The serotonin system in autism

Edwin H. Cook, Jr, MD, and Bennett L. Leventhal, MD

The serotonin system has been implicated as a factor in some cases of autism since the finding in 1961 of elevated serotonin (5-hydroxytryptamine) levels in the blood of patients with autism. This has been clarified as elevation in the platelet content of serotonin. Subjects with elevated whole blood serotonin levels have been shown to have elevated platelet serotonin transport into platelets and decreased serotonin 5-HT_2 receptor binding. Most individuals with autism who are treated with potent serotonin transporter inhibitors have a reduction in ritualistic behavior and aggression. Reduction of central nervous system serotonin, induced by acute tryptophan depletion, causes a worsening of stereotyped behavior. Recent developments in the molecular biology of serotonin receptors are reviewed.

Departments of Psychiatry and Pediatrics, MC3077, University of Chicago, 5841 South Maryland Avenue, Chicago, IL 60637, USA.

Current Opinion in Pediatrics 1996, 8:348–354

Abbreviations

CNS	central nervous system
5-HT	5-hydroxytryptamine
LSD	lysergic acid diethylamide

ISSN 1040–8703

Although the function of serotonin (5-hydroxytryptamine [5-HT]) in the central nervous system is still being clarified, various studies have indicated an important role for serotonin in central nervous system (CNS) development, social behavior, sleep, aggression, anxiety, and affective regulation. Therefore, it is not surprising that serotonin has been the most intensively studied neurochemical in autism over the past three decades. Recent advances in pharmacology, molecular biology, and genetics have increased our knowledge of the serotonin system and hold out the promise of the development of improved pharmacologic treatment for at least some aspects of autism, ritualistic behaviors and aggression.

Hyperserotonemia

In 1961, Schain and Freedman [1] reported elevation of whole blood serotonin levels (hyperserotonemia) in patients with autism. A series of studies were performed to test the hypothesis that increased blood serotonin levels indicated a toxic metabolic accumulation of serotonin. A similar hypothesis was supported for the contemporary finding of phenylketonuria. However, the hypothesis that hyperserotonemia in autism was a reflection of toxic levels of serotonin was not supported. In the process of testing this hypothesis, several possible artifacts, including diet, were excluded [2,3], and the finding of hyperserotonemia in at least 25% of individuals with autism was confirmed.

An assay for platelet-poor plasma serotonin developed by Anderson *et al.* [4] revealed that more than 99% of whole blood serotonin is contained in platelets. Use of this assay revealed that when whole blood serotonin levels were elevated, platelet-poor plasma ultrafiltrate serotonin levels were not elevated in subjects with autism [5]. Because serotonin is both actively transported into platelets and released from platelets, either an increase in serotonin transport into platelets or a decrease in the release of serotonin from platelets would lead to an increased steady-state level of serotonin in platelets.

From a technical perspective, direct measurement of platelet serotonin levels is very difficult, requiring several steps that introduce error, including destruction of platelets, release of serotonin from platelets, or size-dependent and shape-dependent yield of platelets. Review of platelet serotonin studies suggests less consistency in findings when platelets are directly studied by centrifugal separation. Given the technical limitations and the demonstration that virtually all the serotonin in blood

is stored in platelets [4,5] whole blood serotonin and platelet serotonin are essentially interchangeable, and the most accurate method for measureing platelet serotonin levels is to measure it in whole blood. For this review, the two will be considered as synonymous.

Familiality of platelet serotonin

Several studies have shown a positive correlation of platelet serotonin levels between probands with autism and their parents and siblings [6–9]. McBride *et al.* (Paper presented at 34th Annual Meeting of the American Academy of Neuropyschopharmacology, San Juan, 1995) recently presented results from a study that confirmed the importance of controlling for race and ethnicity in studies of platelet serotonin [10]. Black and Hispanic subjects had higher levels of platelet serotonin than did white subjects. There is no evidence for a phsyiologic consequence of this difference, but the finding does prompt the reassessment of many previous studies. For example, it was reported in one study that there was a correlation between whole blood serotonin levels of mothers and fathers of probands with autism [8]. In a subsequent study, this finding was not confirmed [9]. The correlation in the earlier study was accounted for by two "outlier" couples who where black. In contrast, the findings of positive correlations of serotonin between probands and their mothers were present when white subjects were considered separately.

Interestingly, subjects with autism who had a sibling with autism had higher platelet serotonin levels than subjects who did not have a sibling with autism [11]. Dominant or recessive inheritance, rather than oligogenic inheritance, is more likely in families with two autistic children than in families with only one such child. Thus, this finding suggests that hyperserotonemia may be an indicator of autism with a higher risk of sibling recurrence. It would be useful to study families without a proband with autism or attention-deficit hyperactivity disorder to see whether there is general heritability of platelet serotonin levels, independent of diagnosis. Platelet serotonin levels have been demonstrated to be stable after the age of 9 years [12], supporting the hypothesis that platelet serotonin levels are under genetic regulation. Because platelets are fragments of multinucleated megakaryocytes, genetic regulation may occur at the level of the megakaryocyte. Studies of megakaryocytes are obviously not practical. However, it is possible to study genes for regulation of the serotonin transporter and 5-HT_{2A} receptors, because both are expressed in platelets and on pre- and postsynaptic neurons at serotonergic synapses [13,14].

