Review

Pharmacogenetics of Smoking Cessation in General Practice: Results From the Patch II and Patch in Practice Trials

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Abstract

Introduction: Cigarette smoking remains the leading cause of preventable death worldwide. However, the efficacy of available first-line therapies remains low, particularly in primary care practice where most smokers seek and receive treatment. These observations reinforce the notion that ‘one size fits all’ smoking cessation therapies may not be optimal. Therefore, a translational research effort was launched by the Imperial Cancer Research Fund (later Cancer Research UK) General Practice Research Group, who led a decade-long research enterprise that examined the influence of pharmacological hypothesis-driven research into genetic influences on drug response for smoking cessation with transdermal nicotine replacement therapy in general practice.

Methods: New and previously published smoking cessation genetic association results of 30 candidate gene polymorphisms genotyped for participants in two transdermal nicotine replacement clinical trials based in UK general practices, which employed an *intention to analyze approach*.

Results: By this high bar, one of the polymorphisms (COMT rs4680) was robust to correction for multiple comparisons. Moreover, future research directions are outlined; and lessons learned as well as best-practice models for designing, analyzing, and translating results into clinical practice are proposed.

Conclusions: The results and lessons learned from this general practice-based pharmacogenetic research programme provide transportable insights at the transition to the second generation

of pharmacogenetic and genomic investigations of smoking cessation and its translation to primary care.

Introduction

Tobacco smoking remains a leading cause of preventable death internationally, with worldwide smoking-attributable deaths approaching 10 million annually by 2030 (Peto et al., 1996). Most smokers in developed countries present in primary care settings, providing an opportunity for general practitioners (GPs) to treat patients with nicotine dependence, but even with major advances in pharmacotherapy in recent decades, there remains room for improvement (Fiore et al., 2000, 2008). In a coordinated effort to advance the state of the science, a transdisciplinary research team, the General Practice Research Group (GPRG), embarked on an initiative to explore genetic moderation of drug response for smoking cessation.

Objectives

A formative review, published at the outset of the present project, described its conceptual framework (Walton, Johnstone, Munafò, Neville, & Griffiths, 2001). The objectives of this summative article are to (a) provide an historical narrative of the rationale, developmental, and deductive process that guided the GPRG’s pharmacogenetic smoking cessation research initiative; (b) present new and extant pharmacogenetic data in full; and (c) synthesize results and apply theoretical and practical lessons learned toward proposed best-practice research models to guide future translation to clinical practice.

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Rationale for and Development of Smoking Cessation Pharmacogenetic Research

Research concerning genetic influences on drug response for smoking cessation has undergone rapid growth in the last decade. In the two decades preceding this, multiple lines of investigation involving the GPRG and others converged to lay the groundwork for this new field of investigation marked by three major stages of translational research occurring in parallel and in series.

The first stage was the elucidation of the pharmacological substrates of nicotine dependence. A series of preclinical studies from multiple investigators resulted in major advances in our knowledge of the reward pathways implicated in nicotine self-administration and conditioned nicotine-seeking behavior and the complex interplay of multiple neurotransmitter pathways (dopamine [DA], serotonin, nicotinic acetylcholine, opioid, and other) affected by nicotine (Balfour, 2002; Benwell & Balfour, 1992; Corrigan, Franklin, Coen, & Clarke, 1992; Corrigan, Coen, & Adamson, 1994; Di Chiara & Imperato, 1988; Jones, Sudweeks, & Yakel, 1999; Klink, de Kerchove d'Exaerde, Zoli, & Changeux, 2001; Laviolette & van der Kooy, 2004; Marubio et al., 2003; Mifsud, Hernandez, & Hoebel, 1989; Olausson, Engel, & Soderpalm, 2002; USDHHS, 1988).

