Eur Arch Psychiatry Clin Neurosci (2011) 261:567–573
DOI 10.1007/s00406-011-0201-y

ORIGINAL PAPER

Clomipramine-induced serum prolactin as a marker for serotonin and dopamine turnover: results of an open label study

Joachim Cordes · Kai G. Kahl · Christian Werner · Uwe Henning · Gunnar Regenbrecht · Rolf Larisch · Christian Schmidt-Kraepelin · Johanna Thünker · Marcus W. Agelink · Stefan Löffler · Thomas Hohlfeld · Wolfgang Gaebel · Ansgar Klimke

Received: 30 January 2010 / Accepted: 21 February 2011 / Published online: 15 March 2011

Abstract Central nervous system (CNS) monoamine deficits have been linked to a number of pathological conditions such as major depressive disorder. Individual biological variations in 5-hydroxyindoleacetic acid (5-HIAA), homovanillic acid (HVA) and 3-methoxy-4-hydroxyphenylglycol (MHPG) might account for the variation in responses of neurotransmitter systems observed after the administration of clomipramine. The prolactin response to clomipramine has been widely used to assess CNS functioning. This open label study investigates the prolactin response induced by clomipramine in the plasma of healthy volunteers and whether it is related to changes in monoamine metabolites. The effects of clomipramine challenge on prolactin, 5-HIAA, HVA and MHPG were measured in 12 healthy volunteers. Samples were drawn directly before and 50 min after clomipramine infusion. A statistically significant increase in serum prolactin concentrations was measured in women 50 min after CMI infusion, but not in men. We found no significant increases in the serum monoamine metabolite concentrations 50 min after CMI infusion. Changes in HVA and 5-HIAA correlated statistically significantly and positively with the amount of prolactin release in the whole sample.

Patient consent: The study was approved by the ethics committee of the Universitiy of Duesseldorf, Moorenstr. 5, D-40225 Düsseldorf (Germany) and was carried out in accordance with The Code of the World Medical Association (Declaration of Helsinki).

J. Cordes (✉) · U. Henning · G. Regenbrecht · C. Schmidt-Kraepelin · J. Thünker · W. Gaebel
Department of Psychiatry and Psychotherapy, Medical Faculty, Heinrich-Heine-University, Duesseldorf, Germany
e-mail: joachim.cordes@lvr.de

U. Henning
e-mail: uwe.henning@lvr.de

G. Regenbrecht
e-mail: gunnar.regenbrecht@lvr.de

C. Schmidt-Kraepelin
e-mail: Christian.schmidt-kraepelin@lvr.de

J. Thünker
e-mail: Johanna.Thuenker@gmx.de

W. Gaebel
e-mail: wolfgang.gaebel@lvr.de

K. G. Kahl
Department of Psychiatry, Social Psychiatry and Psychotherapy, Hannover Medical School, Hannover, Germany
e-mail: kahl.kai@mh-hannover.de

C. Werner
Medical Department, Otsuka Pharma GmbH, Frankfurt, Germany
e-mail: chrwer@web.de

R. Larisch
Department of Nuclear Medicine, Heinrich-Heine-University, Düsseldorf, Germany
e-mail: rolf.larisch@klinikum-luedenscheid.de

M. W. Agelink
Department of Psychiatry, Psychotherapy and Psychosomatic, Ruhr-University Bochum, Herford, Germany
e-mail: agelink@klinikum-herford.de

S. Löffler · A. Klimke
Department of Psychiatry, Psychotherapy, Offenbach, Germany
e-mail: SLoeffler@klinikum-offenbach.de

A. Klimke
e-mail: ansgar.klimke@klinikum-offenbach.de

T. Hohlfeld
Institut für Pharmakologie und Klinische Pharmakologie, Heinrich-Heine-University, Düsseldorf, Germany
e-mail: hohlfeld@uni-duesseldorf.de

Furthermore, positive correlations were found between $\Delta_{50-0\ min}$ 5-HIAA and $\Delta_{50-0\ min}$ HVA, although we did not find a correlation between $\Delta_{50-0\ min}$ prolactin and $\Delta_{50-0\ min}$ MHPG after clomipramine challenge. The pronounced prolactin release in healthy adult women might indicate a higher physiological sensitivity. Correlations between intra-individual changes in HVA, 5-HIAA and serum prolactin might indicate a central nervous effect of clomipramine on monoamine turnover. We conclude that monoamine changes in relation to prolactin response after clomipramine challenge may be suitable for characterizing the relationship between central serotonergic and dopaminergic function.

