REPLY TO HU ET AL.:

On the interpretation of gasdermin-B expression quantitative trait loci data

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We recently published structural and biochemical studies of gasdermin-B (GSDMB) (1), a protein that regulates the maintenance of the epithelial cell barrier as well as cell proliferation and differentiation processes (2, 3). *GSDMB* amplification and GSDMB overexpression lead to a poor response to HER2-targeted therapy in HER2-positive breast cancer (4). We showed that GSDMB binds to sulfatide and phosphoinositides, components of the epithelial cell membrane. Because sulfatide promotes cell migration and metastasis (5), we speculated that GSDMB might be directly or indirectly involved in the transport of sulfatide to the cell membrane. Genome-wide association studies show that the presence of two GSDMB missense SNPs (dbSNP: rs2305479 and dbSNP:rs2305480) correlate with an increased risk of complex trait inflammatory diseases that impair the integrity of the epithelial cell membrane, for example, inflammatory bowel disease (IBD) and asthma (6). We therefore speculated that SNP-related structural changes we observed in the protein domain containing the disease-risk SNPs might affect GSDMB sulfatide binding affinity and, in turn, sulfatide transport, providing a possible disease-related mechanism.

Hu et al. (7) use a number of expression quantitative trait loci (eQTL) datasets to examine the correlation between these complex trait-related *GSDMB* SNPs and the expression levels of *GSDMB* and its neighboring genes, *ORMDL3* and *GSDMA*. They conclude that the disease-risk rs2305479 A and rs2305480 T alleles reduce *GSDMB* and *ORMDL3* expression in human immune cells and whole blood. Therefore, they attribute the disease mechanism to changes in gene expression and not to changes in protein properties. Unfortunately, almost none of the methodology used to reach this conclusion is included in their letter, and no reference to methods is given. The one methodological detail provided, the expression for calculation of the Z-score, is inconsistent with the data (table 1 of ref. 7) they provide (the Z-score is dependent on the effect size, beta, but Z-scores are only reported for SNPs with no beta value). Therefore, we consider that the conclusions of Hu et al. (7) should not be given much weight unless further details are provided.

On the other hand, at least one already published and reliable study, by the Genotype-Tissue Expression (GTEx) consortium (8), has reported a statistically significant correlation between the presence of these two SNPs and GSDMB RNA message abundance in some tissues (GTEx Portal, https://www.gtexportal.org, accessed on July 28, 2017). The reported effect sizes in the small intestine, the tissue most relevant to IBD, are −0.33 for rs2305479 and −0.40 for rs2305480. For the lungs, the most relevant tissue for asthma, the effect size for rs2305479 is smaller, −0.18, and there is no reported correlation for rs2305480.

Statistical significance is not the same as biological significance. The largest effect size, −0.40, corresponds to an ∼25% and ∼40% decrease in expression for heterogeneous and homozygous SNP genotypes, respectively. These expression differences are small, and are also small compared with the range of expression found in individuals with the same genotype (https://www.gtexportal.org/home/eqtls/bySnp?snpId=rs2305480&tissueName=All); that is, the change appears insignificant compared with variation in expression for other reasons. While these data do not rule out an expression-related disease mechanism, they suggest it is unlikely.

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The authors declare no conflict of interest.

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