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3B but which 3B? And that's just one of the questions: the heterogeneity of human 5-HT₃ receptors

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Abstract

The 5-hydroxytryptamine 3 (5-HT₃) receptor is expressed widely in the central and peripheral nervous systems, where it mediates or modulates a wide range of physiological processes. The receptor is targeted by drugs administered for nausea and/or emesis and irritable bowel syndrome and has been proposed as a potential drug target in various psychiatric disorders. The 5-HT₃ receptor is a pentameric ligand-gated ion channel and belongs to the Cys-loop receptor family. In contrast to the immense heterogeneity characterizing other Cys-loop receptors, native 5-HT₃ receptors historically have been considered a much more homogenous receptor population. However, the recent discovery of additional 5-HT₃ subunits and the dawning realization that central and peripheral 5-HT₃ receptor populations might comprise several subtypes characterized by distinct functional properties has emphasized the complexity of human 5-HT₃ receptor signaling. In this review potential implications of these findings and of the entirely new layer of interindividual diversity introduced to the 5-HT₃ receptor system by genetic variations will be outlined.

Introduction

Serotonin (5-hydroxytryptamine, 5-HT) is a major neuro-transmitter in both central and peripheral nervous systems (CNS and PNS, respectively), where it plays essential roles in numerous basic functions including sleep, mood, libido, appetite, respiration, nociception, cardiovascular function and thermoregulation. In vertebrates serotonin mediates its physiological effects through six classes of 7-transmembrane receptors and a single class of ionotropic receptors, the 5-HT₃ receptors. The 5-HT₃ receptors belong to the 'Cys-loop receptor' superfamily, which also includes nicotinic acetylcholine (nACh) receptors, γ -aminobutyric acid type A (GABA_A) receptors and glycine receptors [1–4]. The receptors are homomeric and heteromeric complexes composed of five subunits, where neurotransmitter binding to the extracellular domain of the complex elicits the opening of a transmembrane

ion channel and the flux of ions. Being cationic ion channels permeable to Na^+ , Ca^{2+} and K^+ , activation of the 5-HT₃ receptors results in depolarization of the cell [1].

The 5-HT₃ receptors are distributed throughout the CNS and PNS, where they mediate or modulate a wide range of processes. In the CNS postsynaptic 5-HT₃ receptors mediate the fast synaptic response to serotonin, and presynaptic receptors regulate the synaptic release of serotonin and several other neurotransmitters [5,6]. The 5-HT₃ receptors are established drug targets because competitive antagonists targeting central and gastrointestinal 5-HT₃ receptors are used in the clinical treatment of the nausea and/or emesis associated with radio- and chemotherapy treatment of cancer, postoperative nausea and vomiting, and irritable bowel syndrome [7,8]. Furthermore, central 5-HT₃ receptors have been proposed as potential targets for the treatment of various psychiatric disorders, cognitive dysfunctions, drug abuse and withdrawal, and certain forms of pain [8].

In contrast to the immense heterogeneity characterizing the nACh and GABA_A receptor systems, where a total of 17 and 19 different subunits, respectively, form a substantial number of physiologically relevant subtypes characterized by distinct expression patterns, synaptic localizations and signaling characteristics [2,3], native 5-HT₃ receptors historically have been considered a much more homogenous receptor population. In this review we will outline how new levels of molecular diversity within the human 5-HT₃ receptor family have been disclosed in recent years and discuss some of the physiological implications of these findings.