Keywords Clomipramine challenge · Homovanillic acid · Prolactin · Monoamine metabolites · Plasma

Introduction

The identification of biological measures for early diagnosis, treatment response and outcome predictors is essential for improving clinical outcome in depression [2, 27, 35]. Central deficits in the monoamines serotonin (5-HT), dopamine (DA) and noradrenaline (NE) have been described in the pathogenesis of major depressive disorder [37], and several findings have suggested a predictive value of peripheral monoamine metabolites for assessing clinical outcome in depression.

3-methoxy-4-hydroxyphenylglycol (MHPG) reflects mainly peripheral noradrenergic functions [11]. Because of its correlation with CNS system turnover, serum 5-hydroxyindoleacetic acid (5-HIAA; the primary serotonin metabolite) has been used to measure CNS serotonin deficits [1, 3, 8, 14, 32, 43, 45]. Another method to assess central serotonergic function is the serum prolactin response to serotonin (5-HT)-releasing agents.

Clomipramine (CMI), a strong serotonin uptake inhibitor [15, 16], may have some advantages in challenge studies, namely a dose–response relationship has been described for the neuroendocrine response to CMI challenge [9, 16]; intravenous administration of clomipramine minimizes the problems seen with oral pharmacologic challenges regarding inter-individual differences in the rate and degree of absorption [18]; and parenteral administration of CMI at a low dose (12.5 mg) is relatively well tolerated and produces no cardiovascular effects [16, 38].

Clomipramine possesses the highest in vitro affinity for the serotonin transporter (Ki = 0.14 nM) of any of the tricyclic antidepressants, a moderate affinity to the norepinephrine transporter (Ki = 54 nM) and a low affinity for the dopamine transporter (Ki = 3.020 nM) [31]. It is therefore plausible that inhibition of the serotonin neurotransmitter transporter might lead to enhanced serotonin levels in the brain [36, 44]. Furthermore, there is evidence that clomipramine increases extracellular dopamine metabolites induced by increases in extracellular serotonin levels and receptor activation [36, 40].

The combined measurements of changes in serum monoamine metabolite levels in relation to prolactin response after clomipramine challenge may be useful for characterizing central monoaminergic function and for improving our understanding of the pathophysiology of affective disorders.

Consistently with this idea, parenteral clomipramine challenge has been found to be able to differentiate adults and adolescents with major depressive disorder from healthy controls [4, 9, 17, 18, 39]. A blunted prolactin response to clomipramine persists in depressed patients after recovery from acute illness and may reflect an underlying biological vulnerability [19, 28]. Clomipramine challenge combined with positron emission tomography (PET) should be useful for studying cerebral serotonergic mechanisms and predictors of outcomes [25, 42].

Several findings suggest a predictive value of peripheral monoamine metabolites for assessing clinical outcome. Clinical improvement of depressive inpatients was statistically significantly associated with changes in both plasma and CSF HVA after 4 weeks of pharmacological treatment [41]. Furthermore, changes in plasma HVA were found to be correlated with changes in CSF HVA [41]. In other studies, the HVA/5-HIAA ratio was found to be associated with symptom severity and treatment outcome in depressed patients and suicide attempters [22, 32]. A recent study demonstrated a positive correlation between the increase in plasma HVA levels and the rate of response to 8-week sertraline treatment in depressed patients [46].

As such, changes in serum prolactin and monoamine metabolites after clomipramine challenge may be useful state markers for depression which have the potential to identify remitted or healthy individuals at a high risk of developing depression.

The purpose of our study was to investigate whether intra-individual changes in 5-HIAA and HVA plasma concentrations reflect the acute effects of an intravenous CMI challenge. We hypothesized a correlation between prolactin changes as a central measurement for CMI effects and serum metabolite level changes if central nervous effects were the main target of CMI action, and no such correlation if peripheral effects are the major target of CMI action on catecholaminergic systems.

Materials and methods

Subjects

Seven healthy Caucasian men and five women with a mean age of 37.1 ± 11.2 years were assessed by a clinical psychiatric- and general medical interview and physical examination. Exclusion criteria were as follows: any acute or lifetime episode of DSM-IV defined axis-1 disorders and any pharmacological treatment in the period of two weeks before examination. Written informed consent was obtained from each subject. The study was approved by the Ethics Committee of the Heinrich-Heine University Duesseldorf (Germany) and was in accordance with the guidelines laid down in the Declaration of Helsinki.

