THE DIFFICULTIES OF REPRODUCING CONVENTIONALLY DERIVED RESULTS THROUGH 500K-CHIP TECHNOLOGY

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A genetically predisposed aberrancy of the inflammatory response system has been linked to various complex diseases. In the field of psychiatry, we have several sources of evidence for the involvement of inflammatory process in the pathogenesis of psychiatric disorders: (1) a population-based study of 7,704 patients with a diagnosis of schizophrenia and 192,590 control subjects without psychiatric history has revealed that a parental history of schizophrenia was associated with a 5-fold risk for autoimmune diseases, whereas a parental history of autoimmune diseases increased this risk only slightly, by a factor of 1.45 [*Eaton et al. 2006*]; (2) several antidepressants and antipsychotics show anti-inflammatory effects [e.g., *Müller and Schwarz 2006a,b*]; and (3) a quantitative (R)-[11C]PK11195 positron emission tomography study demonstrated microglia activation in recent-onset schizophrenia [*van Berckel et al. 2008*].

Since the so-called "natural" antibody IgM in normal serum is often found to bind to specific antigens, even in the absence of prior immunization, and the formation of chronically elevated levels of poly-reactive IgM develops years before the first clinical symptoms occur [*Nielen et al. 2006*], elevated IgM levels have been hypothesized to be related to the patients' genetically predisposed aberrancy of inflammatory response system, and may even be related to the abnormalities of CNS metabolism observed with schizophrenic and affective disorders.

To investigate the extent to which IgM levels can be reproducibly predicted for each individual patient from his/her multilocus genotype, we aimed at carrying out a Neural Network (NN) analysis on a sufficiently large sample (n=1,042; genotyped for 5,728 SNPs of a conventionally designed 0.4 Mb genome scan) under the constraint of a 10-fold cross-validation. Since NN results tend to be over-optimistic, even when using stringent cross-validation approaches, we were interested in the reproducibility of predictors across populations ("training" versus "test" samples) and across SNP sets (conventionally designed genome scan versus anonymous 500k-chip). To address these questions, we relied on independent test samples (n=746; genotyped for 545,080 SNPs of a 500k-chip) along with 6 different SNP sets, each with 5'728 SNPs drawn from the 500k-chip under the constraint of maximum informativeness and compatibility with the training SNPs.

Averaged across the 10-fold cross-validation solutions and applied to the 1,042 probes, weight matrices and classifiers yielded an overall performance for each optimization step. The optimization stopped when a plateau was reached at a rate of 77.3% [±0.636] correctly classified subjects out of the entire sample. In terms of clusters of at least 3 SNPs within a 0.5 Mb region, the training step yielded a configuration of 15 genomic loci (61 SNPs) that served as reference for subsequent investigations into the reproducibility of classifiers across populations and SNP sets. When applying the same algorithm to the 746 test samples with 6 competitive SNP sets, analyses typically yielded relatively reproducible results for 4 out of the 6 SNP sets, whereas the results of the 2 other SNP sets turned out to be largely arbitrary. Tentative analyses of our patients treated with antidepressants or antipsychotics suggest that up to 15% of the observed variation of response under antidepressants and up to 25% under antipsychotics might be explained by inflammatory effects.

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GENE ASSOCIATION STUDY AND META-ANALYSIS OF SUPEROXIDE DISMUTASE 2 (SOD2, MNSOD) GENES

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Introduction: Tardive dyskinesia (TD) is a potentially irreversible side effect of chronic antipsychotic medication that arises in approximately 25% of chronically treated schizophrenia patients. Oxidative stress has been one of the proposed mechanisms influencing TD risk. Hori et al (2000) reported the original positive finding of an association between TD and the *SOD2* (*MnSOD*) Ala9Val polymorphism in a Japanese sample, but most research groups failed to replicate their positive findings.

Methods: We investigated the role of the *SOD2* Ala9Val (rs4880) polymorphism in a group of well-characterized schizophrenia patients (N=223) who have been assessed for TD. We also performed a meta-analysis of all the previously published TD studies, including data from our sample, on the Pro187Ser (N=5 studies) and Ala9Val (N=9 studies) polymorphisms.

