TITLE

Risk-conferring *HLA* variants in an epilepsy cohort: benefits of multifaceted use of whole genome sequencing in clinical practice

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AFILLIATIONS

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ABSTRACT

Background: Whole genome sequencing is increasingly used in healthcare, particularly for diagnostics. However, its clinically multifaceted potential for individually-customised diagnostic and therapeutic care remains largely unexploited. We utilised existing whole genome sequencing data to screen for pharmacogenomic risk factors related to antiseizure medication-induced cutaneous adverse drug reactions (cADRs), such as *HLA-B*15:02*, *HLA-A*31:01* variants.

Methods: Genotyping results, generated from the Genomics England UK 100,000 Genomes Project primarily for identification of disease-causing variants, were used to additionally screen for relevant *HLA* variants and other pharmacogenomic variants. Medical records were retrospectively reviewed for clinical and cADR phenotypes for *HLA* variant carriers. Descriptive statistics and the chi square test were used to analyse phenotype/genotype data for *HLA* carriers and compare frequencies of additional pharmacogenomic variants between *HLA* carriers with and without cADRs, respectively.

Results: 1043 people with epilepsy were included. Four *HLA-B*15:02* and 86 *HLA-A*31:01* carriers were identified. One out of the four identified *HLA-B*15:02* carriers had suffered antiseizure medication-induced cADRs; the point prevalence of cADRs was 16.9% for *HLA-A*31:01* carriers of European origin (*n*=46) and 14.4% for *HLA-A*31:01* carriers irrespective of ancestry (*n*=83).

Conclusions: Comprehensive utilisation of genetic data spreads beyond the search for causal variants alone and can be extended to additional clinical benefits such as identifying pharmacogenomic biomarkers which can guide pharmacotherapy for genetically-susceptible individuals.

INTRODUCTION

The clinical utility of genomics is increasingly recognized: whole genome sequencing (WGS) has been introduced as a first-line diagnostic genetic test in the UK's National Health System [1] and elsewhere [2]. WGS reveals the complete genetic code, making it a powerful tool not only for diagnosis but also for informing personalised approaches to genetically-tailored treatment strategies and comorbidity risk stratification. Pharmacogenomics (PGx), the influence of genetic variation on the outcome of drug treatment, is fundamental to personalisation of drug management. Variants in the genes responsible for pharmacokinetics and pharmacodynamics may serve as clinically-actionable variants [3]. Current progress in WGS and advanced bioinformatics approaches provide opportunities for profiling clinically-actionable PGx variants, focused on new treatments but also on response to existing treatments in several fields, including the epilepsies [4].

The merits of WGS in the epilepsies have emerged mainly in diagnostics and have also revealed genetic associations, through single nucleotide polymorphisms or polygenic risk scores, carrying increased risk of comorbid conditions in epilepsy [5, 6], potentially generating individualised information of value to management, and contributing to a broad interpretation of 'precision medicine'. Drug treatment in the epilepsies is characterized by significant inter-individual variability both in terms of efficacy and susceptibility to adverse drug reactions (ADRs); genetic factors contribute to this variability [4]. Most epilepsy PGx studies have focused on the search for genetic predictors of pharmacoresistance or idiosyncratic ADRs.

ADRs are considered a major problem in healthcare. A large prospective study in the UK estimated that up to 6.5% of all hospital admissions are related to ADRs [7]; additionally they account for up to 30% of emergency department visits and hospital admissions [8]. Moreover, ADRs generate substantial healthcare costs. ADRs may add USD$2284-5640 per individual to hospitalisation costs [9], and account for up to 9.5% of all direct healthcare costs in primary, outpatient and hospital care [10]. Cutaneous ADRs (cADRs) are one of the most common types of ADRs and can manifest with a multitude of phenotypes, ranging from mild self-limiting maculopapular eruptions to serious reactions, such as Stevens-Johnson Syndrome (SJS) and toxic epidermal necrolysis (TEN), associated with high rates of morbidity, mortality and long-term disability [11]. They are common with the use of antiseizure medications (ASMs), with an overall incidence of 10% [12]. Aromatic ASMs are amongst the leading culprit drugs, with carbamazepine (CBZ), phenytoin (PHT) and lamotrigine (LTG) considered to be the principal offenders [13]. In addition to direct health consequences, the burden of cADRs on healthcare expenditure is high. The annual direct costs for ASM-induced severe cADRs range between 2,460 to 7,910 Rupees (~27-77USD$) in India [14] and 3,720-8,061 USD$ in Korea, where the authors estimated that the costs are comparable to other major health problems such as chronic renal disease and lung cancer [15]. There are two main categories of risk factors for cADRs: drug-related factors such as drug exposure, duration of exposure and