The second stage was the development, evaluation, and dissemination of effective smoking cessation therapies to large populations of smokers. Russell, Fowler, and colleagues conducted a series of studies of nicotine replacement therapy (NRT) (Russell, Feyerabend, & Cole, 1976; Russell, Sutton, Feyerabend, Cole, & Saloojee, 1977; Russell, Wilson, Feyerabend, & Cole, 1976) and behavioral therapies in general practice (Fowler, 1979, 1982; Fowler, Mant, Fuller, & Jones, 1989; Jackson, Stapleton, Russell, & Merriman, 1989; Jamrozik & Fowler, 1982; Jamrozik, Fowler, Vessey, & Wald, 1984; Jamrozik, Vessey, et al., 1984; Russell, Merriman, Stapleton, & Taylor, 1983; Russell, Stapleton, Hajek, Jackson, & Belcher, 1988; Russell, Wilson, Taylor, & Baker, 1979; Russell et al., 1993; Stapleton, Lowin, & Russell, 1999; Stapleton et al., 1995). These germinal studies demonstrated that all forms of NRT and clinician brief advice were efficacious for smoking cessation and formed the basis for the subsequent "Patch Trial" (described below) and the establishment of the Cochrane Tobacco Addiction Group. A series of Cochrane reviews by Lancaster and colleagues (Abbot, Stead, White, & Barnes, 1998; Gourelay, Stead, & Benowitz, 1998; Hajek & Stead, 1998; Hughes, Stead, & Lancaster, 1998; Lancaster & Stead, 1998a, 1998b; Silagy & Ketteridge, 1998; Silagy, Lancaster, & Fowler, 1998; Silagy, Mant, Fowler, & Lancaster, 1998; Stead & Hughes, 1998; White, Rampes, & Ernst, 1998) confirmed the efficacy of NRT and informed evidence-based guidelines for GPs and other health professionals in the United States and United Kingdom (Raw, McNeill, & West, 1998, 1999).

The third stage was the establishment of the heritability of cigarette smoking phenotypes and characterization of candidate genes using the classical pharmacogenetic model (Evans & Johnson, 2001; Evans & Relling, 1999). Starting in 1999, the GPRG recontacted participants in the Patch Trial (GPRG, 1993, 1994b), in order to collect blood samples for DNA extraction and genotyping and genotyped participants from a multiple risk factor reduction trial by the OXCHECK Study Group (1989–93) (GPRG, 1991, 1994a, 1995).

The GPRG initially focused on metabolic candidate genes in DA and serotonin pathways (i.e., monoamine oxidase [MAOA/MAOB], dopamine beta hydroxylase [DBH], catechol-O-methyl transferase [COMT]) and smoking quantity (McKinney et al., 2000) in OXCHECK and then examined associations between these and other candidate genes and smoking cessation outcomes in Patch II (Murphy, Johnstone, & Walton, 2004). At this time, Neville and colleagues, working in the GPRG, discovered that the rs1800497 (also known as "DRD2 Taq1A"; C32806T) single nucleotide polymorphism (SNP) is present in a previously unidentified threonine kinase gene ("ankyrin repeat and kinase domain containing 1" [ANKK1]) (Neville, Johnstone, & Walton, 2004). In 2002, the GPRG launched the Patch in Practice (PiP) Trial, which examined different levels of behavioral support in treatment-seeking smokers using transdermal NRT, again in general practice. PiP was designed to study pharmacogenetic associations, and samples for DNA analysis were collected at trial entry (Aveyard et al., 2007). Replication of associations from Patch II was sought, and novel polymorphisms were genotyped, in PiP (Johnstone & Murphy, 2007).

Methods of Patch II and PiP Trials

Additional details about both trials are reported in the debut clinical trial outcome papers (Aveyard et al., 2007; GPRG, 1993, 1994b) and subsequent pharmacogenetic investigation papers (David et al., 2002; David, Johnstone, et al., 2008; David, Munafò, et al., 2008; David, Munafò, Murphy, Walton, & Johnstone, 2007; Johnstone, Clark, Griffiths, Murphy, & Walton, 2002; Johnstone et al., 2007; Johnstone, Yudkin, Griffiths, et al., 2004; McKinney et al., 2000; Munafò, Zetteler, & Clark, 2007; Yudkin et al., 2004).

