Letter to The Editor

Helicobacter Pylori Antibodies in the Aqueous Humor of an Iranian Glaucoma Cohort

Christos Zavos, Jannis Kountouras

Department of Gastroenterology, Second Medical Clinic, Ippokration Hospital, Aristotle University of Thessaloniki, Thessaloniki, Greece

TO THE EDITOR

We read the paper written by Razeghinejad et al. (1), who conducted a study on anti-Helicobacter pylori (H. pylori) IgG antibody levels in an Iranian glaucoma cohort, with considerable interest and based on our original concept (2-5). The authors concluded that a relation between H. pylori infection and primary open-angle glaucoma is not supported by their results, because the aqueous anti-H. pylori IgG antibody concentration did not differ significantly from the cataract control population. However, in the discussion there is little attention to the limitations of their work. Specifically, there is no discussion of: (a) the relatively small sample size, (b) the limited power of the study, (c) the possibility that the control group represents a selected group resulting in bias, (d) the absence of normalization of aqueous antibody titers to another serum protein to serve as control, such as IgG or albumin, and (e) the limited accuracy of using the commercial ELISA technique in the aqueous humor, originally manufactured for serum samples.

It has been reported that the prevalence of H. pylori infection in the city of Shiraz, where the study by Razeghinejad et al. was conducted, is very high (6,7) regardless of the socioeconomic status, an already established significant factor affecting H. pylori prevalence in the European countries. This means that to prove a difference in H. pylori prevalence between any two groups in Shiraz, several hundreds or even a few thousands of participants are required. Instead, Razeghinejad et al. presented their results based on a small number of patients, and therefore the power of their study was too low. The authors failed to comment on the prevalence of H. pylori infection they found in their study groups, which should be very high in both groups according to previous reports and increase with increasing age (6,7).
Furthermore, the authors found significantly higher levels of anti *H. pylori* IgG in the aqueous humor of patients with pseudoexfoliation (XFG) compared with cataract control group and explained their finding by a more severe disruption of blood-aqueous barrier in XFG compared to the patients with cataract (1). However, the fact that they did not normalize aqueous antibody titers to another serum protein to serve as control cannot allow for such definitive conclusions. The proper method of documenting these results is to use Witmer-Goldman coefficient.

From another point of view, the authors comment that the ELISA technique they have used had a sensitivity and specificity of over 95%. This is not correct, because this technique is originally manufactured for serum samples and each laboratory has to establish the normal range of this technique for the aqueous humor samples. The mean anti-*H. pylori* IgG titers reported by Razeghinejad et al. are extremely low, and a predictable error due to mishandling or due to a low accuracy should be discussed. The cut-off value that our lab has established to determine the *H. pylori*-positive cases in the aqueous humor was 4.76 U/ml (2), whereas Razeghinejad et al. found a mean level of 0.24-0.64 U/ml in all their 3 groups (1). This practically means that *H. pylori* antibodies in the aqueous humor were within normal range for all patients.

Finally, the authors discussed that Galloway et al. (8) were also not able to show any association between glaucoma and *H. pylori* infection. However, the authors failed to comment on the methodological flaws that interfere with the interpretation of these results, as highlighted in our subsequent letter to the editor in the same journal (9).

**REFERENCES**