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From genes to psychoses and back: the role of the 5HT2α-receptor and prepulse inhibition in schizophrenia

Abstract Decomposition of schizophrenia into neurobiological vulnerability traits is necessary to understand the complex genetic underpinnings of this phenomenologically defined disorder. This issue is discussed with a focus on prepulse inhibition (PPI) as a neurobiological phenotype and the 5HT2a-receptor as a candidate gene. A series of recent studies illuminates that PPI and 5HT2a-receptors present as vulnerability markers for schizophrenia; a functional sequence variant in the 5HT2a-gene is contributing to this relationship and might consequently contribute to the genetic predisposition to schizophrenia with a very small risk increase.

Key words schizophrenia · endophenotypes · prepulse inhibition · serotonin 2α-receptor (5HT2a) · 5HT2a-gene

From the psychoses to genes?

Psychiatric disorders are defined by clusters of symptoms and other conventional criteria (temporal, cut-off, psychosocial impairment). Their etiological basis is complex, encompassing polygenic and environmental causal factors. The classical approach for unravelling the genetic architecture of psychiatric disorders is to start with the phenotype and to map disease genes in a systematic (e.g. genome-wide) manner. This approach is sometimes called the backward approach. The final goal is to trace these identified susceptibility genes through their molecular, cellular and systemic actions to the behavioural phenotype ("forward genetics") (Fig. 1). Among psychiatric disorders schizophrenia received most extensive etiological research interest in this respect.

Using diagnostic constructs as phenotypes, the search for the molecular genetic determinants of schizophrenia by genome-wide linkage and subsequent candidate region based fine-mapping studies delivered only inconsistent results [14]; yet, meta-analyses based on multiple case-control studies confirmed allele-specific associations to schizophrenia for a few genes (mainly NRG1 and G72/DAOA); the strength of the associations is only very weak—mostly around a relative risk of 1.5 or less specific to genetic markers [16].

The recent methodological progress towards hypothesis-free genome-wide association studies did not strengthen the impact of these postulated susceptibility genes: Although genetic variants meeting the requirements of genome-wide significance-were identified in extensive or less well sized samples or subsamples (e.g. [8]); none of these susceptibility variants fits together with variants identified through the previous approach combining genome-wide linkage studies and candidate region fine-mapping. The diversity of these results points to an enormous complexity of the number and interactions of contributing factors. Alternative approaches are required in order to disentangle this complexity. One currently preferred strategy is the decomposition of the diagnostic phenotype into its neurobiological correlates.

Endophenotypes instead of diagnoses?

The phenomenological basis of the classification of psychiatric disorders is considered as the most impeding obstacle for the success in the search for susceptibility genes. Hence endophenotypes were proposed to substitute diagnoses in the search for genes underlying human diseases. In this context

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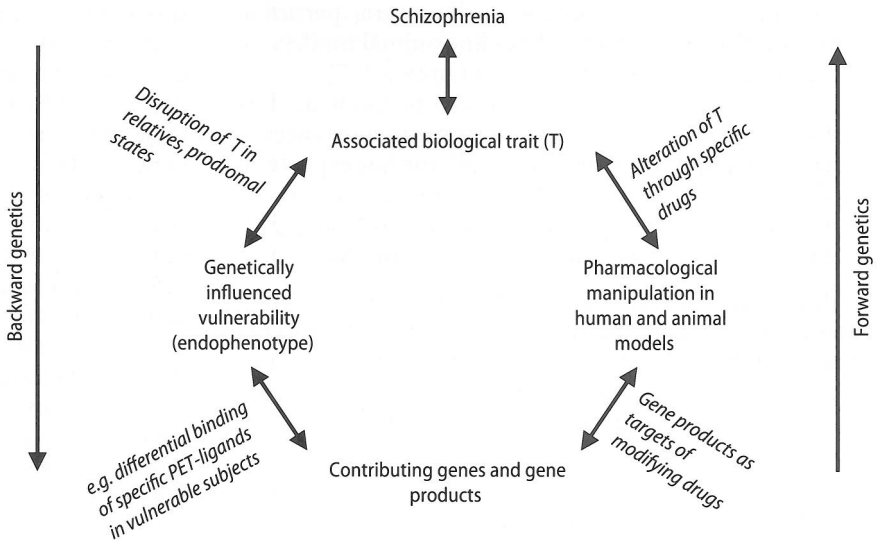

Fig. 1 Illustration of the interplay between backward genetics and forward genetics

endophenotypes present as the phenotypic endpoints of pathogenic variants of susceptibility genes. It is believed that endophenotypes are more closely related to the gene actions than the diagnoses; therefore these intermediate phenotypes might guide the way to the genetic basis of diseases. Neurobiological phenotypes are most promising in this respect [7].