The growing 5-HT₃ receptor family

The cloning in 1991 of a 5-HT₃ subunit exhibiting widespread expression in CNS and PNS and capable of forming functional receptors seemed to fit the notion of a simple receptor system [9]. However, the biophysical properties displayed by the homomeric receptors formed by this 5-HT_{3A} subunit were very different from those of native 5-HT₃ receptors [10]. These discrepancies were largely accounted for with the cloning of a second subunit, 5-HT_{3B}, almost a decade later [11,12]. Although unable to form functional receptors on its own, the 5-HT_{3B} subunit coassembles with the 5-HT_{3A} subunit to form the heteromeric 5-HT_{3AB} receptor subtype. The 5-HT_{3AB} receptor exhibits biophysical properties much more similar to those of native 5-HT₃ receptors: a significantly higher single-channel current conductance (7–16 pS) than the homomeric 5-HT_{3A} receptor (sub-pS) and a significantly lower Ca^{2+} permeability [11,13]. Alternative splicing, and also possibly alternative transcription start sites, contribute to the heterogeneity of the two subunits at a molecular level (Figure 1a). The human 5-HT_{3A} splice variants 5-HT_{3AL} and 5-HT_{3AT} do not form functional homomeric receptors but appear to form heteromeric complexes with 5-HT_{3A} characterized by reduced (5-HT_{3A}/5-HT_{3AL}) and larger (5-HT_{3A}/5-HT_{3AT}) cation fluxes compared to that of the homomeric 5-HT_{3A} receptors [14]. In the case of the 5-HT_{3B} subunit, brain-specific promoters for *HTR3B* recently have been proposed to give rise to two alternative brain transcripts, BT-1 and BT-2 [15]. The predicted BT-1 protein only differs from the ‘canonical’ 5-HT_{3B} subunit identified in peripheral tissues [11,12] when it comes to five of the first six residues of its N-terminal domain, whereas the predicted BT-2 protein lacks a large segment of this domain [15] (Figure 1a). Neither of the two CNS transcripts are

predicted to contain a signal peptide, however, and it remains to be determined whether they are expressed as proteins and capable of forming functional heteromeric complexes with 5-HT_{3A} at the cell surface.

The recent discovery of three human *HTR3*-like genes, *HTR3C*, *HTR3D* and *HTR3E*, encoding the putative 5-HT_{3C}, 5-HT_{3D} and 5-HT_{3E} subunits, has further added to the multiplicity of the human 5-HT₃ system (Figure 1a), whereas genes encoding orthologous subunits have not been found in rodents [16–18]. At present the existence of two 5-HT_{3D} isoforms have been proposed, 5-HT_{3D(short)} [17] and 5-HT_{3D(long)} (GenBank accession number AJ437318; M. Brüss, unpublished), and in addition to the 5-HT_{3E} and 5-HT_{3Ea} isoforms originally identified [17,18], a third 5-HT_{3E} isoform, termed 5-HT_{3Eb} in this review, recently has been reported (GenBank accession number EU165354, J.D. Holbrook *et al.*, unpublished) (Figure 1a). Analogous to 5-HT_{3B}, the 5-HT_{3C}, 5-HT_{3D(short)}, 5-HT_{3E} and 5-HT_{3Ea} subunits are not transported to the cell membrane in heterologous expression systems unless they are coexpressed with the 5-HT_{3A} subunit [19]. The signaling properties of the 5-HT_{3AC}, 5-HT_{3AD(short)}, 5-HT_{3AE} and 5-HT_{3AEa} receptor combinations have been found only to differ slightly from those exhibited by the homomeric 5-HT_{3A} receptor in a rather coarse luminescence-based assay [19]. However, serotonin has been reported to elicit significantly reduced currents in *Xenopus laevis* oocytes coexpressing 5-HT_{3A} and 5-HT_{3C} subunits compared to 5-HT_{3A}-receptor-expressing oocytes [16], and future detailed electrophysiological studies might reveal additional differences between 5-HT₃ receptor complexes containing these new subunits and 5-HT_{3A} and 5-HT_{3AB} receptors.

