Updated overview of the putative role of the serotoninergic system in obsessive-compulsive disorder

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Abstract: The pathophysiology of obsessive-compulsive disorder (OCD) remains unknown. However, increasing attention has been paid to the putative role of the serotoninergic system, the strongest evidence being based on the widely demonstrated efficacy of serotonin (5HT) reuptake inhibitor antidepressants in the treatment of OCD. The therapeutic effects are correlated with changes in peripheral parameters of 5HT function, which have been found to be altered in OCD, suggesting the possibility of reduced 5HT reuptake capacity. This could reflect a compensatory mechanism presumably due to decreased availability of extracellular 5HT, as evidenced by data derived from direct assessment of central 5HT neurotransmission. The development of new neurochemical probes that explore the sensitivity of various 5HT receptor subtypes has provided precious information. m-Chlorophenylpyperazine (m-CPP), an agonist to 5HT1A, 5HT1D, and 5HT2C receptors, and which also blocks 5HT3 receptors, exacerbates OC symptoms. In contrast, neither MK-212 (6-chloro-2-[1-piperazinyl]-pyrazine), a 5HT1A and 5HT2C receptor agonist, nor ipsapirone or buspirone, which acts as an agonist to 5HT1A receptors, have any effect on OC symptom severity. This suggests the potential implication of the 5HT1D receptor, as shown by the aggravation of OC manifestations in response to sumatriptan, a selective 5HT1D receptor agonist. The 5HT3 plays no specific role, given the lack of influence of the 5HT3 antagonist ondansetron, on OC symptom intensity. Further studies are required to elucidate the pharmacological molecular determinants of the putative 5HT1D receptor dysfunction.

Keywords: serotonin, serotonin reuptake inhibitors, receptors, serotonin, 5HT1D receptor agonists, obsessive-compulsive disorder

Introduction

Obsessive-compulsive disorder (OCD) is a relatively common anxiety disorder characterized by recurrent intrusive thoughts and repetitive time-consuming behaviors, with an estimated lifetime prevalence of 2%–3% in the general population (Antony et al 1998). OCD generally has a chronic course and causes severe distress with a significant impairment in quality of life and social and occupational functioning (Koran et al 1996). To date, the pathophysiology of OCD remains unclear. However, during the last decade, an increasing interest among researchers has contributed to the putative involvement of the serotoninergic function. This assumption primarily stems from indirect arguments based on the well established efficacy of the antidepressant agents with serotonin (5HT) reuptake inhibiting properties for treating OCD (Flament and Bisserbe 1997; Goodman 1999; McDougle 1999; Pigott and Seay 1999).

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After general considerations about the anatomical and functional organization of the 5HT system, the present review examines the putative role of 5HT neurotransmission in OCD through separate and complementary approaches that can be summarized as follows: (1) evaluation of 5HT function in response to drug treatment with a view to establishing strong relationships between the anti-obsessional effects of antidepressant agents acting preferentially by blocking 5HT reuptake process and their influence on peripheral markers of 5HT function; (2) assessment of 5HT function based on direct measurements of some peripheral and central parameters; and (3) exploration of 5HT function with diverse pharmacological challenges for studying a relatively large variety of 5HT receptor subtypes and their importance in the production of OC symptoms. Thereafter, 5HT disruption is discussed within the context of a complex anatomo-functional model for OCD emerging from phenomenological aspects. Finally, possible interactions with other neurotransmitter systems, particularly dopamine, are discussed.

General anatomical and functional characteristics of 5HT system

The 5HT-producing neurons are mainly located in the brainstem raphe nuclei that are described as giving rise to two major groups of neurons: (1) the superior group at the interface between the midbrain and the pons; and (2) the inferior group located more caudally in the pons (Azmitia and Whitaker-Azmitia 1995). They form the largest and most complex neurochemical efferent system in the brain. The superior group of 5HT neurons comprising the dorsal and median raphe nuclei is the source of vast projections to various sites in the forebrain. Rich 5HT innervations of telencephalic limbic regions such as the prefrontal and cingulate cortices, the amygdala, hippocampus, and ventral striatum, and diencephalic structures, especially the hypothalamus and thalamus, are found (Bentivoglio et al 1993; Azmitia and Whitaker-Azmitia 1995; Murphy et al 1998; Stahl 1998; Deutch and Roth 1999) (Figure 1). The dorsal and median raphe nuclei differentially innervate the forebrain target regions. For instance, the dorsal raphe nucleus provides projections primarily to the amygdala and