Patch Trial and Patch II Study

The study was a double-blinded, randomized, placebo-controlled trial of NRT patches for smoking cessation ($N = 1,686$). Treatment seeking smokers between the ages of 25 and 65 years who smoked ≥ 15 cigarettes/day were recruited from 19 UK general practices in Oxfordshire and randomized into one of four equal groups to receive active NRT patch in reducing doses over 12 weeks or a placebo patch, in combination with a specific Health Authority smoking cessation support booklet or a standard Health Education Authority leaflet. Abstinence at 1, 4, 8, and 12 weeks was assessed using self-report combined with exhaled carbon monoxide (CO) ≤ 10 ppm to confirm abstinence since the previous visit at 4 or 8 weeks and salivary cotinine ≤ 20 ng/ml at 12, 24, and 52 weeks. Primary outcomes were biochemically confirmed point prevalence abstinence at each follow-up assessment.

Eight years after the completion of the Patch Trial, surviving participants were recontacted. The GPs for 1,533 of the 1,686 original Patch Trial participants were located and provided consent to contact their patients, of whom 1,532 were invited at approximately 8 years from time of trial entry and 840 provided written consent and returned questionnaires. Of these, 767 also gave blood samples (50%). The final pharmacogenetic study sample was composed of 755 European-ancestry individuals who answered smoking history questionnaires at 8-year follow up (Patch II Study). Ethical approval was obtained from the Anglia and Oxford Multicentre Research Ethics Committee and from the 86 Local Research Ethics Committees covering the areas of residence of the patients.

The PiP Trial

This study was an open-label randomized trial of behavioral support intensity for smoking cessation in smokers 18 years and older who smoked more than 10 cigarettes/day using NRT patches ($N = 925$). Participants were recruited from 26 UK general practices in Oxfordshire and Buckinghamshire and randomized to one of two equal groups: Basic support (pre-cessation counseling and support visits by trial nurses in general practice surgeries at 1 and 4 weeks after the initial appointment) or weekly support (basic support plus telephone calls at 10 days and 3 weeks after the initial appointment and an additional visit at 2 weeks to motivate adherence to NRT patch and renew quit attempts). Treatment consisted of 15 mg/16 hr patches for 8 weeks. Abstinence at 1, 4, 12, and 26 weeks from quit day was assessed using self-report combined with exhaled CO < 10 ppm and salivary cotinine < 15 ng/ml. Primary outcomes were biochemically confirmed sustained abstinence at each follow-up assessment. Unlike the Patch Trial, participants underwent phlebotomy at trial entry ($n = 908$), and the final pharmacogenetic study sample consisted of 792 European-ancestry individuals.

DNA Extraction and Genotyping

Blood sample receipt and processing, DNA extraction, assays, primers, PCR, and other genotyping procedures are described in detail elsewhere (David, Johnstone, et al., 2008; David, Munafò, Murphy, Walton, & Johnstone 2007; David et al., 2002; Johnstone et al., 2002, 2007; Johnstone, Yudkin, Griffiths, et al., 2004; Huang, Payne, Ma, & Li, 2008; Lerman et al., 1997; McKinney et al., 2000; Munafò, Elliot, Murphy, Walton, & Johnstone, 2007; Munafò, Johnstone, Guo, Murphy, & Aveyard, 2008; Yudkin et al., 2004). The full list of polymorphisms genotyped is in Supplementary Table 1 and other genotyping details are available upon request.

Data Analysis

Chi-square analyses using two-tailed tests of significance were conducted comparing cotinine-verified 12-week (end of treatment) and 26-week abstinence rates for heterozygous and homozygous genotypes for the minor alleles to homozygous wild-type genotypes for each polymorphism (Supplementary Table 2). Statistical analyses of Patch II and PiP were stratified by genotype (wild type [allele]/wild type; wild type/minor [allele]; or minor/minor) and Patch II was also stratified by treatment (active patch; placebo patch). Based on the recommendations of Gwinn, Guessous, and Khoury (2008), we employed an *intention to analyze* approach to prevent publication bias and minimize Type 1 error (Munafò, Johnstone, Murphy, & Aveyard, 2008).