Studies of the mechanism of increased platelet serotonin

A few studies have examined platelet serotonin function in subjects with elevated platelet serotonin levels. Platelet ^{3}H-lysergic acid diethylamide (LSD)-labeled 5-HT_2 receptor binding was decreased in adults with autistic disorder when compared with binding in normal adult control subjects [15]. There was a relatively strong but insignificant (n = 9) negative correlation between 5-HT_2 receptor binding sites and platelet serotonin levels. In another study, children with autistic disorder did not differ from their parents or siblings when ^{125}I-spiroperidol-labeled platelet 5-HT_2 receptor binding was studied [16]. There was no difference between subjects with normal and high platelet serotonin levels (Cook, Unpublished data). A more recent study found that ^{3}H-LSD-labeled platelet 5-HT_2 receptor binding was lower in hyperserotonemic first-degree relatives of autistic children when compared with normoserotonemic relatives [17]. The difference in outcome between these two studies may be related to several factors, including a putative point mutation in the 5-HT_{2A} receptor altering affinity for agonists but not antagonists [18]. The two studies were performed during different seasons (February vs July–September). LSD has been shown to bind with high affinity to transfected 5-HT_{5A} [19], 5-HT_6 [20], and 5-HT_7 [21] receptors. It is also possible that there was binding to platelet serotonin receptors other than the 5-HT_{2A} receptor by LSD, but not spiroperidol, under the conditions used (Table 1).

A correlation was also found between platelet serotonin levels and rate of platelet serotonin transport [17]. The increase in transport is interesting in light of the therapeutic effect of potent serotonin transporter inhibitors on ritualistic behaviour and irritability in autism (*see* "Pharmacology").

Relationship of elevated platelet serotonin levels to clinical subgroups

No consistent pattern between symptoms of autism and elevated whole blood serotonin levels has emerged [22], but these studies have not used reliable instruments to assess autistic symptoms, such as the Autism Diagnostic Interview [23] or the Autism Diagnostic Observation Schedule [24]. In particular, future studies of serotonin levels in autism should focus on symptoms pertaining to restricted and repetitive interests or aggression, because these symptoms respond partially to treatment with potent serotonin transporter inhibitors. (*See* "Pharmacology").

Pharmacology

The most compelling evidence for the relationship between serotonin levels and autism is the efficacy of antidepressant medications that potently inhibit serotonin transport. Potent serotonin transporter inhibitors include the tricyclic antidepressant clomipramine and the selective serotonin reuptake inhibitors fluoxetine, sertraline, fluvoxamine, and paroxetine. Potent serotonin transporter inhibitors have been shown to reduce rituals

Table 1

Serotonin receptors

Receptor	Signal transduction	Notes	Amino acid variants	Chromosome	Studies
5-HT_{1A}	↓ AC	8-OH-DPAT; autoreceptor; stimulates neurite branching	128V, G22S	5	Sikich et al. [44], Fargin *et al.* [49], Goldman *et al.* [50], Nakhai *et al.* [51], Erdmann *et al.* [52]
5-HT_{1B}	↓ AC	= 5-$HT_{1D\beta}$ aggression in knockout mice; autoreceptor	F124C	6q13	Ramboz *et al.* [53], Hartig *et al.* [54], Lappalainen *et al.* [55], Saudou *et al.* [56], Nöthen *et al.* [57]
5-HT_{1D}	↓ AC	= 5-$HT_{1D\alpha}$ trigeminal ganglia, autoreceptor homologous pseudogene		1p35	Hartig *et al.* [54], Ozaki *et al.* [58]
5-HT_{1E}	↓ AC	= S31		6q14-15	Goldman *et al.* [50], McAllister *et al.* [59], Levy *et al.* [60]
5-HT_{1F}	↓ AC	= 5-$HT_{1E\beta}$ = MR77		3p	Lovenberg *et al.* [61], Cook (Unpublished data)
5-HT_{2A}	↑ PLC	formerly 5-HT_2; sequence identical brain and platelet [14]; LSD	T25N, A439V, H444Y	13q14.1 13q14.2	Cook *et al.* [14], Julius *et al.* [62], Ozaki *et al.* [63], Erdmann *et al.* [64]
5-HT_{2B}	↑ PLC	= 5-HT_{2F}		2q36.3-2q37.1	Schmuck *et al.* [65], Le Coniat *et al.* [66]
5-HT_{2C}	↑ GC	= 5-HT_{1C}; sudden death in knockout mice; choroid plexus; alternative splice variant	C23S	Xq21	Julius *et al.* [67], Lappalainen *et al.* [68], Tecott *et al.* [69], Kaufman *et al.* [70]
5-HT_3	Ion channel	Ondansetron; splice variants		11q23.1-q23.2	Uetz *et al.* [71], Miyake *et al.* [72], Weiss *et al.* [73]
5-HT_4	↑ AC	Zacopride; two splice variants–5-HT_{4L} and 5-HT_{4S}; human 5-HT_{4L} 16 amino acids shorter than rat, autoreceptor		5q	Adham *et al.* [74], Gerald *et al.* [75], Cook (Unpublished data)
5-HT_{5A}	↓ AC	= REC17; colocalized with mouse reeler mutant and human and human holoprosencephaly 3, LSD		7q36.1	Matthes *et al.* [19], Schanen *et al.* [76]
5-HT_{5B}		= MR22; pseudogene in humans		2q11-q13	Matthes *et al.* [19], Grailhe *et al.* [77]
5-HT_6	↑ AC	= St-B17; clozapine, clomipramine, LSD		1p35-36	Kohen *et al.* [78]
5-HT_7	↑ AC	Antagonists: *eg,* clozapine, risperidone; agonists: LSD, 8-OH-DPAT; also 80%–90% homologous pseudogene	P279L	10q	Bard *et al.* [21], Gelernter *et al.* [79], Goldman *et al.* [80]
5-HTT	Transporter	= SLC 6A4		17q11.1-q12	Gelernter and Freimer [81]