polytherapy, and host-related factors such as age, gender, concomitant medical conditions, immune system disorders and genetic factors. Human leucocyte antigen (*HLA*) variants are the most well-established genetic markers for predisposition to cADRs, especially with the use of aromatic ASMs [13]. The *HLA-B*15:02* variant is strongly associated with SJS secondary to CBZ or PHT exposure in Asian populations, where the variant has the highest prevalence [16]. The strongest association has been reported in Han Chinese from Taiwan [17] with odds ratio (OR) of 1357 (95% CI: 193.4–8838.3); an association confirmed in Malay [18], Thai [19] and Indian populations [20]. In contrast to the Asian-specific *HLA-B*15:02*, the *HLA-A*31:01* variant shows population variability. Moreover, it predisposes to various cADR phenotypes, including maculopapular exanthema, SJS, TEN and drug reaction with eosinophilia and systemic symptoms (DRESS). A recent meta-analysis investigating this association showed an increased aggregated risk, with OR of 5.92 (95% CI 4.35–8.05) for *HLA-A*31:01*-mediated cADRs [21], corroborating the results of previous meta-analyses [22,23].

As WGS gains ground in healthcare settings, the potential for mining relevant information which spreads beyond diagnostics, such as PGx variants, is significant. Its clinical value remains largely unexplored, as transformation of such information into clinical tools has been complex and slow. Nonetheless, utilising WGS data comprehensively can add layers of clinically meaningful information, with the prospect of more individually-customised diagnostic and therapeutic clinical care. Here we utilised existing WGS data, generated from the Genomics England UK 100,000 Genomes Project (GEL) to identify causal variants, to additionally screen for *HLA-B*15:02* and *HLA-A*31:01* variants, in a cohort of people with epilepsy from a single tertiary epilepsy centre. We reviewed the medical records for drug history and occurrence of aromatic ASM-induced cADRs. As a secondary aim, we screened additional PGx variants associated with the pharmacokinetics of aromatic ASMs to investigate the modulatory effects of genetics on ASM-induced cADRs in individuals carrying *HLA-B*15:02* or *HLA-A*31:01* variants.

METHODS

Study Cohorts

Individuals with a diagnosis of epilepsy from a single tertiary centre (National Hospital for Neurology and Neurosurgery, University College London NHS Foundation Trust), for whom WGS was performed through GEL, were included in the study. Unrelated individuals with non-neurological rare diseases and no personal diagnosis of epilepsy recruited to GEL were used as controls for comparison of *HLA* variant frequencies.

We defined our cohort of interest based on *HLA* carrier status. We used this cohort to conduct reverse phenotyping and analysis of additional PGx variants.

*HLA-A*31:01* and *HLA-B*15:02* Variant Genotyping

We used *HLA* genotyping results, already available within the GEL Research Environment, to screen for *HLA-B*15:02* and *HLA-A*31:01* variants. The proxy polymorphisms rs10484555 and

rs1061235 were used to confirm the presence of *HLA-B*15:02* and *HLA-A*31:01* respectively [24, 25] and were examined in the Integrative Genomics Viewer (IGV) tool [26].

We selected further genes and single nucleotide polymorphisms reported to be associated with drug and/or metabolite level or maintenance dosing of aromatic ASMs [27-30]. Genotype results of 13 PGx variants involving 8 genes (*CYP2C19* [rs192154563], *CYP3A4* [rs2242480], *EPHX1* [rs2234922; rs1051740], *UGT1A4* [rs2011425; rs6755571], *UGT2B7* [rs7668258; rs7439366], *ABCB1* [rs1045642], *ABCC2* [rs3740066; rs2273697; rs717620], *SCN1A* [rs3812718]) were extracted from WGS data of all *HLA-B*15:02* and/or *HLA-A*31:01* carriers.

