currents. J. Pharmacol. Exp. Ther. 245: 773–778

Teyler, T. J., Discenna, P. (1987) Long term potentiation. Annu. Rev. Neurosci. 4: 131–161

Toral, J., Hu, W., Yi, L., Barrett, J. E., Sokol, P. T., Ziai, M. R. (1994) Use of cultured human neuroblastoma cells in rapid discovery of the voltage-gated potassium-channel blockers. J. Pharm. Pharmacol. 46: 731–734

Tyers, M. B., Kilpatrick, G. J., Ireland, S. J., Oxford, A. W. (1993) Ondansetron (Zofran). Drug News and Perspectives 6: 308–315

Weir, S. W., Weston, A. H. (1986) The effect of BRL 34915 and nicorandil on electrical and mechanical activity and on 86-Rb efflux in rat blood vessels. Br. J. Pharmacol. 88: 121–128

Williams, F. M., Rothaul, A. L., Kane, K. A., Parratt, J. R. (1985) Antiarrhythmic and electrophysiological effects of ICS 205-930, an antagonist of k5-hydroxytryptamine at peripheral receptors. J. Cardiovasc. Pharmacol. 7:550-555

Williams, P. D., Cohen, M. L., Turk, J. A. (1991) Electrocardiographic effects of zatosetron and ondansetron, two 5-HT_3 receptor antagonists, in anesthetized dogs. Drug Dev. Res. 24: 277–284

Wong, E. H. F., Bonhaus, D. W., Lee, J. A., Wu, I., Loury, D. N., Eglen, R. M. (1993) Different densities of 5-HT3 receptors are labeled by [^{3}H] quipazine, [^{3}H] GR 65630 and [^{3}H] granisetron. Neuropharmacol. 32: 869–875