AC–adenylyl cyclase; GC–guanylyl cyclase; LSD–lysergic acid diethylamide; 8-OH-DPAT–8-hydroxdipropylaminotetralin; PLC–phospholipase C.

associated with anxiety and to reduce aggression in more than 50% of children and adults with autism in open [25–29] and double-blind trials [30,31••]. These drugs have also been used successfully to treat self-injurious behavior and stereotypic movements in patients with mental retardation without autism [29,32]. The acute effect of administration of these drugs in healthy adults is a reduction in basolateral limbic system (amygdala and hippocampus) metabolism [33]. Chronic administration in rodents leads to an increase in serotonergic neurotransmission through downregulation of presynaptic terminal autoreceptors [34].

Preliminary evidence of the efficacy of risperidone has been reported in autism [35,36]. The effects appear to be similar to those of potent serotonin transporter inhibitors but are more rapid in onset (Cook *et al.*, Unpublished observation). Risperidone is an antagonist at dopamine D_2 and D_4 receptors [37]. It is also an antagonist at 5-HT_{2A} and 5-HT_7 receptors [38]. Development of more specific antagonists may allow study of the possibility that the rapid action of risperidone in reducing rituals in autism may be due to antagonism of presynaptic 5-HT_7 autoreceptors, which would lead to a more rapid onset of increased serotonin neurotransmission than potent inhibition of the presynaptic serotonin transporter. In addition, absence of D_2 receptor antagonism is likely to reduce or eliminate the risk of tardive dyskinesia.

Further evidence of serotonin involvement in autism comes from a pharmacologic study using tryptophan depletion. Tryptophan depletion leads to reduced serotonin synthesis, release, and neurotransmission. McDougle *et al.* [39••] found exacerbation of behaviors such as whirling, flapping, pacing, banging and hitting self, rocking, toe walking, and anxiety in more than 50% of adults with autism after tryptophan depletion. This finding is consistent with the finding of a decreased ratio of the level of serum tryptophan to the level of large neutral amino acids in idiopathic infantile autism relative to control subjects [40•]. This decreased ratio would lead to a lower basal level of serotonin synthesis, vulnerability to tryptophan depletion, and response to pharmacologic manipulations that increase serotonin neurotransmission (Fig. 1).

