

Response to: 'Correspondence on 'Machine learning algorithms reveal unique gene expression profiles in muscle biopsies from patients with different types of myositis' by Takanashi *et al*

Thank you for your constructive comments.¹ We agree that transcriptomic data from affected muscle tissue have the potential to improve the diagnosis and treatment of inflammatory myopathies.² First, transcriptomic data may allow us to identify the most relevant inflammatory pathways in a particular patient and thereby individualise therapy. For example, patients with marked upregulation of interferon-induced genes may benefit most from treatment with Janus kinase inhibitors. Second, transcriptomic analysis requires very little muscle tissue while providing a large amount of biological information. Thus, transcriptomic analysis using needle muscle biopsies may be as diagnostically useful as conventional surgical muscle biopsies. Finally, visual interpretation of muscle biopsies is a complicated task that, even when performed by experts, has relatively poor interrater reliability.³ In contrast, the analysis of transcriptomic data is objective and can be automated.

In our study, the presence of interstitial lung disease was almost always present in certain myositis subgroups (anti-MDA5⁴ and anti-Jo1⁵) and almost completely absent in others (anti-Mi2,⁶ anti-NXP2,⁷ anti-TIF1g,⁸ anti-SRP,⁹ anti-HMGCR,¹⁰ IBM¹¹). Thus, the sample size of patients with and without interstitial lung disease within each subgroup was not sufficient to make robust comparisons within subgroups.

We agree that biopsies obtained from different muscles may not have identical histological or transcriptomic features. Indeed, we previously showed that deltoid muscle biopsies tend to have more severe histological abnormalities than muscle biopsies from other locations.¹² In the current study, we used muscle biopsies that were obtained for diagnostic purposes by numerous clinicians at several different hospitals. Thus, individual clinicians made decisions about which muscles to biopsy based on factors including the degree of weakness, electromyographic findings and/or imaging features. Since clinicians at these centres do not perform muscle biopsies in completely amyopathic forms of myositis, the muscle biopsies from anti-MDA5 and

anti-TIF1 γ patients included in our study all came from patients who had muscle weakness.

Although anti-MDA5 and anti-TIF1 γ patients are the patients with least myopathic dermatomyositis, 79% of anti-MDA5 patients and 81% of anti-TIF1 γ patients develop clinically detectable weakness during follow-up. Furthermore, we have shown that the level of expression of interferon-inducible genes in anti-MDA5 and anti-TIF1 γ was equivalent to that in the more myopathic forms of dermatomyositis (ie, those with anti-Mi2 and anti-NXP2 autoantibodies).¹³ Also, we have verified that there is a positive correlation between the expression of the interferon-inducible genes and the level of muscle weakness.¹³ Thus, it will be interesting to know if these inflammatory patterns are still detectable in completely amyopathic patients.

In our recent study, about half of muscle biopsies were from the quadriceps, 1/6 from the biceps and 1/3 from the deltoid. Nonetheless, independent of what muscle was biopsied, the key genes used by the machine learning algorithm to classify the muscle biopsies had similar magnitudes of expression (online supplemental figure 1). While further studies will be needed, this observation suggests that transcriptomic data from any of these affected muscles may be adequate for diagnostic purposes and identifying pathologically relevant pathways. Notwithstanding this, there were striking transcriptomic differences between biopsies obtained from different muscles independent of the myositis subgroup (table 1). Perhaps not surprisingly, the biggest differences were founded in homeobox genes that control morphogenesis. In fact, using the differentially expressed genes between muscle biopsy locations, a linear support vector machine model was able to predict the location of the biopsy with an accuracy of 95% (95% CI 87% to 100%). We hypothesise that the transcriptional differences between muscles may be related to the characteristic patterns of weakness observed in the different types of myositis (eg, inclusion body myositis,¹¹ anti-NXP2⁷ and anti-Pm/Sci¹⁴).

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Table 1 Top 10 differentially expressed genes according to the location of the muscle biopsy independent of the type of myositis

Quadriceps versus biceps			Quadriceps versus deltoid			Deltoid versus biceps		
Gene	log2FC	padj	Gene	log2FC	padj	Gene	log2FC	padj
HOXC8	2.4	4.E-26	HOXC8	2.3	1.E-32	HOXA11-AS	-1.6	6.E-11
POU3F3	-2.5	4.E-21	HOXC9	2.3	1.E-26	HOXA11	-1.6	2.E-10
HOXC4	1.6	1.E-18	HOXD9	-2.0	1.E-26	POU3F3	-1.6	3.E-08
HOXC9	2.1	9.E-18	HOXD8	-1.6	6.E-25	TBX1	-1.4	5.E-07
HOXC-AS2	2.0	9.E-14	HOXC4	1.4	2.E-24	ALX4	1.4	2.E-06
HOXC-AS1	1.8	3.E-10	MAB21L1	1.6	2.E-22	UCHL1	1.1	6.E-04
HOXC6	1.3	9.E-10	HOXC-AS2	2.4	1.E-20	HOXA13	-1.2	7.E-04
IRX6	-1.6	2.E-09	HOXC6	1.4	2.E-18	DACT2	-1.2	2.E-03
ZNF385A	-1.3	2.E-09	IRX6	-1.9	2.E-18	CACNA1E	-1.1	7.E-03

log2FC, log2 fold-change.

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Competing interests None declared.

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