Results: We did not observe a significant association of the Ala9Val polymorphism with TD occurrence or AIMS in our Caucasian (n=193; p=0.38) and African American (n=30; p=0.72) samples when analyzed independently. Meta-analysis did not reveal a significant association of the Ala9Val alleles or genotypes with TD occurrence (Odds ratio for the Ala allele=0.98; 95% confidence interval: 0.73-1.32; p=0.89).

Conclusions: The *SOD2* Ala9Val polymorphism does not appear to play a major role in TD risk; however, additional polymorphisms should be tested before the role of *SOD2* in TD can be completely excluded.

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PREDICTION OF RESPONSE TO PHARMACOTHERAPY BY SEROTONIN AND DOPAMINE RELATED GENES IN OBSESSIVE COMPULSIVE DISORDER

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Background: Serotonin reuptake inhibitors (SRIs) are the most effective pharmacological treatment currently available for patients with obsessive-compulsive disorder (OCD). Still, up to 40 to 60% of OCD patients do not respond to pharmacotherapy. Genetic differences in the expression of serotonin and dopamine related functions between patients with OCD might explain the discrepant response to pharmacotherapy.

Method: In a first study, 91 outpatients with primary OCD according to DSM-IV criteria were randomly assigned in a 12-week, double-blind trial to receive dosages titrated upward to 300 mg/day of venlafaxine, or 60 mg/day of paroxetine. Primary efficacy was assessed by the change from baseline on the Yale-Brown obsessive-compulsive scale (Y-BOCS), and response was defined as a ≥ 25 % reduction on the Y-BOCS. In a second study, 64 drug-free or drug-naïve patients meeting DSM-IV criteria for OCD were randomized to 10 weeks double-blind treatment with citalopram (60 mg/day) with quetiapine (300-450 mg/day) or with placebo. Responders and non-responders were stratified according to DRD2 TaqI A and COMT Val(158)Met and 5-HTT, 5-HT_{2A}, and 5-HT_{1B} genotypes.

Results: In the first study, 64% of responders carried the S/L genotype of the 5-HTTLPR polymorphism (χ^2 = 7.17, df=2, p=0.028). In the paroxetine treated patients, the majority of responders carried the G/G genotype of the 5-HT_{2A} polymorphism (χ^2 = 8.66, df=2, p=0.013), whereas in the venlafaxine treated patients, the majority of responders carried the S/L genotype of the 5-HTTLPR polymorphism (χ^2 = 9.71, df=2, p=0.008). In the second study, no significant differences in genotype distribution or allele frequencies of the COMT or DRD2 receptor were found between responders and non-responders to citalopram with quetiapine. However, nearly half of responders to citalopram with placebo carried the Met/Met (48%) genotype of the COMT polymorphism compared to none of the non-responders (χ^2= 10.06, df=2, p=0.007).

Conclusions: The results of these studies suggest that response in drug treated OCD patients is associated with specific genotypes of the serotonin and dopamine system.

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ETHNIC DIFFERENCE IN PHARMACOGENETIC RESULT IN MAJOR DEPRESSION. THE RESULT OF META-ANALYSIS.

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Objective. Major depressive disorder is a severe and increasingly important disease for its high prevalence and association with serious consequences and substantial negative impact on social health. The genetically determined investigation of pharmacological responses would be much helpful to evaluate the best therapeutic tool for each patient. However, heterogeneity across the studies could also make it difficult for these candidates to be translated into treatment recommendations. Among others ethnicity could play an important role as confounder in pharmacogenetic result. Therefore we investigated pharmacogenetic effect of possible candidate variants such as 5-HTTLPR, HTR1A C-1019G, HTR2A, TPH1 and Gbeta3 in both Japanese and Italian population and also performed meta-analysis of these genes with consideration of assessment procedure, duration of treatment and type of antidepressant to detect the difference of genetic influence on antidepressant response between Asian and Caucasian.

Methods. Studies were included in the current meta-analysis if they evaluated the association between response/remission rate or intolerance rate to antidepressants treatments and genetic polymorphism in adult patients diagnosed with major depressive disorder and analyzed by RevMan analysis 1.01.

Results. Clearly different results between ethnicities were observed with HTR1A, HTR2A -1438A/G (102T/C) and TPH1 A218C independent from efficacy of type antidepressant possibly due to efficacy of other polymorphisms, different allele frequency, differential effect depending on specific symptoms, and cultural or social differences between Asians and Caucasians. As for 5-HTTLPR, inconsistent result could not be observed between ethnicities but SSRIs prescribed study and others.