This strategy was, however, not as successful as initially expected. Genetic variants with the strongest impact on endophenotypes (e.g. the COMT-met/val-polymorphism) were up to now not confirmed as susceptibility genes [8]. Yet, the genetic analysis of neurobiological endophenotypes might shed light on the genetic basis of the disorder in question and help to disentangle the genetic complexity. We discuss this possibility with the concept of prepulse inhibition (PPI) as a particularly promising endophenotype.

PPI of the acoustic startle response (ASR) has been firmly established as an operational measure of sensorimotor gating. PPI reflects the regulation of sensory input by filtering out irrelevant or distracting signals; by this means an overflow of sensory stimuli is prevented resulting in a selective and efficient processing of relevant information. PPI is defined as a substantial reduction of the amplitude of the startle reflex that occurs when a prepulse (distinctive non-startling stimulus) is presented 30–500 ms prior to the startling stimulus. The degree of PPI reveals a broad interindividual variation. PPI is mostly expressed as the percent reduction of the startle amplitude between prepulse-pulse trials and pulse-alone trials.

PPI: more than an endophenotype

One source of variation of the PPI is the disease status: In accordance with the filter deficit model of schizophrenia, diminished PPI has been consistently demonstrated in patients with schizophrenia and schizophrenia spectrum disorders as schizotypal personality disorder [6]. This PPI deficit in schizophrenia reflects a core pathogenic pattern of the disease: anatomical and functional perturbations in the cortico-striato-pallido-thalamic (CSPT) circuitry.

PPI is under strong genetic influences:

1. Twin studies suggest that over 50% of PPI variance can be attributed to genetic factors [1].
2. The established genetic predisposition of schizophrenia might also contribute to the PPI deficit of schizophrenia patients; unaffected first-degree relatives of schizophrenia patients also display a PPI deficit [2].

Increased risk for schizophrenia does not only derive from a familial-genetic relationship to an affected biological relative. Prodromal schizophrenia is characterized by psychosis-like intermittent or attenuated symptoms or signs which put a person on a substantially increased risk for conversion to the full blown psychosis. PPI also characterises this high risk state: Individuals with prodromal states reveal PPI deficits [11].

On the basis of these findings PPI disruption has emerged as a particularly promising candidate endophenotype of schizophrenia. Most of the criteria suggested by Gottesman and coworkers for an endophenotype are fulfilled by PPI [4]: it is associated with illness in families in the population, it is inherited, it shows temporal stability, and is more common in non-affected family members. In addition, PPI deficits are common in prodromal states. Yet, the criterion of pharmacological stability of the endophenotype is not fulfilled which may turn out to be an advantage for research purposes. Two aspects favour PPI as a candidate trait beyond alternative endophenotypes:

1. PPI does not only occur in humans but also in rodents [18]; thus it can be widely studied using genetic and

pharmacological techniques; furthermore, perturbation of PPI in rodents might define animal models for a core cognitive aspect of schizophrenia [17];
2. PPI in humans is not stable but can be modulated by drugs including a series of antipsychotic substances as well as psychotomimetic drugs [19]; the target proteins of these drugs define candidate genes underlying the PPI mechanism; these target proteins might also pave the way to investigate involved genetic mechanisms.

PPI shares these two aspects with some other equally well studied neurobiological endophenotypes of schizophrenia (particularly working and episodic memory). These privileged endophenotypes deserve detailed pharmacological and genetic analysis.

Pharmacological basis of PPI

Which neurotransmitter systems are determining PPI? Two major techniques are able to unravel the neurochemical basis of PPI:

1. Pharmacological manipulation by well characterized specific drugs and nicotine in human and animal models. These studies are pointing to four neurotransmitter systems: serotonin, dopamine, glutamate and acetylcholine.
2. Genetic manipulation in animal models using transgenic techniques confirms the impact of these four neurotransmitter systems.

We focus on the serotonin 2a-receptor (5HT2a-R). This receptor is involved in the vulnerability to schizophrenia. Using F-Altanserin as a PET-ligand of the 5HT2a-R, Hurlemann et al. [5] found a progressive reduction of the cortical and subcortical 5HT2a-R density (particularly in the right caudate nucleus) in subjects reporting prodromal symptoms or signs of schizophrenia. This observation was independent of the later conversion to full blown psychosis, but subsequent converters revealed the most distinct deviations.

Agonists of the 5HT2a-R have been extensively studied, e.g. the drug psilocybin [19]; these drugs are psychotomimetic and increase the vulnerability for psychosis. These 5HT2a-R-agonists simultaneously reduce PPI in human volunteers depending on the stimulus characteristics [19]. Dopamine-receptor antagonists such as atypical neuroleptic drugs like olanzapine antagonize the PPI deficit in patients with schizophrenia [10].