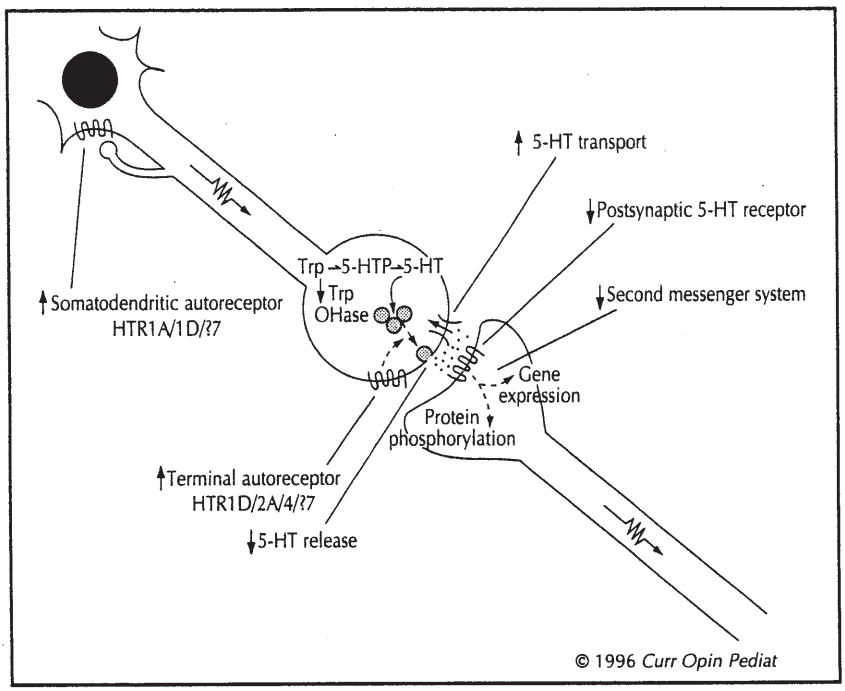

Fig. 1. Several possible ways that functional activity of a serotonin (5-HT) synapse may be reduced in autism, leading to behavioral problems such as the performance of rituals, anxiety and aggresssion in childhood and adolescence, and abnormal central nervous system development. Decreased 5-HT signaling may occur through decreased 5-HT synthesis, increased somatodentric 5-HT autoreceptor sensitivity (HTR1A, HTR1D, or possibly HTR7) or terminals (HTR1D, HTR2A, HTR4, or possibly HTR7), decreased 5-HT release, increased 5-HT transport (reuptake) into the presynaptic terminals, decreased postsynaptic 5-HT receptor sensitivity, or decreased signal transduction in the postsynaptic neuron. In each case of an *arrow* demonstrating the possible mechanism of reduced 5-HT function in autism, pharmacologic strategies would be designed in the opposite direction. For example, potent 5-HT transporter inhibitors such as clomipramine and fluvoxamine decrease 5-HT transport (reuptake). This model also demonstrates the advantage of development of autoreceptor antagonists, because the regulation of serotonergic neurons leads to net decreased 5-HT function if synaptic 5-HT exceeds a level sufficient to trigger autoreceptors, leading to a narrow therapeutic window for potent 5-HT transporter inhibitors. 5-HTP–5-hydroxytryptophan; OHase–hydroxylase; Trp–tryptophan.

Molecular biology of serotonin receptors and the serotonin transporter

Several recent developments in the molecular biology of serotonin receptors are relevant in the study of autism. Past studies of 5-HT_2 receptors must be reconsidered, because the 5-HT_{2B}, 5-HT_{5A}, 5-HT_6, 5-HT_7 receptors have been cloned. Table 1 lists much of the information about serotonin receptors and the serotonin transporter that may be of relevance in developing a better understanding of the role of the serotonin system in autism. The expression of the 5-HT_{2A} receptor and the serotonin transporter in autism provides the most logical connection between platelet findings and the relevant CNS mechanism. Because pharmacologic evidence suggests that more than 50% of patients with autism have an abnormality in serotonergic neurotransmission, it is possible that one or more of these mechanisms (decreased postsynaptic 5-HT_2 receptor binding or increased serotonin transporter function) may lead to platelet findings, but other possible specific mechanisms may not be expressed in platelet changes (*eg*, increased 5-HT_{1A} autoreceptor sensitivity).

Role of serotonin in central nervous system development

Serotonin may have a role in the developmental neuropathologic abnormalities found in the hippocampus, amygdala, and cerebellum in autistic disorder [41]. As a specific example, decreased neurite branching has been observed in the hippocampus in autism [42]. Stimulation of the 5-HT_{1A} receptor by 8-hydroxydipropylaminotetralin (and possibly 5-HT_7 receptor [43]) decreases neurite branching during development of the nervous system [44]. In addition, the 5-HT_{1A} receptor has neurotrophic effects in the hippocampus [45]. The 5-HT_{1A} receptor is also expressed in the fetal and neonatal but not the adult human cerebellum [46]. The 5-HT_{2A} receptor is expressed in the hippocampus and cerebellum [47]. The 5-HT_6 receptor is expressed in the amygdala, hippocampus, and cerebellum [48].

Our laboratory has recently studied expression of serotonin receptor in an amygdala cDNA library and found a pattern of serotonin receptor mRNA of the order 5-HT_{1A} > 5-HT_4 = 5-HT_{5A} = 5-HT_7 > 5-HT_{2C} > 5-HT_3 > 5-HT_{1D} > 5-HT_{1E} > 5-HT_{1B} = 5-HT_{1F} = 5-HT_{2A} = 5-HT_{2B} = 5-HT_6 (Cook *et al.*, Unpublished observation). The serotonin transporter is expressed at all presynaptic serotonin terminals.

Conclusions

Convergent findings from behavioral neuroscience, platelet, pharmacologic, and genetic studies indicate the involvement of serotonin in many of the symptoms of autistic disorder. Over the next decade, as the molecular biology of serotonin-related proteins is elucidated and medications are developed using this knowledge, it will be possible to assist people with autism to more effectively control the disabling symptoms of aggression, anxiety, and inflexible rituals and routines. As the molecular biology of the role of serotonin in CNS development is elucidated, it may become possible to treat or prevent the development of the social and cognitive dysfunction associated with autism through rational pharmacologic or dietary intervention.

Acknowledgment

The authors would like to acknowledge the inspiration, encouragement, and constant push to understand the mechanism of hyperserotonemia provided by Daniel X. Freedman.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- • Of special interest
- •• Of outstanding interest

