



Neuronal localization of the 5-HT₂ receptor family in the amygdaloid complex

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The amygdaloid complex (or amygdala), a heterogeneous structure located in the medial portion of the temporal lobe, is composed of deep, superficial, and “remaining” nuclei. This structure is involved in the generation of emotional behavior, in the formation of emotional memories and in the modulation of the consolidation of explicit memories for emotionally arousing events. The serotonergic fibers originating in the dorsal and medial raphe nuclei are critically involved in amygdalar functions. Serotonin (5-hydroxytryptamine, 5-HT) regulates amygdalar activity through the activation of the 5-HT₂ receptor family, which includes three receptor subtypes: 5-HT_{2A}, 5-HT_{2B}, and 5-HT_{2C}. The distribution and the functional activity of the 5-HT₂ receptor family has been studied more extensively than that of the 5-HT_{2A} receptor subtypes, especially in the deep nuclei. In these nuclei, the 5-HT_{2A} receptor is expressed on both pyramidal and non-pyramidal neurons, and could play a critical role in the formation of emotional memories. However, the exact role of the 5-HT_{2A} receptor subtypes, as well as that of the 5-HT_{2B} and 5-HT_{2C} receptor subtypes, in the modulation of the amygdalar microcircuits requires additional study. The present review reports data concerning the distribution and the functional roles of the 5-HT₂ receptor family in the amygdala.

Keywords: amygdaloid complex, pyramidal neurons, GABAergic interneurons, GABAergic projection neurons, serotonin, 5-HT_{2A} receptor, 5-HT_{2B} receptor, 5-HT_{2C} receptor

INTRODUCTION

The amygdaloid complex (or amygdala), a heterogeneous structure located in the medial portion of the temporal lobe, is involved in multiple tasks, such as the generation of emotional behavior, formation of emotional memories related to fear and anxiety and modulation of the consolidation of explicit memories for emotionally arousing events (Aggleton, 2000; Whalen and Phelps, 2009). Several neuromodulators, including serotonin, are critical for amygdalar functions. Many neurological and psychiatric diseases, especially affective disorders, are characterized by a dysfunction of the amygdaloid complex and the serotonergic system (Sanders and Shekhar, 1995; Jasnow and Huhman, 2001; Manji et al., 2001; Amaral, 2002; Braga et al., 2002; Hariri et al., 2002; Pralong et al., 2002; Rainnie et al., 2004; Canli et al., 2005; Keele, 2005; Kim et al., 2005; Rodrigues Manzanares et al., 2005; Hariri and Holmes, 2006; Shin et al., 2006; Van Nobelen and Kokkinidis, 2006). Selective serotonin reuptake inhibitors (SSRIs) are effective in the treatment of a variety of psychiatric diseases, such as anxiety disorders, in which the amygdaloid complex may play a role. Accordingly, it has been shown that the acquisition of auditory fear conditioning in the rat was enhanced by the SSRI citalopram when administered in acute cases and reduced when administered in chronic cases; indeed, fear conditioning is known to be a model of emotional learning in which amygdaloid circuits play an important role (Burghardt et al., 2004). Selective serotonin reuptake inhibitors also reduce conditioned fear through its effect on the amygdala (Inoue et al., 2004). Moreover, co-administration of serotonin receptor agonists with paroxetine and

venlafaxine could enhance the therapeutic effects of these drugs (Dhonnchadha et al., 2005).

ANATOMICAL ORGANIZATION AND MAJOR CELL TYPES OF THE AMYGDALA

The amygdala is composed of numerous nuclei and areas with different cytoarchitectonic, chemoarchitectonic, and connectional characteristics. In particular, this structure is composed of deep, superficial, and “remaining” nuclei (or areas) (Pitkänen, 2000; Pitkänen and Kempainen, 2002). The deep nuclei include the lateral nucleus, the basal nucleus, the accessory basal nucleus, and the paralaminar nucleus (especially in primates). The lateral, basal, and accessory basal nuclei constitute the basolateral amygdala. The superficial nuclei include the anterior cortical nucleus, the nucleus of the lateral olfactory tract, the bed nucleus of the accessory olfactory tract, the medial nucleus and the posterior cortical nucleus. The remaining nuclei consist of the anterior amygdaloid area, the central nucleus, the intercalated nuclei and the amygdalohippocampal area (Pitkänen, 2000; Pitkänen and Kempainen, 2002). Each nucleus can be partitioned into different subdivisions, as reported in **Table 1** and **Figure 1**.

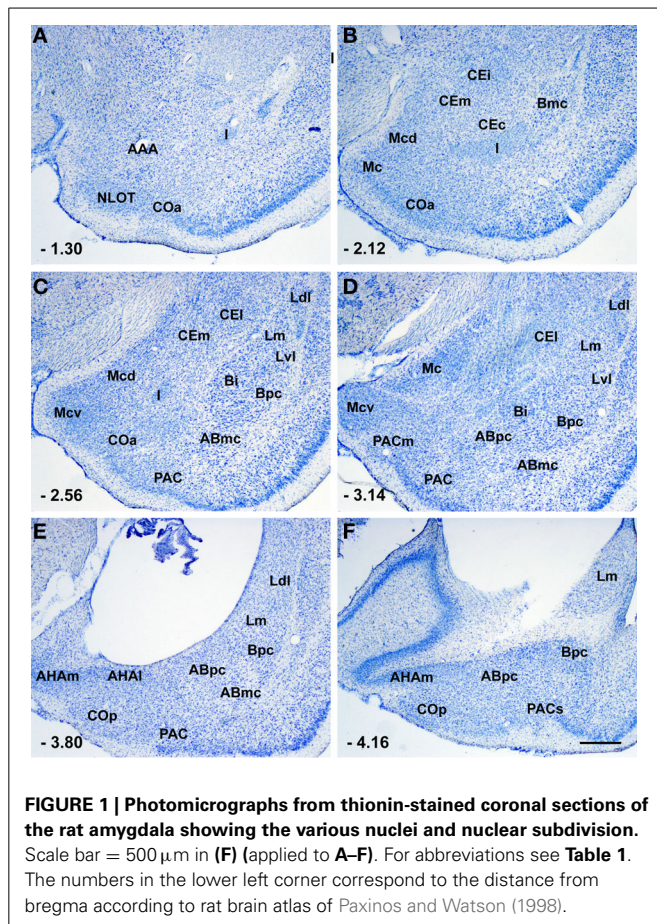
The deep nuclei consist of two types of neurons: excitatory (glutamatergic) pyramidal cells and inhibitory (GABA[γ-aminobutyric acid]ergic) non-pyramidal neurons (McDonald, 1992, 1998; Sah et al., 2003; Spanpanato et al., 2011). Pyramidal cells have spiny dendrites, form nearly 80% of the total cell population, and act as projection neurons (McDonald, 1992, 1998; Sah et al., 2003). Non-pyramidal neurons are spine-sparse or aspiny

Table 1 | Nuclei and nuclear subdivisions of the rat, the monkey and the human amygdala (modified from Pitkänen and Kempainen, 2002).

Nuclei	Rat subdivisions	Monkey subdivisions	Human subdivisions
Lateral nucleus (L)	Dorsolateral (Ldl) Medial (Lm) Ventrolateral (Lvl)	Dorsal Dorsal intermediate Ventral intermediate Ventral	Lateral Medial
Basal nucleus (B)	Magnocellular (Bmc) Intermediate (Bi) Parvicellular (Bpc)	Magnocellular Intermediate Parvicellular	Magnocellular Intermediate Parvicellular
Accessory basal nucleus (AB)	Magnocellular (ABmc) Parvicellular (ABpc)	Magnocellular Parvicellular Ventromedial	Magnocellular Parvicellular Ventromedial
Paralamina nucleus	Absent	No subdivisions	Lateral Medial
Bed nucleus of the accessory olfactory tract	No subdivisions	Absent	Absent
Medial nucleus (M)	Rostral (Mr) Central dorsal (Mcd) Central ventral (Mcv) Caudal (Mc)	No subdivisions	No subdivisions
Nucleus of the lateral olfactory tract (NLOT)	No subdivisions	No subdivisions	No subdivisions
Anterior cortical nucleus (COa)	No subdivisions	No subdivisions	No subdivisions
Periamygdaloid cortex (PAC)	Periamygdaloid cortex (PAC) PAC medial (PACm) PAC sulcal (PACs)	PAC oral PAC1 PAC2 PAC3 PAC sulcal	PAC oral PAC1 PAC3 PAC sulcal
Posterior cortical nucleus (COp)	No subdivisions	No subdivisions	No subdivisions
Anterior amygdaloid area (AAA)	No subdivisions	No subdivisions	No subdivisions
Central nucleus (CE)	Capsular (CEc) Lateral (CEl) Intermediate (CEi) Medial (CEm)	Lateral Medial	Lateral Medial
Intercalated nuclei (I)	No subdivisions	No subdivisions	No subdivisions
Amygdalohippocampal area (AHA)	Lateral (AHA1) Medial (AHAm)	Dorsal Ventral	Lateral Medial

cells, represent approximately 20% of all neurons, and constitute local circuits. These cells have axon collaterals restricted to the deep nuclei, acting as interneurons (McDonald, 1992, 1998; Sah et al., 2003; Spampanato et al., 2011). However, recent tracing studies combined with immunohistochemistry have demonstrated that some GABAergic neurons in the deep nuclei originate

long projections directed to the prefrontal cortex (McDonald, 1987), basal forebrain (McDonald et al., 2012) and mediodorsal thalamic nucleus (McDonald, 1987; McDonald and Mascagni, 2007). As in the neocortex and hippocampal region, interneurons in the deep nuclei can be classified into distinct subpopulations on the basis of their content of calcium binding proteins