Conclusion. Ethnicity was found to play an important role as a confounder in pharmacogenetic result and therefore we should pay attention to ethnicity when we deal with these variants as possible clinical predictor, however heterogeneity of protocol in each study also make it difficult for us to know actual impact of such variants.

EFFECT OF 5-HT1A GENE POLYMORPHISMS ON ANTIDEPRESSANT RESPONSE IN MAJOR DEPRESSIVE DISORDER

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Variability in antidepressant response is due to genetic and environmental factors. Among genetic factors, the ones controlling for availability of the drug at the target site are interesting candidates. Rs6295C/G SNP in the 5-HT1A gene (HTR1A) has been found to affect the expression and function of HTR1A. In fact rs6295C/G is in strong linkage disequilibrium with other polymorphisms of HTR1A suggesting that those functional effects could be associated with polymorphisms other than or together with the synonymous rs6295C/G. In the present study we examined the possible association of a panel of markers in strong linkage disequilibrium of the HTR1A with SSRI/SNRI response in 137 Japanese major depression subjects followed for 6 weeks. We observed a significant association of better response to antidepressant in rs10042486C/C (P<0.0001), rs6295G/G (P<0.0001) and rs1364043T/T (P=0.018) genotype carriers (minor allele homozygotes), independently from clinical variables. Furthermore minor allele homozygous carriers in all these three SNPs were associated with treatment response by various assessment such as HAM-D score change over time (P=0.001), week 2 (P<0.0001), 4 (P=0.007), and 6 (P=0.048) as well as response rate (P=0.0005) and remission rate (P=0.004). We also pointed out the genotyping mis-definition of rs6295C/G in the previous four articles. In conclusion, this is the first study that reports a significant association of antidepressant response with rs10042486C/T and rs1364043T/G variants ofHTR1Aandalso with rs10042486–rs6295–rs1364043 combination. This finding adds important information for the pathway of detecting the genetics of antidepressant response even if results must be verified on larger samples.

INFLUENCE OF TAAR6 POLYMORPHISMS ON RESPONSE TO ARIPIPRAZOLE

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Background: There is some evidence suggesting a role of TAAR6 in schizophrenia. The aim of the present study is to investigate possible influences of a panel of markers in TAAR6 (rs8192625, rs4305745, rs4305746, rs6903874, rs6937506) on clinical outcomes and side effects in a sample of Korean schizophrenic aripiprazole treated patients.

Methods: Efficacy was assessed at baseline and weeks 1, 2, 4, 6, 8 using CGI-S, CGI-I, BPRS and SANS. Side effects were evaluated through SAS, BAS and AIMS. Multivariate analysis of covariance (MANCOVA) was used to test possible influences of single SNPs on clinical and safety scores. Haploview 3.2 was used to generate a linkage disequilibrium (LD) map and to test for Hardy-Weinberg equilibrium. Tests for associations using multi-marker haplotypes were performed using the statistics environment "R".

Results: A significant time per genotype interaction was found between rs4305746 and repeated measures of ANOVA on BPRS scores ($F=2.45$, d.f.$=10,365$, $p=0.007$). In particular G/A and A/A genotype patients were more likely to improve over time. Haplotype analysis did not revealed any significant association with clinical and safety scores at any time.

Conclusion: A possible association could exist between some genotypes in TAAR6 and response to aripiprazole. However, several limitations as the small sample size, the finding related to a single scale and the possibility of false positive findings require further investigations.

HARM AVOIDANCE MODERATES THE INFLUENCE OF SEROTONIN TRANSPORTER GENETIC VARIANTS ON DEPRESSIVE TREATMENT OUTCOME IN BIPOLAR PATIENTS

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Response to pharmacological treatments is moderated by both genetic and environmental factors. The contribution of such factors is relatively small and complex interactions are likely to be involved. Serotonin transporter gene (SLC6A4) is a major candidate gene associated to response to antidepressant treatment. Moreover, 5-HTTLPR has been associated with anxious-related such as neuroticism and harm avoidance (HA), which are known to influence the risk to develop Mood disorders and response to treatments. Accordingly, in a previous analysis, we found high HA as a predictor of poor outcome in Bipolar (BP) spectrum patients, particularly after three-six months of treatment. Recently, the temperamental trait of neuroticism has been hypothesized to partially mediate the effect of 5-HTTLPR on the risk to develop Major depression. Given this preliminary evidence, in the present study we aimed to investigate the interaction between 5-HTTLPR and HA on medium term antidepressant response on our sample of BP patients.