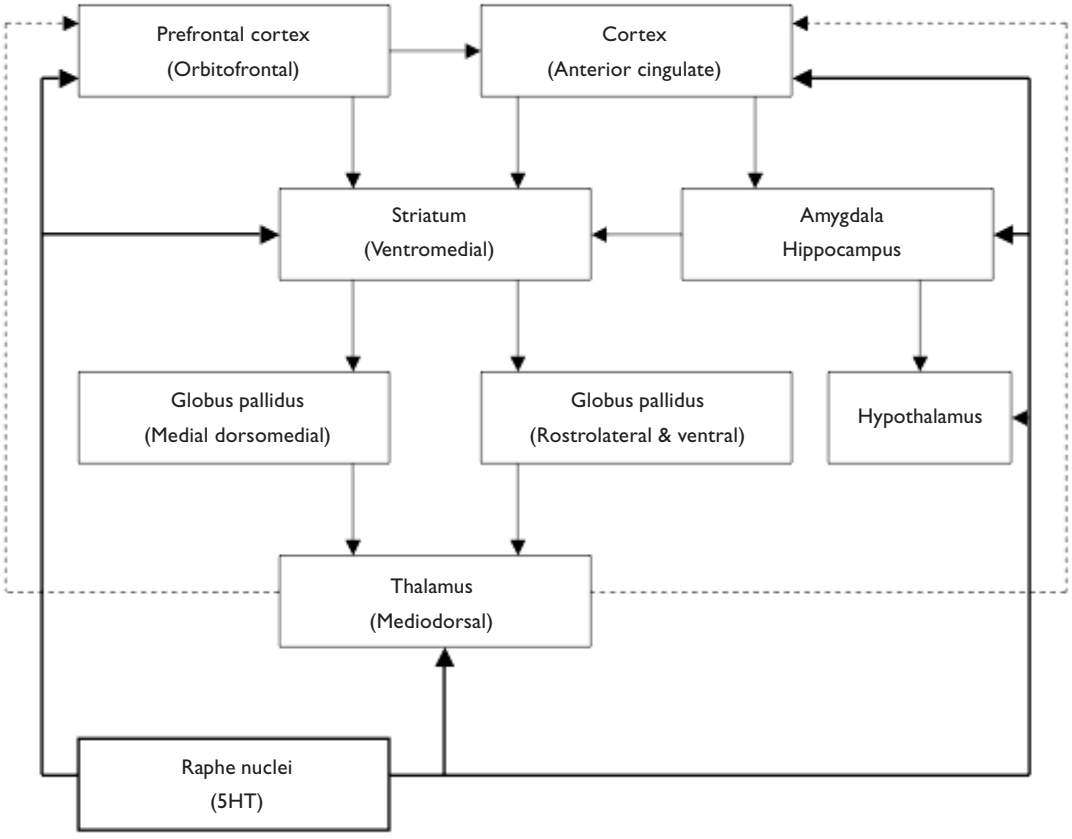

Figure 1 Schematic representation of the serotoninergic projections from the brainstem raphe nuclei to the forebrain and relationship with the cortico-subcortical loops. The cell bodies of the serotoninergic neurons are present in the brainstem raphe nuclei. They provide important innervations to a wide range of limbic target regions in the forebrain, comprising the orbital prefrontal and anterior cingulate cortices, ventral striatum, and thalamus belonging to the orbitofrontal and anterior cingulate loops, and other related structures, especially the amygdala, hippocampus, and hypothalamus.

ventral striatum, whereas the median raphe nucleus preferentially innervates the prefrontal and cingulate cortices and the hippocampus. The lowest levels of 5HT fibers are seen in the motor regions of the frontal lobe (Azmitia and Whitaker-Azmitia 1995). The inferior group of 5HT-containing neurons sends abundant descending spinal projections (Azmitia and Whitaker-Azmitia 1995; Deutch and Roth 1999).

The 5HT is formed via a two-step pathway following the initial active uptake of the essential amino acid precursor tryptophan into the neuron (Deutch and Roth 1999; Sanders-Bush and Mayer 2001). The first step of this synthesis involves the enzyme tryptophan hydroxylase that converts tryptophan to 5-hydroxytryptophan, the intermediate precursor of 5HT. The brain tryptophan hydroxylase is not saturated with substrate. This is very important for explaining the influence of the levels of tryptophan in dietary sources (and thus in brain) on 5HT synthesis (Barr et al 1992; Deutch and Roth 1999; Sanders-Bush and Mayer 2001). The second step is the decarboxylation of 5-hydroxytryptophan in 5HT by the enzyme L-amino acid decarboxylase, which also participates in catecholamine synthesis. 5HT is stored in the vesicles before their fusion to the neuronal membrane and is then released into the synaptic cleft, a process called exocytosis, in a Ca^{2+}-dependent manner in response to depolarizing stimuli. The 5HT actions are mainly terminated by reuptake into presynaptic neurons through 5HT transporter proteins present in the outer membrane of the axon terminals (Deutch and Roth 1999; Sanders-Bush and Mayer 2001). The 5HT transporter surface expression is controlled through kinase-linked pathways particularly involving protein kinase C (PKC) (Qian et al 1997; Blakely et al 1998; Ramamoorthy et al 1998; Ramamoorthy and Blakely 1999). Autoradiographic studies using antidepressant drugs with potent inhibitor profiles for 5HT transporter as radioactive ligands have found high densities of 5HT uptake sites in the brain regions receiving large inputs from the raphe nuclei, including the prefrontal cortex, amygdala, hippocampus, ventral striatum, hypothalamus, and thalamus (Bentivoglio et al 1993; Barker and Blakely 1995; Meneses 1999). The 5HT uptake system has also been identified in a number of specialized non-neuronal cells, especially in platelet and lymphocyte membranes taking up 5HT from the blood, thereby keeping the 5HT levels in blood low (Barker and Blakely 1995; Sanders-Bush and Mayer 2001). In addition, a metabolic degradation of 5HT occurs primarily involving the enzyme monoamine oxidase type A (MAO-A) present on the outer surface of mitochondria, which metabolizes 5HT to form 5-hydroxyindole acetaldehyde. This aldehyde is then converted to 5-hydroxyindole acetic acid (5-HIAA) by the ubiquitous mitochondrial enzyme aldehyde dehydrogenase (Sanders-Bush and Mayer 2001).