Heterogeneity of native 5-HT₃ receptors

The complexity introduced to the 5-HT₃ receptor system in 1999 with the discovery of 5-HT_{3B} raised the question whether native 5-HT₃ receptors were a heterogeneous population composed of several subtypes with distinct physiological roles. Whereas the 5-HT_{3A} and 5-HT_{3B} subunits are coexpressed in several tissues in the PNS, making the heteromeric 5-HT_{3AB} receptor a major peripheral subtype, the molecular composition of central 5-HT₃ receptors surprisingly still is a matter of controversy. In their 2002 *Trends in Pharmacological Sciences* Update entitled ‘5-HT₃ receptors in the CNS: 3B or not 3B?’, parodying the soliloquy of a famed gloomy Danish prince, van Hooft and Yakel set forth a rather black-and-white proposition regarding 5-HT₃ receptors in CNS [20]. The 5-HT_{3B} subunit was proposed not to be a major determinant of 5-HT₃ receptor signaling in the CNS, and, thus, the central 5-HT₃ receptor population was claimed to be composed predominantly of homomeric 5-HT_{3A} receptors with the possible addition of complexes between 5-HT_{3A} and other 5-HT₃ subunits not identified at the time or other assembly partners, such as the α4 nACh receptor subunit [20,21]. However, most of the studies published at the time and in the following years strongly indicate that the 5-HT_{3B} subunit indeed is expressed in several CNS regions (summarized in Table 1). The ‘3B or not 3B in CNS’ controversy has been ascribed to species differences because reverse transcriptase-polymerase chain reaction (RT-PCR) and *in situ* hybridization studies using rat brain tissue have been unable to detect 5-HT_{3B} transcripts, whereas the subunit consistently has been detected in human CNS regions (Table 1). However, studies failing to detect 5-HT_{3B} transcripts in specific rat brain regions have nevertheless detected the subunit in whole-brain tissue [22,23], and furthermore the

absence of 5-HT_{3B} mRNA from the rat hippocampus [22–24] is contrasted by the detection of the rat 5-HT_{3B} protein in three immunohistochemistry studies using different 5-HT_{3B} antibodies [25–27] (Table 1). Studies attempting to identify the native 5-HT₃ receptor in rodent hippocampal neurons [23,28] and in murine neuro blastoma cell lines [29–32] based on its conductance properties in single-channel recordings have observed sub-pS conductances indicative of 5-HT_{3A} receptors as well as conductances similar to those displayed by recombinant 5-HT_{3AB} receptors (Table 1). The differences in the conductance properties displayed by native 5-HT₃ receptors in these studies are somewhat reminiscent of the discrepancies observed between the various studies of 5-HT_{3B} expression (Table 1).

We believe that the data presented in Table 1 convincingly make the case that 5-HT_{3B} is indeed expressed in the CNS. All experimental evidence suggests that the subunit is expressed in the human brain, and the observed discrepancies in studies of the rodent CNS are likely to arise from differences in the protocols used for the RT-PCR, *in situ* hybridization and immunohistochemistry experiments in these studies and from different sensitivities of these techniques. Thus, the discrepancies could just as well be a reflection of the presence of both 5-HT_{3A} and 5-HT_{3AB} receptors as of absence of the 5-HT_{3B} subunit from the CNS. In particular the presence of 5-HT_{3B} in the hippocampus is well documented, whereas the reproducibility between different RT-PCR studies when it comes to other human CNS regions expressing 5-HT_{3B} mRNA admittedly has not been overwhelming (Table 1). These discrepancies could arise from low expression levels of the mRNA, from the subunit being expressed in subpopulations of neurons only or from different sensitivities of the RT-PCR protocols used in the various studies. The expression pattern of the 5-HT_{3B} protein in the hippocampus overlaps to a great extent with that of the 5-HT_{3A} subunit [25–27,33], but at the subcellular level expression of the 5-HT_{3A} subunit in the absence of 5-HT_{3B} also has been observed [27]. Furthermore, although the demonstration of high-conductance channels in single-channel recordings on rodent hippocampal neurons cannot be taken as an unequivocal proof of 5-HT_{3B} expression, the divergent findings of the electrophysiological recordings summarized in Table 1 seem to support the notion of both 5-HT_{3A} and ‘non-5-HT_{3A}’ receptors, that is 5-HT_{3AB} receptors and/or other heteromeric 5-HT₃ receptor complexes, being formed in the rodent CNS. Considering the distinct biophysical properties of 5-HT_{3A} and 5-HT_{3AB} receptors, differential subcellular distribution of two subtypes having specialized functions could be nature's way of fine-tuning 5-HT₃ receptor signaling in certain neurons. Hopefully, future studies will be able to unravel the respective roles of these subtypes in a synaptic setting.

