

# Interaction of Pipamperone Augmentation of Citalopram and Genetic Variables in the Prediction of Antidepressant Response

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## Abstract

**Background:** Individual variability observed in therapeutic response to antidepressant drug treatment is thought to be in part genetically determined. In a search for genetic predictors of antidepressant response in 1953 patients treated with citalopram (CIT) in the STAR\*D trial, McMahon et al., reported that variation in the gene encoding the serotonin 2<sub>A</sub> receptor (5HT<sub>2A</sub>) was associated with antidepressant treatment outcome. Pipamperone (PIP) is an antipsychotic drug that is not available in the US, but is widely used in Germany, Belgium and other EU countries. PIP is a highly potent and selective antagonist at the 5-HT<sub>2A</sub> and D<sub>4</sub> receptors when administered at very low doses (5 to 15 mg/day). In a randomized, double-blind proof-of-concept (POC) study in major depression (DSM-IV), PIP at those low doses was found to both accelerate and augment the antidepressant effects of CIT with no additional side effect burden.

**Methods:** As part of a separate protocol, approved by the responsible IRB, patients participating in the POC study were asked to participate in pharmacogenetic testing. Of the 165 patients enrolled into the POC study, 89 consented to this additional protocol, 44 of them receiving CIT and 45 PIP/CIT. Pharmacogenetic assessments included polymorphisms in 2 genes influencing the pharmacokinetics of CIT and PIP: *CYP2D6* and *ABCB1*, as well as polymorphisms in 5 genes known to be related to pharmacodynamic effects of selective serotonin receptor inhibitors: the serotonin transporter, *BDNF*, *COMT*, *5HT<sub>2A</sub>* and *FKBP5*. In addition, the analysis included three putatively functional polymorphisms in genes regulating dopaminergic transmission: *DRD4*, *DRD3* and the dopamine transporter gene. Here we present the results of an exploratory analysis on the relation of the assessed genetic variants and the onset of antidepressant effect, quantified by means of T<sub>50</sub> values. T<sub>50</sub> was defined as the time to 50% of the maximum observed antidepressant effect, i.e. 50% of the maximum drop in MADRS score and was obtained by means of population pharmacokinetic/pharmacodynamic (PK/PD) modeling.

**Results:** In this subsample, augmentation with PIP led to a highly significant reduction of T<sub>50</sub> (p<0.001), with the T<sub>50</sub> being over 5 days shorter in patients treated with PIP/CIT than patients treated with CIT alone. Of the 19 tested polymorphisms, only the *BDNF* Val66Met polymorphism had a significant main effect on T<sub>50</sub> (p=0.04, genotypic model). Using a general linear model, we then tested the interaction of the genetic predictors with the treatment arm on T<sub>50</sub> and observed a significant treatment by SNP interaction for the *BDNF* Val66Met polymorphism (p=0.038) as well as for rs7997012 within the *5HT<sub>2A</sub>* gene (p=0.024). In both cases, significant effects of the polymorphisms were only seen in CIT but not the PIP/CIT group. In fact, for carriers of the *BDNF* Met risk allele, the improvement of T<sub>50</sub> by the augmentation was 7.7 days on average vs. 4.2 days for the Val/Val carriers. For carriers of the rs7997012 risk allele, the improvement was 6.6 days, vs 1.6 days for the carriers of the rarer beneficial AA genotype. *CYP2D6* metabolizer status did not have an effect on T<sub>50</sub> in either treatment group.

**Discussion:** Augmentation with PIP led to significant reduction in T<sub>50</sub>. Polymorphisms in *BDNF* and *5HT<sub>2A</sub>*, previously reported to be associated with response to antidepressant treatment, predicted T<sub>50</sub> only in the CIT but not the PIP/CIT group. Improvement of T<sub>50</sub> with PIP augmentation was much more pronounced in carriers of the previously defined non response alleles. This suggests that patients carrying genetic polymorphisms known to be associated with poorer response to traditional antidepressant drugs would especially profit from PIP augmentation.