The current, widely accepted classification proposes at least seven distinct recognition sites for 5HT (Sanders-Bush and Canton 1995; Murphy et al 1998; Stahl 1998; Sanders-Bush and Mayer 2001). All members of the 5HT1 receptor subtype belong to the superfamily of G protein-coupled receptors. They generally inhibit adenylate cyclase, leading to decreased cyclic adenosine monophosphate (cAMP) production. The 5TH1A receptor represents a somato-dendritic autoreceptor on the cell body of 5HT neurons in the brainstem raphe nuclei. Another subtype, the 5HT1B receptor (and its human homolog, 5HT1D) has been found to function as an autoreceptor on axon terminals. When activated, these two receptors decrease the intrinsic firing of the raphe cells, thereby inhibiting 5HT release. The 5HT1A receptors have also been characterized at post-synaptic sites. A significant amount of 5HT1B receptors are present on postsynaptic structures, yet their function is still unknown (Sanders-Bush and Canton 1995; Murphy et al 1998; Marek and Aghajanian 1999; Sanders-Bush and Mayer 2001). The 5HT1 receptor subtypes are mainly located in the brainstem raphe nuclei and various cortical and subcortical limbic target regions (Boschert et al 1994; Glennon and Dukat 1995; Stahl 1998; Meneses 1999). The so-called 5HT2 receptor family comprises characterized receptors that have been referred to as 5HT2A and 5HT2C receptors coupled to G proteins. They are associated primarily with phospholipase C that catalyzes the hydrolysis of phosphatidylinositol bisphosphate (PIP2) and generates the second messenger molecules inositol triphosphate (IP3) (which mobilizes calcium from intracellular stores resulting in PKC activation) and diacylglycerol (which potentiates PKC activation). PKC regulates numerous processes of cell function. For example, PKC activation causes a reduction in 5HT uptake capacity by phosphorylation and sequestration of 5HT transporter proteins (Qian et al 1997; Blakely et al 1998; Ramamoorthy et al 1998; Ramamoorthy and Blakely 1999). The 5HT2 receptor subcategories have a widespread distribution in the brain (Glennon and Dukat 1995; Meneses 1999; Sanders-Bush and Mayer 2001). The 5HT3 receptors are members of the large family of ligand-operating ion channels that regulate the permeability of cation channels. They are primarily located on nerve terminals, where they facilitate 5HT release (Sanders-Bush

and Canton 1995; Murphy et al 1998; Marek and Aghajanian 1999; Sanders-Bush and Mayer 2001). In the central nervous system, these receptors are present in the prefrontal and entorhinal cortices, the amygdala and hippocampus, or in the area postrema. They have also been identified in the gastrointestinal tract, so they are involved in the emetic response (Glennon and Dukat 1995; Stahl 1998; Meneses 1999; Sanders-Bush and Mayer 2001). Four additional 5HT receptor subcategories have more recently been characterized (5HT4, 5HT5, 5HT6, and 5HT7). Although little is still known about the pharmacology and functional role of these receptors, most are coupled to G proteins and stimulate adenylate cyclase, resulting in cAMP accumulation and activation of protein kinase A (PKA) (Sanders-Bush and Canton 1995; Murphy et al 1998; Sanders-Bush and Mayer 2001). However, unlike PKC, PKA-triggered phosphorylation seems to have no modulating effect on 5HT transporter function (Blakely et al 1998).

The 5HT system has been widely demonstrated to be involved in the pathogenesis of diverse mental illnesses such as OCD. Several lines of evidence suggest that the dysfunction of 5HT neurotransmission, and especially an altered sensitivity of the 5HT receptor subtype, may constitute a crucial factor in the pathophysiology of OCD.

Drug treatment responses

The strongest evidence for the putative role of the 5HT system is provided by the increasing number of clinical studies pointing to the efficacy of 5HT reuptake inhibitor antidepressants (SRIs) for the pharmacological treatment of OCD. The earlier investigations were performed with the tricyclic antidepressant clomipramine, which has a preferential serotoninergic profile. It has extensively been shown that clomipramine consistently reduces OC symptomatology (Flament and Bisserbe 1997; Goodman 1999; McDougle 1999; Pigott and Seay 1999). A positive correlation has been found between plasma clomipramine levels and the anti-obsessional response (Stern et al 1980; Mavissakalian et al 1990). In contrast, plasma levels of its major metabolite N-desmethylclomipramine, which exerts a potent blockade of norepinephrine reuptake, were not related to the clinical outcome (Stern et al 1980; Mavissakalian et al 1990). Many studies have also documented the importance of non-tricyclic selective 5HT reuptake inhibitors (fluvoxamine, fluoxetine, paroxetine, sertraline, and citalopram) as anti-obsessional medications (Flament and Bisserbe 1997; Goodman 1999; McDougle 1999; Pigott and Seay 1999). The efficacy of these agents contrasts with the lack of therapeutic effects of anti-depressants such as desipramine, which has a preponderant blocking action on norepinephrine reuptake (Ananth et al 1981; Volavka et al 1985; Goodman et al 1990; Hoehn-Saric et al 2000).

The central role of 5HT neurotransmission is illustrated by the change in peripheral markers of 5HT function associated with the improvement in OCD symptoms after treatment with SRIs.

In the early study by Flament et al (1987), platelet 5HT contents in OCD patients exhibited a profound decrease after a five-week period of treatment with clomipramine, which was positively correlated with the clinical improvement. However, a lesser reduction in 5HT content in whole blood after one week of treatment with paroxetine or clomipramine was more recently found to be associated with a better anti-obsessional response at 12 weeks (Humble et al 2001). Treatment with clomipramine for three weeks was also reported to reduce the concentrations of 5-HIAA, the major central metabolite of 5HT, in the lumbar cerebrospinal fluid (CSF) (as a measure of central 5HT turnover) of OCD patients. This progressive decline in CSF levels of 5-HIAA was positively correlated with an alleviation in OCD symptoms (Thoren et al 1980). Similar results were found in OCD children and adolescents treated with clomipramine over a longer period of time ranging from 8.5 to 34 months (Altemus et al 1994). These findings support the correlation found between the decrease in CSF 5-HIAA levels and the 5HT reuptake inhibiting activity determined in vitro in plasma of depressed patients receiving clomipramine (Asberg et al 1977). Thus, it can be postulated that an enhanced availability of 5HT at the synapse, due to presynaptic reuptake inhibition, may be a central factor in the clinical efficacy of antidepressant agents in OCD.