(calbindin-D28k [CB], parvalbumin [PV] and calretinin [CR]), and peptides (somatostatin [SOM], cholecystokinin [CCK], neuropeptide Y [NPY], and vasoactive intestinal peptides [VIP]). The CB- and CR-immunoreactive (IR) interneurons are the predominant interneuronal subpopulations in the deep nuclei (Kemppainen and Pitkänen, 2000; McDonald and Mascagni, 2001). The CB-IR neurons can express PV (CB+/PV+) or CCK (CB+/CCK+) or SOM (CB+/SOM+) (McDonald and Betette, 2001; McDonald and Mascagni, 2002; Mascagni and McDonald, 2003). Similarly, the CR-IR interneurons can be immunopositive for CCK and/or VIP (Mascagni and McDonald, 2003; Muller et al., 2003). Microcircuits located in the deep nuclei are tightly regulated through the activity of the interneurons, which can innervate pyramidal cells as well as other interneurons. In particular PV-IR interneurons form symmetrical synapses with perisomatic (cell body, axon initial segment and thick proximal dendrites) and distal dendritic (small-caliber dendrites and dendritic spines) domains of pyramidal cells (Muller et al., 2006). Interestingly, pyramidal cells constitute synapse-like contacts with the perisomatic and distal dendritic domains of the PV-IR interneurons, in this way constituting a reciprocal connection (McDonald et al., 2005). The PV-IR interneurons also form interneuronal networks interconnected by electrical (gap junctions) and chemical synapses (Muller et al., 2005; Woodruff and Sah, 2007). PV-interneurons of the basolateral amygdala

can be subdivided into four functionally distinct subpopulations. Fast spiking cells are the most common PV-IR functional cell type. Interestingly, these cells innervate the perisomatic domain of pyramidal neurons (Woodruff and Sah, 2007). The CCK-IR interneurons form synapses with the somata and the proximal dendrites of the pyramidal cells. The SOM-IR neurons provide an inhibitory innervation (symmetrical synapses), especially of the distal dendritic domain (small-caliber dendrites and dendritic spines) of pyramidal cells (Muller et al., 2007a). In addition, SOM-IR axon terminals also contact SOM-, VIP- and PV-IR interneurons (Muller et al., 2007a). The VIP-IR interneurons do not innervate pyramidal cells, but form synapses with other interneurons, especially CCK-positive interneurons (Mascagni and McDonald, 2003; Muller et al., 2003).

The superficial nuclei exhibit two major cell classes: spiny pyramidal cells and spine-sparse or aspiny non-pyramidal neurons (McDonald, 1992, 1998; Sah et al., 2003). Pyramidal cells are glutamatergic projection neurons whereas non-pyramidal neurons represent local inhibitory GABAergic interneurons. These cells are not randomly organized, as in the deep nuclei, but exhibit a laminar organization (layers I, II and III) (McDonald, 1992, 1998; Sah et al., 2003). Interestingly, GABAergic projection neurons are also distributed in the superficial nuclei (McDonald et al., 2012). The medial nucleus does not contain pyramidal and non-pyramidal neurons but small- to medium-sized ovoid neurons which possess spiny dendrites (McDonald, 1992, 1998; Sah et al., 2003).

In the amygdalohippocampal area, pyramidal and non-pyramidal neurons similar to those located in the deep nuclei are the two main cell types (McDonald, 1992, 1998). Similarly, the anterior amygdaloid area contains spiny projection neurons and aspiny interneurons (McDonald, 1992, 1998). Central nucleus and intercalated masses exhibits striatal-like GABAergic neurons (McDonald, 1992, 1998; Sah et al., 2003). Moreover, neurons located in the central nucleus can be subdivided into distinct subpopulations based on their expression of neuropeptides (neurotensin, corticotropin-releasing factor, enkephalin, galanin, SOM, substance P, CCK, and VIP) (Cassell et al., 1986; Cassell and Gray, 1989).

SEROTINERGIC INNERVATION OF THE AMYGDALA AND SEROTONIN RECEPTOR

Serotonin (5-hydroxytryptamine, 5-HT) is a molecule located in the central nervous system which has the role of a neurotransmitter/neuromodulator. Serotonergic somata are located along the midline of the brainstem in cell body groups designated raphe nuclei. The amygdala receives substantial serotonergic innervation originating mainly from the dorsal raphe nucleus and, to a lesser extent, from the median raphe nucleus (Pralong et al., 2002; Hensler, 2006; Asan et al., 2013). Within the rat amygdala, serotonergic fibers are directed especially in the lateral nucleus, basal nucleus (magnocellular division) and amygdalohippocampal area (Steinbusch, 1981). On the contrary, in the monkey amygdala, the highest density of serotonergic fibers is located in the central nucleus, the nucleus of the lateral olfactory tract, the paralaminar nucleus, the anterior amygdaloid area, and the amygdalohippocampal area (Bauman and Amaral, 2005). In the

rat basal nucleus (magnocellular and intermediate divisions), an ultrastructure study has demonstrated that serotonin terminals contact pyramidal as well as non-pyramidal (PV-IR and VIP-IR) neurons (Muller et al., 2007b).

The different physiological effects of serotonin are mediated by seven families of receptors (5-HT₁–5-HT₇). With the exception of the 5-HT_{3A/3B} receptors, which are a ligand-gated ion channel, the serotonin receptors are metabotropic receptors and belong to the G-protein coupled receptor (GPCR) superfamily (Barnes and Sharp, 1999; Hoyer et al., 2002). The 5-HT₂ receptor family contains three receptor subtypes, 5-HT_{2A} (471 amino acids), 5-HT_{2B} (479–504 amino acids) and 5-HT_{2C} (458–460 amino acids), which exhibit a 46–50% overall sequence identity and couple preferentially to Gq/11 to increase the hydrolysis of inositol phosphates and elevate intracellular calcium. 5-HT₂ receptors may also couple to G12/13 which are known to mediate long term structural changes in cells (Barnes and Sharp, 1999; Hoyer et al., 2002; Hannon and Hoyer, 2008).

The 5-HT_{2A} receptor is coupled to G-protein and stimulates phosphoinositide-specific phospholipase C with a consequent increment of inositol triphosphate (Raymond et al., 2001; Hoyer et al., 2002; Hannon and Hoyer, 2008). This serotonin receptor also activates phospholipase D and phospholipase A2 by interacting with additional G-proteins. The 5-HT_{2A} receptor activation also closes potassium channels, producing neuronal depolarization (Aghajanian, 1995; Barnes and Sharp, 1999). In addition, the activations of this receptor subtype increases also cGMP levels by means of a mechanism dependent on N-methyl-D-aspartate (NMDA) receptor activation (Regina et al., 2003, 2004). Interestingly, the 5-HT_{2A} and 5-HT_{2C} receptors are paradoxically regulated by agonists and antagonists (Gray and Roth, 2001; Van Oekelen et al., 2003).

EXPRESSION OF 5-HT₂ RECEPTOR SUBTYPES IN THE AMYGDALOID COMPLEX

The amygdaloid complex expresses moderate to high density of serotonergic receptors including 5-HT_{1A}, 5-HT₂, 5-HT₃, 5-HT₄, and 5-HT₆ (Pralong et al., 2002).

Using autoradiography, *in situ* hybridization and immunohistochemistry, it has been demonstrated that 5-HT₂ receptor family mRNA and protein are present in the amygdala. Interestingly, the expression of the 5-HT_{2A} and 5-HT_{2C} receptors varied during postnatal development in the rat amygdaloid complex (Li et al., 2004).

