

Elevated percentage of HLA-DR⁺ and ICAM-1⁺ conjunctival epithelial cells in active Graves' orbitopathy

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Dear Editor,

We read with interest the article by Pawlowski et al. describing an elevated percentage of HLA-DR⁺ and ICAM-1⁺ conjunctival epithelial cells in active Graves' orbitopathy [1]. Although the study is mostly well designed and analyzed, we have the following questions and comments. The authors concluded that the percentage of HLA-DR⁺ and ICAM-1⁺ conjunctival epithelial cells in patients with active Graves' orbitopathy (GO) may serve as a topical inflammation marker in GO (from the results in Table 2 and Fig. 2); their data showed differences between the active GO vs. Graves' disease (GD) without active GO and vs. normal control, respectively, using two separate Mann–Whitney U tests.

Their conclusions could be more convincing if they were obtained by a Kruskal–Wallis test with Dunn's nonparametric multiple comparisons or with a method (e.g., Bonferroni adjustment) to avoid the increase of the statistical significance level due to multiple hypothesis tests [2, 3]. Furthermore, data obtained from fifteen eyes of ten healthy volunteers (normal control) do not satisfy the assumption of the Mann–Whitney test that the observations in a sample are independent. In this study, violation of the measurement independence assumption principle partly occurred; all observations both in active GO and GD without active GO are independent, while two-thirds

of the measurements in normal controls are dependent. Thus we recommend that only ten observations (one eye per person) should be used in the statistical test. An excellent reference for the statistical methods regarding the dependence of observations in ophthalmology was given by Armstrong [4].

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Conflicts of interest No authors have any financial/conflicting interests to disclose.

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