Procedures

A history of physical health and psychiatric disorders was carefully assessed in all subjects. Psychological testing included the structured clinical interview for DSM-IV (SCID-1/2) [12]. After overnight fasting, CMI challenges and blood collection were performed between 9 and 10 a.m. utilizing an antecubital vein catheter. Baseline samples were taken directly before administration of CMI (12.5 mg in 100 ml NaCl 0.9% over a period of 15 min), and blood samples were collected 50 min after CMI infusion while the subjects remained seated.

Hormone and amine assays

Blood samples were centrifuged for 10 min at 2,500*g,* and separated serum samples were stored at −75°C. Serum prolactin concentrations were determined using a commercial antibody radioimmunoassay (PRL-Serozyme, supplied by Biochem Immunosystems, Freiburg/Germany, detection limit 0.2 ng/ml (4μIU/ml) serum; intra-assay and inter-assay coefficients of variation 4.5 and 6.1%, respectively). Serum HVA, 5-HIAA and MHPG concentrations were assayed by high-pressure liquid chromatography (HPLC). HVA and 5-HIAA were quantified using a modified procedure based on the method described by Gupta and Whelton [21]. Modifications were made as followed: (1) 4-hydroxy-3-methoxybenzyl-alcohol was used as internal standard; (2) Fast solid phase extraction (SPE) was done on Isolute ENV columns (Biotage, Sweden); (3) Samples were separated on an Inertsil-ODS analytical column (250 mm × 4.6 mm, 3 μm) purchased from MZ-Analysentechnik (Germany). The retrieval rate was 76.6% for HVA and 80% for 5-HIAA. The limit of detection was 1.25 ng/ml. Intra- and inter-assay coefficients of variation were HVA 7.1/19.8%, 5-HIAA 6.0/7.3% and MHPG 6.9/18.1%, respectively. For the quantification of MHPG, the method of Minegishi and Ishizaki was used [30] and modified: (1) SPE with Isolute ENV columns (Biotage, Sweden); (2) Analytical separation with a Hypersil-ODS column (250 mm × 4 6 mm, 3 μm) from MZ-Analysentechnik (Germany). The retrieval rate for MHPG was 74.8%, and the limit of detection was 1.0 ng/ml. The measurement of all metabolites was taken using the digital electrochemical amperometric detector (Decade) from ANTEC (Netherlands).

Statistical analysis

Firstly, an exploratory analysis of baseline HVA, 5-HIAA and MHPG measures (HVA 0 min, 5-HIAA 0 min, MPHG 0 min), post-infusion measurements (HVA 50 min, 5-HIAA 50 min, MPHG 50 min) and their differences ($\Delta_{50–0\ min}$ HVA, $\Delta_{50–0\ min}$ 5-HIAA, $\Delta_{50–0\ min}$ MHPG) was performed before prolactin measurements were analysed in the same way ($\Delta_{50–0\ min}$ prolactin, calculated as fifty minutes post-infusion change in serum prolactin concentration from the pre-infusion baseline value). The comparison of data at the two different measurement time points was made using the paired *t*-test for women and men separately. Pearson's correlation coefficients were determined for the differences of the four variables, where data from both genders were pooled. The level of significance was set at $P = 0.05$ for all tests.

Results

The incidence of gastrointestinal complaints rated on the 3-item scale was low (mean = 0.15, SD = 0.49).

Effects of gender on serum prolactin changes

A repeated measures ANOVA with the factors time and gender revealed an effect of gender (F = 5,283, $df = 1$, $P = 0.044$) and an interaction between time and gender (F = 6,454, $df = 10$, $P = 0.029$). Women showed both higher serum prolactin concentrations and higher increases in serum prolactin concentrations.

Effects of clomipramine challenge on serum prolactin release

A statistically significant increase in serum prolactin concentrations was measured in women 50 min after CMI infusion (T = −2.969, $df = 4$, $P = 0.041$), but not in men (T = −2.322, $df = 6$, $P = 0.059$, paired *t*-test, Fig. 1).

