Reviewer’s report

Title: Peripheral blood lymphocytes immunophenotyping predicts disease activity in clinically isolated syndrome patients

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Reviewer: Ming-Feng Liao

Reviewer's report:

In this study, the authors checked the immune-phenotype of peripheral leukocyte of multiple sclerosis patients at the different times. According to author, only individual differences between baseline values and values at the time of a new relapse were analyzed. The author stated that higher levels of B lymphocytes predicted relapse-free status, decrease of NK cells and the naïve subset of cells (CD45RA+ in CD4+) were associated with disease progression. This is an interesting idea, which may apply to clinical practices. However, I think the methodology of statistical analysis is complicate and difficult to be understood by general reader. It is better to clarify the detailed statistical methods and simplify the figures.

1. For example, how to choose the threshold of 0.808 (ratio v.s. last year), 1.638 (ratio v.s. baseline), 9.050 (value) in Figure 1? Why choose those three indicators (value, ratio v.s. last year, and ratio v.s. baseline)? Please explain and compare which indicators is better.

2. The absolute number (or ratio?) of different subtypes of leukocyte seems didn't have significant difference between patients with and without disease progression at different time points (table 1 and 2). Is there any significant difference if we only directly compared the ratio (ratio v.s. last year or ratio v.s. baseline) of different subtypes of leukocyte between patients with and without disease progression at different time points? Can the author show the origin data?

There are some minor suggestions:

1. In introduction: please add discussions about previous studies and applications of peripheral leukocyte immune-phenotype in multiple sclerosis in introduction.

2. In methods and materials (in the study population of CIS patients), may add the reference of inclusion criteria of CIS patients and "presence of >= two oligo clonal band". Did any patient have spinal cord lesions or optic neuritis?
3. In methods and materials (in study outcome), add reference of "Confirmed disability progression (CDP) was defined as an increase in EDSS by 1.0 point (if baseline EDSS > 0) or 1.5 points (if baseline EDSS = 0) confirmed after 6 months".

4. In methods and materials (in statistical methods), please clarify the statistical methods more detail as suggestions.

5. In result: table 1 and 2 need to add the unit of results (ex: %?). may summarized positive findings in one table.

6. In result: how many patients have relapse or disease progression at 6, 12, 24, 36, 48 months, respectively? If any patient had 2 relapse during those periods?

7. In results: all patients received IV steroid and interferon treatment. How many patients also received other treatments (fingolimod, copaxone)? If we can analyze the effect of different treatment to immune-phenotype of leukocyte?

Are the methods appropriate and well described?
If not, please specify what is required in your comments to the authors.

No

Does the work include the necessary controls?
If not, please specify which controls are required in your comments to the authors.

Yes

Are the conclusions drawn adequately supported by the data shown?
If not, please explain in your comments to the authors.

Yes

Are you able to assess any statistics in the manuscript or would you recommend an additional statistical review?
If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

I recommend additional statistical review

Quality of written English
Please indicate the quality of language in the manuscript:

Needs some language corrections before being published
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I declare that I have no competing interests except that I have few multiple sclerosis patients.

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