Published and New Results From Patch II and PiP Trials

Descriptive Data

Patch II study. There were 755 European-ancestry participants for whom DNA was available for genotyping. The sample was 59% female ($n = 447$), mean age of 43 ($SD = 10$) and mean Horn–Russell (Russell, Peto, & Patel, 1974) nicotine dependency score of 15 ($SD = 5$). There were 378 participants randomized to active patch (50%) and 377 to placebo patch.

PiP study. There were 792 European-ancestry participants for whom DNA was available for genotyping. The sample was

59% female ($n = 410$), mean age of 44 ($SD = 12$), and mean Fagerström Test of Nicotine Dependence (FTND) (Heatherton, Kozlowski, Frecker, & Fagerström, 1991) nicotine dependency score of 5 ($SD = 2$). There were 378 participants randomized to active patch (50%) and 377 to placebo patch.

The Horn–Russell measure was not administered to PiP participants and the FTND was not assessed in Patch II participants. Study characteristics for nonparticipants, those participants who were not available at 8-year follow-up to provide DNA samples (Patch) or those who refused to participate (Patch II), are presented elsewhere (David, Johnstone, et al., 2008; Johnstone, Yudkin, Hey, et al., 2004). Briefly, in Patch II, participants providing DNA samples were more likely to be female (59% vs. 52%), older (mean age 43 years [$SD = 10$] vs. $M = 42$ [10]), more likely to have attended all follow-up visits, and confirmed abstinent at each visit ($ps < .01$). There were no significant differences in nicotine dependence or baseline cigarette consumption. In PiP, the consent process included providing blood samples for DNA extraction/genotyping and blood samples, and DNA extraction was successfully performed for 98% of participants. In all, there were 30 polymorphisms genotyped in Patch II and PiP.

Eight polymorphisms (present within or in flanking regions of *ANKK1*, *DRD2*, *DRD4*, *SLC6A4*, *DBH*, and *OPRM1* genes) were genotyped in both studies in order to replicate Patch II results using PiP samples, seven polymorphisms were genotyped solely in Patch II samples (present within or in flanking regions of *DRD2*, *DRD4*, *SLC6A4*, *MAOA*, and *CLOCK* genes) and were not genotyped in PiP because they were determined to be low priority and/or relatively less informative, and 15 were genotyped uniquely in PiP samples (present within or in flanking regions of *HTR1A*, *TPH1*, *TH*, *DBH*, *CHRNA4*, *CHRNA7*, and *CHRNA2* genes) building on hypotheses generated in Patch II and advances in the state of the science during the course of the project.

Association With Smoking Cessation Outcomes

Published results. Replication was observed for only one polymorphism, *COMT* rs4680. *COMT* rs4680 AA genotype (vs. AG/GG) was associated with greater efficacy of NRT patch (vs. placebo) at 12 weeks in Patch II (Johnstone et al., 2007) and participants with AA genotypes were also more likely to be abstinent through treatment for the first 12 weeks and beyond extending to 52 weeks in PiP (Munafò, Johnstone, Guo, et al., 2008). The observations of association between rs1800497 12-week abstinence outcomes in Patch II (Johnstone, Yudkin, Hey, et al., 2004) were not replicated in PiP (Munafò, Johnstone, et al., 2009). Moreover, the gene $\times$ sex interaction reported for Patch II (Yudkin et al., 2004)—indicating that females (but not males) with CT/TT genotypes were more likely to be confirmed abstinent on active versus placebo patch at all follow-ups—was not observed in PiP (Munafò, Johnstone, et al., 2009). As previously reported, there was no association between the *SLC6A4* 5-HTTLPR and smoking cessation in Patch II or PiP—even when pooled analyses were conducted with samples from a third trial (David, Johnstone, et al., 2008; David, Munafò, Murphy, Walton, & Johnstone, 2007; Munafò et al., 2006).