The sample was composed by 86 patients fulfilling DSM-IV criteria for a major depressive episode and evaluated for personality traits by the Temperament and Character Inventory, revised scale (TCI-R) at intake. Depressive severity was evaluated by the Hamilton rating scale for depression (HAMD) at intake and after 2, 6 and 12 months of treatment. Three serotonin transporter gene variants were genotyped: the tri-allelic 5-HTTLPR $L_A/L_G/S$ variant (rs25531), rs25533 T/C and the variable number tandem repeat (VNTR) STin2 12/10/9 polymorphism.

Contrary to expectations, SLC6A4 variants did significantly influence neither the course of depressive symptoms nor HA scores. However, a significant interaction was observed between HA and 5-HTTLPR genotype. Indeed, a high HA worsened more significantly the outcome in patients carrying the L_G/S or the S/S genotype than L_A/L_A patients. Though a number of limitations characterize the present study, particularly the small sample size and its naturalistic characterization, our results indicate HA as a potential moderator of the effect of 5-HTTLPR on the outcome of depression, other than influencing the individual risk to develop a depressive disorder as previously reported. Given that many factors may influence response to pharmacological treatments, studies that consider personality and other individual characteristics are warrant also in pharmacogenetic investigations.

ASSOCIATION STUDY OF INSULIN INDUCED GENE 2 (INSIG2) WITH ANTIPSYCHOTIC INDUCED WEIGHT GAIN.

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Introduction: Schizophrenia is a debilitating mental disorder affecting 1% of the general population. Treatment of schizophrenia is primarily through antipsychotic drugs (APD). Of the two classes of drugs, typical and atypical APDs, the latter are associated with metabolic side effects such as weight gain, diabetes mellitus, dyslipidemia, hypertension, coronary heart disease and stroke, together termed *metabolic syndrome*. The development of antipsychotic induced weight gain in only a subset of schizophrenia patients (~ 30%) and concordance amongst monozygotic twins, suggest a genetic basis. Recently, Le Hellard et al (2008) reported a significant association of three polymorphisms in Insulin induced gene 2 (INSIG2) with antipsychotic induced weight gain (rs17587100, rs10490624 and rs17047764). In this study we tried to replicate their findings in our patient population who had received either clozapine or olanzapine (n=92).

Results: We did not observe any significant allelic, genotypic or haplotypic association of the polymorphisms with antipsychotic weight gained in the European American population (n=63; $p>0.05$). In the African American patient sample (n=29) a significant association was observed with the rs17047764 G>C polymorphism present downstream of the INSIG2 gene. Carriers of the heterozygous genotype gained more weight compared to the homozygous genotypic groups (GC vs GG +CC; 13.11 ± 8.78 vs 5.56 ± 4.26; $p=0.022$).

Conclusion: We were unable to replicate the association that was reported by Le Hellard et al., (2008) in our relatively smaller European American sample set. However, we observed a significant association of the polymorphism in the African American sample. These observations have been made in a relatively small sample set and require replication in a larger sample set.

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TRIALLELIC FUNCTIONAL SEROTONIN TRANSPORTER POLYMORPHISM AND TREATMENT-RESISTANT DEPRESSION

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Clinical and preclinical studies support a dysfunction of serotonin metabolism in treatment-resistant depression (TRD). Functional polymorphism in the promoter region of the serotonin transporter gene (5-HTTLPR) is believed to be involved in the pathogenesis and treatment of major depression disorder (MDD). Here we investigated the influence of functional 5-HTTLPR in the triallelic system (S, L_A and L_G alleles; A/G variant rs25531) on the risk of TRD.

A total of 287 DSM-IV MDD patients judged treatment-resistant and 267 healthy volunteers were enrolled and genotyped for the 5-HTTLPR S/L and rs25531 polymorphisms.

Difference in the clustered allele frequencies (L_A vs. L_G+S) was observed between the two populations (p=0.032), and the L_AL_A homozygotes were more represented in the controls group (p=0.016, OR=0.64, 95%CI: 0.44-0.92).