Phenotypic Information

Demographic and clinical variables for the *HLA* carriers were obtained from medical records and included age, ancestry, type of epilepsy [31], drug history specifically including exposure to aromatic ASMs (CBZ, PHT, LTG, oxcarbazepine (OXC), eslicarbazepine (ESL)), and presence of cADRs including rash (maculopapular or unspecified), SJS, TEN and DRESS. Clinical variables to explore systemic involvement in conjunction with cADRs other than the previously described syndromes were also collected where available, including presence of fever, liver or haematological abnormalities. For *HLA-A*31:01* and *HLA-B*15:02* carriers with ASM-induced cADRs, additional phenotypic features were included as relevant risk factors, where available, comprising age at time of exposure to culprit ASM, duration of exposure to culprit ASM, monotherapy/polytherapy at time of cADR (including ASM and non-ASM medications if on polytherapy), allergy to non-ASMs and presence and type of comorbidities.

Statistical Analyses

Descriptive statistics were employed to analyse phenotypic data as well as genotype data for *HLA-B*15:02* and/or *HLA-A*31:01* and the 13 additional PGx variants. The chi squared test was used to compare the prevalence of *HLA-A*31:01* between people with epilepsy and controls, and to compare the frequency of 13 PGx variants in *HLA-A*31:01* carriers of European ancestry with or without ASM-induced cADRs. Bonferroni correction was applied to *P*-values to correct for multiple testing.

RESULTS

A total of 1043 individuals with epilepsy were included (558 female, 53.5%). The control cohort included 1187 individuals (722 female, 60.8%). We identified 86 individuals with epilepsy and 76 controls who carried the *HLA-A*31:01* variant. Four individuals with epilepsy, and no controls, carried the *HLA-B*15:02* variant.

All *HLA-A*31:01* and *HLA-B*15:02* carriers were heterozygotes. The IGV analysis confirmed the presence of both proxies (rs1061235 for all 86 *HLA-A*31:01* carriers and rs10484555 variant for all four *HLA-B*15:02* carriers). Comparison between the individuals with epilepsy and controls of European ancestry showed no significant difference in the *HLA-A*31:01* allele

frequency (AF) distribution (AF: 0.036 vs 0.032 respectively, *P*=0.47). Formal statistical comparison for the *HLA-B*15:02* cohort was not undertaken given the very small sample size.

The clinical characteristics of *HLA-A*31:01* and *HLA-B*15:02* carriers with epilepsy are summarised in Table 1. 62/86 *HLA-A*31:01* carriers (72.1%) were Europeans. Most *HLA-A*31:01* carriers had been exposed to at-risk aromatic ASMs (83/86, 96.5%); among those, 68/83 (82%) were exposed to CBZ alone or in combination with other aromatic or non-aromatic ASMs. Twelve out of 83 *HLA-A*31:01* carriers had developed cADRs, yielding a point prevalence of 14.4%.

177 **Table 1.** Demographic and Clinical Characteristics of *HLA-A*31:01* and *HLA-B*15:02* carriers
178 with epilepsy

	HLA-A*31:01	**HLA-B*15:02**
HLA carriers; N	86	4
Age (in years) [a]	48.1 (SD 14.4)	49.2 (SD 15.1)
Sex; N: Female/Male	41/45	2/2
Ancestry; N (%) [b]		
European/White background	62 (72.1)	0
Black background	3 (3.5)	0
Black African	2 (2.3)	0
Black Caribbean	1 (1.1)	0
Asian background	12 (13.9)	3 (75)
Asian/Indian	8 (9.3)	1 (25)
Asian/Bangladeshi	2 (2.3)	1 (25)
Asian/Pakistani	2 (2.3)	0
Asian/Chinese	0	1 (25)
Mixed background	4 (4.7)	1 (25)
Background not specified	5 (5.8)	0
Epilepsy type; N (%)		
Focal epilepsy	78 (90.7)	3 (75)
Generalised epilepsy	5 (5.8)	1 (25)
Unknown	3 (3.5)	0
Exposure to aromatic ASMs; N (%)	83 (96.5)	4 (100)
cADR Phenotypes; N (%) [c]		
Rash alone	10 (12)	1 (25)
Or with		
Systemic involvement		
Fever	1 (1.2)	0
Liver abnormalities	0	0
Haematological	0	0
abnormalities	0	0
Other organ involvement		
SJS	1 (1.2)	0
TEN	0	0
DRESS	0	0