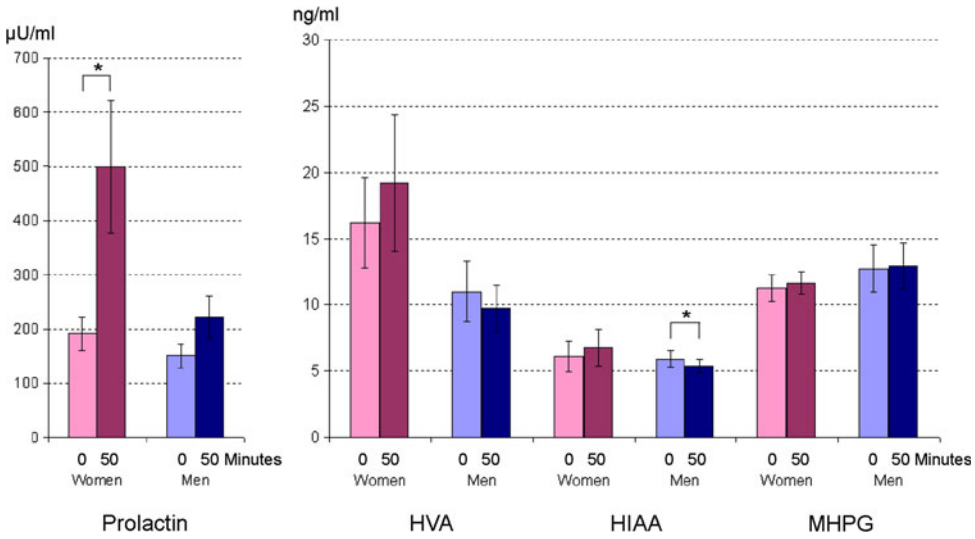

Fig. 1 Effects of clomipramine challenge on serum levels of prolactin and monoamine metabolites

Effects of clomipramine challenge on monoamine metabolites

We usually detected non-significant changes in the serum monoamine metabolite concentrations 50 min after CMI infusion (women: HVA: T = −1.297, *df* = 4, *P* = 0.264; 5-HIAA: T = −1.079, *df* = 4, *P* = 0.341; MHPG: T = −0.338, *df* = 4, *P* = 0.752; men: HVA: T = 0.865, *df* = 6, *P* = 0.420; 5-HIAA: T = 2.869, *df* = 6, *P* = 0.028; MHPG: T = −0.495, *df* = 6, *P* = 0.638; Fig. 1).

Correlations between changes in prolactin and monoamine metabolite plasma levels

When applying a Kolmogorov–Smirnov test, no deviation from a normal distribution could be found for $\Delta_{50-0\ min}$ prolactin (Z = 0.781, N = 12, *P* = 0.576), $\Delta_{50-0\ min}$ HVA (Z = 0.519, N = 12, *P* = 0.951), $\Delta_{50-0\ min}$ 5-HIAA (Z = 1.139, N = 12, *P* = 0.149) and $\Delta_{50-0\ min}$ MHPG (Z = 0.495, N = 12, *P* = 0.967).

The parametric correlation analysis (Pearson) revealed a positive correlation between $\Delta_{50-0\ min}$ prolactin and $\Delta_{50-0\ min}$ HVA after CMI challenge (R = 0.657, *P* = 0.020, Fig. 2a). Linear regression analysis allowed the following equation to be derived for describing the correlation: [$\Delta_{50-0\ min}$ HVA] = −2.187 + 0.016 * [$\Delta_{50-0\ min}$ Prolactin].

Furthermore, positive correlations were found between $\Delta_{50-0\ min}$ prolactin and $\Delta_{50-0\ min}$ 5-HIAA (R = 0.829, *P* = 0.001, Fig. 2b) and between $\Delta_{50-0\ min}$ 5-HIAA and $\Delta_{50-0\ min}$ HVA (R = 0.878, *P* < 0.001, Fig. 2c). Linear regression analysis allowed the following equations to be derived to describe the correlations: [$\Delta_{50-0\ min}$ 5-HIAA] = −0.842 + 0.005* [$\Delta_{50-0\ min}$ prolactin]; [$\Delta_{50-0\ min}$ HVA] = 0.677 + 3.745* [$\Delta_{50-0\ min}$ 5-HIAA]. We found no correlation between $\Delta_{50-0\ min}$ prolactin und $\Delta_{50-0\ min}$ MHPG after the clomipramine challenge (*R* = −0.298, *P* = 0.347).

Discussion

Consistently with previous findings, we found an increase in serum prolactin levels within 50 min of clomipramine infusion in women, indicating a central nervous effect of