New results. Supplementary Table 2 presents abstinence rates at 12 weeks and 26 weeks and chi-square p -values for all SNPs or variable number of tandem repeats (VNTRs), including new data. Only the *COMT* rs4680 SNP was statistically

significantly associated with smoking cessation, when correcting for multiple comparisons ($\alpha = .05/30$). Rs4680 was associated with smoking cessation at end of treatment ($p < .001$) and 26 weeks in the PiP trial ($p < .001$). In Patch II, the association between rs4680 and smoking cessation approached statistical significance in smokers treated with active patch ($p = .062$). Statistical trends were observed between *DRD2* rs6276 and rs6277, *CHRNA* rs2273502, *TH VNTR*, and *OPRM1* rs1799971.

Discussion

Interpretation and Clinical Translation of Patch II and PiP Results

The ultimate goal of pharmacogenetics is to “provide new strategies for optimizing drug therapy based on each patient’s genetic determinants of drug efficacy and toxicity” (Evans & Johnson, 2001). The results of the Patch II and PiP, when considered in the context of this ultimate goal, have informed and partially advanced the field of smoking cessation pharmacogenetics. However, the conceptual synthesis of the data from these and other pharmacogenetic investigations must take into account the potential transportability of the findings to future personalized treatments. There are, by this metric, several constraints to the external validity of these data.

First, at the patient care level, genetic tailoring of medications is a choice between different medications, rather than between an active drug and a placebo. Therefore, while it is essential to compare groups defined by either active or placebo or gradations in drug dose to demonstrate a pharmacogenetic effect, this does not address the clinically relevant question of whether or not one genetic subgroup is more responsive to a medication than another. Second, many pharmacogenetic investigations compare wild-type homozygous genotypes to those who carry one or more variant alleles, based on assumptions of autosomal dominant inheritance of pharmacogenetic traits and/or to maximize statistical power. However, medical decisions are made on an individual level and one cannot assume that heterozygosity and homozygosity are equivalent in penetrance of pharmacodynamic and pharmacokinetic phenotypes. Third, while the Patch II and PiP investigations were based in general practice primary care settings, most smoking cessation clinical trials are based in academic medical centers in tertiary care settings, limiting generalizability to community-based general practice. Clinical trials based in tertiary academic medical centers may be more likely to incorporate intensive behavioral therapy, whereas smokers in the population seeking treatment are more likely to receive brief advice from health care professionals, with limited follow-up, or use telephone quit lines. Fourth, drug response for smoking cessation is polygenic and any one genetic variant will explain only a fraction of the genetic variance. Finally, retrospective association analyses do not address the question of the effects of genetic tailoring on treatment outcomes. Therefore, best-practice models are needed for pharmacogenetic and pharmacogenomic clinical trials of smoking cessation.

One solution to the problem of the small impact of individual variants is to investigate multiple genes and their interactions. However, this would require several thousand participants to achieve sufficient power (Cardon, Idury, Harris, Witte, & Elston, 2000). Recently, Wang and Li (2010) published a path-

way analysis in which they identified 75 genes associated with smoking cessation. Genes associated with smoking cessation were over-represented in pathways involved in DA receptor signaling, cyclic AMP receptor signaling, calcium signaling, G protein-coupled receptor signaling, tyrosine metabolism, and other functions. There was substantial overlap with genes associated with smoking cessation in Patch II and/or PiP (i.e., *COMT*, *DRD2*, *DRD4*, *DBH*, *OPRM1*) (Wang & Li, 2010). Taken together, these data support a putative role for DA and mu-opioid pathway genes in smoking cessation, but more randomized controlled trial (RCT)-based genome-wide association studies are needed to advance the field toward personalized genomic medicine. GPRG investigators are contributing to this process, but research is still in its early days (Uhl et al., 2008, 2010).