Furthermore, other analyses are in progress in order to compare alleles frequencies of this polymorphism in additional MDD patients groups, responders or remitted to specific antidepressant drugs treatment.

In conclusion, this preliminary study represents the first evidence of the protective effect of the L_A allele of the 5-HTTLPR/rs25531 polymorphism on the TRD.

THE 5-HT-2C GENE AND ANTIPSYCHOTIC INDUCED WEIGHT GAIN: AN OLD TARGET BACK IN FOCUS

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Associations between gene variants of the 5-HT2C gene and antipsychotic induced weight gain are the most established findings in this area of research (reviewed by Mueller and Kennedy, 2006). However, the 5-HT2C gene has not been studied extensively and much of the genetic variance has not been studied yet. Thus our study aims to investigate three polymorphisms (including Cys23Ser, -759 C/T, -697G/C) using four different samples (total n = 222). Samples were collected in the US (A, B and C; total n = 139) and Germany (D; n = 83). Sample A and B (n = 80) was treated exclusively with clozapine and weight gain was assessed after 6 weeks. Sample C (n = 59) was treated with four antipsychotics (clozapine, olanzapine, haloperidol and risperidone) and weight gain was assessed on average for 11 weeks. Sample D was assessed for 6 weeks using a variety of antipsychotics. Since the 5-HT2C gene is X-linked, we performed separate analyses in females and males.

Our preliminary analyses indicate no association between males and any of the 5-HT2C variants. In females, we found a borderline significant finding with the -759C/T polymorphism and antipsychotic induced weight gain (p = .04). Carriers with the T/T-genotype did gain less weight, a finding that is consistent with previous findings in the literature (eg Reynolds et al., 2002; Ellingrod et al, 2005).

We are currently expanding our analyses including other polymorphisms and will perform haplotype analyses.

MODULATION OF ANXIETY-LIKE BEHAVIORS BY DIPEPTIDE ANALOGUE OF ENDOGENOUS CHOLECYSTOKININ DEPENDS ON EMOTIONAL STRESS REACTION PHENOTYPE.

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Background: Novel strategies for pharmacological intervention include the use of drugs that interact with brain neuropeptide systems. Of these neuropeptides, cholecystokinin (CCK) has been extensively studied and appears involved in the neuropathology of stress-related disorders. On the basis of structure of endogenous CCK_4 a series of retropeptides was designed. Behavioral experiments in rodents have shown that dipeptide GB-115 ($Ph(CH_2)_5CO$-Gly-D-Trp-NH_2) demonstrated selective anxiolytic effects in classical models of anxiety such as "open field" and "elevated plus-maze" (L.Kolik, 2006).

Objective: The present study investigated the modulatory action of GB-115 on emotional processes in Balb/c and C57Bl/6 mice with opposite emotional stress reaction phenotypes. Methods: The tests used include the forced swimming to reveal antidepressant effects and exposure of mice to a natural predator (cat) as the most anxiogenic stress for rodents followed by subsequent benzodiazepine binding assay.

Results: In the forced swim test GB-115 (0.025 mg/kg/, IP) was demonstrated to increase the latency period and decrease the immobilization time in both C57Bl/6 mice with active type of behavior and in Balb/c mice with "freezing" reaction to emotional stress. Diazepam (1.0 mg/kg, IP) was inactive in this paradigm. Taking into account the pronounced decrease in benzodiazepine binding at the mice brains under induced stressful conditions, GB-115 and Diazepam effects on binding of [^{3}H] flunitrazepam to $GABA_A$ receptors were studied *ex vivo* after cat exposure. GB-115 prevented the decrease in binding only at Balb/c (3493,5±230,5 DPM/μgp after stress compare to 3705,7±317,7 DPM/μgp in control) and didn't have such effect at C57Bl/6 (2957+232 DPM/μgp after stress compare to 3862,0±133,1 DPM/μgp in control). Diazepam was active in both mice strains.

Conclusion: It was found that GB-115 produced antidepressant effects in both mice with different levels of emotionality. The data obtained support and extend our previous findings that the anxiolytic-like action of GB-115 depends on genetically controlled emotional stress reaction phenotype. This work indicates that novel dipeptide analogue of CCK may have potential in the clinical management of anxiety and depression.

