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LETTERS

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Association of interleukin-1 receptor antagonist and interleukin-6 polymorphisms with osteolysis after total hip arthroplasty: comment on the article by Gordon et al

To the Editor:

We read with great interest the article by Gordon and colleagues on the association of interleukin-1 receptor antagonist (IL-1Ra) and IL-6 genes with osteolysis in patients who have undergone total hip arthroplasty (THA) (1). The authors described the association of the *IL1RA* +2018C allele with decreased risk, and the *IL6* haplotype with increased risk, of osteolysis after THA. We would like to offer some comments regarding the methodology of the study and the interpretation of its results.

First, in our experience (unpublished observations) and also as reported by others (2,3), there is little agreement between radiographic assessment and intraoperative visualization of osteolysis, with the former having a tendency to underrepresent both the incidence and magnitude of the condition. Though anteroposterior and lateral radiographs may reveal no osteolysis in the hips being evaluated, this does not necessarily mean that no osteolysis is present. Therefore, using their criteria for the assessment of osteolysis (1), the authors cannot be absolutely sure that they did not enroll some patients with osteolysis into their control group.

Second, the osteolysis-free interval used by Gordon et al to distinguish between the study groups can be questioned. Osteolysis is a dynamic process that can originate anytime and anywhere at the bone–implant interface, including those areas hidden by the prosthesis. Theoretically, even if an "ideal" method for osteolysis detection were available, only a prospectively conducted study involving repeated regular imaging could answer the question of osteolysis origination. Without such study design and sensitive diagnostic tools, the term "osteolysis-free interval" has only limited validity. Surprisingly, when the authors examined a significant genetic variant (IL-1Ra) in multiple logistic regression analysis after adjustment for the effects of other significant covariates, osteolysis-free interval was one of the covariates they included.

Third, the report by Gordon et al does not contain relevant information on a range of factors that could play a role in the development of osteolysis, such as the brand of implants used, the number of participating surgeons and their experience, details on the polyethylene/polymethylmethacrylate used in the implants, and other factors (4,5). Without this information, it could be speculated that the early THA failure and osteolysis development observed by Gordon and colleagues may be the result of poor implant choice or surgical inexperience in the cementing technique, which can lead to the type of premature failure associated with osteolysis, regardless of the genotype (6,7).

Fourth, the authors claim that the results of their in vitro experiments with peripheral blood mononuclear cells stimulated with titanium particles provide a mechanistic explanation for the observed association between the *IL1RA* +2018C allele and a decreased risk of osteolysis, because up-regulation of IL-1Ra messenger RNA (mRNA) was observed in C allele carriers (1). Despite the meticulous approach of Gordon et al with regard to experiment set-up and statistical interpretation, we do not think the data obtained substantiate their conclusion.

The first problem with this interpretation by Gordon and colleagues is that it requires in vitro data on systemic blood cells to be extrapolated and applied to a much more complex in vivo situation, where mesenchymal cells interact predominantly with polyethylene and polymethylmethacrylate particles. The second and more critical issue is their use of β-actin as a reference (housekeeping) gene in real-time polymerase chain reaction (PCR). Even commonly used housekeeping genes may vary in stability depending on the cell type or the disease being studied (8). Thus, as has been observed at our laboratory (9) and by others (8,10), it is necessary to identify appropriate housekeeping-type genes that show sample-independent stability. Therefore, utilization of carefully validated reference genes other than β-actin is an absolute prerequisite for obtaining clinically relevant and valid information from quantitative real-time PCR (8,9).

Finally, apart from the concerns mentioned above regarding study design and interpretation of mRNA expression data, the authors neither discussed the need for replication of their data nor mentioned the possibility of obtaining false-positive results, a problem inherent to all association studies that do not correct for the testing of multiple polymorphic sites. Therefore, in accordance with the rules for genetic association studies (11), unless the observation that carriage of the *IL1RA* +2018C allele protects against osteolysis after cemented THA is verified in other centers/populations, the conclusion by Gordon et al is an overstatement and should be considered preliminary.

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Effect of ciprofloxacin-mediated inhibition on cell proliferation in rat tendon cells: comment on the article by Tsai et al

To the Editor:

We read with interest the recent article by Tsai et al in which the authors provided new insight into a possible mechanism of ciprofloxacin-associated tendinopathy (1). Though the study by Tsai and colleagues produced interesting results, we would like to highlight some points that require further clarification.

First, since the results reported by Tsai et al were achieved using the ciprofloxacin concentration at which the exposed tendon cells showed a significant cell cycle response, it would be important to know the serum concentrations that would achieve a similar effect clinically, and whether the concentration of the drug used in Tsai and colleagues' study is higher than the serum level achieved with currently prescribed therapeutic doses (2,3). Second, we would also like to know what the effects on the other proliferating cells in the body would be with the use of ciprofloxacin at a higher concentration than was used in the study, regardless of whether the serum and tendon cell concentrations achieved clinically are equal to or different from those studied by Tsai et al. This was not clarified in their report.

Even with these points in need of clarification, the study by Tsai et al provides useful information with regard to the long-unexplained mechanism of ciprofloxacin-associated tendinopathy.

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Reply

To the Editor:

We thank Dr. Medhi and colleagues for their interest in our study. They raise important points related to ciprofloxacin concentration and the effects of standard dosing regimens on tendon tissue. However, based on the current experimental settings, these questions could not be answered using the results of our study. The peak serum concentrations of ciprofloxacin given orally or intravenously have been reported to range from 0.5 to 10 μg/ml (1–4), which are within the range of concentrations used in our study. Inhibition of tendon cell proliferation was noted with ciprofloxacin at a concentration of 5 μg/ml, although the degree of inhibition was not statistically significant. Although the concentration of ciprofloxacin in tendon tissue after standard dosing regimens remains unknown, the results of our study suggest a potential link between ciprofloxacin-associated tendinopathy and increased doses of ciprofloxacin.

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