DEEP NUCLEI

An autoradiographic study has demonstrated a 5-HT₂ receptor binding site in rat deep nuclei, especially in the lateral nucleus (Pazos et al., 1985). In rodents, the presence of the 5-HT₂ receptor in the lateral, basal and accessory basal nuclei was also verified with *in situ* hybridization experiments (Wright et al., 1995). Autoradiography and *in situ* hybridization studies have reported that binding sites and 5-HT_{2A} receptor mRNA are present in the lateral (dorsomedial division) and basal (magnocellular division) nuclei (Lopez-Gimenez et al., 2001). Pompeiano et al. (1994) have reported the presence of the 5-HT_{2C} receptor mRNA in rat deep nuclei, with the highest levels in the lateral nucleus.

Interestingly, these Authors failed to find 5-HT_{2A} receptor mRNA in the same nuclei. Radioactive *in situ* hybridization studies on the rat (Greenwood et al., 2012) and the mouse (Li et al., 2003) amygdala have shown that the 5-HT_{2C} receptor mRNA is located in the lateral nucleus and, to a lesser extent, in the basal nucleus. Using non-radioactive *in situ* hybridization procedures, the highest number of cells containing 5-HT_{2C} receptor mRNA in the rat amygdala has been observed in the lateral and accessory basal nuclei. On the contrary, only a few 5-HT_{2C} receptor mRNA-reactive cells have been reported in the rat basal nucleus (Bonn et al., 2012, 2013). 5-HT_{2C} receptor mRNA has been reported in the deep nuclei of the human amygdala (Pasqualetti et al., 1999).

In rat deep nuclei, immunoreactivity for the 5-HT_{2A} receptor is located in pyramidal and non-pyramidal neurons (Morilak et al., 1993; Cornea-Hébert et al., 1999; Xu and Pandey, 2000; McDonald and Mascagni, 2007; Jiang et al., 2008; Bombardi, 2011; Bombardi and Di Giovanni, 2013). In the rat, 100% of the pyramidal cells express the 5-HT_{2A} receptor (McDonald and Mascagni, 2007; Bombardi, 2011). The 5-HT_{2A} receptor is abundant in the apical dendrites of pyramidal cells (McDonald and Mascagni, 2007; Bombardi, 2011) where it may amplify the impact of excitatory synaptic currents.

In rat deep nuclei, 5-HT_{2A} receptor immunoreactivity has been observed in GABAergic interneurons (in somata and dendrites) and GABAergic projection neurons (Morilak et al., 1993; McDonald and Mascagni, 2007; Bombardi, 2011). The GABAergic interneurons are present in the lateral, basal and accessory basal nuclei where the 5-HT_{2A} receptor is expressed by 66.3, 70.6, and 66.4% of interneurons, respectively (Bombardi, 2011). These interneurons are particularly abundant in the medial subdivision of the lateral nucleus (74.7% of interneurons) and in the parvicellular and magnocellular subdivisions of the basal nucleus (73.8 and 71.9% of interneurons, respectively) (Bombardi, 2011). In the rat amygdala, 59.8% of PV-IR neurons in the medial subdivision of the lateral nucleus, and 75.6% of PV-IR neurons in the magnocellular subdivision of the basal nucleus exhibit the 5-HT_{2A} receptor (McDonald and Mascagni, 2007). On the contrary, only 33.1% of SOM-IR neurons in the lateral nucleus (medial subdivision), and 32.6% of SOM-IR neurons in the basal nucleus (magnocellular subdivision), express the 5-HT_{2A} receptor (McDonald and Mascagni, 2007). The GABAergic/5-HT_{2A} receptor-IR projection neurons are especially distributed near the external and internuclear borders of the rat basolateral amygdala and project to the mediodorsal thalamus (McDonald and Mascagni, 2007).

5-HT_{2C} receptor-IR neurons, possibly pyramidal cells, have been observed in rat lateral and basal nuclei (Clemett et al., 2000).

Superficial nuclei

In the rat, 5-HT₂ receptor mRNA levels are moderate in every superficial nuclei (Wright et al., 1995). In rat superficial nuclei, 5-HT_{2A} receptor mRNA is detectable only in the bed nucleus of the accessory olfactory tract where it is strongly expressed (Pompeiano et al., 1994). On the contrary, 5-HT_{2C} receptor mRNA is located in different superficial nuclei, such as the anterior cortical nucleus, the bed nucleus of the accessory olfactory tract and the medial nucleus. In particular, 5-HT_{2C} receptor

mRNA levels are high in the bed nucleus of the accessory olfactory tract, intermediate in the medial nucleus and low in the anterior cortical nucleus (Pompeiano et al., 1994). Autoradiographic analyses of the rat brain have demonstrated the presence of the 5-HT₂ receptor binding sites especially in the anterior cortical nucleus, but also in other superficial nuclei (Pazos et al., 1985). Immunoreactivity for the 5-HT_{2A} receptor has been observed in every superficial nucleus of the rat amygdala. However, a high density of immunopositive neurons is present, especially in the nucleus of the lateral olfactory tract and in the bed nucleus of the accessory olfactory tract (Morilak et al., 1993; Cornea-Hébert et al., 1999; Bombardi, 2011). Using *in situ* hybridization procedures, high levels of 5-HT_{2C} receptor mRNA have been observed in the medial nucleus and in the anterior cortical nucleus of the rodent amygdala (Li et al., 2003; Bonn et al., 2012, 2013; Greenwood et al., 2012). Accordingly, many 5-HT_{2C} receptor-IR neurons are located in the rat medial nucleus (Clemett et al., 2000). A moderate level of 5-HT_{2C} receptor-IR neurons has also been observed in the posterior cortical nucleus of the rat amygdala (Clemett et al., 2000).

As in the deep nuclei, the 5-HT_{2A} receptor is also expressed in pyramidal and non-pyramidal neurons in the rat superficial nuclei (Bombardi, 2011). Pyramidal cells are especially distributed in the nucleus of the lateral olfactory tract (layer II), the anterior cortical nucleus (layers II and III), the periamygdaloid cortex (layers II and III) and the posterior cortical nucleus (layers II and III) (Bombardi, 2011). In these cells, the 5-HT_{2A} receptor is strongly expressed in the apical dendrites where it may induce excitatory synaptic currents. The 5-HT_{2A} receptor-IR non-pyramidal neurons are distributed in the nucleus of the lateral olfactory tract, the anterior cortical nucleus, the periamygdaloid cortex and the posterior cortical nucleus. These cells are heterogeneous in shape (multipolar and fusiform) and size (from small to large), and are particularly abundant in layers II and III (Bombardi, 2011). Since the cell types in the medial nucleus are not cortex-like as in the other superficial nuclei, 5-HT_{2A} receptor-IR pyramidal and non-pyramidal neurons are not present in this nucleus (McDonald, 1992, 1998; Sah et al., 2003). Accordingly, the rat medial nucleus contains 5-HT_{2A} receptor-IR principal neurons with ovoid cell bodies (Bombardi, 2011). The rat medial nucleus is the only amygdaloid area containing 5-HT_{2B} receptor-IR neurons. These cells are numerous and show a multipolar and bipolar morphology (Duxon et al., 1997a).

Remaining nuclei

In the rat amygdalohippocampal area, the presence of the 5-HT_{2A} receptor has been demonstrated only with immunohistochemical procedures which have revealed many 5-HT_{2A} receptor-IR neurons with angular- and ovoid-shaped cell bodies (Bombardi, 2011). The rat amygdalohippocampal area also contains a high density of 5-HT_{2C} receptor-IR neurons (Clemett et al., 2000). A moderate level of 5-HT_{2A} receptor mRNA has been revealed in the rat central nucleus (Wright et al., 1995). Accordingly, immunohistochemical procedures have demonstrated the presence of many 5-HT_{2A} receptor-IR ovoid somata in the different subdivisions of the rat central neurons (Cornea-Hébert et al., 1999; Bombardi, 2011). These cells could be GABAergic local

neurons as well as GABAergic projecting neurons (Bombardi, 2011). Cells containing 5-HT_{2C} receptor mRNA have been observed in the rat central nucleus where they are particularly numerous in the lateral capsular subdivision (Bonn et al., 2012, 2013). Both 5-HT_{2A} and 5-HT_{2C} receptor mRNA are present at low density in the rat amygdalohippocampal area. Accordingly, pyramidal and non-pyramidal neurons of the rat amygdalohippocampal area contain the 5-HT_{2A} receptor (Bombardi, 2011). In the rat intercalated nuclei, 5-HT_{2C} receptor mRNA is present at a low density while 5-HT_{2A} receptor mRNA has not been detected (Pompeiano et al., 1994). A different distribution of the 5-HT_{2A} receptor has been observed using immunohistochemical procedures. In fact, Xu and Pandey (2000), and Bombardi (2011) have observed that small and large neurons in the rat intercalated nuclei express the 5-HT_{2A} receptor. The rat intercalated nuclei contain only weak 5-HT_{2C} receptor mRNA-reactive cells (Bonn et al., 2012, 2013). These data are in disagreement with immunohistochemical studies showing that intercalated nuclei contain a high density of 5-HT_{2C} receptor-IR neurons (Clemett et al., 2000).