COMT and Smoking Cessation

There are several reasons why of all the polymorphisms evaluated, *COMT* rs4680 would be the most robust association with NRT response for smoking cessation. Given its role in the degradation of extraneuronally released DA to pharmacologically inert metabolites (dihydroxyphenylacetic acid and homovanillic acid), the *COMT* enzyme tightly regulates DA tone (Akil et al., 2003; Chen et al., 2004). The rs4680 polymorphism in exon 3 of the *COMT* gene results in substitution of an A (methionine) for a G (valine) at codon 108/158 (Lachman et al., 1996). The A (met) allele results in a three- to fourfold reduction in *COMT* enzyme activity (Shield, Thome, Eckloff, Wieben, & Weinshilboum, 2004). *COMT* is a ubiquitous metabolic enzyme in DA brain regions—particularly prefrontal cortex, where there is a relative scarcity of DA transporters.

Converging data from functional neuroimaging studies indicates that compared to the A(met) allele, the G(val) allele is associated with greater sensitivity to nicotine abstinence on verbal working memory in the prefrontal cortex and anterior cingulate gyrus (Loughead et al., 2009), reduced extraneuronal DA tone (Bilder, Volavka, Lachman, & Grace, 2004), but increased phasic DA firing and greater nicotine-induced striatal DA release (Brody et al., 2006). Therefore, a plausible biobehavioral hypothesis to explain why smokers possessing the G(val) allele would be that individuals possessing the *COMT* rs4680 A allele being more sensitive to the cognitive effects of abstinence and at the same time more sensitive to nicotine replacement for positive reinforcement (i.e., increased striatal DA release) and negative reinforcement (i.e., increased phasic DA release in prefrontal cortex/ACC). Colilla et al. (2005) also reported an association between the A allele and NRT response among females. and Berrettini et al. (2007) reported an association of the A allele with bupropion efficacy—for smoking cessation. Given the pharmacology of *COMT*, well-characterized functional nature of the *COMT* rs4680 polymorphism, and patterns of replication for association with smoking cessation in treatment-seeking smokers, we were not surprised to find that its association with smoking cessation in NRT-treated smokers was robust.

Differences Between Patch II and PiP Methodologies

Differences in trial methodologies resulted in an inability to harmonize data to the degree necessary for precise replication. The PiP trial was intended to examine the effectiveness of different

levels of behavioral support in treatment-seeking smokers using NRT in real-world general practice. It was not explicitly designed to replicate Patch II. The strengths of the PiP design, such as generalizability to practice in community settings, resulted in limitations with regard to replicating Patch II.

Patch II reported point prevalence abstinence while PiP reported sustained abstinence. The difference in ascertainment of abstinence between studies is important because continuous abstinence rates are generally lower than point prevalence abstinence because participants can have smoking lapses but successfully abstain later in follow-up and still be considered abstinent using point prevalence criteria. Furthermore, in Patch II participants initially received a higher dose of NRT (21 mg/day) for the first 4 weeks, followed by tapering patch doses over the subsequent 8 weeks. This was combined with in-person behavioral support, assessment of patch adherence, and exhaled CO monitoring from study nurses at multiple follow-up visits in general practices. PiP participants received a lower dose of patch (16 mg), and follow-up support was telephone based. While the quality of telephone support received through the NHS Stop Smoking service is considered to be very high (Coleman et al., 2009), the relatively more intensive behavioral support and higher patch dose in Patch II, compared with PiP, may also have contributed to the lower observed abstinence rates in PiP.

Best-Practice Models for Smoking Cessation Pharmacogenetics and Genomics

Analytic Framework

The National Office of Public Health Genomics at the U.S. Centers for Disease Control and Prevention established the Evaluation of Genomic Applications in Practice and Prevention (EGAPP) initiative in efforts to apply evidence-based medicine precepts to the development of practice guidelines for use of genetic testing in clinical settings (Teutsch et al., 2009).