## Background

- Citalopram (CIT) is a selective serotonin reuptake inhibitor (SSRI) used as a first-line agent for the treatment of major depressive disorder (MDD). A genetic study of phenotypes measuring outcome of CIT treatment in the Sequenced Treatment Alternatives to Relieve Depression (STAR\*D) study detected a significant association with a variant located in the serotonin 2A receptor gene (*HTR2A*).<sup>1</sup>
- Pipamperone (PIP) is a mild antipsychotic used in the treatment of schizophrenia in a number of EU countries. At recommended doses (120–360 mg/d), PIP is only a moderate dopamine D<sub>2</sub> receptor antagonist; at low doses (5–15 mg/d), PIP is a highly selective 5-HT<sub>2A</sub> and D<sub>4</sub> receptor antagonist.

- A randomized, double-blind proof-of-concept study in 165 patients with moderate to severe MDD showed that the addition of low-dose PIP (5 mg twice daily [BID]) augmented and accelerated the antidepressant effect of CIT.
- Patient response is difficult to predict because of the highly variable rates of efficacy and tolerability to antidepressant drugs among individuals.
  - Contributors to this variability include clinical heterogeneity and psychophysiological, psychosocial, and genetic factors.

## Objective

- To investigate whether genetic variation predicts the onset of response to PIP/CIT combination therapy or CIT monotherapy in patients with MDD

## Methods

### Patients

- Participants (n=89) were patients with moderate to severe MDD without psychotic symptoms (as defined by DSM-IV) who participated in a phase II trial (n=165) that tested the efficacy and tolerability of PIP/CIT combination therapy in a randomized, double-blind study (ClinicalTrials.gov identifier: NCT00672659).
- For each participant, 20 mL of whole peripheral blood was obtained by venipuncture.
- Patients received combination treatment with CIT 40 mg once daily (QD) plus PIP 5 mg BID or CIT 40 mg QD plus placebo BID in a fixed-dose titration design.
- The study was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice; all patients provided written informed consent.

### Primary Outcomes

- Change from baseline in the Montgomery-Åsberg Depression Rating Scale (MADRS) score at 8 weeks
- The onset of antidepressant effect, defined as the time to reach 50% of the maximum decrease in total MADRS score (T<sub>50</sub>), identified by a population pharmacokinetic (PK)/pharmacodynamic (PD) model

### Genetic Variants and Genotyping

- 25 single nucleotide polymorphisms (SNPs) were selected from 11 genes that were involved in the PK of PIP or CIT (*ABCB1* and *CYP2D6*), the PD effects of SSRIs (*BDNF*, *COMT*, *FKBP5*, *HTR2A*, and *SLC6A4*), or the regulation of dopamine transmission (*DRD3*, *DRD4*, and *SLC6A3*), or were reported to be associated with antidepressant treatment outcomes (CRHR1).
- DNA was extracted from whole blood using a BioRobot M48 automated extraction system (QIAGEN, Valencia, CA). Variants were genotyped using Taq-Man® assays on a 7900HT Fast Real-Time PCR System (Applied Biosystems, Foster City, CA).
- To distinguish between the long and short alleles of the variant in the promoter of the serotonin receptor (*5HT<sub>1PR</sub>*), fluorescent-labeled primers and polymerase chain reaction products were electrophoresed with an ABI Prism 3730 DNA sequencer (Applied Biosystems, Foster City, CA).
- Metabolizer status was inferred from the genotyping of 6 SNPs in the *CYP2D6* gene; individuals were classified as extensive, intermediate, or poor metabolizers based on the identified haplotypes.

## Statistical Analyses

- A general linear model with T<sub>50</sub> as the dependent variable was used to measure the main effect of each SNP and the interaction between each SNP and the treatment received.
- Both genotypic and dominant models were tested, combining patients who carried the risk allele for a worse response and noncarriers.
- For 2 significant SNPs, interaction analyses assessed the effect of carrying both the *BDNF* (Met/Met or Val/Met) and *HTR2A* risk genotypes (GG or AG) vs beneficial genotypes on T<sub>50</sub> in each treatment group.

## Results

- Demographic and clinical characteristics were similar between groups (Table 1).