EFFECT OF 5-HT₂ RECEPTOR FAMILY ACTIVATION ON AMYGDALAR NEURONS AND MICROCIRCUITS

Serotonin influences amygdalar information processing by activating multiple 5-HT₂ receptor subtypes. Inasmuch as the amygdaloid microcircuits are complex and the expression patterns of the 5-HT₂ receptor subtypes are not fully characterized, the mechanisms by which 5-HT₂ receptor subtypes modulate amygdalar neurotransmission remains poorly understood. This modulation is complex and has been studied especially for 5-HT_{2A} and 5-HT_{2C} receptors.

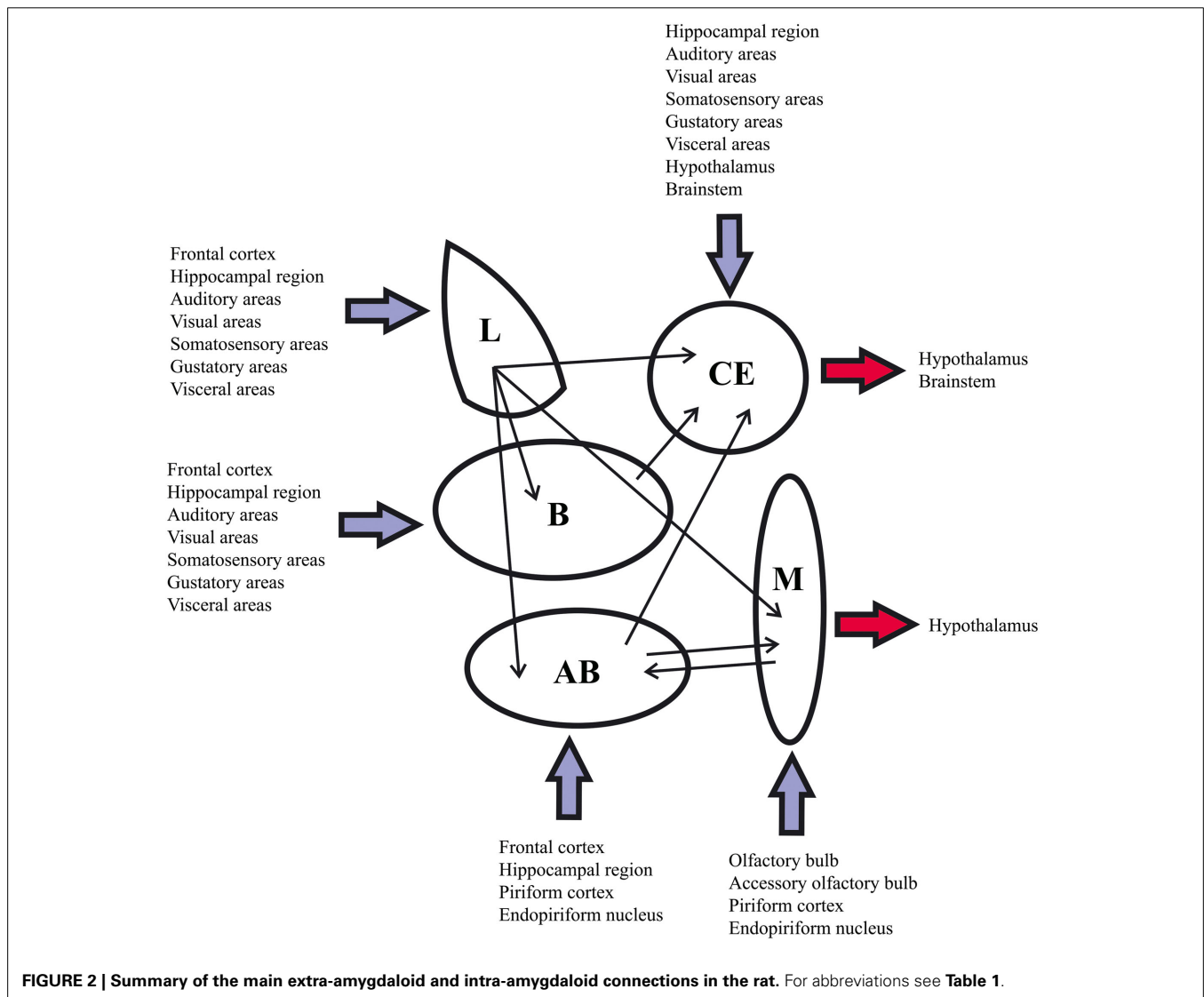
Electrophysiological studies have demonstrated that the 5-HT_{2A} receptor activates the pyramidal cells of the deep nuclei. In fact, the local injection of 1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane (DOI), a 5-HT_{2A}/5-HT_{2C} receptors agonist, increases the discharge rate (Stein et al., 2000) and facilitates synaptic plasticity via an NMDA-mediated mechanism (Chen et al., 2003) in presumptive pyramidal neurons of the rat basolateral amygdala.

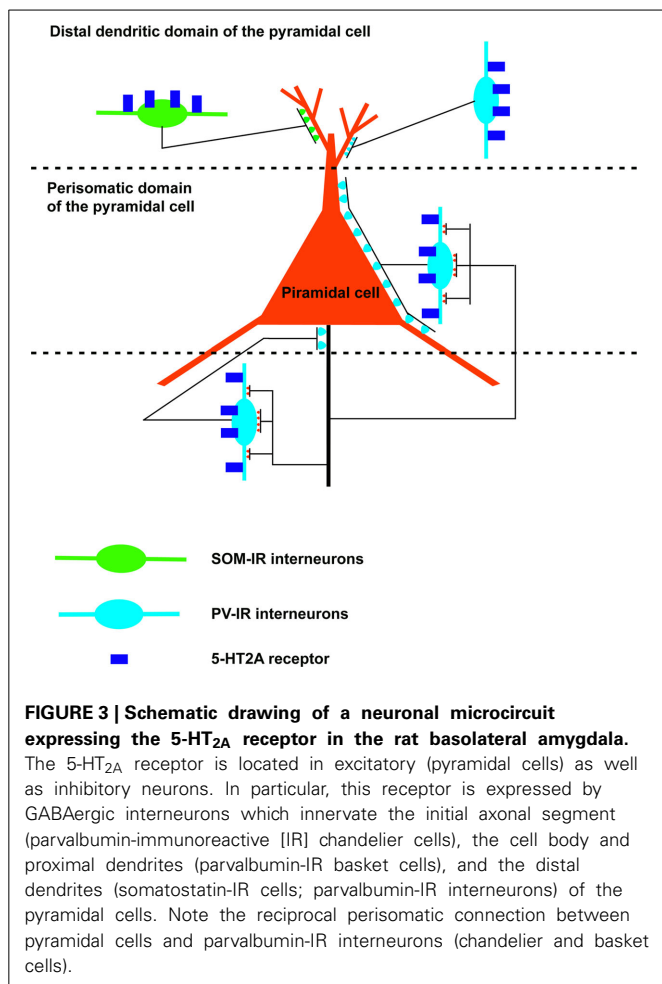
The 5-HT₂ receptor family also modulates the excitability of GABAergic interneurons in the deep nuclei. In fact, electrophysiological studies have demonstrated that the application of α -methyl-5-hydroxytryptamine (a 5-HT₂ receptor agonist) and DOI (a 5-HT_{2A}/5-HT_{2C} receptor agonist), induces the activation of GABAergic interneurons of the rat basolateral amygdala (Rainnie, 1999; Stein et al., 2000; Sokal et al., 2005). In addition, the stimulation of the 5-HT_{2A} receptor increases the frequency and amplitude of spontaneous inhibitory postsynaptic currents (sIPSCs) recorded from the pyramidal neurons of the juvenile rat basolateral amygdala (Jiang et al., 2008). Accordingly, the inhibition of pyramidal cell firing in the lateral nucleus of the rat amygdala obtained after the local application of serotonin is blocked by the simultaneous application of bicuculline methiodide, a GABA antagonist (Stutzmann and LeDoux, 1999). The activation of GABAergic interneurons of the corticomедial amygdala has been demonstrated by iontophoretic injections of DOI (Stein et al., 2000).