NOVEL PSYCHOTROPIC DRUG LADASTEN - THE MECHANISM OF ACTION

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INTRODUCTION

Ladasten (N-(2-adamanthyl)-N-(parabromphenyl)amine) was developed as psychostimulant and anxiolytic drug at the Institute of Pharmacology RAMS. Clinical trial phase II has confirmed Ladasten psychostimulant and anxiolytic properties at treatment of psychogenic disorders with the diagnosis "Neurasthenia F48.0" according to ICD-10.

OBJECTIVE

The primary aim of the research was to to investigate the mechanism of psychotropic effects of Ladasten.

METHODS

1. For pharmacogenomic study and bisulfate sequencing white outbred male rats weighing 200–250 g were used. For HPLC and radioligand binding experiments Maudsley reactive (MR) and non-reactive (MNRA) rats weighing 200–250 g were used.
2.Benzodiazepine binding was estimated by [3H]-diazepam binding technique.
3. Monoamine levels were analysed in the hypothalamus, striatum and nucleus accumbens by HPLC with electrochemical detection after rats exposure in the "Open Field" test.
4. Atlas TM Rat cDNA expression array (BD Bioscience, USA) was used for pharmacogenomic study.
5. Bisulfate sequencing was used to analyze the methylation character of 5'-flanking TH gene region, which is expressed in rat hypothalamus.

RESULTS

The pharmacogenomic study demonstrated that Ladasten modulates the expression of multiple genes involved in neural plasticity. Particularly Ladasten increased the gene expression of tyrosine hydroxylase and inhibited that of GABA transporter.
Ex vivo experiments on stress-induced decrease of benzodiazepine binding model on MR rats with "freezing" reaction in "Open Field" test (OF) demonstrated that Ladasten restores ligand-receptor interaction in benzodiazepine site of GABA-A receptor.

Using HPLC it was shown that Ladasten at a dose of 30 mg/kg increases the level of dopamine and serotonin in the nucleus accumbens and striatum of MNRA rats and does not produce such effect on MR rats with active behavior in OF.The data obtained from this study allow to conclude that the prevalence of psychostimulating or anxiolitic action of Ladasten depends on phenotype of response to emotional stress and can be mediated by monoamine and $GABA_A$ systems.

NEURAL NETWORK MODEL FOR GENE-GENE INTERACTION ANALYSIS: ANALYSIS OF SEROTONERGIC SYSTEM GENES IN ANTIPSYCHOTIC RESPONSE

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Predicting the outcome of antipsychotic treatment by genetic features would be of great usefulness for clinicians as many of schizophrenics may not have a satisfactory response in spite of adequate trials of antipsychotic drugs. In the present work we analyze the serotonergic genes with a few artificial neural network (ANN) models differing from one another by outcome definition and validation procedure. The sample consisted of a reanalysis of a sample of 200 schizophrenics included in trials with different antipsychotics. With the original outcome definition (responders/non-responders), ANN performed better than other methods in terms of gene-gene interaction analysis. In this model we included only genetic factors to test the reliability of ANN in gene-gene interaction analysis.

The ANN approach is as valid as traditional multivariate techniques for the analysis of psychopharmacogenetics studies. The genetic interactions modelled through ANN may be eventually applied at the clinical level for the individualized therapy. However the prediction is still far from satisfactory from a clinical point of view.

ASSOCIATION STUDY OF SEROTONIN-RELATED GENES WITH CLOZAPINE-INDUCED WEIGHT GAIN

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Weight gain is a frequently observed side-effect with many antipsychotic treatments and seems to be underreported and under-recognized in many schizophrenia patients. A number of human and animal studies implicate serotonin in the body weight control and feeding behavior. Interestingly, the serotonin system is an important target of atypical drugs and patients taking atypical antipsychotics are more likely to present weight gain than those taking typical drugs. As it has been shown that clozapine-induced weight gain can present a genetic component, we hypothesized whether genes related with the serotonergic system would be associated with this side-effect. We examined 49 markers across the serotonergic system (5HTR1A, 1B, 1D, 1E, 2A, 3A, 3B, 4, 5, 6, TPH2) in 84 schizophrenia patients treated with clozapine for 6 months. Analyzes have shown that 5HTR2A_rs4941570 allele ($p = 0.01$) and genotype ($p = 0.03$) were associated with higher weight gain. The haplotype composed by 5HTR2A_rs2296972 and rs2760651 were also associated with higher weight gain ($p = 0.03$). Our results suggest that 5HTR2A polymorphisms appear to play a significant role in clozapine-induced weight gain.