1. Schain RJ, Freedman DX: **Studies on 5-hydroxyindole metabolism in autistic and other mentally retarded children.** *J Pediatr* 1961, **58**:315–320.
2. Anderson GM, Freedman DX, Cohen DJ, Volkmar FR, Hoder EL, McPhedran P, Minderaa RB, Hansen CR, Young JG: **Whole blood serotonin in autistic and normal subjects.** *J Child Psychol Psychiatry* 1987, **28**:885–900.
3. Cook EH: **Autism: review of neurochemical investigation.** *Synapse* 1990, **6**:292–308.
4. Anderson GM, Feibel FC, Cohen DJ: **Determination of serotonin in whole blood, platelet-rich plasma, platelet-poor plasma and plasma ultrafiltrate.** *Life Sci* 1987, **40**:1063–1070.
5. Cook EH, Leventhal BL, Freedman DX: **Free serotonin in plasma: autistic children and their first-degree relatives.** *Biol Psychiatry* 1988, **24**:488–491.
6. Kuperman S, Beeghly JH, Burns TL, Tsai LY: **Serotonin relationships of autistic probands and their first-degree relatives.** *J Am Acad Child Psychiatry* 1985, **24**:186–190.
7. Abramson RK, Wright HH, Carpenter R, Brennan W, Lumpuy O, Cole E, Young SR: **Elevated blood serotonin in autistic probands and their first-degree relatives.** *J Autism Dev Disord* 1989, **19**:397–407.
8. Leventhal BL, Cook EH, Morford M, Ravitz A, Freedman DX: **Relationships of whole blood serotonin and plasma norepinephrine within families of autistic children.** *J Autism Dev Disorder* 1990, **20**:499–511.
9. Cook EH, Leventhal BL, Heller W, Metz J, Wainwright M, Freedman DX: **Autistic children and their first-degree relatives: relationships between serotonin and norepinephrine levels and intelligence.** *J Neuropsychiatry Clin Neurosci* 1990, **2**:268–274.
10. Cuccaro ML, Wright HH, Abramson RK, Marsteller FA, Valentine J: **Whole-blood serotonin and cognitive functioning in autistic individuals and their first-degree relatives.** *J Neuropsychiatry Clin Neurosci* 1993, **5**:94–101.
11. Piven J, Tsai G, Nehme E, Coyle JT, Folstein SE: **Platelet serotonin, a possible marker for familial autism.** *J Autism Dev Disord* 1991, **21**:51–59.
12. Ritvo E, Yuwiler A, Geller E, Plotkin S, Mason A, Saeger K: **Maturational changes in blood serotonin levels and platelet counts.** *Biochem Med* 1971, **5**:90–96.
13. Lesch KP, Wolozin BL, Murphy DL, Reiderer P: **Primary structure of the human platelet serotonin uptake site: identity with the brain serotonin transporter.** *J Neurochem* 1993, **60**:2319–2322.
14. Cook EH, Fletcher KE, Wainwright M, Marks N, Yan S-Y, Leventhal BL: **Primary structure of the human platelet serotonin 5-HT_{2A} receptor: identity with frontal cortex serotonin 5-HT_{2A} receptor.** *J Neurochem* 1994, **63**:465–469.
15. McBride PA, Anderson GM, Hertzig ME, Sweeney JA, Kream J, Cohen DJ, Mann JJ: **Serotonergic responsivity in male young adults with autistic disorder.** *Arch Gen Psychiatry* 1989, **46**:205–212.
16. Perry BD, Cook EH, Leventhal BL, Wainwright MS, Freedman DX: **Platelet 5-HT_2-serotonin receptor binding sites in autistic children and their first-degree relatives.** *Biol Psychiatry* 1991, **30**:1–10.
17. Cook E, Arora R, Anderson G, Berry-Kravis E, Yan S-Y, Yeoh H, Sklena P, Charak D, Leventhal B: **Platelet serotonin studies in hyperserotonemic relatives of children with autistic disorder.** *Life Sci* 1993, **52**:2005–2015.
18. Surprenant A, Horstman D, Akbarali H, Limbird L: **A point mutation of the α_2-adrenoceptor that blocks coupling to potassium but not calcium currents.** *Science* 1992, **257**:977–980.
19. Matthes H, Boschert U, Amlaiky N, Grailhe R, Plassat J, Muscatelli F, Mattei M, Hen R: **Mouse 5-hydroxytryptamise$_{5A}$ and 5-hydroytryptamise$_{5B}$ receptors define a new family of serotonin receptors: cloning, functional expression, and chromosomal localization.** *Mol Pharmacol* 1993, **43**:313–319.
20. Monsma F JR, Shen Y, Ward R, Hamblin M, Sibley D: **Cloning and expression of a novel serotonin receptor with high affinity for tricyclic psychotropic drugs.** *Mol Pharmacol* 1993, **43**:320–327.
21. Bard J, Zgombick J, Adham N, Vaysse P, Branchek T, Weinshank R: **Cloning of a novel human serotonin receptor (5-HT_7) positively coupled to adenylate cyclase.** *J Biol Chem* 1993, **268**:23422–23426.
22. Kuperman S, Beeghly JHL, Burns TL, Tsai LY: **Association of serotonin concentration to behavior and IQ in autistic children.** *J Aut Dev Disord* 1987, **17**:133–140.
23. Lord C, Rutter M, Le Couteur A: **Autism Diagnostic Interview-Revised: a revised version of a diagnostic interview for caregivers of individuals with possible pervasive developmental disorders.** *J Autism Dev Disord* 1994, **24**:659–685.
24. Lord C, Rutter M, Goode S, Heemsbergen J, Jordan H, Mawhood L, Schopler E: **Autism Diagnostic Observation Schedule: a standardized observation of communicative and social behavior.** *J Autism Dev Disord* 1989, **19**:185–212.
25. Cook EH, Terry EJ, Heller W, Leventhal BL: **Fluoxetine treatment of borderline mentally retarded adults with obsessive-compulsive disorder.** *J Clin Psychopharmacol* 1990, **10**:228–229.
26. Mehlinger R, Scheftner WA, Poznanski E: **Fluoxetine and autism.** *J Am Acad Child Adolesc Psychiatry* 1990, **29**:985.
27. McDougle CJ, Price LH, Goodman WK: **Fluvoxamine treatment of coincident autistic disorder and obsessive-compulsive disorder: a case report.** *J Autism Dev Disord* 1990, **20**:537–543.
28. Todd RD: **Fluoxetine in autism.** *Am J Psychiatry* 1991, **148**:1089.
29. Cook E, Rowlett R, Jaselskis C, Leventhal B: **Fluoxetine treatment of patients with autism and mental retardation.** *J Am Acad Child Adolesc Psychiatry* 1992, **31**:739–745.
30. Gordon C, State R, Nelson J, Hamburger S, Rapoport J: **A double-blind comparison of clomipramine, desipramine, and placebo in the treatment of autistic disorder.** *Arch Gen Psychiatry* 1993, **50**:441–447.
31. •• McDougle C, Naylor S, Cohen D, Volkmar F, Heninger G, Price L: **A double-blind, placebo-controlled study of fluvoxamine in adults with autistic disorder.** *Arch Gen Psychiatry* 1996, **53**:in press.