Abbreviations: ASMs (antiseizure medications), SJS (Stevens-Johnson Syndrome), TEN (Toxic Epidermal Necrolysis), DRESS (drug reaction with eosinophilia and systemic symptoms)

[a]Age (in years) at the time of inclusion to the study: mean and standard deviation (SD).

[b]Ancestry information based on self-report as documented in electronic health records.

[c]Percentages for cADR phenotypes were calculated using only the number of HLA carriers exposed to aromatic ASMs, rather than all HLA carriers, as denominator: n=83 for HLA-A*31:01 carriers and n=4 for HLA-B*15:02 carriers).

Demographic and phenotypic characteristics of *HLA-A*31:01* and *HLA-B*15:02* carriers with aromatic ASM-induced cADRs are listed in Table 2. All 12 *HLA-A*31:01* carriers with cADRs had been exposed to CBZ. The majority of them (10/12, 83.3%) had a cADR as a result of CBZ exposure (rash; *n*=8, rash with fever; *n*=1 and SJS; *n*=1), whereas 2/12 (16.7%) had a cADR secondary to LTG exposure but not to CBZ. In two individuals with CBZ-induced cADRs (patients #3 and #7), the same type of cADR (rash) was reported additionally with the separate use of PHT or OXC, respectively. The majority of *HLA-A*31:01* carriers were on monotherapy at the time of reported cADRs; 7/12 (53.8%). Only 2/12 (16.7%) had comorbidities at the time of reported cADR.

For 7/12 (58.3%) *HLA-A*31:01* carriers who had developed a cADR on exposure to an at-risk ASM, no allergy information was stated in the dedicated allergy section of their electronic or previous paper health records, even after the documented occurrence of cADR.

Table 2. Characteristics of *HLA-A*31:01* and *HLA-B*15:02* carriers with ASM-induced cADRs

ID	Gender	Age[a]	Ancestry	cADR phenotype	Culprit ASM(s)[b]	Treatment duration (ASM)[c]	Monotherapy/ Polytherapy[d]	Aromatic ASM with no cADRs (If any)	Allergy to other ASMs	Comorbidities/ Concomitant non-ASMs[d]
HLA-A*31:01										
1[e]	F	20-29	European	Rash	CBZ	4 weeks	Monotherapy	LTG	No	No / No
2[e]	M	50-59	Asian/Indian	Rash	LTG	3 months	Polytherapy (with CLB)	CBZ	No	Psychiatric, Cardiovascular / Yes
3[e]	F	20-29	European	Rash	CBZ PHT	NA	NA NA	OXC LTG	No	No / No
4[e]	F	40-49	European	Rash	CBZ	NA	Monotherapy	LTG	No	No / No
5	M	40-49	Not specified	Rash	CBZ	1 month	Monotherapy	LTG	No	No / No
6[e]	M	20-29	European	Rash	CBZ	NA	Monotherapy	LTG	No	No / No
7[e]	F	10-19 (CBZ, OXC)	European	Rash and fever	CBZ OXC	NA	NA NA	LTG PHT	No	No / No
8	M	10-19	European	Rash	CBZ	NA	Monotherapy	LTG PHT	No	Endocrinological / Yes
9[e]	F	40-49	European	Rash	CBZ	NA	NA	LTG	No	No / No
10	F	10-19	European	SJS	CBZ	NA	Monotherapy	LTG PHT	Contrast media	No / No

11	F	<10	European	Rash	CBZ	3 months	Monotherapy	LTG PHT	Co-trimoxazole	No / No
12	F	20-29	European	Rash	LTG	3 weeks	NA	CBZ PHT	No	No / No
						HLA-B*15:02				
13	F	10-19	Asian/Indian	Rash	CBZ	NA	Monotherapy	OXC	CLB	No/No
		10-19			PHT	NA	Monotherapy		CLZ	
		10-19			PB	1 week	Monotherapy			
		30-39			LTG	3 weeks	Monotherapy			

