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Commentary

Differences in common heritable blood immune cell populations may underlie MS susceptibility and progression

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Abstract

A promising new avenue of MS research that may lead to a better understanding of pathogenesis, progression and therapeutic response, and to development of new therapies, comes from the recent identification of defined immune cell populations that are highly heritable. Such stable populations have been identified in three recent papers using extensive flow cytometric panels to investigate twin and family cohorts. They showed that while most of the variation in immune cell populations between individuals was not heritable, some was. This heritability was sometimes very high, and the authors concluded that it likely contributes to variability in response among individuals for disease and drug response traits.

***Keywords*:** Multiple sclerosis, MS risk gene, heritability

Multiple sclerosis (MS) susceptibility is known to be affected by genetic factors, many now known from genome-wide association studies[1] and associated with environmental factors such as latitude of childhood, serum vitamin D levels, Epstein–Barr virus (EBV) infection, salt intake, and smoking.[2] However, the events initiating disease and controlling progression remain elusive.

Modern therapies have greatly improved in efficacy, although with increased adverse reactions.[3] They all target the immune cells of blood, indicating these cells are important in disease activity, even though the cellular focus of the autoimmune response is in the central nervous system, behind the blood-brain barrier. The therapies have different immune cell molecular ligands, and are effective for only a proportion of individuals, suggesting the immune cell dysregulation driving MS may vary among individuals. Initial choice of therapy may be crucial, since any neuronal damage in the time taken to institute effective therapeutic intervention may be irreversible.[4] Minimising this time is therefore a major goal of MS research. Currently no biomarkers are available to indicate which therapy is likely to be successful for individual patients, or cause adverse reactions; and no therapy provides a cure for all patients. To date, no immune cell biomarker of pathogenesis or drug response has been demonstrated unequivocally. However, there is evidence that regulatory T (Treg) cells[5] and natural killer (NK) cells[6] are under-represented in MS, and that MS risk genes such as *eomesodermin* (*EOMES*) and zinc finger, MIZ-type containing 1 (*ZMIZ1*) tag immune cell population differences that may drive disease.[7]

A promising new avenue of MS research that may lead to a better understanding of pathogenesis, progression and therapeutic response, and to development of new therapies, comes from the recent identification of defined immune cell populations that are highly heritable. Such stable populations have been identified in three recent papers by Orru et al.,[8] Roederer et al.[9] and Brodin et al.[10] using extensive flow cytometric panels to investigate twin and family cohorts. They based their studies on cluster of differentiation (CD) markers, which are proteins presented on the surface of immune and other cell types, recognised by antigens tagged to fluors or metal ions. They showed that while most of the variation in immune cell populations among individuals was not heritable, some was. This heritability was sometimes very high, and the authors

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concluded that it likely contributes to variability in response among individuals for disease and drug response traits. Also notable was the finding that the difference in response of immune cell subsets to cytokines was mostly affected by non-heritable factors, except for interleukin (IL)7 and IL2 of the cytokines tested. Polymorphisms in the genes for IL7 and IL2 and/or their receptors are MS genetic risk factors.[11] These genes, encoding the CD markers CD127 and CD25, also define heritable immune cell subsets, and there are many other candidate MS risk genes encoding CD markers that potentially define heritable immune cell populations (Table 1). The consilience of association with pathogenesis (risk gene) and their defining of heritable immune cell subsets makes these CD markers good candidates for studying the basis for MS-associated immune cell subset dysregulation and therapeutic response. Note this dysregulation is not likely to be due to single nucleotide polymorphisms (SNPs). These only have a small effect on MS risk, so that it would be unlikely that they would control immune cell subset differences that were themselves large risk factors – rather any difference in cell populations will likely be a net effect of multiple SNPs and other genetic risk factors. In this context, it is notable that the percentage of lymphocytes in peripheral blood expressing CD4 is highly heritable and predicts response to fingolimod.[12] The MS risk genes which encode CD markers are expressed in a range of blood immune cell subsets (Figure 1), not just T cells, consistent with MS susceptibility being due to complex immune system dysregulation.

Table 1. MS risk genes, CD names and known heritability.

MS risk gene	CD	Known heritable marker
VCAM1	CD106	
TNFRSF1A	CD120a	
IL7R	CD127	Brodin et al.,[10] 2015; Roederer et al.,[9] 2015
CSF2RB	CD131	
CTLA4	CD152	
PVR	CD155	
CXCR4	CD184	
CXCR5	CD185	
CCR4	CD194	
CD226	CD226	
IL2RA	CD25	Brodin et al.,[10] 2015; Roederer et al.,[9] 2015
TNFSF14	CD258	
CD27	CD27	Roederer et al.,[9] 2015
CD28	CD28	Roederer et al.,[9] 2015
FCRL3	CD307c	
CD37	CD37	
CD40	CD40	
CD48	CD48	
CD5	CD5	
CD58	CD58	
CD6	CD6	
CD86	CD86	

MS: multiple sclerosis; CD: cluster of differentiation; Gene symbols are from the HUGO Nomenclature committee. Risk genes are those identified in Beecham et al[11] and papers therein.