Table 1. Demographic and Clinical Characteristics of Patients Included in the Pharmacogenetic Study

Characteristics	Treatment Group	
	CIT (n=44)	PIP/CIT (n=45)
Mean ± SD age (range)	44.7±11.3 (20–64)	43.3±11.8 (20–68)
Men/Women	11/33	5/40
White, n (%)	44 (100)	44 (100)
Mean ± SD duration of current MDD episode, d (range)	104±44 (12–220)	96±39 (31–188)
Duration >12 wk, n (%)	27 (61)	23 (51)
Mean ± SD MADRS total score (range)	32.8±5.8 (19–44)	32.2±5.0 (21–42)
MADRS ≥30, n (%)	34 (77)	28 (62)
Any previous psychiatric history, n (%)	28 (64)	38 (84)
Any ongoing psychiatric diagnosis in addition to MDD, n (%)	4 (9)	2 (4)

- The time to reach onset of antidepressant action averaged 5.6 days earlier for patients receiving PIP/CIT (P<0.001).
- All SNPs were successfully genotyped, with a mean call rate of 98.7%, and all of them were in Hardy-Weinberg equilibrium (P>0.05).
- There was no significant effect of variants in *ABCB1*, *COMT*, *FKBP5*, *DRD3*, *DRD4*, serotonin and dopamine transporters, the CRH receptor 1, or the metabolizer status with the onset of the antidepressant action (T<sub>50</sub>).

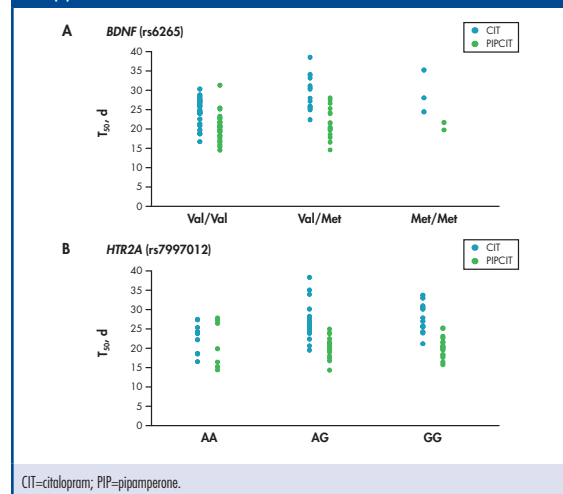
Table 2. Results for the 2 Significant SNPs Modifying the Onset of Antidepressant Response in Each Treatment Group

dbSNP ID	Good Response Allele	Genotype	Number of Patients (CIT/PIP/CIT)	Mean T <sub>50</sub> , d (95% CI)		P Value
				CIT	PIP/CIT	
rs6265	Val	Val/Val	52 (26/26)	24.8 (23.3–26.2)	20.5 (19.0–21.9)	0.038
		Met carriers (Val/Met or Met/Met)	37 (18/19)	29.2 (27.5–31.0)	21.5 (19.8–23.3)	
rs7997012	A	AA	16 (9/7)	23.0 (20.4–25.5)	21.3 (18.4–24.3)	0.024
		G carriers (AG or GG)	72 (34/38)	27.5 (26.1–28.8)	20.9 (19.6–22.1)	
rs6265*rs7997012	Val+A	Val/Val + AA, Val/Val + G carriers	58 (27/31)	24.6 (23.1–30.0)	21.0 (19.6–22.3)	0.002
		Met carriers + AA, Met carriers + G carriers	30 (16/14)	29.9 (28.0–31.7)	20.9 (18.9–22.8)	

CIT=citalopram; PIP=pipamperone; SNP=single nucleotide polymorphisms; T<sub>50</sub>=time to reach 50% of the maximum decrease in total Montgomery-Åsberg Depression Rating Scale score. \*Interaction.

- The SNP associated with T<sub>50</sub> was the functional Val66Met polymorphism in the *BDNF* gene (rs6265). Patients carrying the Met allele showed an average delay of 2.6 days in their improvement in T<sub>50</sub> (P=0.005; genotypic model).
- Results from the interaction of each SNP with the treatment group showed that
  - 2 SNPs (rs6265 in *BDNF* and rs7997012 in *HTR2A*) modified the mean T<sub>50</sub> depending on the treatment type (P=0.038 and P=0.024, respectively).
  - Patients carrying the allele known to be associated with worse antidepressant response (Met for rs6265 and G for rs7997012) had a significantly later onset of antidepressant action when treated with CIT (Table 2; Figure 1) but benefited most from the PIP augmentation.