Such a phenomenon might lead to a variety of adaptive changes in 5HT transporter function, as suggested by a gradual elevation of the number of 3H-imipramine binding sites labeling the 5HT transporter in blood platelets until normalization after an eight-week trial with either fluvoxamine or clomipramine. This was paralleled by an improvement in OC manifestations (Marazziti et al 1997). An enhanced 5HT reuptake rate with a decrease in the inhibitory effect of PKC occurred after six months of treatment of OCD with various SRIs (Marazziti et al 2002). This could be a possible consequence of a reduction in the platelet levels of IP3, an intracellular product of the 5HT2 receptor-mediated phosphoinositide hydrolysis stimulating PKC, before returning to normal values after an eight-week

SRI treatment (Delorme et al 2004). This decrease in the platelet IP3 content was positively correlated with a reduction in the number of 5HT2A receptor binding sites (Delorme et al 2004), which could be reflective of postsynaptic receptor down-regulation caused by SRI administration (Maes and Meltzer 1995; Duman 1999).

Consequently, these data suggest that 5HT function appears to mediate the reducing action of SRI medications on OC symptoms. They raise the essential question of putative disturbances in the availability of extracellular 5HT in OCD, which might affect: (1) presynaptic release regulated by 5HT autoreceptors; (2) reuptake into nerve terminals; and/or (3) metabolic inactivation, before being reversed during successful treatments with SRIs.

Peripheral and central markers

Over the last decade, a growing body of literature has explored the functioning of the 5HT system in OCD. Some studies have shown abnormalities of several markers, thereby reflecting the functional activity of 5HT neurotransmission.

Although no change was found in platelet (Flament et al 1987), whole blood (Hanna et al 1991; Delorme et al 2004), 5HT content, or in platelet MAO activity (Flament et al 1987), enhancement of CSF levels of 5-HIAA, possibly reflecting increased brain 5HT turnover, was observed in OCD patients (Insel et al 1985). However, these results have not been confirmed in other investigations that reported normal CSF concentrations of 5-HIAA (Thoren et al 1980; Leckman et al 1995). A positive correlation was found between the CSF levels of 5-HIAA in children with primary OCD and only one of the eight baseline measures of clinical symptom severity (NIMH Global OCD scale score), and the three Leyton Obsessional Inventory-Child Version improvement scores after five weeks of treatment with clomipramine (Swedo et al 1992). Therefore, these reports fail to establish any clear association between 5HT turnover characteristics and OC symptoms.

Many investigations have documented an alteration of the 5HT reuptake process in OCD, as shown by modifications of three distinct parameters of the 5HT transporter protein function, ie, number, affinity, and velocity. Most of the studies to date have used 3H-imipramine or the more selective ligand 3H-paroxetine as an index of the 5HT reuptake sites in platelets or lymphocytes. A reduction in 3H-impramine binding was found in OCD patients (Weizman et al 1986). Several research groups confirmed these results by finding fewer platelet or lymphocyte 3H-impramine or 3H-paroxetine binding sites in OCD (Bastani et al 1991; Marazziti et al 1992, 1996, 2003; Sallee et al 1996; Delorme et al 2004). Nevertheless, others failed to replicate these findings and reported a normal density in platelet 5HT transporter (Insel et al 1985; Black et al 1990; Kim et al 1991; Vitiello et al 1991). A decreased affinity for 5HT reuptake was found (Bastani et al 1991). Although normal (Weizman et al 1986; Bastani et al 1991; Marazziti et al 1992) or increased velocity of 5HT uptake has been observed (Vitiello et al 1991), a lower 5HT reuptake velocity at baseline and in response to PKC activation in platelets of OCD patients was recently found compared with normal controls (Marazziti et al 2000). This is consistent with the inhibitory effects of PKC leading to the internalization of 5HT transporter and loss of functional transport capacity (Qian et al 1997; Blakely et al 1998; Ramamoorthy et al 1998; Ramamoorthy and Blakely 1999), and suggests an elevated PKC activity in OCD. These findings strictly parallel those by Delorme et al (2004) showing a disturbance in the second messenger pathway coupled to 5HT2 receptors with enhanced IP3 concentrations in the platelets of OCD patients. This might increase the release of calcium from internal storage sites and therefore PKC activity. Abnormalities in the PKA have also been reported with decreased c-AMP-stimulated PKA activity (Perez et al 2000) and a reduced phosphorylation state of the PKA substrate Rap1 (Tardito et al 2001) in the platelets of OCD patients. However, changes in PKA activity and 5HT uptake kinetic have still not been demonstrated to be linked. On the other hand, although no correlation was found between platelet 3H-impramine binding site density and clinical measures of symptom severity (Black et al 1990), the number of 3H-paroxetine sites was negatively correlated with OC symptom intensity (Marazziti et al 1996). Therefore, these observations are, in general, suggestive of relationships between decreased capacity of presynaptic 5HT reuptake and OC manifestations.