In view of the prevalence of 5-HT_{3A} and 5-HT_{3AB} receptors in the human CNS and PNS, it is important to keep in mind that both 5-HT_{3A} and 5-HT_{3B} subunits are subjected to splice variation, and, thus, the specific molecular composition of these two major physiological subtypes might vary from tissue to tissue. Being essential for the assembly of functional 5-HT₃ receptors, the presence of the canonical 5-HT_{3A} subunit in a given cell is an absolute prerequisite for signaling. In contrast to the widespread expression of 5-HT_{3A}, however, the 5-HT_{3AL} and 5-HT_{3AT} isoforms display more restricted expression patterns. Whereas transcripts of both isoforms have been detected in the hippocampus and amygdala, they appear to be absent from small intestine tissue, and only 5-HT_{3AT} mRNA has been detected

in placenta [14]. Considering that 5-HT_{3AL} and 5-HT_{3AT} subunits confer altered cation flux properties to the 5-HT_{3A} receptor in heterologous expression systems [14], the differential expression of these ‘minor’ isoforms might constitute a refined mechanism modulating the contributions of the ‘5-HT_{3A}-component’ to 5-HT₃ receptor signaling in different tissues. Interestingly, the recently proposed existence of two CNS-specific 5-HT_{3B} isoforms suggests that isoforms of this subunit might be even more differentially expressed than in the case of the 5-HT_{3A} subunit [15]. Based on quantification of the alternative brain transcripts, BT-1 has been proposed to be the predominant 5-HT_{3B} isoform in the CNS, and because the predicted BT-1 protein is almost identical to the canonical 5-HT_{3B}, the putative BT-2 isoform constitutes the major potential molecular difference between central and peripheral 5-HT_{3B} subunits [15]. Because of its lack of a segment of the N-terminal domain known to be important for Cys-loop receptor assembly and function, it has been proposed that the BT-2 protein could exert a dominant-negative function on 5-HT₃ receptor signaling [15]. However, considering that the 5-HT_{3D(short)} subunit appears able to form functional receptors with the 5-HT_{3A} subunit [19], BT-2 possibly also could be integrated into receptors complexes expressed at the cell surface. In any case, elaborate speculations about the roles of the BT-1 and BT-2 5-HT_{3B} isoforms in the CNS should await studies confirming that the transcripts are indeed translated into proteins.

The potential impact of the newly identified 5-HT_{3C}, 5-HT_{3D} and 5-HT_{3E} subunits on human 5-HT₃ receptor pharmacology obviously also is very dependent on whether or not they are expressed as proteins *in vivo*. Analogous to the 5-HT_{3A} and 5-HT_{3B} transcripts, 5-HT_{3C} mRNA appears to be expressed throughout the body, including adult brain, gastrointestinal, muscle and lung tissues [16,17]. By contrast, 5-HT_{3E} transcripts have been found exclusively in the colon and intestine, and 5-HT_{3D} also is predominantly localized in gastrointestinal tissues [17,18]. The gut-specific expression of the 5-HT_{3D} and 5-HT_{3E} subunits has prompted speculations about their potential roles in intestinal disorders like irritable bowel syndrome [17,18]. However, further investigations into the physiological functions of the 5-HT_{3C}, 5-HT_{3D} and 5-HT_{3E} subunits seem to be complicated by the absence of orthologs of these subunits in rodents.