Amygdala microcircuitry has not been studied as extensively as that of the neocortex and hippocampal region. However, numerous studies report that the amygdala circuit organization combines cortex-like (deep nuclei, the majority of the superficial nuclei and the amygdalohippocampal area) and striatum-like structures (central nucleus and intercalated nuclei) (McDonald, 1992, 1998; Sah et al., 2003). Since these amygdaloid areas provide numerous intra-amygdaloid and extra-amygdaloid connections, the amygdala is considered to be the interface of the information exchange between the various functional systems of the brain (Pitkänen, 2000). Traditionally, the extra-amygdaloid afferents (all the modalities of sensory inputs and polymodal inputs) target the input side of the amygdala (deep and superficial nuclei) where they are processed locally and then directed, by intra-amygdaloid connections, to the medial and central nuclei which act as an output station. The medial nucleus especially projects to the hypothalamic neuroendocrine zone whereas outputs from the central nucleus especially innervate the hypothalamic and

brainstem nuclei which regulate autonomic functions (Pitkänen, 2000; **Figure 2**).

The distribution of the 5-HT₂ receptor previously reported indicates that this receptor could modulate amygdala activity acting on projection neurons (pyramidal neurons of cortex-like structures and GABAergic projection neurons of the basolateral amygdala and striatum-like structures) as well as on inhibitory interneurons (in particular, GABAergic interneurons of the cortex-like structures). The distribution of the 5-HT₂ receptor family has been studied more extensively for 5-HT_{2A} receptor subtype, especially in the rat basolateral amygdala (**Figure 3**). In the microcircuits of the rat deep nuclei, the 5-HT_{2A} receptor is located on both pyramidal and non-pyramidal neurons. The non-pyramidal neurons containing this receptor express PV and SOM (McDonald and Mascagni, 2007). As previously reported, the PV-IR interneurons innervate the perisomatic domain (cell body and proximal dendrites) of pyramidal cells (Muller et al., 2006). Moreover, these interneurons are connected by gap junctions and





constitute an inhibitory network which synchronizes the firing of pyramidal cells (Woodruff and Sah, 2007). Interestingly, most of the pyramidal neurons form intimate synapse-like contacts with the somata and dendrites (especially proximal dendrites) of the PV-IR interneurons (McDonald et al., 2005), in this way, constituting a reciprocal perisomatic connection which may be important in modulating the synchronized rhythmic activity associated with the formation of emotional memories (Paré and Collins, 2000; Paré et al., 2002; Rainnie et al., 2006). In the rat basolateral amygdala, SOM-IR interneurons innervate the distal dendritic domain of pyramidal cells and could modulate synaptic mechanisms related to emotional learning, including fear conditioning (Paré et al., 2002; Muller et al., 2007a). Since the 5-HT_{2A} receptor is located on PV-IR and SOM-IR interneurons, this receptor subtype could play an important role in the formation of emotional memories.

5-HT₂ RECEPTOR FAMILY AND AMYGDALA-MEDIATED BEHAVIOR

The involvement of the 5-HT₂ receptor family in numerous amygdala-mediated behavioral and physiological effects has been described in several reports. This receptor family plays a crucial role, especially in fear and anxiety. Local infusion of ketanserin (a 5-HT₂ receptor family antagonist) induces an anxiolytic effect

in the conflict test (Hodges et al., 1987). Microinjections of nefazodone (a 5-HT₂ receptors antagonist) into the basolateral nucleus of the rat amygdala enhances the aversive responses induced by NMDA activation of the neural substrates of aversion in the inferior colliculus (Maisonnette et al., 2000). In different mouse models of anxiety, the 5-HT_{2A} receptor mediates different anxiolytic-like effects (Dhonnchadha et al., 2003a,b). Moreover, bilateral injections of ketanserin (a 5-HT_{2A} and 5-HT_{2C} receptors antagonist) into the rat basolateral/medial amygdala produces an anxiogenic profile in an elevated plus-maze (Zangrossi and Graeff, 1994).

The 5-HT_{2A} receptor is also implicated in kindling development from the rat amygdala since the subcutaneous injection of DOI, an agonist of 5-HT_{2A/2C} receptors, facilitates kindling development and reduces the number of amygdaloid stimulations necessary to obtain generalized seizures (Wada et al., 1997).

It is known that direct or indirect projections from the central nucleus of the amygdala to the paraventricular nucleus of the hypothalamus mediate a stress response. *In vivo* microdialysis studies have demonstrated that there is an increase in serotonin release in the amygdala during stress (Kawahara et al., 1993). Accordingly, the 5-HT_{2A} receptor located in the central nucleus of the rat amygdala is able to activate the hypothalamo-pituitary-adrenocortical axis (Feldman et al., 1998). Finally, in the rat basolateral amygdala, the 5-HT_{2A} receptor-mediated serotoninergic facilitation of GABAergic synaptic transmission is impaired by stress (Jiang et al., 2008).

The 5-HT_{2B} receptor is also involved in amygdala-mediated behavior. In fact, the activation of this receptor subtype causes anxiolysis in social interaction tests in the rat (Duxon et al., 1997b).

CONCLUSIONS

The present review reported that the 5-HT₂ receptor family plays a crucial role in regulating the activity of amygdalar microcircuits and projections. In fact, as in the cerebral cortex and the hippocampal regions (Willins et al., 1997; Hamada et al., 1998; Jakab and Goldman-Rakic, 1998, 2000; Cornea-Hébert et al., 1999; Clemett et al., 2000; Xu and Pandey, 2000; Jansson et al., 2001; Miner et al., 2003; Lüttgen et al., 2004; Bombardi, 2012), excitatory as well as inhibitory neurons in the rat amygdala express the 5-HT₂ receptor family. Nevertheless, the exact role of the 5-HT₂ receptor family in the modulation of amygdala activity is still poorly understood and requires additional study. In this way, detailed knowledge of the cellular mechanism underlying the modulation of amygdalar activity mediated by the 5-HT₂ receptor family could provide valuable information for better understanding the pathogenesis of affective disorders and for utilizing a more specific pharmacological treatment.

REFERENCES

- Aggleton, J. P. (2000). *The Amygdala: a Functional Analysis*. 2nd Edn. New York, NY: Oxford University Press.
- Aghajanian, G. K. (1995). "Electrophysiology of serotonin receptor subtypes and signal transduction pathways," in *Psychopharmacology: The Fourth Generation of Progress*, eds F. E. Bloom and D. J. Kupfer (New York, NY: Raven Press), 51–460.