5HT transporter dysfunction appears to occur in platelets, which have become the most widely used peripheral index of the central 5HT system. However, this raises the crucial question of whether such abnormalities are also present in the central 5HT system. For this reason, brain 5HT transporter availability has recently been investigated in OCD patients using functional neuroimaging techniques. It has been shown that 5HT transporter availability is increased in the midbrain-pons. This could result from a decreased competition by lower endogenous 5HT levels leading to a greater number of available reuptake binding sites (Pogarell

et al 2003). In contrast, there was normal 5HT transporter availability in the limbic and subcortical regions, including the anterior cingulate cortex, hippocampus, amygdala, ventral striatum, and thalamus (Simpson et al 2003).

To summarize, indirect evidence for the involvement of the 5HT system has emerged from numerous pharmacological studies showing the therapeutic effects of SRIs in OCD, which thus appear to be associated with changes in peripheral indices of 5HT function. Direct analysis of peripheral and central markers of 5HT neurotransmission suggests that OCD could be associated with decreased availability of extracellular 5HT. This hypothesis leads to the consideration that specific abnormalities of 5HT release and/or 5HT receptors could exist in OCD. From this perspective, a reduction in 5HT reuptake capacity could be considered as a compensatory mechanism in the attempt to maintain the availability of extracellular 5HT.

Pharmacological challenges

Another powerful technique for evaluating the role of 5HT in OCD is investigating whether pharmacological challenges acting on 5HT receptors or 5HT subsystems exacerbate or improve the symptoms of OCD. Together with neuroendocrine measures, this procedure is useful for obtaining important information about the responsivity of the 5HT system in OCD.

m-CPP (m-chlorophenylpiperazine) has been the most frequently used probe to examine 5HT receptor function in the studies of OCD. m-CPP is a non-selective agonist to 5HT1A, 5HT1D, 5HT2C receptor subtypes that antagonizes stimulation of 5HT3 receptors (Gross et al 1998; McDougle 1999). It binds equipotently and with the greatest affinity to 5HT2C and 5HT3 receptor subtypes, and to a lesser extent to 5HT1A and 5HT1D receptors (Gross et al 1998; McDougle 1999). Zohar et al (1987) were the first to examine the behavioral responses to m-CPP in OCD. Oral m-CPP at the dose of 0.5 mg/kg, but not placebo, resulted in a brief and significant exaggeration of OC symptoms in 11/12 (92%) OCD patients. Hollander et al (1992) confirmed these results with a transient worsening of OC symptoms in 11/20 (55%) OCD patients in similar experimental conditions. Erzegovesi et al (2001) reported an increase in OC symptom severity in 6/12 (50%) OCD patients with 0.25 mg/kg oral administration of m-CPP, whereas the usual dose of 0.5 mg/kg caused an aggravation of OC symptoms in only 1/12 (8%) OCD patients. Other authors found no significant effect of oral m-CPP at the standard dose of 0.5 mg/kg on OC symptoms (Pigott et al 1993; Goodman et al 1995; Ho Pian et al 1998a; Khanna et al 2001). Moreover, there was also no exacerbation of OC symptoms after intravenous challenge with m-CPP (0.1 mg/kg) (Charney et al 1988), although a significant symptom aggravation was more recently described (Pigott et al 1993; Broocks et al 1998). The clinical heterogeneity of the patient populations studied and differences in the rate of intravenous m-CPP infusion might account for these discrepancies (McDougle 1999).

Concordant results have been reported regarding the neuroendocrine effects of m-CPP in OCD. A blunted response of cortisol (Zohar et al 1987; Khanna et al 2001) or prolactin (Charney et al 1988; Hollander et al 1992; Khanna et al 2001) to oral or intravenous m-CPP administration was reported in OCD patients compared with normal healthy volunteers.

Despite the disparate findings, the exaggerated behavioral responses to m-CPP documented in some of the previous studies support the hypothesis that OCD patients may be hypersensitive at the level of the 5HT receptor subtype in the brain regions involved in the production of OC symptoms, among which the orbitofrontal and anterior cingulate cortices or the caudate nucleus are the most often cited (Saxena et al 1998; Baxter 1999; Aouizerate et al 2004). This contrasts with the possible hyposensitivity of the 5HT receptor subcategory present at the level of the hypothalamic-pituitary-adrenal axis of OCD patients, as shown by attenuated neuroendocrine responses to m-CPP. There is evidence that the neuroendocrine properties of m-CPP are related to direct agonist effects on postsynaptic 5HT receptors by stimulation of 5HT1A and 5HT2C receptor subtypes (Mueller et al 1986; Hamik and Peroutka 1989; Cowen et al 1990). The actions of the drug are probably mostly due to activation of 5HT2C receptors because of its lower affinity to 5HT1A receptor subtype (Gross et al 1998; McDougle 1999). Therefore, the observations of blunted effects of m-CPP on cortisol or prolactin secretion are more indicative of 5HT2C receptor dysfunction in the brain regions that mediate hormonal responses. Platelet 5HT2 receptors have been studied in OCD (Pandey et al 1993). The number and affinity of 5HT2 receptor binding sites did not differ in OCD patients from those of normal healthy volunteers, and there was no correlation with the clinical severity of the illness. Thus, a disruption in 5HT2 receptor-mediated signal transduction might occur, as supported by enhanced platelet IP3 concentrations in OCD (Delorme et al 2004).

These functional abnormalities of 5HT receptors appear to be reversed in part by effective treatments of OCD. Zohar

et al (1988) reexamined the behavioral and hormonal effects of oral m-CPP in OCD patients following four months of successful treatment with clomipramine. m-CPP failed to significantly increase OC symptoms. However, the hormonal responses remained unchanged. Similar findings were reported after at least 12 weeks of effective fluoxetine treatment (Hollander et al 1991). The inability to normalize neuroendocrine effects of m-CPP is presumably due to the hyporesponsivity of postsynaptic 5HT2C receptors, which would not be reversed by SRI administration.