Implication of the 5HT2a-gene in PPI

Converging evidence for the role of the 5HT2a-R in the vulnerability to schizophrenia comes from molecular genetic investigations. The 5HT2a-R gene carries a DNA-sequence A/G-variation at position 1438 just upstream of the promoter influencing the promoter activity of the 5HT2a gene in in vitro models. In an experimental study by Parsons et al., this polymorphism resulted in a differential transcription of the gene (with the G allele related to underexpression of the gene); putatively, altered receptor density in vivo is the consequence [9]. Interestingly, this site is in linkage disequilibrium with another very frequently investigated SNP in the 5HT2a-R-gene at position 102 T/C. [15] This allelic variation is without a known direct functional effect. The 1438 G allele is segregating together with the 102 C allele. The linked functional 1438 A/G and the synonymous 102 T/C polymorphisms were recently considered together in order to investigate their impact on PPI. Indeed, the degree of PPI varied with the genotype: patients with schizophrenia who revealed homozygosity for C at position 102 together with homozygosity for 1438 G presented with the most pronounced PPI deficits [12]. Taken together with the gene expression studies by Parsons et al. [9], a reduced level of 5HT2a-R-mRNA or eventually also 5HT2a-protein can be expected among cases who are particularly impaired by PPI deficits. Yet, the relationship of PPI to the 5HT2a-R and the functional genetic variations is not unique. For example, Quednow et al. [13] recently described an association of the *val* variant of the COMT-gene to PPI deficits in schizophrenia; a similar relationship was described for healthy volunteers. COMT is the key catabolic enzyme for dopamine metabolism in the prefrontal cortex.

Relationship to the disorder schizophrenia?

The association of specific 5HT2a-genotypes with the endophenotype of PPI deficits as well as a reduction of gene expression proposes an association of the same genotype with schizophrenia. This hypothesis has received some limited although not fully satisfactory support. Several years ago the 102 T/C polymorphism was investigated in many genetic association studies for a relationship with schizophrenia (meta-analysis [20], and more recently [3]). These investigations propose a borderline relationship between the 5HT2a-R-gene 102 T/C polymorphism and schizophrenia with the C allele increasing the risk; the functional 1438 A/G polymorphism was detected later and received only insufficient attention in diagnosis-driven genetic association studies. Yet, a high prevalence of the 102 G homozygous genotype among patients with schizophrenia matches well with the underexpression of 5HT2a-R in vulnerable subjects as emerging from the F-Altanserin binding study by Hurlemann et al. [5].

Conclusion

It becomes increasingly apparent that the genetic architecture of schizophrenia is substantially more

complex than imagined. The phenomenological nature of the diagnostic concept might be a major contributor to this constellation. Decomposition into neurobiological traits and unravelling of the trait-specific genetic determinants is likely to produce more consistent results. We discuss this possibility by focussing on PPI as the intermediate phenotype in relation to the 5HT2a-R gene.

PPI deficits as well as reduced 5HT2a-R binding in PET-studies both define vulnerability markers for schizophrenia. A functional DNA-sequence variant in the promoter region of the 5HT2a-R-gene modifies gene expression; this variant is linked to another polymorphism extensively studied in schizophrenia. This constellation proposes the hypothesis that genetic variations in the 5HT2a-gene have an impact on the increased frequency of impaired PPI in schizophrenia as well as on the reduced 5HT2a-R binding in subjects at risk for schizophrenia.

This relationship between 5HT2a-R genetic polymorphisms and the PPI-intermediate phenotype carries over to the diagnostic phenotype only in a diluted manner. The statistical significance for the relationship between this 5HT2a-R-risk allele and schizophrenia reaches only borderline magnitude and remains undetectable by hypothesis-free genome-wide search strategies.

Conflict of interest statement The authors have no conflict of interest to declare.

References:

1. Anokhin AP, Heath AC, Myers E, Ralano A, Wood S (2003) Genetic influences on prepulse inhibition of startle reflex in humans. Neurosci Lett 353:45–48
2. Cadenhead KS, Swerdlow NR, Shafer KM, Diaz M, Braff DL (2000) Modulation of the startle response and startle laterality in relatives of schizophrenic patients and in subjects with schizotypal personality disorder: evidence of inhibitory deficits. Am J Psychiatry 157:1660–1668
3. Golimbet VE, Lavrushina OM, Kaleda VG, Abramova LI, Lezheiko TV (2007) Supportive evidence for the association between the T102C 5-HTR2A gene polymorphism and schizophrenia: a large-scale case-control and family-based study. Eur Psychiatry 22:167–170
4. Gottesman II, Gould TD (2003) The endophenotype concept in psychiatry: etymology and strategic intentions. Am J Psychiatry 160:636–645
5. Hurlemann R, Matusch A, Kühn K-U, Berning J, Elmenhorst D, Winz O, Kölsch H, Zilles K, Wagner M, Maier W, Bauer A (2008) 5-HT2A receptor density is decreased in the at-risk mental state. Psychopharmacology 195:579–590
6. Kumari V, Soni W, Mathew VM, Sharma T (2000) Prepulse inhibition of the startle response in men with schizophrenia: effects of age of onset of illness, symptoms, and medication. Arch Gen Psychiatry 57:609–614
7. Meyer-Lindenberg A, Weinberger DR (2006) Intermediate phenotypes and genetic mechanisms of psychiatric disorders. Nature 7:818–827
8. O'Donovan MC, Craddock N, Norton N, Williams H, Peirce T, Moskvina V, Nikolov I, Hamshere M, Carroll L, Georgieva L, Dwyer S, Holmans P, Marchini JL, Spencer CC, Howie B, Leung HT, Hartmann AM, Möller HJ, Morris DW, Shi Y, Feng G, Hoffmann P, Propping P, Vasilescu C, Maier W, Rietschel M, Zammit S, Schumacher J, Quinn EM, Schulze TG, Williams NM, Giegling I, Iwata N, Ikeda M, Darvasi A, Shifman S, He L, Duan J, Sanders AR, Levinson DF, Gejman PV; Molecular Genetics of Schizophrenia Collaboration, Gejman PV, Sanders AR, Duan J, Levinson DF, Buccola NG, Mowry BJ, Freedman R, Amin F, Black DW, Silverman JM, Byerley WF, Cloninger CR, Cichon S, Nöthen MM, Gill M, Corvin A, Rujescu D, Kirov G, Owen MJ. (2008) Identification of loci associated with schizophrenia by genome-wide association and follow-up. Nat Genet 30 Jul 2008. [Epub ahead of print]
9. Parsons MJ, D'Souza UM, Arranz M-J, Kerwin RW, Makoff AJ (2004) The -1438A/G polymorphism in the 5-hydroxytryptamine type 2A receptor gene affects promoter activity. Biol Psychiatry 56:406–410
10. Quednow BB, Wagner M, Westheide J, Beckmann K, Bliesener N, Maier W, Kühn K-U (2006) Sensorimotor gating and habituation of the startle response in schizophrenic patients randomly treated with amisulpride or olanzapine. Biol Psychiatry 59:536–545
11. Quednow BB, Frommann I, Berning J, Kühn KU, Maier W, Wagner M (2008) Impaired sensorimotor gating of the acoustic startle response in the prodrome of schizophrenia. Biol Psychiatry 29 May 2008 [Epub ahead of print]
12. Quednow BB, Kühn K-U, Mössner R, Schwab SG, Schuhmacher A, Maier W, Wagner M (2008) Sensorimotor gating of schizophrenia patients is influenced by 5-HT2A receptor polymorphisms. Biol Psychiatry 64:434–437
13. Quednow BB, Wagner M, Mössner R, Maier W, Kühn K-U (2008) Sensorimotor gating of schizophrenia patients depends on catechol O-methyl-transferase Val158Met polymorphism. Schizophr Bull 17 July 2008 [Epub ahead of print]
14. Sanders AR, Duan J, Levinson DF, Shi J, He D, Hou C, Burrel GJ, Rice JP, Nertney DA, Olincy A, Rozic P, Vinogradov S, Buccola NG (2008) No significant association of 14 candidate genes with schizophrenia in a large European ancestry sample: Implications for psychiatric genetics. Am J Psychiatry 165:497–506
15. Serretti A, Drago A, De Ronchi D (2007) HTR2A gene variants and psychiatric disorders: A review of current literature and selection on SNPs for future studies. Curr Med Chem 14:2053–2069
16. Sullivan PF (2005) The genetics of schizophrenia. PLoS Med 2:e212
17. Swerdlow NR, Geyer MA (1998) Using an animal model of deficient sensorimotor gating to study the pathophysiology and new treatments of schizophrenia. Schizophr Bull 24:285–301
18. Swerdlow NR, Braff DL, Geyer MA (2000) Animal models of deficient sensorimotor gating: what we know, what we think we know, and what we hope to know soon. Behav Pharmacol 11:185–204
19. Vollenweider FX, Csomor PA, Knappe B, Geyer MA, Quednow BB (2007) The effects of the preferential 5-HT2A agonist psilocybin on prepulse inhibition of startle in healthy human volunteers depend on interstimulus interval. Neuropsychopharmacology 32:1876–1887
20. Williams J, McGuffin P, Nothen M, Owen MJ, Spurlock G (1997) Meta-analysis of association between the 5-HT2a receptor T202C polymorphism and schizophrenia. Lancet 349:1221