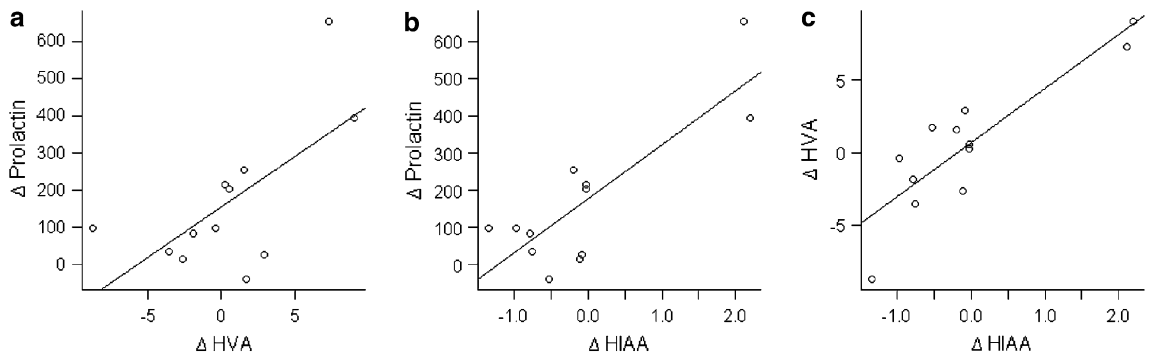

Fig. 2 **a** Correlation between $\Delta_{50-0\ min}$ prolactin and $\Delta_{50-0\ min}$ HVA plasma levels. **b** Correlation between $\Delta_{50-0\ min}$ prolactin and $\Delta_{50-0\ min}$ 5-HIAA plasma levels. **c** Correlation between $\Delta_{50-0\ min}$ 5-HIAA and $\Delta_{50-0\ min}$ HVA plasma levels

clomipramine [4, 17, 18]. A statistically significant increase in serum prolactin concentrations could not be measured in men. There are conflicting views on the role of gender in the neuroendocrine response to acute intravenous challenge with the serotonin reuptake inhibitor clomipramine (CMI) [20, 39]. Sallee et al. found a more increased prolactin release in adolescent healthy women compared to men after clomipramine challenge [39]. Golden et al., in a study involving thirty-seven healthy subjects, found that the maximum change in plasma prolactin concentrations from baseline was not significantly related to gender after controlling for age [20]. However, our result showing a pronounced prolactin release in adult healthy women may indicate a higher physiological sensitivity of women compared to men [29].

We found no significant increases in serum monoamine metabolite concentrations after CMI infusion within a period of 50 min. In order to investigate the central neurotransmitter systems mediating the clomipramine-induced hormonal responses indirectly, we examined the association between acute serum endocrine and amine responses to this agent. Since our aim was not to show differences between low and high responders, but rather to show a correlation between prolactin responses and intra-individual changes in plasma concentrations of 5-HIAA and HVA, we avoided any splitting of the sample for correlation analyses. We initially found positive correlations between 5-HIAA intra-individual alterations and prolactin release in response to CMI infusion. Clomipramine is a strong serotonin reuptake inhibitor. Our results underscore evidence for the serotonergic influence of prolactin release resulting from clomipramine administration.

The second main finding of our study was the significant positive correlation between intra-individual changes in serum HVA and serum prolactin. Clomipramine has been demonstrated to increase striatal levels of dopamine and HVA in control rats [34, 40]. Human studies on acute amine responses to clomipramine have not been carried out. However, the response after D,L-fenfluramine confirmed a significant correlation between acute increases in HVA and prolactin in healthy subjects [33]. Dopamine released by neuroendocrine dopaminergic neurones located in the arcuate and periventricular nuclei of the hypothalamus tonically inhibits prolactin secretion from the anterior gland [5, 6, 13]. As such, the positive correlation between intra-individual changes in serum HVA and serum prolactin is contra-intuitive. However, the secretory activity of the lactotrophs reflected a balance between local and distant releasing and inhibiting factors. The expected reciprocal relationship between dopamine and prolactin is often masked by the action of different factors that control prolactin release [10]. The tonic inhibition of prolactin secretion by dopamine is counteracted by the stimulatory actions of neuropeptides, steroids and growth factors. Prolactin regulates its own release by affecting the dopaminergic neurons via a short-loop rapid negative feedback. By altering the dopamine release, clomipramine may act indirectly via a short feedback mechanism. At very low concentrations, dopamine has been reported to stimulate, rather than inhibit, prolactin secretion [6]. The potential dissociation between central and pituitary dopamine receptor occupancy should be another determinant of the observed degree of prolactin and HVA elevation [7, 23]. However, our findings suggest the possibility that the changes in HVA levels may be secondary to dopamine receptor blockade, since a true functional increase in dopaminergic activity would lead to a reduction in prolactin levels.

Thirdly, intra-individual changes in the noradrenergic system regarding prolactin release, as measured by MHPG plasma level changes, were not observed in our study. Our results are consistent with others who found no effect of CMI and its metabolites on MHPG during clomipramine infusion [16]. However, it should be mentioned at this point that our study design minimized CMI effects on noradrenergic metabolites, since intravenously applied clomipramine avoids the 'first pass' effect of hepatic metabolism. As such, the formation of demethylated clomipramine—a potent noradrenaline uptake inhibitor—was markedly delayed and should not have influenced catecholamine metabolites during the course of our study.