MK-212 (6-chloro-2-[1-piperazinyl]-pyrazine) is a direct-acting 5HT agonist that possesses high affinity for 5HT1A and 5HT2C receptor subtypes (Gross et al 1998). Bastani et al (1990) assessed the behavioral and hormonal responses to MK-212 (20 mg orally [os]) in OCD patients. There was no change in the intensity of OC symptoms, while attenuated cortisol and prolactin responses were observed.

Ipsapirone is an azapirone derivative with total presynaptic 5HT1A receptor agonist activity and partial agonist properties at postsynaptic 5HT1A receptor sites. It is more selective for the 5HT1A receptor subtype compared with buspirone, the prototypical agent in this class, which also possesses a dopamine antagonistic activity (Barr et al 1992; McDougle 1999). Administered orally at the dose of 0.3 mg/kg in OCD patients, ipsapirone had no influence on OC symptom intensity. Normal ACTH and cortisol responses were found (Lesch et al 1991). Similarly, no behavioral and neuroendocrine effects of buspirone (30 mg os) were found (Lucey, Butcher, et al 1992; Norman et al 1994). Therefore, dysfunction of the 5HT1A receptor subtype does not seem to be implicated in the pathophysiology of OCD.

Fenfluramine exerts a complex action on the brain 5HT system by inhibiting 5HT reuptake and facilitating presynaptic 5HT release from storage granules. It also has direct postsynaptic 5HT agonist effects. However, the d- and l-isomers of fenfluramine have been shown to differ in their activity profile. The d-isomer preponderantly interacts with the 5HT system compared with the l-isomer, which modulates dopamine function (Barr et al 1992; McDougle 1999). Fenfluramine challenges in OCD patients have yielded relatively consistent findings. Racemic dl-fenfluramine (60 mg os) had no effect on OC symptom severity (Hollander et al 1992; McBride et al 1992), and prolactin and cortisol responses were normal (Hollander et al 1992, 1993; McBride et al 1992). However, lowered prolactin (Lucey, O'Keane, et al 1992; Monteleone, Catapano, Bortolotti, et al 1997) and cortisol responses (Lucey, O'Keane, et al 1992; Monteleone, Catapano, Tortorella, et al 1997) to d-fenfluramine have been found, although only in female patients, with normalization occurring after 10 weeks of effective treatment with fluvoxamine (Monteleone, Catapano, Bortolotti, et al 1997).

L-tryptophan is the initial dietary 5HT precursor. Because the enzyme tryptophan hydroxylase is not saturated with substrate at physiologic concentrations, addition of exogenous tryptophan stimulates 5HT synthesis (Barr et al 1992; Deutch and Roth 1999; Sanders-Bush and Mayer 2001). Studies of the behavioral and neuroendocrine effects of the L-tryptophan in OCD have not been numerous. No influence of the 5HT precursor (7 g intravenous [IV]) was found on OC symptoms (Charney et al 1988), while the prolactin response was significantly greater in OCD patients compared with normal healthy controls (Charney et al 1988), although the latter finding was not subsequently replicated (Fineberg et al 1994).

Tryptophan depletion induced by drinking a tryptophan-deficient amino acid mixture is an experimental procedure that temporarily and markedly reduces blood tryptophan, brain tryptophan, 5HT, and 5-HIAA in laboratory animals with similar lowering in plasma available tryptophan in humans (McDougle 1999; Van der Does 2001). When applied in OCD patients, tryptophan depletion did not induce any worsening of OC symptoms (Smeraldi et al 1996). Moreover, no effect of tryptophan depletion was found on OC symptom severity after successful treatment with various SRIs, while a rapid return of depressed mood was observed (Barr et al 1994). This suggests that acute depletion in 5HT availability has no influence on OC symptoms. However, reduction in brain 5HT that tryptophan depletion challenge produces could be insufficient to aggravate OC manifestations. Furthermore, the therapeutic effects of SRIs in OCD, but not in depression, could not depend on the short-term availability of 5HT (Barr et al 1994).

Methergoline is a non-selective antagonist to 5HT1/5HT2 receptor subtypes that has been shown to block physiological and hormonal responses to m-CPP in normal healthy volunteers (Mueller et al 1986). No increase in OC symptom severity was found following the oral administration of methergoline (4 mg) in OCD patients (Zohar and Insel 1987), but oral pretreatment with methergoline at the same dose completely abolished the m-CPP-induced exacerbation of OC symptoms (Pigott et al 1991, 1993). However, OC symptoms emerged after administration of methergoline to OCD patients who favorably responded to treatment with clomipramine (2.5–24 months) (Benkelfat

et al 1989). These observations support the central role of 5HT neurotransmission in the m-CPP-elicited induction of OC symptoms and their improvement by treatment with SRIs.