- Amaral, D. G. (2002). The primate amygdala and the neurobiology of social behavior: implications for understanding social anxiety. *Biol. Psychiatry* 51, 11–17. doi: 10.1016/S0006-3223(01)01307-5
- Asan, E., Steinke, M., and Lesch, K. P. (2013). Serotonergic innervation of the amygdala: targets, receptors, and implications for stress and anxiety. *Histochem. Cell. Biol.* 139, 785–813. doi: 10.1007/s00418-013-1081-1
- Barnes, N. M., and Sharp, T. (1999). A review of central 5-HT receptors and their function. *Neuropharmacology* 38, 1083–1152. doi: 10.1016/S0028-3908(99)00010-6
- Bauman, M. D., and Amaral, D. G. (2005). The distribution of serotonergic fibers in the macaque monkey amygdala: an immunohistochemical study using antisera to 5-hydroxytryptamine. *Neuroscience* 136, 193–203. doi: 10.1016/j.neuroscience.2005.07.040
- Bombardi, C. (2011). Distribution of 5-HT_{2A} receptor immunoreactivity in the rat amygdaloid complex and colocalization with γ -aminobutyric acid. *Brain Res.* 1370, 112–128. doi: 10.1016/j.brainres.2010.11.055
- Bombardi, C. (2012). Neuronal localization of 5-HT_{2A} receptor immunoreactivity in the rat hippocampal region. *Brain Res. Bull.* 87, 259–273. doi: 10.1016/j.brainresbull.2011.11.006
- Bombardi, C., and Di Giovanni, G. (2013). Functional anatomy of 5-HT_{2A} receptors in the amygdala and hippocampal complex: relevance to memory functions. *Exp. Brain Res.* 230, 427–439. doi: 10.1007/s00221-013-3512-6
- Bonn, M., Schmitt, A., and Asan, E. (2012). Double and triple *in situ* hybridization for coexpression studies: combined fluorescent and chromogenic detection of neuropeptide Y (NPY) and serotonin receptor subtype mRNAs expressed at different abundance levels. *Histochem. Cell. Biol.* 137, 11–24. doi: 10.1007/s00418-011-0882-3
- Bonn, M., Schmitt, A., Lesch, K. P., Van Bockstaele, E. J., and Asan, E. (2013). Serotonergic innervation and serotonin receptor expression of NPY-producing neurons in the rat lateral and basolateral amygdaloid nuclei. *Brain Struct. Funct.* 218, 421–435. doi: 10.1007/s00429-012-0406-5
- Braga, M. F., Aroniadou-Anderjaska, V., Post, R. M., and Li, H. (2002). Lamotrigine reduces spontaneous and evoked GABA_A receptor-mediated synaptic transmission in the basolateral amygdala: implications for its effects in seizure and affective disorders. *Neuropharmacology* 42, 522–529. doi: 10.1016/S0028-3908(01)00198-8
- Burghardt, N. S., Sullivan, G. M., McEwen, B. S., Gorman, J. M., and LeDoux, J. E. (2004). The selective serotonin reuptake inhibitor citalopram increases fear after acute treatment but reduces fear with chronic treatment: a comparison with tianeptine. *Biol. Psychiatry* 55, 1171–1178. doi: 10.1016/j.biopsych.2004.02.029
- Canli, T., Congdon, E., Gutknecht, L., Constable, R. T., and Lesch, K. P. (2005). Amygdala responsiveness is modulated by tryptophan hydroxylase-2 gene variation. *J. Neural. Transm.* 112, 1479–1485. doi: 10.1007/s00702-005-0391-4
- Cassell, M. D., and Gray, T. S. (1989). Morphology of peptide-immunoreactive neurons in the rat central nucleus of the amygdala. *J. Comp. Neurol.* 281, 320–333. doi: 10.1002/cne.902810212
- Cassell, M. D., Gray, T. S., and Kiss, J. Z. (1986). Neuronal architecture in the rat central nucleus of the amygdala: a cytological, hodological, and immunocytochemical study. *J. Comp. Neurol.* 246, 478–499. doi: 10.1002/cne.902460406
- Chen, A., Hough, J., and Li, H. (2003). Serotonin type II receptor activation facilitates synaptic plasticity via N-Methyl-D-Aspartate-mediated mechanism in the rat basolateral amygdala. *Neuroscience* 119, 53–63. doi: 10.1016/S0306-4522(03)00076-9
- Clemett, D. A., Punhani, T., Duxon, M. S., Blackburn, T. P., and Fone, K. C. (2000). Immunohistochemical localisation of the 5-HT_{2C} receptor protein in the rat CNS. *Neuropharmacology* 39, 123–132. doi: 10.1016/S0028-3908(99)00086-6
- Cornea-Hébert, V., Riad, M., Wu, C., Singh, S. K., and Descarries, L. (1999). Cellular end subcellular distribution of the serotonin 5-HT_{2A} receptor in the central nervous system of adult rat. *J. Comp. Neurol.* 409, 187–209. doi: 10.1002/(SICI)1096-9861(1999)0628
- Dhonnchadha, B. A. N., Bourin, M., and Hascoët, M. (2003a). Anxiolytic-like effects of 5-HT₂ ligands on three mouse models of anxiety. *Behav. Brain Res.* 140, 203–214. doi: 10.1016/S0166-4328(02)00311-X
- Dhonnchadha, B. A. N., Hascoët, M., Jolliet, P., and Bourin, M. (2003b). Evidence for a 5-HT_{2A} receptor mode of action in the anxiolytic-like properties of DOI in mice. *Behav. Brain Res.* 147, 175–184. doi: 10.1016/S0166-4328(03)00179-7
- Dhonnchadha, B. A. N., Ripoll, N., Clénet, F., Hascoët, M., and Bourin, M. (2005). Implication of 5-HT₂ receptor subtypes in the mechanism of action of antidepressants in the four plates test. *Psychopharmacology* 179, 418–429. doi: 10.1007/s00213-004-2044-y
- Duxon, M. S., Flanigan, T. P., Reavley, A. C., Baxter, G. S., Blackburn, T. P., and Fone, K. C. F. (1997). Evidence for expression of the 5-hydroxytryptamine-2B receptor protein in the rat central nervous system. *Neuroscience* 76, 323–329. doi: 10.1016/S0306-4522(96)00480-0
- Duxon, M. S., Kennett, G. A., Lightowler, S., Blackburn, T. P., and Fone, K. C. (1997). Activation of 5-HT_{2B} receptors in the medial amygdala causes anxiolysis in the social interaction test in the rat. *Neuropharmacology* 36, 601–608. doi: 10.1016/S0028-3908(97)00042-7
- Feldman, S., Newman, M. E., Gur, E., and Weidenfeld, J. (1998). Role of serotonin in the amygdala in hypothalamo-pituitary-adrenocortical responses. *Neuroreport* 9, 2007–2009.
- Gray, J. A., and Roth, B. L. (2001). Paradoxical trafficking and regulation of 5-HT_{2A} receptors by agonists and antagonists. *Brain Res. Bull.* 56, 441–451. doi: 10.1016/S0306-9230(01)00623-2
- Greenwood, B. N., Strong, P. V., Loughridge, A. B., Day, H. E., Clark, P. J., Mika, A., et al. (2012). 5-HT_{2C} receptors in the basolateral amygdala and dorsal striatum are a novel target for the anxiolytic and antidepressant effects of exercise. *PLoS ONE* 7:e46118. doi: 10.1371/journal.pone.0046118
- Hamada, S., Senzaki, K., Hamaguchi-Hamada, K., Tabuchi, K., Yamamoto, H., Yamamoto, T., et al. (1998). Localization of 5-HT_{2A} receptor in rat cerebral cortex and olfactory system revealed by immunohistochemistry using two antibodies raised in rabbit and chicken. *Mol. Brain Res.* 54, 199–211. doi: 10.1016/S0169-328X(97)00322-7
- Hannon, J., and Hoyer, D. (2008). Molecular biology of 5-HT receptors. *Behav. Brain Res.* 195, 198–213. doi: 10.1016/j.bbr.2008.03.020
- Hariri, A. R., and Holmes, A. (2006). Genetics of emotional regulation: the role of the serotonin transporter in neural function. *Trends Cogn. Sci.* 10, 182–191. doi: 10.1016/j.tics.2006.02.011
- Hariri, A. R., Mattay, V. S., Tessitore, A., Kolachana, B., Fera, F., Goldman, D., et al. (2002). Serotonin transporter genetic variation and the response of the human amygdala. *Science* 297, 400–403. doi: 10.1126/science.1071829
- Hensler, J. G. (2006). Serotonergic modulation of the limbic system. *Neurosci. Biobehav. Rev.* 30, 203–214. doi: 10.1016/j.neubiorev.2005.06.007
- Hodges, H., Green, S., and Glenn, B. (1987). Evidence that the amygdala is involved in benzodiazepine and serotonergic effects on punished responding but not discrimination. *Psychopharmacology* 92, 491–504. doi: 10.1007/BF00176484
- Hoyer, D., Hannon, J. P., and Martin, G. R. (2002). Molecular, pharmacological and functional diversity of 5-HT receptors. *Pharmacol. Biochem. Behav.* 71, 533–554. doi: 10.1016/S0091-3057(01)00746-8
- Inoue, T., Li, X. B., Abekawa, T., Kitaichi, Y., Izumi, T., Nakagawa, S., et al. (2004). Selective serotonin reuptake inhibitor reduces conditioned fear through its effect in the amygdala. *Eur. J. Pharmacol.* 497, 311–316. doi: 10.1016/j.ejphar.2004.06.061
- Jakab, R. L., and Goldman-Rakic, P. S. (1998). 5-hydroxytryptamine_{2A} serotonin receptors in the primate cerebral cortex: possible site of action of hallucinogenic and antipsychotic drugs in pyramidal cell apical dendrites. *Proc. Natl. Acad. Sci. U.S.A.* 95, 735–740. doi: 10.1073/pnas.95.2.735
- Jakab, R. L., and Goldman-Rakic, P. S. (2000). Segregation of serotonin 5-HT_{2A} and 5-HT₃ receptors in inhibitory circuits of the primate cerebral cortex. *J. Comp. Neurol.* 417, 337–348. doi: 10.1002/(SICI)1096-9861(2000)417
- Jansson, A., Tinner, B., Bancila, M., Vergé, D., Steinbusch, H. W. M., Agnati, L. F., et al. (2001). Relationships of 5-hydroxytryptamine immunoreactive terminal-like varicosities to 5-hydroxytryptamine-2A receptor-immunoreactive neuronal processes in the rat forebrain. *J. Chem. Neuroanat.* 22, 185–203. doi: 10.1016/S0891-0618(01)00133-8
- Jasnow, A. M., and Huhman, K. L. (2001). Activation of GABA(A) receptors in the amygdala blocks the acquisition and expression of conditioned defeat in Syrian hamster. *Brain Res.* 920, 142–150. doi: 10.1016/S0006-8993(01)03054-2
- Jiang, X., Xing, G., Yang, C., Verma, A., Zhang, L., and Li, H. (2008). Stress impairs 5-HT_{2A} Receptor-mediated serotonergic facilitation of GABA release in juvenile rat basolateral amygdala. *Neuropsychopharmacology* 33, 1–14. doi: 10.1038/npp.2008.71
- Kawahara, H., Yoshida, M., Yokoo, H., Nishi, M., and Tanaka, M. (1993). Psychological stress increases serotonin release in the rat amygdala and prefrontal cortex assessed by *in vivo* microdialysis. *Neurosci. Lett.* 162, 81–84. doi: 10.1016/0304-3940(93)90565-3