The EGAPP Working Group is incorporating an analytic framework and key questions to determine if specific genetic tests result in “clinically meaningful results” or are “useful in medical decision making” and to evaluate the following components of evaluation as “links in a possible chain of evidence”: analytic validity, clinical validity, clinical utility, and ethical, legal, and social implications (ACCE). *Analytic validity* relates to technical test performance (e.g., predicting drug response for smoking cessation and sustained abstinence), *clinical validity* pertains to the strength of association between genotype and clinical phenotype (e.g., nicotine metabolism or nicotine craving), and *clinical utility* is the degree to which the genetic test is beneficial or potentially harmful to patients (e.g., reduced nausea or nicotine toxicity or increased drug adherence because of more precise drug selection and/or dosing, or might revelation of test results lead to poor psychological outcomes or reduced drug adherence). The ACCE framework also addresses the *Ethical, Legal, and Social Implications* of genetic and genomic research in keeping with National Human Genome Research Institute guidelines (<http://www.genome.gov/10001618>).

In the determination of whether or not a polymorphism or other genetic variant, or series thereof, should be used in clinical practice, one can assess the level of evidence for each of the

ACCE components along a hierarchy of data sources and study designs. For example, a “gold standard” (Level I evidence) pharmacogenetic test would demonstrate high analytic validity based on data from a large panel of well-characterized samples from multiple sites and well-designed phenotype harmonization, high clinical validity based on well-designed clinical trials with rigorous and uniform assessment of abstinence and other behavioral process and outcome measures, and clinical utility drawing from meta-analyses of *prospective* RCTs of genetically tailored smoking cessation interventions.

In addition, the ethical, legal, and social implications of introducing specific PGx tests should be carefully considered and scrutinized. Most candidate gene and genome-wide association studies have been limited to samples of European ancestry. While it may be necessary to stratify analyses according to ancestry, pharmacogenetic research in non-European smokers must be greatly expanded to prevent a predictable new health disparity in personalized medicine. Furthermore, there is a paucity of prospective, genetically tailored treatment studies for smoking cessation, and caution is needed to refrain from moving directly from retrospective genetic investigations of RCTs to adoption of a genetic test for drug selection and/or dosing, given that the behavioral impact of clinical use of pharmacogenetics is largely unknown. Additional research is needed on cost-effectiveness and comparative clinical effectiveness of personalized smoking cessation treatment.

Based on the effect size, replication of association with treatment response and by meeting the statistical criterion for intention to analyse, the *COMT* rs4680 polymorphism would appear to have high analytic validity. However, without prospective treatment studies examining the efficacy of genetically tailored treatment informed by this polymorphism, its clinical validity and clinical utility cannot as yet be assessed. The ethical justification for introducing smoking cessation PGx informed by *COMT* rs4680 genotype is limited by virtue of the fact that there is no published evidence that it would improve treatment outcomes. This observation reinforces the need for the next generation of smoking cessation PGx studies to incorporate methods and metrics that will permit evaluation using an ACCE analytic framework.

Study Design

Methodological standards for the next phase of PGx smoking cessation interventions should be set in accordance with lessons learned from the many limitations of first stage of PGx investigations. Future trials should be designed to adequately test interactions between multiple genetic variants and multiple drugs (gene $\times$ gene $\times$ drug) and ranges of drug dosages. As pharmacological mechanisms of current and emerging pharmacotherapies become better understood by convergence of basic research, preclinical studies, and pharmacogenomic investigations, a priori selection of candidate genes should proceed systematically with attention to genes representing pharmacological pathways and putative cellular and biochemical ontologies that mediate drug response. An argument could be made to focus attention on pharmacokinetic genes with well-defined effects of enzymatic activity and pharmacodynamic genes with *in vitro* or *in vivo* evidence of effects on drug or drug receptor binding, signaling, or transport, based on the most robust associations from PGx and pharmacogenomic investigations to date (David & Munafo, 2008; Murphy et al., 2004), to inform specificity and dose selection when genetically tailoring therapies. Future studies

will also need to incorporate clinical settings such as community-based general practice or primary care to maximize generalizability to the majority of patients in populations. Non-European ancestry studies using ancestry-informative markers are needed to better identify informative haplotypes and their impact on drug response, which is an essential step toward ensuring equity in the delivery of personalized medicine for smoking cessation. In addition, until prospective, genetically tailored clinical trials have been conducted, the actual influence of genotype on smoking cessation efficacy cannot be determined.