The response of OCD patients to a series of 5HT pharmacological challenges has been extensively studied. Importantly, OC symptoms worsened with m-CPP, which acts as an agonist to 5HT1A, 5HT1D, 5HT2C receptors and as antagonist to the 5HT3 receptor. In contrast, MK-212, which demonstrates an affinity for 5HT1A and 5HT2C receptor subtypes, and the 5HT1A receptor agonists ipsapirone or buspirone, have no effect on OC symptom intensity. These observations support the hypothesis that 5HT1D and/or 5HT3 receptor subtypes may be involved in the production of OC symptoms. In this respect, oral sumatriptan, a selective 5HT1D receptor agonist, produced a transient and significant aggravation of OC symptoms at the dose of 100 mg (Gross et al 1998), a result confirmed in more recent investigations (Stein et al 1999; Koran et al 2001). However, these provocative findings failed to be replicated in other studies (Ho Pian, Westenberg, van Megan, et al 1998; Boshuisen and den Boer 2000) using sumatriptan or zolmitriptan, a recent selective 5HT1D receptor agonist, which has better brain-penetrating properties than sumatriptan. In addition, the 5HT3 receptor subtype does not mediate m-CPP-induced worsening of OC symptoms. Ondansetron, a potent 5HT3 antagonist, administered alone (0.15 mg/kg IV) or in combination with m-CPP (0.08 mg/kg IV) in OCD patients, failed to induce any significant change in OC symptom severity (Broocks et al 1998). Therefore, a 5HT1D receptor hyperresponsivity appears to be associated with the emergence of OC manifestations. Such a dysfunction of the terminal 5HT1D autoreceptor may be expected to reduce presynaptic release and thus lead to decreased availability of extracellular 5HT. Another example of evidence for the so-called "5HT1D receptor hypothesis" (Zohar and Kindler 1992) is illustrated by a family-based association study of the 5HT1Dβ receptor gene (Mundo et al 2000, 2002). G and C alleles of this gene differ by a single base pair at nucleotide 861 of the coding region (Pato et al 2002). Significant linkage disequilibrium between the G861C variant of the 5HT1Dβ receptor gene and OCD was found, with preferential transmission of the G allele to the affected subjects (Mundo et al 2000, 2002). Although this association has not been confirmed (Di Bella et al 2002; Camarena et al 2004), subjects with a preferential transmission of the G allele experienced more severe OC symptoms than those carrying the C allele (Camarena et al 2004). Thus, it can be concluded that there is a putative involvement of the 5HT1Dβ receptor gene in the etiopathogenesis of OCD.

Perspectives

Abundant neurochemical data obtained with various approaches suggest the central role of the 5HT system in OCD. In this respect, we will first seek to establish strong relationships with the anatomo-functional model of OCD that has emerged due to the existence of functional abnormalities in the frontal-subcortical loops, and second, to define the importance of possible interactions with other neurotransmitter systems, particularly dopamine, in the genesis of OC symptomatology.

Toward a relationship with the anatomo-functional model of OCD

Although the pathophysiology of OCD is still far from resolved, functional neuroimaging studies have provided important information about the brain regions involved in OCD. A consensus is emerging regarding an enhanced metabolic activity in several brain areas including the orbitofrontal and anterior cingulate cortices, the ventral striatum (head of the caudate nucleus) and the thalamus (Saxena et al 1998; Baxter 1999; Aouizerate et al 2004), and our knowledge of the physiology of the cortico-subcortical functional loops involving these brain regions in the expression of OC symptoms is increasing.

Phenomenologically, the central point of the obsessional symptomatology is the subjective impression that "something is wrong" (Schwartz 1998, 1999). In other words, obsessions may be considered as resulting from the recurrent perception of a mistake and/or error in response to specific environmental stimuli. Compulsions are defined as behavioral responses performed to alleviate the internal tension or anxiety produced by exposure to the stimuli. This relief may be thought as a form of reward. Nevertheless, it is usually brief and immediately followed by a resurgence of profound discomfort. This results in the urge to carry out reward-directed behaviors in an unreasonable and excessive way on the basis of an internal, emotional, and motivational drive. These phenomenological aspects are of major significance in terms of processes disrupted within OCD, including: (1) error recognition; and (2) emotion and motivation and their activational aspects (eg, activations for initiation and sustaining behavioral reactions and tendency to work for reward) (Aouizerate et al 2004). Therefore, it

can be assumed that a dysfunction of the brain regions mediating the processes of error detection and/or management of the reward and emotion occurs in OCD. In this respect, it has been shown that the orbitofrontal cortex (OFC) is involved in appraisal in determining the emotional and motivational values of environmental information, and in integrating the subject's prior experience, which is crucial in decision-making (Charney and Bremner 1999; Tremblay and Schultz 1999, 2000a, 2000b; Krawczyk 2002; Ramnani and Owen 2004). The OFC also contributes to the selection, comparison and judgment of stimuli, and error detection (Rosenkilde et al 1981; Thorpe et al 1983; Ramnani and Owen 2004). The anterior cingulate cortex (ACC) is divided into: (1) a ventral or affective region that could keep attention on the internal emotional and motivational status and participates in the regulation of autonomic responses; and (2) a dorsal and cognitive region that serves a wide range of functions such as attention, working memory, error detection, conflict monitoring, response selection, and anticipation of incoming information (Niki et Watanabe 1979; Devinsky et al 1995; Shima and Tanji 1998; Bush et al 2000, 2002; Akkal et al 2002; Shidara and Richmond 2002; Ito et al 2003; Kerns et al 2004). The ventral striatum, which is intimately connected to the OFC and ACC, is primarily involved in the preparation, initiation, and execution of behavioral responses oriented toward reward delivery after cognitive and emotional integration of behaviorally relevant information at the cortical level (Hollerman et al 1998; Tremblay et al 1998; Hassani et al 2001). Thus, these observations indicate the crucial role of the brain regions belonging to the orbitofrontal and anterior cingulate loops in the pathophysiology of OCD in the light of phenomenological evidence.