Finally, a positive correlation was found between $\Delta_{50-0\ \mathrm{min}}$ 5-HIAA and $\Delta_{50-0\ \mathrm{min}}$ HVA. One may speculate as to whether this finding may have reflected an interaction between dopaminergic and serotonergic processes. The observed correlation may be interpreted as a net effect of different central interactions between serotonergic and dopaminergic effects in neurons of adenohypophysial tissue, the locus coeruleus, striatal dopaminergic neurons, the hypothalamic pituitary axis and serotonergic neurons in the raphe nuclei. Literature on interactions between dopaminergic and serotonergic systems is extensive. The histaminergic, catecholaminergic, glutaminergic, cholinergic and dopaminergic systems interact with the serotonergic system in their regulation of the release of hypothalamic and pituitary gland hormones, possibly via neuronal connections in the hypothalamic paraventricular nucleus [41]. Dopamine and serotonin systems are morphologically interconnected in the midbrain. Several studies have also demonstrated a functional relationship between these two monoamine systems [41]. The positive relationship between central dopaminergic and serotonergic activity that was demonstrated in our study may be secondary to an increase in serotonergic function. The observed individual changes in plasma concentrations of 5-HIAA and HVA in relation to prolactin might be an indicator for the acute

effects of intravenous clomipramine challenge on the central nervous system (CNS). These findings may be interpreted as a sign of the central nervous effects of CMI.

Amongst the methodological limitations include the study's open label design, which did not include a placebo treatment, and which may also have been underpowered due to the small size of the population sample. A comparison with non-medicated depressive patients should confirm the correlations observed. The accumulation of monoamines within the cerebrospinal fluid should exclude any peripheral influences on the monoamine metabolite turnover.

More empirical data are needed in order to improve our knowledge about the neurobiological effects of the challenges used in this study. This should improve our understanding of the pathophysiology of affective disorders, the ability to diagnose them in the early stages of the psychiatric disease and the ability to predict individual treatment responses and prognoses [24, 26].

Acknowledgments The authors thank Klaus Krieger, Ph.D. and Margit Geisler for performing the biochemical analyses and Raimund Schneider, Ph.D. and Prof. Erlo Lehmann, Ph.D. for their helpful suggestions. The authors further acknowledge the essential statistical support provided by Wolfgang Meyer, Ph.D. (Zentralinstitut für angewandte Mathematik, Research Center Jülich). The present work was supported by the *Pinguin Stiftung*. This funding source had no influence on the study design, the collection, analysis and interpretation of data, the writing of the report or on the decision to submit the paper for publication.

Conflict of interest Pharmaceutical companies were involved in supporting the speakers' honoraria, travel funds, advisory panel payments and research grants. There were, however, no competing interests related directly to the subject of this paper. All authors declare that they have no conflicts of interest.