Provided that the three novel 5-HT₃ subunits are expressed *in vivo* and are able to participate in the formation of functional receptors, native 5-HT₃ receptor populations could be highly heterogeneous (Figure 1b). In addition to the possible formation of ‘simple’ heteromeric complexes between 5-HT_{3A} and each of the complementary subunits in the 5-HT₃ receptor family, the existence of more than two 5-HT₃ subunits opens up for the formation of more ‘complex’ 5-HT₃ receptor pentamers, analogously to what is known from nACh and GABA_A receptors [2,3]. In an atomic force microscopy study, the 5-HT_{3A}:5-HT_{3B} ratio of 5-HT_{3AB} receptors expressed in mammalian cells has been determined to be 2:3 [34], and although the presence of two 5-HT_{3A} subunits probably is required to form a functional 5-HT₃ receptor, the three complementary subunits do not necessarily have to be identical. Thus, the 5-HT_{3ABC} receptor could be a physiologically relevant sub-type in numerous tissues, and a plethora of subtypes such as 5-HT_{3ABE}, 5-HT_{3ACE} and 5-HT_{3ABD} receptors or even 5-HT_{3ABDE} and 5-HT_{3ABCE} receptors could exist in the gut (Figure 1b). Finally, heteromeric 5-HT₃ receptors with stoichiometries other than 2:3 could exist in native tissues,

as exemplified for the 5-HT_{3AB} receptor in Figure 1b. Interestingly, (α 4)₂(β 2)₃ and (α 4)₃(β 2)₂ nACh receptors have exhibited significantly different agonist sensitivities, desensitization kinetics and Ca²⁺ permeabilities in heterologous expression systems, a molecular diversity claimed to have physiological relevance in nicotine addiction [35,36]. Analogously, the existence of alternative stoichiometries of heteromeric 5-HT₃ receptors with distinct signaling properties would increase the heterogeneity of native receptor populations.

Interindividual differences in the 5-HT₃ receptor system

The sequencing of the human genome has revealed an extensive genetic variability of the human race and has helped to unveil causal relations between genetic variations and diseases or therapeutic treatment efficacies. In the 5-HT₃ receptor field, variations in the regulatory regions of *HTR3A* and *HTR3B* have been shown to alter expression levels of the 5-HT_{3A} and 5-HT_{3B} subunits [37,38], and several single-nucleotide polymorphisms (SNPs) in the coding regions of the two genes also impact 5-HT₃ receptor expression and/or signaling significantly (K. Krzykowski *et al.*, unpublished) [39–41]. Most of these variants are found in one or a few ethnic groups and in low frequencies worldwide. The impact of pharmacogenetics has always been a numbers game, however, which makes the *HTR3B* variant encoding an Y129S variation in the 5-HT_{3B} subunit (c.386C>A, NCBI dbSNP rs1176744) highly interesting. This variant has been found in such high allelic frequencies (0.17–0.43) in all ethnic groups investigated so far that a significant fraction of any ethnic group will be carriers of the variant allele (Figure 2a). We recently have reported that 5-HT_{3AB} receptors composed of 5-HT_{3A} and 5-HT_{3B}(Y129S) subunits display 20- and 10-fold slower deactivation and desensitization kinetics, respectively, than wild-type (WT) 5-HT_{3AB} receptors (Figure 2c), properties that most likely arise from a 7-fold prolonged mean opening duration of the variant-containing channels [41]. Furthermore, coexpression of the 5-HT_{3A} subunit with both WT 5-HT_{3B} and 5-HT_{3B}(Y129S) subunits also yields receptors with significantly enhanced signaling characteristics compared with those of the WT 5-HT_{3AB} receptor, indicating that 5-HT_{3AB} receptor signaling in heterozygous carriers of the variant allele (Y/S) also could be significantly augmented compared to homozygous ‘WT’ allele carriers (Y/Y) [41].