- Keele, N. B. (2005). The role of serotonin in impulsive and aggressive behaviors associated with epilepsy-like neuronal hyperexcitability in the amygdala. *Epilepsy Behav.* 7, 325–335. doi: 10.1016/j.yebeh.2005.06.014
- Kemppainen, S., and Pitkänen, A. (2000). Distribution of parvalbumin, calretinin, and calbindin-D(28k) immunoreactivity in the rat amygdaloid complex and colocalization with gamma-aminobutyric acid. *J. Comp. Neurol.* 426, 441–467. doi: 10.1002/1096-9861(20001023)
- Kim, J. J., Koo, J. W., Lee, H. J., and Han, J. S. (2005). Amygdalar inactivation blocks stress-induced impairments in hippocampal long-term potentiation and spatial memory. *J. Neurosci.* 25, 1532–1539. doi: 10.1523/JNEUROSCI.4623-04.2005
- Li, Q. H., Nakadate, K., Tanaka-Nakadate, S., Nakatsuka, D., Cui, Y., and Watanabe, Y. (2004). Unique expression patterns of 5-HT_{2A} and 5-HT_{2C} receptors in the rat brain during postnatal development: Western blot and immunohistochemical analyses. *J. Comp. Neurol.* 469, 128–140. doi: 10.1002/cne.11004
- Li, Q., Wichems, C. H., Ma, L., Van de Kar, L. D., Garcia, F., and Murphy, D. L. (2003). Brain region-specific alterations of 5-HT_{2A} and 5-HT_{2C} receptors in serotonin transporter knockout mice. *J. Neurochem.* 84, 1256–1265. doi: 10.1046/j.1471-4159.2003.01607.x
- Lopez-Gimenez, J. F., Vilaro, M. T., Palacios, J. M., and Mengod, G. (2001). Mapping of 5-HT_{2A} receptors and their mRNA in monkey brain: [3H]MDL100,907 autoradiography and *in situ* hybridization studies. *J. Comp. Neurol.* 429, 571–589. doi: 10.1002/1096-9861(20010122)
- Lüttgen, M., Ögren, S. O., and Meister, B. (2004). Chemical identity of 5-HT_{2A} receptor immunoreactive neurons of the rat septal complex and dorsal hippocampus. *Brain Res.* 1010, 156–165. doi: 10.1016/j.brainres.2004.03.016
- Maisonnette, S., Villela, C., Carotti, A. P., and Landeira-Fernandez, J. (2000). Microinfusion of nefazodone into basolateral nucleus of the amygdala enhances defensive behavior induced by NMDA stimulation of the inferior colliculus. *Physiol. Behav.* 70, 243–247. doi: 10.1016/S0031-9384(00)00256-0
- Manji, H. K., Drevets, W. C., and Charney, D. S. (2001). The cellular neurobiology of depression. *Nat. Med.* 7, 541–547. doi: 10.1038/87865
- Mascagni, F., and McDonald, A. J. (2003). Immunohistochemical characterization of cholecystokinin containing neurons in the rat basolateral amygdala. *Brain Res.* 976, 171–184. doi: 10.1016/S0006-8993(03)02625-8
- McDonald, A. J. (1987). Organization of amygdaloid projections to the mediadorsal thalamus and prefrontal cortex: a fluorescence retrograde transport study in the rat. *J. Comp. Neurol.* 262, 46–58. doi: 10.1002/cne.902620105
- McDonald, A. J. (1992). “Cells types and intrinsic connections of the amygdala,” in *The Amygdala: Neurobiological Aspects of Emotion, Memory and Mental Dysfunction*, ed J. P. Aggleton (New York, NY: Wiley-Liss), 67–96.
- McDonald, A. J. (1998). Cortical pathways to the mammalian amygdala. *Prog. Neurobiol.* 55, 257–332. doi: 10.1016/S0301-0082(98)00003-3
- McDonald, A. J., and Bette, R. L. (2001). Parvalbumin-containing neurons in the rat basolateral amygdala: morphology and co-localization of Calbindin-D(28k). *Neuroscience* 102, 413–425. doi: 10.1016/S0306-4522(00)00481-4
- McDonald, A. J., and Mascagni, F. (2001). Colocalization of calcium-binding proteins and GABA in neurons of the rat basolateral amygdala. *Neuroscience* 105, 681–693. doi: 10.1016/S0306-4522(01)00214-7
- McDonald, A. J., and Mascagni, F. (2002). Immunohistochemical characterization of somatostatin containing interneurons in the rat basolateral amygdala. *Brain Res.* 943, 237–244. doi: 10.1016/S0006-8993(02)02650-1
- McDonald, A. J., and Mascagni, F. (2007). Neuronal localization of 5-HT type 2A receptor immunoreactivity in the rat basolateral amygdala. *Neuroscience* 146, 306–320. doi: 10.1016/j.neuroscience.2007.01.047
- McDonald, A. J., Mascagni, F., Mania, I., and Rainnie, D. (2005). Evidence for a perisomatic innervation of parvalbumin-containing interneurons by individual pyramidal cells in the basolateral amygdala. *Brain Res.* 1035, 32–40. doi: 10.1016/j.brainres.2004.11.052
- McDonald, A. J., Mascagni, F., and Zaric, V. (2012). Subpopulations of somatostatin-immunoreactive non-pyramidal neurons in the amygdala and adjacent external capsule project to the basal forebrain: evidence for the existence of GABAergic projection neurons in the cortical nuclei and basolateral nuclear complex. *Front. Neural Circuits* 6:46. doi: 10.3389/fncir.2012.00046
- Miner, L. A. H., Backstrom, J. R., Sanders-Bush, E., and Sesack, S. R. (2003). Ultrastructural localization of serotonin_{2A} receptors in the middle layers of the rat prelimbic prefrontal cortex. *Neuroscience* 116, 101–117. doi: 10.1016/S0306-4522(02)00580-8
- Morilak, D. A., Garlow, S. J., and Ciaranello, R. D. (1993). Immunocytochemical localization and description of neurons expressing serotonin₂ receptors in the rat brain. *Neuroscience* 54, 701–717. doi: 10.1016/0306-4522(93)90241-7
- Muller, J. F., Mascagni, F., and McDonald, A. J. (2003). Synaptic connections of distinct interneuronal Subpopulations in the rat basolateral amygdala nucleus. *J. Comp. Neurol.* 456, 217–236. doi: 10.1002/cne.10435
- Muller, J. F., Mascagni, F., and McDonald, A. J. (2005). Coupled networks of parvalbuminimmunoreactive interneurons in the rat basolateral amygdala. *J. Neurosci.* 25, 7366–7376. doi: 10.1523/JNEUROSCI.0899-05.2005
- Muller, J. F., Mascagni, F., and McDonald, A. J. (2006). Pyramidal cells of the rat basolateral amygdala: synaptology and innervation by parvalbumin-immunoreactive interneurons. *J. Comp. Neurol.* 494, 635–650. doi: 10.1002/cne.20832
- Muller, J. F., Mascagni, F., and McDonald, A. J. (2007a). Postsynaptic targets of somatostatin-containing interneurons in the rat basolateral amygdala. *J. Comp. Neurol.* 500, 513–529. doi: 10.1002/cne.21185
- Muller, J. F., Mascagni, F., and McDonald, A. J. (2007b). Serotonin-immunoreactive axon terminals innervate pyramidal cells and interneurons in the rat basolateral amygdala. *J. Comp. Neurol.* 505, 314–335. doi: 10.1002/cne.21486
- Paré, D., and Collins, D. R. (2000). Neuronal correlates of fear in the lateral amygdala: multiple extracellular recordings in conscious cats. *J. Neurosci.* 20, 2701–2710.
- Paré, D., Collins, D. R., and Pelletier, J. G. (2002). Amygdala oscillations and the consolidation of emotional memories. *Trends Cogn. Sci.* 6, 306–314. doi: 10.1016/S1364-6613(02)01924-1
- Pasqualetti, M., Ori, M., Castagna, M., Marazziti, D., Cassano, G. B., and Nardi, I. (1999). Distribution and cellular localization of the serotonin type 2C receptor messenger RNA in human brain. *Neuroscience* 92, 601–611. doi: 10.1016/S0306-4522(99)00011-1
- Paxinos, G., and Watson, C. (1998). *The Rat Brain in Stereotaxic Coordinates, 4th Edn.* New York, NY: Academic Press.
- Pazos, A., Cortés, R., and Palacios, J. M. (1985). Quantitative autoradiographic mapping of serotonin receptors in the rat brain. II. Serotonin-2 receptors. *Brain Res.* 346, 231–249. doi: 10.1016/0006-8993(85)90857-1
- Pitkänen, A. (2000). “Connectivity of the rat amygdaloid complex,” in *The Amygdala: a Functional Analysis*. 2nd Edn., ed J. P. Aggleton (New York, NY: Oxford University Press), 31–115.
- Pitkänen, A., and Kemppainen, S. (2002). Comparison of the distribution of calcium-binding proteins and intrinsic connectivity in the lateral nucleus of the rat, monkey, and human amygdala. *Pharmacol. Biochem. Behav.* 71, 369–377. doi: 10.1016/S0091-3057(01)00690-6
- Pompeiano, M., Palacios, J. M., and Mengod, G. (1994). Distribution of serotonin 5-HT₂ receptor family mRNAs: comparison between 5-HT_{2A} and 5-HT_{2C} receptors. *Mol. Brain Res.* 23, 163–178. doi: 10.1016/0169-328X(94)90223-2
- Pralong, E., Magistretti, P., and Stoop, R. (2002). Cellular perspectives on the glutamate-monoamine interactions in limbic lobe structures and their relevance for some psychiatric disorders. *Prog. Neurobiol.* 67, 173–202. doi: 10.1016/S0301-0082(02)00017-5
- Rainnie, D. G. (1999). Serotonergic modulation of neurotransmission in the rat basolateral amygdala. *J. Neurophysiol.* 82, 69–85.
- Rainnie, D. G., Bergeron, R., Sajdyk, T. J., Patil, M., Gehlert, D. R., and Shekhar, A. (2004). Corticotrophin releasing factor-induced synaptic plasticity in the amygdala translates stress into emotional disorders. *J. Neurosci.* 24, 3471–3479. doi: 10.1523/JNEUROSCI.5740-03.2004
- Rainnie, D. G., Mania, I., Mascagni, F., and McDonald, A. J. (2006). Physiological and morphological characterization of parvalbumin-containing interneurons of the rat basolateral amygdala. *J. Comp. Neurol.* 498, 142–161. doi: 10.1002/cne.21049
- Raymond, J., Mukhin, Y., Gelasco, A., Turner, J., Collinsworth, G., Gettys, T., et al. (2001). Multiplicity of mechanisms of serotonin receptor signal transduction. *Pharmacol. Ther.* 92, 179–212. doi: 10.1016/S0163-7258(01)00169-3
- Regina, M. J., Bucelli, R. C., Winter, J. C., and Rabin, R. A. (2004). Cellular mechanism of serotonin 5-HT_{2A} receptor-mediated cGMP formation: the essential role of glutamate. *Brain Res.* 1003, 168–175. doi: 10.1016/j.brainres.2004.01.014
- Regina, M. J., Winter, J. C., and Rabin, R. A. (2003). Characterization of a novel effect of serotonin 5-HT_{1A} and 5-HT_{2A} receptors: increasing cGMP levels in rat frontal cortex. *Neuropharmacology* 45, 1041–1049. doi: 10.1016/S0028-3908(03)00287-9

