Reviewer's report

**Title:** Significant difference between three observers in the assessment of intraepidermal nerve fiber density in skin biopsy

**Version:** 3  **Date:** 31 December 2008

**Reviewer:** Giuseppe Lauria

**Reviewer's report:**

I am still concerned by the fact that the authors have first declared to follow the protocol proposed by EFNS guidelines and, when emphasized by the reviewer, changed the methods reporting to have followed that published by Vlckova-Moravcova in Muscle Nerve 2008. The authors should make a statement on this in the covering letter. Besides the differences between the two protocols, this is a major point regarding the reliability of the study.

The authors should temper the considerations on the lack of reliability of the statistical approach used in previous studies.

The primary aim of this study remains quite unclear. Since the interlab agreement is a very important achievement, was it a test for internal errors?

It seems that the 3 examiners just applied the published counting rules but did not undergo joint training sessions in which the rules were discussed. To have the same approach means to settle possible discrepancies in the interpretation of some rules. Indeed, the primary achievement of such a lab should be having an internal agreement on counting. In clinical practise, what examiner among the 3 who participated in the study should be trusted?

Moreover, for each lab a quality control is mandatory. It includes comparing slides and results of IENF quantification with at least another lab.

Normal reference values for each site are also needed. Conversely, the authors compare results from foot biopsies to those from distal leg biopsies. I don't understand the meaning of comparing results “with published control data (ref. 13) as done in a previous study (ref. 4) and classified as pathologic in case of IENFD less than 9 fibers/mm”.

**Level of interest:** An article of insufficient interest to warrant publication in a scientific/medical journal

**Quality of written English:** Needs some language corrections before being published

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:**
I declare that I have no conflict of competing interests' below.