Being a subunit in a monoamine-activated Cys-loop receptor, there is plenty of circumstantial evidence suggesting that a high-frequency 5-HT_{3B} variant could be of therapeutic interest. Several SNPs in adrenoceptor genes are associated with cardiovascular and/or asthma phenotypes, and genetic variations in receptors, transporters and enzymes in the dopamine and serotonin systems seem to be contributing factors to the etiology of psychiatric disorders [42,43]. Furthermore, nonsynonymous SNPs in genes for other Cys-loop receptors have been found to be causative of channelopathies such as epilepsies and startle disease [44]. When it comes to the 5-HT₃ receptor field, several polymorphisms in *HTR3A*, *HTR3B* and *HTR3C* have been shown to be associated with specific phenotypes and/or drug responses [45–57]. Considering its high allelic frequency, the 5-HT_{3B}(Y129S) variant has been the focus of relatively few association studies in which the link between the variant and the nausea and/or emesis or gastrointestinal side effects induced by antidepressants or chemotherapy [48,54,57–59], various psychiatric disorders [51,53,60] and

fibromyalgia [61] have been investigated. Some of these studies have identified an association or a trend toward an association between the variant and a certain disorder or drug response [48,51,53,54,59], whereas other studies have not [57,58,60,61]. In view of the limited experimental data available we will refrain from speculations about potential links between the variant and different disorders. However, pronounced differences in the signaling properties of 5-HT_{3B}-containing 5-HT₃ receptors between individuals could potentially translate into differential effects on the various physiological functions regulated by the receptors, and, as such, the contributions of this individual factor to 5-HT₃ receptor functions should be considered. For example, could the enhanced signaling characteristics of 5-HT_{3B}(Y129S)-containing 5-HT₃ receptors cause altered thresholds for development of nausea and/or emesis or altered responses to 5-HT₃ receptor-based antiemetics in homozygous and heterozygous carriers of the variant allele (S/S and Y/S) compared to individuals homozygous for the 'WT' allele (Y/Y) (Figure 3)? And how will slowly desensitizing 5-HT_{3B}(Y129S)-containing 5-HT₃ receptors influence the activity in the serotonergic synapse? The actual impact of the observed signaling differences between WT 5-HT_{3B}- and 5-HT_{3B}(Y129S)-containing 5-HT_{3AB} receptors *in vitro* on 5-HT₃ receptor signaling *in vivo* obviously needs to be elucidated before such questions can be addressed. Until then the sheer possibility that this HTR3B variant could have implications for the overall serotonin-mediated neurotransmission in different individuals is highly intriguing.

Conclusions

The widespread expression of 5-HT₃ receptors in the human body and the considerable therapeutic prospects in the serotonin system make it important to understand the molecular composition of native 5-HT₃ receptors and their contributions to serotonergic signaling. Although the concept of a single major CNS subtype and one or two subtypes in peripheral tissues certainly would be refreshing in its simplicity compared to the plethora of 7-trans-membrane serotonin receptors, it seems that 5-HT₃ receptor signaling, at least in humans, also might be orchestrated by numerous subtypes expressed throughout the body. The newfound molecular diversity in the human 5-HT₃ receptor system calls for investigations into the physiological functions maintained by the respective sub-types, and it will be important to establish the roles, if any, of the newly identified additional 5-HT₃ subunits and the putative brain-specific 5-HT_{3B} isoforms *in vivo*. This, in turn, raises the same need for truly subtype-selective ligands as craved for in the nACh and GABA_A receptor fields for decades. Furthermore, the observed dissimilarities when it comes to the identity and the distribution of the subunits forming the native 5-HT₃ receptors in different species underlines the need for caution when drawing conclusions about human 5-HT₃ receptor pharmacology from rodent studies. Finally, the realization that 5-HT₃ receptor signaling might even vary significantly within the human race as a result of genetic variation introduces an entirely new level of complexity to the field.

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Figure 1.