This paper represents the first double-blind study in which both efficacy and the absence of long-term irreversible side effects (*ie*, neuroleptics) have been replicated for a class of medications–potent serotonin transporter inhibitors–in the treatment of core symptoms of autistic disorder. This paper is also significant because it is the first double-blind study showing efficacy for a selective serotonin reuptake inhibitor in autistic disorder, confirming an earlier systematic open trial with fluoxetine. Although the core social and communicative impairments are not addressed by this class of medications, symptoms such as the performance of rituals, preoccupations, and routines associated with anxiety and aggression often respond.

32. Lewis M, Bodfish J, Powell S, Golden R: **Clomipramine treatment for stereotypy and related repetitive movement disorders associated with mental retardation.** *Am J Ment Retard* 1995, **100**:299–312.
32. Cook E, Metz J, Leventhal B, Lebovitz M, Nathan M, Semerdjian S, Brown T, Cooper M: **Fluoxetine effects on cerebral glucose metabolism.** *Neuroreport* 1994, **5**:1745–1748.
32. Chaput Y, de Montigny C, Blier P: **Presynaptic and postsynaptic modifications of the serotonin system by long-term administration of antidepressant treatments: an in vivo electrophysiologic study in the rat.** *Neuropsychopharmacology* 1991, **5**:219–229.
35. Purdon SE, Lit W, Labelle A, Jones BD: **Risperidone in the treatment of pervasive developmental disorder.** *Can J Psychiatry* 1994, **39**:400–405.
36. Casaer P, Walleghem D, Vandenbussche I, Huang M-L, De Smedt G: **Pharmacokinetics and safety of risperidone in autistic children.** *Pediatr Neurol* 1994, **11**:89.
37. Seeman P, Van Tol H: **Dopamine receptor pharmacology.** *Trends Pharmacol Sci* 1994, **15**:264–270.
38. Roth B, Craigo S, Choudhary S, Uluer A, Monsma Jr F, Hen Y, Meltzer H, Sibley D: **Binding of typical and atypical antipsychotic drugs to 5-**

hydroxytryptamine-6 and 5-hydroxytryptamine-7 receptors. *J Pharmacol Exp Ther* 1994, **268**:1403–1410.

39. • McDougle C, Naylor S, Cohen D, Aghajanian G, Heninger G, Price L: **Effects of tryptophan depletion in drug-free adults with autism.** *Arch Gen Psychiatry* 1996, **53**: in press.

This study shows worsened stereotypic behavior in subjects with autism who had undergone acute dietary depletion of tryptophan (which acutely reduces serotonin synthesis).

40. • D' Eufemia P, Finocchiaro R, Celli M, Viozzi L, Monteleone D, Giardini O: **Low serum tryptophan to large neutral amino acids ratio in idiopathic infantile autism.** *Biomed Pharmacother* 1995, **49**:288–292.

Reduction in the ratio of tryptophan levels to large neutral amino acid levels has been shown in other studies to lead to decreased tryptophan transport across the blood-brain barrier and subsequently to decreased serotonin synthesis. This is an interesting study in relationship to the finding of worsening of stereotypic symptoms after tryptophan depletion.

41. Bauman M, Kemper T: **Neuroanatomic observations of the brain in autism.** In *The Neurobiology of Autism.* Edited by Bauman M, Kemper T. Baltimore: Johns Hopkins University Press; 1994:119–145.

42. Raymond GV, Bauman ML, Kemper TL: **Hippocampus in autism: a Golgi analysis.** *Acta Neuropathol* 1996, **91**:117–119.

43. Lovenberg T, Baron B, de Lecea L, Miller J, Prosser R, Rea M, Foye P, Racke M, Slone A, Siegel B, Danielson P, Sutcliffe J, Erlander M: **A novel adenylyl cyclase-activating serotonin receptor ($5\text{-}HT_7$) implicated in the regulation of mammalian circadian rhythms.** *Neuron* 1993, 11:449–458.

44. Sikich L, Hickok JM, Todd RD: **5-HT1A receptors control neurite branching during development.** *Brain Res Dev Brain Res* 1990, **56**:269–274.