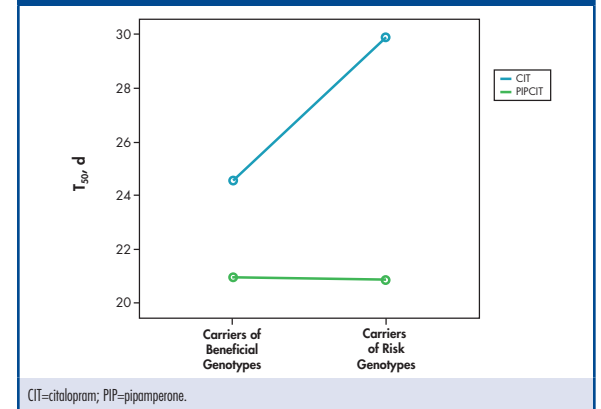
Figure 1. Time to Reach 50% of the Maximum Decrease in Total Montgomery-Åsberg Depression Rating Scale score (T<sub>50</sub>) as a Function of Treatment Received and Genotype in 2 Significant SNPs: (A) Functional Val66Met Variant in *BDNF* and (B) *HTR2A*



- Patients who received PIP augmentation and carried the Met risk allele in *BDNF* improved in T<sub>50</sub> by a mean average of 7.7 days compared with those patients who only received CIT, and by 6.6 days if they were carriers of the G risk allele of rs7997012 in *HTR2A* (Table 2). Patients with other genotypes benefited from the PIP augmentation to a lesser extent (4.3 d earlier response in the case of *BDNF* and 1.7 d in the case of *HTR2A*).

- Patients who received PIP augmentation and carried the risk genotypes of both the *BDNF* and *HTR2A* SNPs improved 9 days earlier than patients with the same genotype configuration but who received CIT alone (Table 2; Figure 2).

Figure 2. Plot of the Time to Reach 50% of the Maximum Decrease in Total Montgomery-Åsberg Depression Rating Scale score (T<sub>50</sub>) as a Function of Treatment Received and Being a Carrier of Beneficial or Risk Genotypes at Val66Met Variant in *BDNF* and *HTR2A*



## Conclusions

- Both *BDNF* and *HTR2A* were previously reported to be associated with response to antidepressant treatment. The rs7997012 AA genotype in *HTR2A* is associated with a better response to CIT,<sup>2</sup> whereas both the Val and the Met allele in *BDNF* have been related to a better response to different antidepressants.
- PIP/CIT produced overall greater improvements in the clinical response to treatment compared with CIT monotherapy.
- Genetic variants in *BDNF* and *HTR2A* can predict a slower response to CIT treatment.
- Patients carrying the nonresponse alleles for these SNPs might benefit most from combination therapy with PIP and CIT.

## References

- McMahon FJ, et al. *Am J Hum Genet.* 2006;78(5):804-14.
- Horstmann S, Binder EB. *Pharmacol Ther.* 2009;124(1):57-73.

## Disclosures

Dr. Elisabeth Binder, Dr. Kees Bol, Prof. Didier de Chaffoy, Dr. Ludo Haazen are consultants to PharmaNeuroBoost. Prof. Charles Nemeroff, Prof. Alan Schatzberg, and Prof. Thomas Schlaepfer are members of the Scientific Advisory Board of PharmaNeuroBoost and are also shareholders in the company. Prof. Nemeroff additionally serves on the Scientific Advisory Board for AstraZeneca and CeNeRx. He owns equity or is stock holder in Corcept; Revaax; NovaDel Pharma and CeNeRx. Prof. Schatzberg also owns equity in Corcept; Amnestix; BrainCells; CeNeRx; Somaxon Forest; Pfizer; Merck; Neurocrine; and Amylin. Prof. Schatzberg is a consultant for Corcept; Pfizer; Lilly; Neurocrine; BrainCells; Amnestix; Sanofi; Takeda; CeNeRx; CNS Response; Vivus; Roche; Xytis; Lundbeck; Forest Laboratories and GSK. Prof. Schatzberg receives income from Stanford University. Prof. Schlaepfer is a consultant for Lundbeck. Prof. Didier de Chaffoy is Chief Scientific Officer and Dr. Ludo Haazen is Chief Medical Officer of PharmaNeuroBoost. Dr. Erik Buntinx is psychiatrist, Chief Executive Officer and Managing Director of PharmaNeuroBoost, and is also a shareholder in PharmaNeuroBoost.

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