The human 5-HT₃ receptor family. **(a)** The human 5-HT₃ subunits. **5-HT_{3A}**: compared to the canonical 5-HT_{3A}, 5-HT_{3AL} has a 32-amino-acid-residue insertion into the extracellular loop between transmembrane regions 2 and 3, and 5-HT_{3AT} has a 57-residue insertion (followed by a stop codon) after transmembrane region 1. **5-HT_{3B}**: the 5-HT_{3B} BT-1 isoform only differs from the canonical 5-HT_{3B} in five of the first six amino acid residues of its N-terminal domain, whereas the 5-HT_{3B} BT-2 isoform lacks the first 77 residues of this domain compared to the mature canonical 5-HT_{3B} protein. Neither BT-1 nor BT-2 is predicted to contain a signal peptide. The localization of the Y129 residue in 5-HT_{3B} is indicated. **5-HT_{3D}**: The 5-HT_{3D(short)} and 5-HT_{3D(long)} isoforms possess N-terminal domains of 57 and 230 amino acid residues, respectively. **5-HT_{3E}**: 5-HT_{3Ea} only differs from the 5-HT_{3E} in the first 22 amino acid residues of its N-terminal domain, whereas 5-HT_{3Eb} has a 15-amino acid residue deletion as well as a 26-amino acid residue insertion in its N-terminal domain compared to 5-HT_{3E}. **GenBank accession numbers:** D49394 (canonical 5-HT_{3A}), AJ003078 (5-HT_{3AL}), AJ003080 (5-HT_{3AT}), AF080582 (canonical 5-HT_{3B}), AF459285 (5-HT_{3C}), AJ437318 (5-HT_{3D(long)}), AY159812 (5-HT_{3D(short)}), AY159813 (5-HT_{3E}), DQ644022 (5-HT_{3Ea}) and EU165354 (5-HT_{3Eb}). **(b)** The

heterogeneity of human 5-HT₃ receptors. Examples of ‘simple’ heteromers, ‘complex’ heteromers composed of more than two different subunits and 5-HT_{3AB} receptor heteromers characterized by different subunit arrangements and stoichiometries. For simplicity, the various isoforms of the different subunits are not included in this figure. Abbreviations: amino acids, aa; transmembrane 1, TM1.

Figure 2. Interindividual 5-HT₃ receptor diversity arising from genetic variation: the 5-HT_{3B}(Y129S) variant. **(a)** Distribution of individuals homozygous for the 'WT' and variant alleles (Y/Y and S/S, respectively) and heterozygous individuals (Y/S) in five major populations. The figure is based on NCBI's dbSNP build 127, submission numbers ss48404756 and ss23605381. CEPH, Centre d'Etude du Polymorphisme Humain. **(b)** The heteromeric 5-HT_{3AB} receptor complexes formed in individuals homozygous for the 'WT' and variant alleles (Y/Y and S/S, respectively) and in heterozygous individuals (Y/S). **(c)** 5-HT_{3B}(Y129S)-containing 5-HT_{3AB} receptors exhibit significantly slower deactivation (left) and desensitization (right) kinetics than WT 5-HT_{3AB} receptors. Traces are from whole-cell recordings on HEK293 cells transiently expressing the two receptors and are reproduced from Ref. [41] with permission [Copyright (2008) National Academy of Sciences, U.S.A.]

Figure 3.

The roles of 5-HT₃ receptors in the neurocircuitry associated with emesis and the potential impact of the 5-HT_{3B}(Y129S) variant. **(a)** Serotonin is released from enterochromaffin (EC) cells in the intestinal mucosa as a result of different luminal stimuli, including chemotherapy and radiation treatment, and activates 5-HT₃ receptors (and other serotonin receptors) in abdominal vagal afferents. **(b)** Activation of abdominal vagal afferents stimulates neurons in the area postrema (AP) and nucleus tractus solitarius (NTS), the emetic center of the medulla oblongata of the hindbrain [7]. Carriers of the 5-HT_{3B}(Y129S) variant potentially could have altered signals coming from 5-HT_{3AB} receptors expressed in vagal afferent neurons, in nodose ganglia neurons (known to express 5-HT_{3B}) that innervate NTS and in the AP and NTS. Part (b) of the figure is inspired by a similar figure in Ref. [7].

