UK NEQAS
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Histocompatibility & Immunogenetics

Results from the UK NEQAS for H&I Scheme 8: HLA Genotyping for Coeliac and Other HLA Associated Diseases

Amy De'Ath and Tracey Rees

Introduction

UK NEQAS for H&I's scheme 8 HLA Genotyping for Coeliac and Other HLA Associated Diseases assesses participants' ability to HLA type samples to aid in the diagnosis of Class I and Class II HLA associated diseases such as Coeliac Disease and Birdshot Retinopathy.

10 blood samples are distributed each year in 2 distributions of 5 samples. Participants can register for a number of HLA associated diseases and are required to report their HLA genotype findings for each disease they have registered for, using the correct HLA nomenclature. Assessment is made against a reference type allowing labs to report at a resolution that is applicable to the needs of their clinical users. Laboratories can also report interpretative comments although these are not currently assessed.

Satisfactory performance is obtaining 10 sample in agreement with the reference genotype in a year for each disease registered for.

Participation

In 2018, 10 samples were distributed. During this time 52 labs participated, 10 from the UK and Ireland (UK&I) and 42 from the rest of the world (RoW). Table 1 shows a breakdown of Participants registered for each disease and the number of labs with unacceptable performance. Performance in HLA typing for Coeliac Disease was a particular concern and investigated further.

Disease	HLA Association	Number of Participants	No. of Participants with Unacceptable Performance
Coeliac	DQ2, DQ8, DQA	50	11
Narcolepsy	DQB1*06:02	21	3
Actinic Prurigo	DRB1*04:07	4	1
Birdshot Retinopathy	A*29	9	1
Behçet's	B*51	6	0
Rheumatoid Arthritis	DRB1*04	2	0
Diabetes	DR3, DR4	4	1

Table 1: Breakdown of Participant Numbers and Performance in Scheme 8

Results

22% (11/50) of labs in Coeliac Disease category reported an incorrect result during the 2018 distribution year. Of these labs only one participated solely in Scheme 8 suggesting this UK lab may have a limited H&I remit. In total 17 errors were made (4 by UK based labs and 13 in the rest of the world): 7 labs made 1 error, 2 labs made 2 errors and 2 labs made 3 errors. Only 2 of the 10 samples distributed were reported correctly by all participants.

The 17 errors made could be grouped into three broad categories (see Table 2) depending on the type of error made:

- 7/17 41% - Incorrect HLA Type 1st field
- 6/17 35% - Incorrect HLA Type 2nd field
- 4/17 24% - Multiple Errors in HLA Type

Laboratories with unacceptable performance were asked to submit a corrective and preventative action investigation. Upon review of the responses it was found the errors were caused by a number of issues.

Error Type	No. of Errors (n=17)	Example Error	
		Consensus Type	Reported Error
Incorrect HLA Type 1st Field	7 (41%)	DRB1*04:01, 07:01/34/72; DQB1*02:02, 03:01; DQA1*02:01, 03:03	Negative for ***DQ2*** and DQ8
Incorrect HLA Type 2nd Field	6 (35%)	DRB1*03:01/124/132/137, 04:08; DQB1*02:01, 03:01; DQA1*03:03, 05:01	DRB1*03, DRB1*04, ***DQA1*03:02***, DQA1*05:01, DQB1*02:01, DQB1*03:01
Multiple Errors in HLA Type	4 (24%)	DRB1*04:01, 07:01; DQB1*02:02, 03:02; DQA1*02:01, 03:01	***DQ2: NEG DQ8: POS (A1*05: NEG, B1*02: POS, B1*0302: POS)***

Table 2: Type and Examples of Errors Made in Reporting for Coeliac Disease

The errors were as a result of a sample handling or technical error (2 labs (18%)), a misinterpretation of a correct HLA type (3 labs (27%)), ambiguous kit results or resolution issue (4 labs (37%)), 2 labs (18%) did not respond.

Many of the errors were a result of the misinterpretation of a correct HLA type. Many labs struggled with understanding DQB and DQA subunits and their association. For example, a lab noted that their current guidelines are *"to not report DQ2.2 as DQ2 positive but to report it as "DQ2 negative DQB1*02 positive" and that risk of coeliac disease cannot be excluded based on genotype."* Likewise, a further lab reported that DQ2 was present or "Half DQ2 positive" when they had detected HLA-DQB1*03:01, DQA1*05:05 as they wanted to report they had detected the DQA1*05 subunit.

Also, these investigations have highlighted some deficiencies in the resolution of the HLA type that some commercial kits can detect and their reporting capability in terms of interpretation of the result achieved. Table 3 shows the insert from a commercial kit in relation to result interpretation and how misleading it can be (yellow highlight) especially for labs with limited H&I experience.

Signal	Mix DQA1*05	Mix DQB1*02	Mix DQB1*03:02	HLA-Genotype
	-	-	+	DQ8
	-	+	+	DQ8
	+	-	+	DQ8
	+	+	+	DQ2 and DQ8
	+	+	-	DQ2
	+	-	-	A genetic predisposition for Coeliac Disease is unlikely
	-	+	-	
	-	-	-	

Table 3: Example of a Commercial Kit Package Insert Result Interpretation Section

Comment

It is important that laboratories are able to perform accurate HLA typing to support the diagnosis of HLA associated diseases. The relatively low overall rate of errors is encouraging, however, further work is required to eliminate errors that could impact on patient care.

UK NEQAS for H&I have contacted manufacturers to address the issues identified. The UK NEQAS for H&I Steering Committee are in the process of collating some guidelines in collaboration with BSHI and the British Society of Gastroenterology on reporting for Coeliac disease and stratifying risk to aid participants and standardise reporting.

Further Information

Full information on all UK NEQAS for H&I schemes is available at **www.neqashandi.org** or contact **ukneqashandi@wales.nhs.uk** .

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