References

1. Ågren H, Terenius L (1985) Hallucinations in patients with major depression: interaction between CSF monoaminergic and endorphinergic indices. J Affect Disord 9:25–34
2. Altamura AC, Dell'Osso B, Berlin HA, Buoli M, Bassetti R, Mundo E (2010) Duration of untreated illness and suicide in bipolar disorder: a naturalistic study. Eur Arch Psychiatry Clin Neurosci 260(5):385–391
3. Amin F, Davidson M, Davis KL (1986) Homovanillic acid measurement in clinical research: a review of methodology. Neuropsychobiology 16(2–3):85–87
4. Anderson IM, Ware J, Daroza Davis JM, Cowen PJ (1992) Decreased 5-HT-mediated prolactin release in major depression. Br J Psychiatry 160:372–378
5. Ben-Jonathan N, LaPensee CR, LaPensee EW (2008) What can we learn from rodents about prolactin in humans? Endocr Rev 29(1):1–41
6. Ben-Jonathan N, Olivaer C, Weiner HJ, Mical RS, Porter JC (1977) Dopamine in hypophysial portal plasma of the rat during the estrous cycle and throughout pregnancy. Endocrinology 100:452–458
7. Bressan RA, Erlandsson K, Spencer EP, Ell PJ, Pilowsky LS (2004) Prolactinemia is uncoupled from central D2/D3 dopamine receptor occupancy in amisulpride treated patients. Psychopharmacology 175(3):367–373
8. Carpenter LL, Anderson GM, Siniscalchi JM, Chappell PB, Price LH (2003) Acute changes in cerebrospinal fluid 5-HIAA following oral paroxetine challenge in healthy humans. Neuropsychopharmacology 28(2):339–347
9. Cordes J, Larisch R, Henning U, Thünker J, Werner C, Orozco G, Mayoral F, Rivas F, Auburger G, Tosch M, Rietschel M, Gaebel W, Müller H-W, Klimke A (2009) Abnormal neuroendocrine response to clomipramine in hereditary affective psychosis. Depression Anxiety 26(8):E111–E119
10. Demarest KT, Riegle GD, Moore KE (1984) Adenohypophysial dopamine content during physiological changes in prolactin secretion. Endocrinology 115(6):2091–2097
11. Elsworth JD, Redmond DE Jr, Roth RH (1982) Plasma and cerebrospinal fluid 3-methoxy-4-hydroxy-phenylethyleneglycol (MHPG) as indices of brain norepinephrine metabolism in primates. Brain Res 235:115–124
12. Endicott J, Spitzer RL (1978) A diagnostic interview: The schedule for affective disorders and schizophrenia. Arch Gen Psychiatry 35:837–844
13. Freeman ME, Kanyicska B, Lerant A, Nagy G (2000) Prolactin: structure, function, and regulation of secretion. Physiol Rev 80(4):1523–1631
14. Fujita K, Kobayashi A, Suzuki S, Nakazawa K (1991) Changes of serotonin and catecholamines are related to pharmacokinetic alterations of clomipramine in rat brain. Eur J Pharmacol 12; 204(3):227–233
15. Gillman PK (2007) Tricyclic antidepressant pharmacology and therapeutic drug interactions updated. Br J Pharmacol 151(6): 737–748
16. Golden RN, Hsiao J, Lane E, Hicks R, Rogers S, Potter WZ (1989) The effects of intravenous clomipramine on neurohormones in normal subjects. J Clin Endocrinol Metab 68:632
17. Golden RN, Hsiao J, Lane E, Ekstrom D, Rogers S, Hicks R, Potter WZ (1990) Abnormal neuroendocrine responsivity to acute intravenous clomipramine challenge in depressed patients. Psychiatry Res 31(1):39–47
18. Golden RN, Ekstrom D, Brown TM, Ruegg R, Evans DL, Haggerty JJ Jr, Garbutt JC, Pedersen CA, Mason GA, Browne J, Carson SW (1992) Neuroendocrine effect of intravenous clomipramine in depressed patients and healthy subjects. Am J Psychiatry 149(9):1168–1175
19. Golden RN, Heine AD, Ekstrom RD, Bebchuk JM, Leatherman ME, Garbutt JC (2002) A longitudinal study of serotonergic function in depression. Neuropsychopharmacology 26(5):653–659
20. Golden RN, Gilmore JH, Ekstrom RD, Knight B, Ruegg RG, Miller HL, Carson SW (1996) The effects of age, gender, and season on serotonergic function in healthy subjects. Prog Neuropsychopharmacol Biol Psychiatry 20(8):1315–1323
21. Gupta RN, Whelton C (1992) Determination of plasma homovanillic acid by liquid chromatography with electrochemical detection. J Chromatogr 582:236–241
22. Jokinen J, Nordström AL, Nordström P (2007) The relationship between CSF HVA/5-HIAA ratio and suicide intent in suicide attempters. Arch Suicide Res 11(2):187–192
23. Kapur S, Langlois X, Vinken P, Megens AA, De Coster R, Andrews JS (2002) The differential effects of atypical antipsychotics on prolactin elevation are explained by their differential blood-brain disposition: a pharmacological analysis in rats. J Pharmacol Exp Ther 302(3):1129–1134
24. Kemp AH, Gordon E, Rush AJ, Williams LM (2008) Improving the prediction of treatment response in depression: integration of