NICOTINIC RECEPTOR POLYMORPHISMS IN NICOTINE REPLACEMENT TREATMENT

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Tobacco addiction is the major cause of worldwide disability and premature death (about 5 million deaths per year). The World Health Organization estimates that if current trends continue, the annual number of deaths from tobacco-related disease, including heart disease, lung disease, cancer and increased variety of infectious disease, will double from five million in the year 2000 to 10 million in 2020. Nicotine replacement therapies (NRTs), such as nicotine gum, patch, spray, inhaler and bupropion are currently the principal U.S. Food and Drug Administration - approved first-line pharmacological treatments for smoking cessation, although varenicline has gained approval. Although current guidelines recommend transdermal nicotine as a first-line treatment for nicotine dependence, the vast majority of smokers receiving transdermal nicotine relapse to their former smoking practices. In that regard, pharmacogenetic research may help to identify smokers that are good responders to nicotine replacement therapy. We hyphothesized that variants located in the gene for the alpha7 nicotinic acetylcholine receptor subunit (CHRNA7) and a related gene (CHRFAM7A) which arises from a partly duplicated portion of CHRNA7 are among the factors conferring the interindividual variability in NRT response. The DNA samples were taken from 308 study subjects participating in the STOP (Stop Smoking Therapy, for Ontario Patients) Study, began in January 2006. The subjects were categorized as responders and non-responders based on quit success at the end of 10-weeks of treatment with NRT. To genotype each sample, we have performed PCR reactions, using 1 µl or 20 ng of template DNA. The SNPs were genotyped using automated methods (Q-PCR; ABI PRISM 7000, Applied Biosystems Inc. Foster City CA). Allelic discrimination was carried out on the ABI Prism 7000 Sequence Detection System using allele specific fluorescent labeled probes. Basic analyses were used chi-square to compare genotypes at each marker with the responder / non-responder phenotype. Then as a second phase of analyses the genetic data are analyzed using Helix Tree and logistic regression for post hoc interaction analysis. We have not found significant association between analyzed variants and response to NRTs. Our results suggest that CHRNA7 and CHRFAM7A do not play a major role in the susceptibility to quit smoking after NRT.

THE 5-HTTLPR INTERACTS WITH STRESSFUL LIFE EVENTS TO PREDICT RESPONSE IN GENDEP

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The serotonin transporter promoter linked polymorphic region (5-HTTLPR) is the variant with the most evidence to date for association with antidepressant response (Serretti et al, 2007; Mrazek et al, 2008). Environmental factors such as stressful life events (SLEs) prior to treatment have also been shown to predict treatment response (Monroe et al, 1983; Monroe et al, 1992; Mazure et al, 2000). Wilhelm et al. (2006) investigated the interaction between SLEs, 5-HTTLPR genotype, and baseline severity of depression in a longitudinal follow-up study. However, the effect of an interaction between genetic variants and SLEs in moderating response to treatment with antidepressants is relatively unexplored.

We here report analysis of data from GENDEP, a part-randomised pharmacogenomics trial, on the outcome of 811 subjects with major depression undergoing treatment with either escitalopram or nortriptyline. The occurrence of SLEs predicted response to escitalopram (maximum likelihood estimate, $P<0.05$), and the 5-HTTLPR (maximum likelihood estimate, $P<0.05$) moderated these effects. Gene-environment interactions including life events may therefore be important not only in the aetiology of depression, but also in predicting response to antidepressants.

Funding: GENDEP was funded by a European Commission Framework 6 grant, EC Contract LSHB-CT-2003-503428. Lundbeck provided both nortriptyline and escitalopram free of charge for the GENDEP study. Roche Molecular Systems provided the AmpliChip CYP450 Test® and associated support. GlaxoSmithKline contributed by funding an add-on project in the London centre, and latterly the London centre also received additional funding from the Biomedical Research Centre for Mental Health at the Institute of Psychiatry, King's College London and South London and Maudsley NHS Foundation Trust (funded by the National Institute for Health Research, Department of Health, UK).