- Rodrigues Manzanares, P. A., Isoardi, N. A., Carrer, H. F., and Molina, V. A. (2005). Previous stress facilitates fear memory, attenuates GABAergic inhibition, and increases synaptic plasticity in the rat basolateral amygdala. *J. Neurosci.* 25, 8725–8734. doi: 10.1523/JNEUROSCI.2260-05.2005
- Sah, P., Faber, E. S., Lopez De Armentia, M., and Power, J. (2003). The amygdaloid complex: anatomy and physiology. *Physiol. Rev.* 83, 803–834. doi: 10.1152/physrev.00002.2003
- Sanders, S. K., and Shekhar, A. (1995). Regulation of anxiety by GABA_A receptors in the rat amygdala. *Pharmacol. Biochem. Behav.* 52, 701–706. doi: 10.1016/0091-3057(95)00153-N
- Shin, L. M., Rauch, S. L., and Pitman, R. K. (2006). Amygdala, medial prefrontal cortex, and hippocampal function in PTSD. *Ann. N.Y. Acad. Sci.* 1071, 67–79. doi: 10.1196/annals.1364.007
- Sokal, D. M., Giarola, A. S., and Large, C. H. (2005). Effects of GABA_B, 5-HT_{1A}, and 5-HT₂ receptor stimulation on activation and inhibition of the rat lateral amygdala following medial geniculate nucleus stimulation *in vivo*. *Brain Res.* 1031, 141–150. doi: 10.1016/j.brainres.2004.10.035
- Spampanato, J., Polepalli, J., and Sah, P. (2011). Interneurons in the basolateral amygdala. *Neuropharmacology* 60, 765–773. doi: 10.1016/j.neuropharm.2010.11.006
- Steinbusch, H. W. M. (1981). Distribution of serotonin-immunoreactivity in the central nervous system of the rat cell bodies and terminals. *Neuroscience* 6, 557–618. doi: 10.1016/0306-4522(81)90146-9
- Stein, C., Davidowa, H., and Albrecht, D. (2000). 5-HT_{1A} receptor-mediated inhibition and 5-HT₂ as well as 5-HT₃ receptor-mediated excitation in different subdivisions of the rat amygdala. *Synapse* 38, 328–337. doi: 10.1002/1098-2396(20001201)38:3%3C328::AID-SYN12%3E3.3.CO;2-K
- Stutzmann, G. E., and LeDoux, J. E. (1999). GABAergic antagonist block the inhibitory effects of serotonin in the lateral amygdala: a mechanism for modulation of sensory inputs related to fear conditioning. *J. Neurosci.* 19, 1–4.
- Van Nobelen, M., and Kokkinidis, L. (2006). Amygdaloid GABA, not glutamate neurotransmission or mRNA transcription controls footshock-associated fear arousal in the acoustic startle paradigm. *Neuroscience* 137, 707–716. doi: 10.1016/j.neuroscience.2005.08.061
- Van Oekelen, D., Luyten, W. H., and Leysen, J. E. (2003). 5-HT_{2A} and 5-HT_{2C} receptors and their atypical regulation properties. *Life Sci.* 72, 2429–2449. doi: 10.1016/S0024-3205(03)00141-3
- Wada, Y., Shiraishi, J., Nakamura, M., and Koshino, Y. (1997). Role of serotonin receptor subtypes in the development of amygdaloid kindling in rats. *Brain Res.* 747, 338–342. doi: 10.1016/S0006-8993(96)01322-4
- Whalen, P. J., and Phelps, E. A. (2009). *The Human Amygdala*. New York, NY: The Guilford Press.
- Willins, D. L., Deutch, A. Y., and Roth, B. L. (1997). Serotonin 5-HT_{2A} receptors are expressed on pyramidal cells and interneurons in the rat cortex. *Synapse* 27, 79–82. doi: 10.1002/(SICI)1098-2396(199709)27:1%3C79::AID-SYN8%3E3.0.CO;2-A
- Woodruff, A. R., and Sah, P. (2007). Network of parvalbumin-positive interneurons in the basolateral amygdala. *J. Neurosci.* 27, 553–563. doi: 10.1016/j.neuron.2012.04.022
- Wright, D. E., Seroogy, K. B., Lundgren, K. H., Davis, B. M., and Jennes, L. (1995). Comparative localization of serotonin 1A, 1C, and 2 receptor subtype mRNAs in rat brain. *J. Comp. Neurol.* 351, 357–373. doi: 10.1002/cne.903510304
- Xu, T., and Pandey, S. C. (2000). Cellular localization of serotonin_{2A} (5-HT_{2A}) receptors in the rat brain. *Brain Res. Bull.* 51, 499–505. doi: 10.1016/S0361-9230(99)00278-6
- Zangrossi, H. Jr., and Graeff, F. G. (1994). Behavioral effects of intra-amygdala injections of GABA and 5-HT acting drugs in the elevated plus-maze. *Braz. J. Med. Biol. Res.* 27, 2453–2456.

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