Table 1

The findings of studies of 5-HT_{3B} mRNA and protein expression in CNS and of electrophysiology studies of cultured rodent hippocampal neurons and murine neuroblastoma cell lines

Species	Findings	Technique	Refs
5-HT_{3B} mRNA			
Rat	Not detected in hippocampus (CA1 interneurons) but detected in whole brain	Single-cell RT-PCR	[23]
Rat	Not detected in pyramidal neurons and interneurons from neocortex but detected in whole brain	Single-cell RT-PCR	[22]
Rat	Not detected in whole brain, anterior olfactory nucleus, frontal cortex, striatum or hippocampus	RT-PCR	[24]
Rat	Not detected in hippocampus, visual cortex or entorhinal cortex	<i>In situ</i> hybridization	[24]
Rat	Detected in amygdala	–	[13]
Mouse	Detected in neuroblastoma cell lines NG108–15, NCB-20 and N1E-115	Northern blot	[13]
Mouse	Detected in whole brain	RT-PCR	[62]
Human	Detected in whole brain, amygdala, caudate nucleus, hippocampus and thalamus	Northern blot	[11]
Human	Detected in whole brain, cerebral cortex, amygdala and hippocampus but not in cerebellum, caudate nucleus, putamen, medulla, corpus callosum substantia nigra or thalamus	Semiquantative PCR	[12]
Primates	Detected in amygdala, entorhinal cortex and cerebral cortex	RT-PCR <i>in situ</i> hybridization	[12]
Human	Detected in caudate putamen but not in whole brain, amygdala, hippocampus or thalamus	RT-PCR	[17]
Human	Detected in hippocampus, striatum and thalamus but not in amygdala	RT-PCR	[63]
Human	Detected in hippocampus	RT-PCR	[33]
Human	Detected in whole brain, amygdala, hippocampus and caudate nucleus but not in pons, hypothalamus or medulla oblongata	Real-time RT-PCR	[15]
5-HT_{3B} protein			
Rat	Detected in hippocampus, predominantly interneurons	Immunocytochemistry	[25]
Rat	Detected in hippocampus, predominantly in pyramidal and molecular layers	Immunohistochemistry	[27]
Rat	Detected in CA1 and dentate gyrus (but not in CA2 and CA3) in hippocampus and in scattered neurons in cerebral cortex	Immunohistochemistry	[26]
Human	Detected in hippocampus, predominantly pyramidal neurons in CA2 and CA3, presumably glutamatergic neurons, identical distribution pattern as 5-HT _{3A}	Immunohistochemistry	[33]
Electrophysiology			
Mouse/rat	Conductances of 5-HT ₃ receptors in hippocampal neurons were similar to those obtained in peripheral neurons (and to those of recombinant 5-HT _{3AB} receptors)	Single-channel recordings	[28]
Rat	Absence of observable single-channel currents in rat hippocampal interneurons (indicative of 5-HT _{3A} receptors)	Single-channel recordings	[23]
Mouse	Small inward currents induced by serotonin in N1-E115 and NCB-20 cells, which could not be resolved into discrete single channel events. Conductance estimated to be 0.3 pS (by fluctuation analysis of the currents)	Single-channel recordings	[29]
Mouse	Small inward currents induced by serotonin in N1-E115 cells, which could not be resolved into discrete single channel events. Conductance estimated to 0.4–0.6 pS (by fluctuation analysis of the currents)	Single-channel recordings	[30]
Mouse	Channel conductances of ~12 pS and ~4 pS in undifferentiated and differentiated NG108-15 cells, respectively	Single-channel recordings	[32]
Mouse	Channels conductances of 6–27 pS in N1E-115 cells, depending on the phosphorylation state of the receptor	Single-channel recordings	[31]