Abbreviations: cADR(s): cutaneous adverse reaction(s), ASM(s): antiseizure medication(s), CBZ: carbamazepine, LTG: lamotrigine, OXC: oxcarbazepine, PHT: phenytoin, PB: phenobarbital, CLB: clobazam, CLZ: clonazepam, SJS: Stevens- Johnson Syndrome, NA: information not available

[a]Age (decadal range, in years) at time of exposure to culprit ASM.

[b]If more than one ASM: each ASM caused a cADR separately.

[c]Duration of treatment with culprit aromatic ASM.

[d]At the time of cADR.

[e]Individuals in whom no allergy information was found in electronic health records.

European- and CBZ-specific Cohort

The majority of *HLA-A*31:01* carriers of European ancestry (59/62, 95.2%) had been exposed to at-risk aromatic ASMs, with reported cADRs in 10/59, yielding a point prevalence of 16.9%. Most of the European *HLA-A*31:01* carriers (46/59, 77.9%), had been exposed to CBZ alone or in combination with other aromatic or non-aromatic ASMs. 9/46 (19.6%) had CBZ-induced cADRs, whereas 37/46 (80.4%) did not (CBZ-tolerant). The CT genotype of the *ABCC2*: c.3972C>T variant (RR=7.73; 95% CI=1.04-57.24; unadjusted-*P*=0.011) was the only nominally-enriched variant in European *HLA-A*31:01* carriers with CBZ-induced cADRs (77.78%) compared to the CBZ-tolerant carriers (32.3%), when comparing the 13 additional PGx variants (Supplementary Table 1) , but significance was not sustained after correcting for multiple comparisons. Analysis of the additional PGx variants was not conducted in other ancestry-specific groups due the small sample size.

DISCUSSION

In this retrospective study, we utilised existing WGS data from a cohort of people with epilepsy, which were primarily produced for diagnostic purposes, to look for well-established PGx markers of ASM-induced cADRs. Out of 1043 individuals with epilepsy, we identified 86 carriers of *HLA-A*31:01* and four carriers of *HLA-B*15:02* variants. *HLA-A*31:01* prevalence varies across populations, with the highest prevalence in populations of south and central America (ranging from 2.6% to 28.8%) [22], variable frequencies across other populations such as Europeans (1-5.9%), Japanese (7-11%), south Koreans (5.6%) and with the lowest prevalence in African populations (0.4–1.1%) [21]. In our cohort, *HLA-A*31:01* carriers were of different ancestral backgrounds, with the majority being of European descent (72.1%), most likely reflecting the make-up of individuals seen at our centre. In contrast, the prevalence of *HLA-B*15:02* is highest in Asian populations, found in up to 6.9% and 8% of East and Southeast Asians respectively, whereas it is rare in Europeans, South American or African populations (<1%) [16]. All four *HLA-B*15:02* carriers in our cohort were of Asian/mixed ancestry. We did not expect our epilepsy cohort to be enriched or protected with respect to the *HLA* variants as selection (natural, clinical or ADR-related) of participants was not based on these variants. Indeed, on formal comparison there was no significant difference in the *HLA-A*31:01* distribution between individuals with epilepsy and controls of European ancestry (*P* =0.42). Formal statistical comparison was not undertaken for the *HLA-B*15:02* cohort given the very small sample size.

Our cohort was selected based on available genetic data, and irrespective of the presence of cADRs. We performed reverse phenotyping, using the presence of the risk-conferring *HLA* variants as a starting point to identify genetically-susceptible individuals and then search for cADR phenotypes from medical records. We identified a cADR point prevalence of 14.4% for all *HLA-A*31:01* carriers exposed to aromatic ASMs irrespective of ancestry (*n*=83) and 16.9% for European *HLA-A*31:01* carriers (*n*=46). The majority of the carriers on aromatic ASMs,