45. Whitaker-Azmitia PM, Murphy R, Azmitia EC: **Stimulation of astroglial 5-HT1A receptors releases the serotonergic growth factor, protein S-100, and alters astroglial morphology.** *Brain Res* 1990, **528**:155–158.

46. del Olmo E, Diaz A, Guirao-Pineyro M, del Arco C, Pascual J, Pazos A: **Transient localization of 5-HT1A receptors in human cerebellum during development.** *Neurosci Lett* 1994, **166**:149–152.

47. Morilak D, Somogyi P, Lujan-Miras R, Ciaranello R: **Neurons expressing $5\text{-}HT_2$ receptors in the bat brain: neurochemical identification of cell types by immunocytochemistry.** *Neuropsychopharmacology* 1994, 11:157–166.

48. Ward RP, Hamblin MW, Lachowicz JE, Hoffman BJ, Sibley DR, Dorsa DM: **Localization of serotonin subtype 6 receptor messenger RNA in the rat brain by in situ hybridization histochemistry.** *Neuroscience* 1995, **64**:1105–1111.

49. Fargin A, Raymond J, Lohse M, Kobilka B, Caron M, Lefkowitz R: **The genomic clone G-21 which resembles the β-adrenergic receptor sequence encodes the $5\text{-}HT_{1A}$ receptor.** *Nature* 1988, **335**:358–360.

50. Goldman D, Lappalainen J, Ozaki N, Nakhai B, Pesonen U, Koulu M, Giblin B, Nielsen D, Linnoila M, Dean M: **Polymorphism and genetic mapping of six human serotonin receptor genes.** *Soc Neurosci Abstr* 1994, **20**:1266.

51. Nakhai B, Nielsen D, Linnoila M, Goldman D: **Two naturally occurring amino acid substitutions in the human 5HT1A receptor: 5HT1A-22 Gly-Ser and 5HT1a-28 Ile-Val.** *Biochem Biophys Res Commun* 1995, **210**:530–536.

52. Erdmann J, Shimron-Abarbanell D, Cichon S, Albus M, Maier W, Lichtermann D, Minges J, Reuner U, Franzek E, Ertl MA, *et al.*: **Systematic screening for mutations in the promoter and the coding region of the 5-HT1A gene.** *Am J Med Genet* 1995, **60**:393–399.

53. Ramboz S, Saudou F, Amara DA, Belzung C, Segu L, Misslin R, Buhot MC, Hen R: **5-HT1B receptor knock out: behavioral consequences.** *Behav Brain Res* 1995, **73**:305–312.

54. Hartig PR, Branchek TA, Weinshank RL: **A subfamily of $5\text{-}HT_{1D}$ receptor genes.** *Trends Pharmacol Sci* 1992, **13**:152–159.

55. Lappalainen J, Dean M, Charbonneau L, Virkkunen M, Linnoila M, Goldman D: **Mapping of the serotonin 5-HT1DBeta autoreceptor gene on chromosome 6 and direct analysis for sequence variants.** *Am J Med Genet* 1995, **60**:157–161.

56. Saudou F, Amara D, Dierich A, LeMeur M, Ramboz S, Segu L, Buhot M-C, Hen R: **Enhanced aggressive behavior in mice lacking $5\text{-}HT_{1B}$ receptor.** *Science* 1994, **265**:1875–1878.

57. Nöthen M, Erdmann J, Shimron-Abarbanell D, Propping P: **Identification of genetic variation in the human serotonin 1Dβ receptor gene.** *Biochem Biophys Res Comm* 1994, **205**:1194–1200.

58. Ozaki N, Lappalainen J, Dean M, Virkkunen M, Linnoila M, Goldman D: **Mapping of the serotonin 5-HT1DAlpha autoreceptor gene (HTR1D) on chromosome 1 using a silent polymorphism in the coding region.** *Am J Med Genet* 1995, **60**:162–164.

59. McAllister G, Charlesworth A, Snodin C, Beer M, Noble A, Middlemiss D, Iversen L, Whiting P: **Molecular cloning of a serotonin receptor from human brain ($5\text{-}HT_{1E}$): a fifth $5\text{-}HT_1$-like subtype.** *Proc Natl Acad Sci U S A* 1992, **89**:5517–5521.

60. Levy F, Gudermann T, Birnbaumer M, Kaumann A, Birnbaumer L: **Assignment of the gene encoding the 5-HT1E serotonin receptor (S31) (locus HTR1E) to human chromosome 6q14-q15.** *Genomics* 1994, **22**:637–640.

61. Lovenberg T, Erlander M, Baron B, Racke M, Slone A, Siegel B, Craft C, Burns J, Danielson P, Sutcliffe J: **Molecular cloning and functional expression of rat and human $5\text{-}HT_{1E}$-like-5-hydroxytryptamine receptor genes.** *Proc Natl Acad Sci U S A* 1993, **90**:2184–2188.

62. Julius D, Huang KN, Livelli TJ, Axel R, Jessell TM: **The 5HT2 receptor defines a family of structurally distinct but functionally conserved serotonin receptors.** *Proc Natl Acad Sci U S A* 1990, **87**:928–932.