The three landmark studies by Orru et al.,[8] Roederer et al.[9] and Brodin et al.[10] have several limitations which may have led the authors to underestimate the degree of heritability of immune cell populations. Firstly, only a subset of known CD markers were tested, based on markers already in use for characterising immune cell subsets. Secondly, as cell subsets are usually defined by marker combinations (e.g. CD25hi CD127low of CD4positive cells for Tregs), and marker panels are of limited size (e.g. typically 8–12 for flow cytometry), the best combinations may not have been tested. Improved marker combinations may be identified with technologies such as time of flight mass spectrometry (CYTOF), which can examine combinations of up to 40 markers in a panel. Thirdly, only proteins expressed on the cell surface (CD markers) were examined – expression of intracellular protein may define important immune cell subsets. The MS risk genes encode proteins that are predominantly located in the intracellular space. Transcription factors which control immune cell lineages are over-represented among the risk factors. We have identified underexpression of MS risk factors which are also transcription factors: *EOMES* and *T-box 21* (*TBX21*); and *ZMIZ1/ZFP36* ring finger protein-like 2 (*ZFP36L2*),[7] and higher expression of a gene activated by the MS risk factor ribosomal protein S6 kinase, 70kDa, polypeptide 1 (*RPS6KB1*).[14] Expression of these genes is stable over time, consistent with their expression being under tight genetic control. Their expression is altered by therapies used in MS. Intracellular proteins can be assayed using flow cytometry if suitable tagging antibodies are known.

Overall, we suggest that highly heritable variance in immune cell populations, particularly variance associated with disease risk genes, contributes to differences in MS susceptibility, progression and response to therapy. As such, measurement of these populations, or more simply the expression of their tagging risk genes in blood, when clearly defined and validated in large clinical studies, is likely to be useful in clinical management of MS.

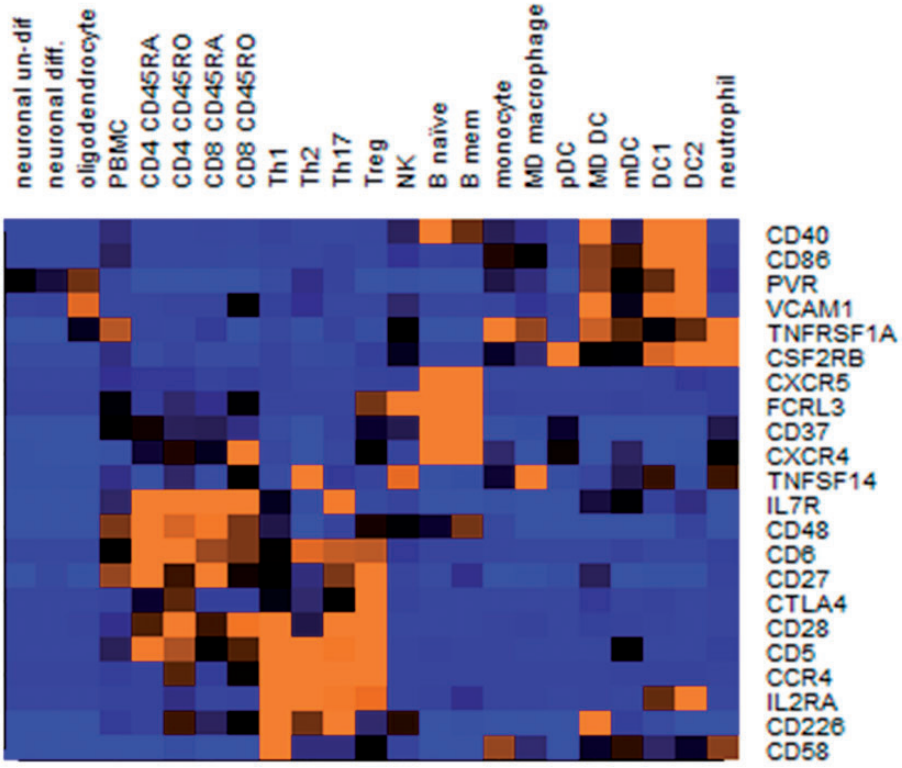

Figure 1. Heat map indicating relative blood immune cell subset expression of MS risk genes from Table 1. High expression is orange, low expression blue. Columns are immune cell subsets, rows MS risk genes. Details of immune cell transcriptome interrogation are in Shahijanian et al., 2014.[13]
MS: multiple sclerosis; PBMC: peripheral blood mononuclear cells; CD: cluster of differentiation; Treg: regulatory T cells; NK: natural killer cells; pDC: plasmacytoid dendritic cells; mDC: myeloid dendritic cells; Gene symbols are as defined by the HUGO Nomenclature Commmittee.

Conflict of interest

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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