68/83 (82%), were exposed to CBZ monotherapy or polytherapy. *HLA-A*31:01* risk association is well-established for CBZ and OXC-induced cADRs [22]; data are less robust for PHT or LTG-induced cADRs [32]. 10/12 (83.3%) *HLA-A*31:01* carriers who had developed any cADR, did so with exposure to CBZ. The single *HLA-B*15:02* carrier had suffered recurrent cADRs after exposure to several aromatic ASMs, despite clinical evidence of previous cADRs. Had the information about PGx risk been available earlier, repeated trials of aromatic, potentially offending, ASMs may have been avoided. This reverse phenotyping approach differs from other studies investigating the *HLA* association, which start with the phenotype (of cADRs) and therefore formal comparison of our prevalence results with other studies is difficult. Defining the prevalence of cADRs in *HLA* carriers could help understand why only a proportion of carriers develop cADRs.

Besides the *HLA-A*31:01* and *HLA-B*15:02* association with cADRs, emerging data have highlighted the potential role of additional PGx variants. Combining different PGx biomarkers can shed light on the nuances of inter-individual variability related to cADR susceptibility. In a recent study by Mullan et al. [33], five novel genetic variants were found to increase the risk of cADRs irrespective of *HLA* carrier status, in Chinese individuals with epilepsy. Moreover, in the same study, a non-protein coding variant was shown to reduce the risk of CBZ-induced cADRs in *HLA-B*15:02* carriers. Here, we sought additional PGx variants to explore why some of the *HLA-B*15:02* and/or *HLA-A*31:01* carriers developed or did not develop aromatic ASM-induced cADRs. We identified an enrichment of the *ABCC2*:c.3972C>T variant in *HLA-A*31:01* carriers with CBZ-induced cADRs compared to CBZ-tolerant carriers, which was not significant after correcting for multiple comparisons. Variants in the *ABCC2* gene have been reported to affect CBZ pharmacokinetics, potentially contributing to inter-individual variability in response to CBZ treatment. The presence of the *ABCC2*:c.3972C>T variant has been associated with higher serum levels of CBZ 10,11-epoxide, the active CBZ metabolite, and the need for higher CBZ maintenance doses in Caucasian and Han Chinese individuals with epilepsy [34], whereas it was not reported to affect CBZ clearance in an epilepsy cohort from the UK, for which ancestry was not specified [27]. Further work is needed in this area.

In none of the individuals with cADRs had post-cADR genetic screening for *HLA* carrier status been undertaken. Moreover, for 7/12 (53.8%) *HLA-A*31:01* and the single *HLA-B*15:02* carriers with reported cADRs, no relevant information was available in the dedicated allergy section of their medical records. Proper documentation of drug hypersensitivity in electronic health records is of crucial importance as it not only supports more informed treatment strategies, but it may also prevent re-administration of the offending medication. Re-administration of cADR-provoking drugs has been reported in 27% of patients who have previously suffered ADRs in the hospital setting, solely due to insufficient documentation [35]. Following our findings, allergy information was updated in the electronic health records of the individuals with clinically-established cADRs stating the offending medication.

As genetic information is becoming increasingly available in the clinical setting, identification of clinically relevant PGx variants will accelerate. In our study, identification of *HLA* variants in individuals with epilepsy became available *en passant*, and indeed revealed 90 *HLA* variant carriers; 14.4% had already suffered a relevant cADR. Had this genetic information been readily available to the treating clinical teams, it could translate to potential treatment benefits, as it could assist clinicians adopt a more informed approach on treatment selection and ensure more rigorous monitoring for genetically-susceptible individuals. Notwithstanding the potential clinical benefits for our cohort, the prospect of large-scale implementation of such PGx screening in the clinical setting could help prevent cADRs by identifying genetically-susceptible individuals and thus could facilitate more effective mitigation strategies for cADR-related adverse outcomes and related healthcare costs. A recent study comparing the cost-effectiveness between PGx-guided (including studies on PGx-guided CBZ treatment) and conventional pharmacological treatment based on published economic evaluations estimated that, sequencing and genotyping costs aside, 75% of economic evaluations would support PGx-guided treatment, with 50% of these considering it the preferred, and even cost-saving, treatment strategy [36]. As genetic testing costs decrease and data become more widely-available, incorporating genetic information for all individuals as part of their healthcare may become a realistic prospect. Where WGS has been undertaken to search for a cause, PGx data may be available effectively for free.