63. Ozaki N, Rosenthal N, Pesonen U, Lappalainen J, Felman-Naim S, Schwartz P, Turner E, Goldman D: **Identification of two naturally occurring amino acid substitutions of the $5\text{-}HT_{2A}$ receptor in patients with seasonal affective disorder and in contols.** *Biol Psychiatry* 1996, in press.

64. Erdmann J, Shimron-Abarbanell D, Rietschel M, Albus M, Maier W, Koerner J, Bondy B, Chen K, Shih JC, Knapp M, Propping P, Noethen MM: **Systematic screening for mutations in the human serotonin-2A (5-HT2A) receptor gene: identification of two naturally occurring receptor variants and association anaylysis in schizophrenia.** *Hum Genet* 1996, **97**:614–619.

65. Schmuck K, Ullmer C, Engels P, Lübbert H: **Cloning and functional characterization of the human $5\text{-}HT_{2B}$ serotonin receptor.** *FEBS Lett* 1994, **342**:85–90.

66. Le Coniat M, Choi DS, Maroteaux L, Launay JM, Berger R: **The 5-HT2B receptor gene maps to 2q36.3-2q37.1.** *Genomics* 1996, **32**:172–173.

67. Julius D, MacDermott A, Axel R, Jessel T: **Molecular characterization of a functional cDNA encoding the serotonin 1C receptor.** *Science* 1988, **241**:558–564.

68. Lappalainen J, Zhang L, Dean M, Oz M, Ozaki N, Yu DH, Virkkunen W, Weight F, Linnoila M, Goldman D: **Identification, expression, and pharmacology of a $Cys_{23}\text{-}Ser_{23}$ substitution in the human $5\text{-}HT_{2C}$ receptor gene (HTR2C).** *Genomics* 1995, **27**:274–279.

69. Tecott L, Sun L, Akana S, Strack A, Lowenstein D, Dallman M, Julius D: **Eating disorder and epilepsy in mice lacking $5\text{-}HT_{2C}$ serotonin receptors.** *Nature* 1995, **374**:542–546.

70. Kaufman MJ, Hartig PR, Hoffman BJ: **Serotonin 5-HT2C receptor stimulates cyclic GMP formation in choroid plexus.** *J Neurochem* 1995, **64**:199–205.

71. Uetz P, Adelatty F, Villaroel A, Rappold G, Weiss B, Koenen M: **Organization of the murine 5-HT3 receptor gene and assignment to human chromosome 11.** *FEBS Lett* 1994, **339**:302–306.

72. Miyake A, Mochizuki S, Takemoto Y, Akuzawa S: **Molecular cloning of human 5-hydroxytryptamine3 receptor: heterogeneity in distribution and function among species.** *Mol Pharmacol* 1995, **48**:407–416.

73. Weiss B, Mertz A, Schroeck E, Koenen M, Rappold G: **Assignment of a human homolog of the mouse HTr3 receptor gene to chromosome 11q23.1-q23.2.** *Genomics* 1995, **29**:304–305.

74. Adham N, Gerald C, Vaysse PJ-J, Weinshank R, Branchek T: **Pharmacological characterization of two splice variants of the cloned**

rat 5-HT$_4$ receptor coupled to stimulation of adenylate cyclase. *Soc Neurosci Abstr* 1994, **20**:1266.

75. Gerald C, Adham N, Kao HT, Olsen MA, Laz TM, Schechter LE, Bard JA, Vaysse PJJ, Hartig PR, Branchek TA, Weinshank RL: **The 5-HT4 receptor: molecular cloning and pharmacological characterization of two splice variants.** *EMBO J* 1995, **14**:2806–2815.

76. Schanen NC, Scherer SW, Tsui LC, Francke U: **Assignment of the 5-hydroxytryptamine (serotonin) receptor 5A gene (HTR5A) to human chromosome band 7q36.1.** *Cytogenet Cell Genet* 1996, **72**:187–188.

77. Grailhe R, Amlaiky N, Ghavami A, Ramboz S, Yocca F, Mahle C, Margouris C, Perrot F, Hen R: **Human and mouse 5-HT$_{5A}$ and 5-HT$_{5B}$ receptors: cloning and functional expression.** *Soc Neurosci Abstr* 1994, **20**:1160.

78. Kohen R, Metcalf MA, Khan N, Druck T, Huebner K, Lachowicz JE, Meltzer HY, Sibley DR, Roth BL, Hamblin MW: **Cloning, characterization, and chromosomal localization of a human 5-HT6 serotonin receptor.** *J Neurochem* 1996, **66**:47–56.

79. Gelernter J, Rao P, Pauls D, Hamblin M, Sibley D, Kidd K: **Assignment of the 5HT7 receptor gene (HTR7) to chromosome 10q and exclusion of linkage with Tourette syndrome.** *Genomics* 1995, **26**:207–209.

80. Goldman D, Nielsen D, Okada M, Adamson M, Lappalainen J, Nalhotra N, Pesonen U, Koulu M, Eggert M, Virkkunen M, Ozaki N, Linnoila M: **Molecular genetics of impaired impulse control and early onset alcoholism [abstract].** *Biol Psychiatry* 1996, **39**:212.

81. Gelernter J, Freimer M: **PstI RFLP at the SERT locus.** *Hum Mol Genet* 1994, **3**:383.