clinical, cognitive, psychophysiological, neuroimaging, and genetic measures. CNS Spectr 13(12):1066–1086
25. Larisch R, Klimke A, Hamacher K, Henning U, Estalji S, Hohlfeld T, Vosberg H, Tosch M, Gaebel W, Coenen HH, Müller-Gärtner HW (2003) Influence of synaptic serotonin level on [18F]altanserin binding to 5HT2 receptors in man. Behav Brain Res 17; 139(1-2):21–9
26. MacQueen GM (2009) (2009) Magnetic resonance imaging and prediction of outcome in patients with major depressive disorder. J Psychiatry Neurosci 34(5):343–349
27. Mann JJ, Currier D (2007) A review of prospective studies of biologic predictors of suicidal behavior in mood disorders. Arch Suicide Res 11(1):3–16
28. Markianos M, Hatzimanolis J, Lykouras L (2002) Serotonergic and dopaminergic neuroendocrine responses of male depressive patients before and after a therapeutic ECT course. Eur Arch Psychiatry Clin Neurosci 252(4):172–176
29. McBride PA, Tierney H, DeMeo M, Chen JS, Mann JJ (1990) Effects of age and gender on CNS serotonergic responsivity in normal adults. Biol Psychiatry 15; 27(10):1143–55
30. Minegishi A, Ishizaki T (1984) Determination of free 3-Methoxy-4-Hydroxyphenylglycol with several other monoamine metabolites in plasma by high-performance liquid chromatography with ampherometric detection. J Chromatogr 311:51–57
31. Millan MJ, Gobert A, Lejeune F, Newman-Tancredi A, Rivet JM, Auclair A, Peglion JL (2001) S33005, a novel ligand at both serotonin and norepinephrine transporters: I. Receptor binding, electrophysiological, and neurochemical profile in comparison with venlafaxine, reboxetine, citalopram, and clomipramine. J Pharmacol Exp Ther 298(2):565–580
32. Mitani H, Shirayama Y, Yamada T, Kawahara R (2006) Plasma levels of homovanillic acid, 5-hydroxyindoleacetic acid and cortisol, and serotonin turnover in depressed patients. Prog Neuropsychopharmacol Biol Psychiatry 30(3):531–534
33. Mitchell PB, Smythe GA (1991) Endocrine and amine responses to D, L-fenfluramine in normal subjects. Psychiatry Res 39(2): 141–153
34. Palencia G, Rios C, Sotelo J (2001) Clomipramine increases the striatal concentrations of dopamine and homovanillic acid in rats chronically consuming alcohol. J Stud Alcohol 62(6):724–729
35. Pompili M, Serafini G, Innamorati M, Möller-Leimkühler AM, Giupponi G, Girardi P, Tatarelli R, Lester D (2010) The hypothalamic-pituitary-adrenal axis and serotonin abnormalities: a selective overview for the implications of suicide prevention. Eur Arch Psychiatry Clin Neurosci 260(8):583–600
36. Ravna AW, Sylte I, Dahl SG (2009) Structure and localisation of drug binding sites on neurotransmitter transporters. J Mol Model 15(10):1155–1164
37. Ruhé HG, Mason NS, Schene AH (2007) Mood is indirectly related to serotonin, norepinephrine and dopamine levels in humans: a meta-analysis of monoamine depletion studies. Mol Psychiatry 12(4):331–359
38. Sallee FR, Vrindavanam NS, Deas-Nesmith D, Carson SW, Sethuraman G (1997) Pulse intravenous clomipramine for depressed adolescents: double-blind, controlled trial. Am J Psychiatry 154(5):668–673
39. Sallee FR, Vrindavanam NS, Deas-Nesmith D, Odom AM, Carson SW, Sethuraman G (1998) Parenteral clomipramine challenge in depressed adolescents: mood and neuroendocrine response. Biol Psychiatry 1; 44(7):562–7
40. Santiago M, Matarredona ER, Machado A, Cano J (1998) Influence of serotoninergic drugs on in vivo dopamine extracellular output in rat striatum. J Neurosci Res 52(5):591–598
41. Sharma R, Javaid JI, Janicak P, Faull K, Comaty J, Davis JM (1989) Plasma and CSF HVA before and after pharmacological treatment. Psychiatry Res 28(1):97–104
42. Smith DF, Geday J (2001) PET neuroimaging of clomipramine challenge in humans: focus on the thalamus. Brain Res (16) 892(1):193-7
43. Stanley M, Traskman-Bendz L, Dorovini-Zis K (1985) Correlations between aminergic metabolites simultaneously obtained from human CSF and brain. Life Sci 7;37(14):1279-86
44. Suhara T, Takano A, Sudo Y, Ichimiya T, Inoue M, Yasuno F, Ikoma Y, Okubo Y (2003) High levels of serotonin transporter occupancy with low-dose clomipramine in comparative occupancy study with fluvoxamine using positron emission tomography. Arch Gen Psychiatry 60(4):386–391
45. Sweeney D, Nelson C, Bowers M, Maas J, Heninger G (1978) Delusional versus non-delusional depression. Neurochemical differences. Lancet 100–101
46. Umene-Nakano W, Yoshimura R, Ueda N, Suzuki A, Ikenouchi-Sugita A, Hori H, Otani K, Nakamura J (2009) Predictive factors for responding to sertraline treatment: views from plasma catecholamine metabolites and serotonin transporter polymorphism. J Psychopharmacol PMID:19825907