The responsibility of 5HT neurotransmission for the dysfunction of the frontal-subcortical loops that emanate from the OFC and ACC remains to be established (Figure 1). However, there are several strands of indirect evidence particularly implicating the 5HT1D receptor in the mediation of these functional abnormalities. First, in various animal species, there is an abundant 5HT innervation along with an expression of the terminal autoreceptor 5HT1B (termed 5HT1D in humans) in the cortex, especially in the limbic areas and related structures (eg, ventral striatum, amygdala, hippocampus, and thalamus) (Boschert et al 1994; Glennon and Dukat 1995; Stahl 1998; Meneses 1999). Second, antidepressant drugs that predominantly inhibit 5HT reuptake have largely proven effective for treating OCD (Flament and Bisserbe 1997; Goodman 1999; McDougle 1999; Pigott and Seay 1999), resulting in a progressive decline in the metabolic activity of the brain regions cited above (Saxena et al 1998; Baxter 1999; Aouizerate et al 2004). Third, the anti-obsessional response of SRIs appears to be related to a desensitization of the terminal 5HT autoreceptor in the orbitofrontal cortex, disinhibiting neuronal firing and then increasing 5HT tonus (Bergqvist et al 1999). Fourth, pharmacological agents that conversely reduce 5HT function by acting as agonists to the terminal autoreceptor 5HT1D have been shown to aggravate the severity of OC manifestations (Zohar et al 1987; Hollander et al 1992; Pigott et al 1993; Broocks et al 1998; Erzegovesi et al 2001), thus suggesting that the 5HT1D receptor subtype may be hypersensitive in OCD. This is particularly important for explaining the delayed onset of the optimal beneficial effects of SRIs in OCD and the long-time course for this desensitization of the terminal 5HT autoreceptor, which also depends on the brain region (Stahl 1998). Preclinical studies have found that the delay to obtain desensitization of the 5HT autoreceptor is longer in the orbitofrontal cortex than in other cortical regions (El Mansari et al 1995). Thus, although there is no direct conclusion of a clearly specific 5HT abnormality to date, a 5HT component particularly involving the 5HT1D receptor subtype seems to exist in the pathophysiology of OCD.

Toward a possible interaction with the dopaminergic system in OCD

5HT function cannot be considered as solely implicated in the pathogenesis of OCD. Among the putative neurotransmitter systems that are thought to play a major role in the expression of OC symptoms, the dopamine system with which 5HT interacts, probably represents the most likely candidate. First, antidopaminergic agents have favorable effects in the management of SRIs-resistant forms of OCD with and without comorbid tics (McDougle et al 1994, 1995, 2000; Saxena et al 1996). Second, a disturbance in dopaminergic function seems to be observed in OCD, as supported by recent neuroimaging studies showing an increase in dopamine transporter binding along with lowered levels of dopaminergic D2 receptor binding in the basal ganglia of patients with OCD (Kim et al 2003; Denys et al 2004). This might reflect a compensatory mechanism resulting from higher synaptic concentrations of dopamine in the striatum, leading to an elevation in the dopamine transporter and to a down-regulation of the D2 receptor. Third, many studies have underlined the fundamental

importance of dopamine in reinforcement (Kiyatkin 1995; Le Moal 1995; Piazza and Le Moal 1996, 1997; Koob and Le Moal 1997; Schultz 1998, 2000, 2002; Horvitz 2000; Vallone et al 2000; Nieoullon 2002; Salamone and Correa 2002). Mesocorticolimbic dopamine neurons originating in the ventral tegmental area and projecting largely to the nucleus accumbens with other limbic ventral striatal regions and cortical areas, especially the OFC and ACC (Horvitz 2000; Vallone et al 2000; Nieoullon 2002), are essential for the learning and motivational processes, two critical aspects of behaviors oriented to reaching goals or obtaining rewards (Salamone and Correa 2002). Dopamine neurons encode a reward error-prediction rule and provide information about the predictability of the reward, which is central in reward-directed learning (Schultz 1998, 2000, 2002). They are modeled as the "critic" that produces evaluative feedback by observing the consequences of a given action on the environment, and which generates error signal when differences are perceived between predictions and reality (Bar-Gad and Bergman 2001). Dopamine neurons are also involved in the activational aspects of motivation (eg, activation to obtain and effort in working for reward delivery) (Salamone and Correa 2002). In other words, overactivity of the mesocorticolimbic system might be reflected by the occurrence of recurrent obsessions resulting from excessive and inappropriate feedback error signals upon exposure to behavior-inducing stimuli. It may also be essential in the emergence of compulsive behaviors as reinforcing behavioral approach intended to reduce distress related to intrusive thoughts (Aouizerate et al 2004). Thus, the existence of anatomical and functional 5HT-dopamine interactions at both cortical and subcortical levels (Kapur and Remington 1996) and the modulating influence of 5HT on motor behavior and its flexibility (Lucki 1998; Clarke et al 2004) suggest that 5HT might act through the dopaminergic system in the production of OC symptoms. However, the exact significance of such interactions between 5HT and dopamine functions remains to be more elucidated.

Conclusion

The widely demonstrated efficacy of SRIs for the treatment of OCD suggests the important role of the 5HT system in the pathogenesis of OCD. Clinical studies based on pharmacological challenges designed to manipulate the functional activity of the 5HT system and its receptor subtypes have failed to establish firm conclusions about the induction of OC symptoms by 5HT mechanisms. However, the use of m-CPP and sumatriptan remains an important issue as shown by the worsening of clinical symptoms that these agents induce. Investigations into this issue might throw light on the potential importance of the 5HT1D receptor subtype underlying the symptomatic expression of OCD. Further research into developing more selective probes of the 5HT1D receptor subsystem would provide more information about this 5HT receptor subcategory associated with the production of OC symptoms and about the pharmacological molecular bases of the putative 5HT1D receptor subtype malfunction.

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