However, we also note that for *HLA* variants, there is a translational gap that hinders large-scale clinical implementation mainly attributed to economic barriers, conflicting data on cost-effectiveness, test availability and the variable predictive values emerging from studies investigating *HLA* associations with cADRs [13]. There have been considerable advances in this context. *HLA-B*15:02* screening before initiation of CBZ treatment has been proven to be cost-effective for several Asian populations [37]. *HLA-B*15:02* pre-emptive screening prior to CBZ treatment is a requirement for Asians according to the FDA and is routinely conducted in Thailand's national healthcare system [38]. Screening for *HLA-B*15:02* variants irrespective of ancestry may double the number of identified at-risk individuals compared to screening individuals of Asian ancestry alone [39]. Nonetheless, its application in healthcare systems remains limited, even for Asian populations. In the UK although the risk association with *HLA-B*15:02* and cADRs is acknowledged, *HLA-B*15:02* screening, even for Asian CBZ-naive individuals, is only stated as a recommendation and not a requirement [40]. Although highly likely to be so at a global level [37], the cost-effectiveness of routine screening for *HLA-A*31:01* has not yet been established. Nevertheless, official recommendations in the UK state that if people or European or Japanese ancestry are known carriers of the *HLA-A*31:01*, the use of CBZ should be considered only if the benefits outweigh the risks [40].

Our study had a number of limitations. Given the retrospective study design, phenotypes of cADRs could not be assessed based on established diagnostic criteria but rather based on the relevant documentation available in medical records, mainly because access to the

contemporary primary clinical data was not possible. Therefore, further classification of the reported rashes was generally not possible. In addition, we were unable to collect and analyse data on serum concentrations for either the parent drug or drug metabolites, which could be valuable parameters in investigating the lack of cADRs in the majority of *HLA* carriers. Moreover, despite the clinical relevance and potential clinical utility of our PGx findings, we were unable to implement them into the clinical setting for our cohort, such as incorporating the HLA carrier status along with the relevant guidance into the medical records, mainly due to lack of clear mechanisms within the local national health system for streamlining clinical testing of PGx markers and incorporating them into clinical/treatment algorithms (the tests were unavailable on the UK National Genomic Test Directory).

In conclusion, we emphasise that comprehensive utilisation of WGS data spreads beyond the search for causal variants alone and can be extended to additional clinical benefits such as identifying PGx markers, which can guide pharmacotherapy for genetically-susceptible individuals who are at risk for ASM-induced cADRs, some of which are life-threatening. As WGS is entering health systems as a routine test, PGx data will become increasingly accessible to clinicians with the realistic prospect of a meaningful multifaceted shift towards precision medicine. Nevertheless, as highlighted by the limitations in our study, allied work between the research community, regulatory agencies and national health systems is necessary for the successful incorporation of PGx procedures into medical practice, which could translate into meaningful benefits for quality of care, healthcare costs and optimisation of pharmacotherapy.

CONTRIBUTORS

AV and RB contributed equally to this paper. AV, RB contributed to study design, data collection, data analysis, data interpretation and writing. MG and HMC contributed to data interpretation and writing. SB contributed to critical revision of the manuscript. SMS contributed to study design and critical revision of the manuscript.

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COMPETING INTERESTS

None of the authors has any conflict of interest to disclose.

ETHICS APPROVAL

This study was approved by the Camden & Kings Cross Research Ethics Committee (reference 11/LO/2016, under which approval all phenotypic information was collected). Analysis of genotypic information was conducted within the Research Environment of Genomics England UK and export of genotypic information was done in accordance with Genomics England UK 100,000 Genomes Project Ethics Protocols. Further information can be found in supplementary material.

DATA AVAILABILITY STATEMENT

Access to genetics data of the 100,000 Genomes Project may be obtained via membership of the Genomics England Clinical Interpretation Partnership (GeCIP; https://www. genomicsengland.co.uk/about-gecip/joining-research- community/). Data are available on reasonable request. The authors confirm that the data supporting the findings of this study are available from the corresponding author, on reasonable request and subject to protocol approvals.

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