Clinical Best Practice

Clinical information systems and protocols that support clinician decision making and processes of care for personalized medicine are needed (Kawamoto, Lobach, Willard, & Ginsburg, 2009). Cost effectiveness and comparative effectiveness research must be expanded to convince payers, policy makers, and opinion leaders that funding specific genetic testing for nicotine dependence treatment is a high priority public investment and to compel industry to develop novel genetically tailored therapies (Aspinall & Hamermesh, 2007). There has been controversy among opinion leaders regarding the potential clinical utility of pharmacogenetics for smoking cessation (Berrettini et al., 2004; Bierut et al., 2007; Burke & Psaty, 2007; Merikangas & Risch, 2003), and more work needs to be done to translate promising results in terms of replication and cost-effectiveness to even more compelling and clinically applicable data for diffusion into policy and practice.

The clinician workforce needs to be trained in both communication skills and medical indications for genetic testing in the treatment of nicotine dependence. Moreover, health care organizations and patients need to be educated to promote genetic literacy and ensure protection of patient confidentiality and high standards for clinical benchmarks to evaluate clinician performance. Finally, the care of patients in provision of personalized smoking cessation treatment must be patient centered and informed by local knowledge and clinical competency. Patients' ideas, concerns, and expectations should be sought in a shared decision making model (Kasper, Mulley, & Wennberg, 1992) to maximize the potential for positive health outcomes and minimize the risk of harm. Genetic determinism or fatalism, if such ideations are present in patients, has the potential to result in psychological harm and/or higher risk of treatment nonadherence (Marteau & Lerman, 2001). In addition, patients' genetic literacy should be assessed and appropriate education provided such that true informed consent in genetic tailoring of treatment is possible (Roter, Erby, Larson, & Ellington, 2009).

Future Directions

The GPRG has more recently participated in collaborative efforts with other research groups providing DNA for genome-wide association studies (Uhl et al., 2008). Pharmacogenetic investigations of non-nicotine pharmacotherapies such as bupropion, nortriptyline, and varenicline in primary care and general practice settings are in various stages of completion by GPRG members in the United Kingdom and the United States. Cost-effectiveness analyses (Welton, Johnstone, David, & Munafò, 2008) and meta-analyses (Munafò, Clark, Johnstone, Murphy, & Walton, 2004; Munafò, Timpson, David, Ebrahim, & Lawlor, 2009; Munafò, Zetteler, et al., 2007) of smoking cessation candidate genes have been published and the range of candidate genes for these dissemination tools will be expanded in future publications. Moreover, DNA will continue to be avail-

able as a resource for future investigations and extant data for meta-analyses, cost-effectiveness studies, and other secondary analyses. In keeping with the ultimate goal of the GPRG's decade of PGx research, our investigators are using careful, mixed-methods approaches to dissemination into clinical practice. One prospective PGx clinical trial of NRT has been completed in the United Kingdom (Marteau et al., 2010) and a second, genetic tailoring trial of NRT or bupropion in the United States (David, 2010) by GPRG investigators and trans-Atlantic collaborators.

The field of smoking cessation PGx is in its adolescence but still has a few years of difficult growing pains before it matures to the point of enhancing the state of the science of smoking cessation in general practice and primary care. However, the corpus of primary care-based research conducted by the GPRG contributed to a strong foundation for this ambitious yet vitally important element of promoting public health.

Supplementary Material

Supplementary Tables 1 and 2 can be found online at <http://www.ntr.oxfordjournals.org>

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Declaration of Interests

Paul Aveyard has done consultancy for McNeil, Pfizer, and Celtic Biotechnology and Sean David has done consultancy with Pfizer—both with regard to smoking cessation.

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