Alternative Macrophage Activation Proteins as Candidate Serum Markers in Periprosthetic Osteolysis

INTRODUCTION:
Periprosthetic loosening and osteolysis are recognized as the major long term complications of total joint arthroplasty, often resulting in the need for revision surgery. The development of periprosthetic osteolysis is characterized by wear debris accumulation along the periprosthetic membrane and phagocytosis by macrophages. Increasing evidence suggests that periprosthetic osteolysis may be characterized by alternative activation of macrophages similar to other disorders that have been linked to macrophage accumulation of non disposable material. Expression profiling of periprosthetic osteolysis tissue revealed elevated levels of the alternative macrophage activation markers CCL18 and chitotriosidase (CHIT1) (Fig 1). In light of the fact that CCL18 and CHIT1 have proved to be valuable serum markers for other disorders of macrophage function, the goal of this study is to assess these as possible serum biomarkers of periprosthetic loosening.

METHODS:
To evaluate CCL18 and CHIT1 as potential biomarkers for periprosthetic loosening, serum from patients requiring revision surgery due to periprosthetic loosening was compared to healthy donors. Serum CCL18 levels were measured in 26 osteolysis patients and 20 healthy controls using a sandwich enzyme-linked immunosorbent assay (ELISA) from R&D Systems consisting of an anti-human CCL18 capture antibody, a biotinylated detection antibody, streptavidin conjugated horseradish-peroxidase and recombinant human CCL18 to construct a standard curve. Serum was diluted 1:1000. Serum CHIT1 enzyme activity was measured in 29 patients with periprosthetic loosening and 18 healthy controls. CHIT1 activity was determined using the method previously described by Hollak et al. The Mann-Whitney U test was used to compare osteolysis and healthy control groups. The study was approved by the Hospital for Special Surgery IRB.

RESULTS:
Figure 2 demonstrates CCL18 serum levels from 26 osteolysis patients and 20 healthy controls. Serum CCL18 levels are elevated in osteolysis patients compared to healthy controls (Fig 2,4). The mean serum CCL18 level in controls is 41.9 ng/mL, whereas the mean level in osteolysis is 78.9 ng/mL (Fig 4). CHIT1 activity is also elevated in 29 osteolysis patients with a mean of 325.4 nmol/mL/hr while mean activity in 20 healthy controls is 92.0 nmol/mL/hr (Fig 3,4). Differences were found to be statistically significant for both CCL18 (P<0.002) and CHIT1 (P<0.001) using the Mann-Whitney U test.

DISCUSSION:
Serum CCL18 and CHIT1 levels were elevated in periprosthetic loosening, confirming their value as plausible biomarkers for periprosthetic osteolysis. Although mean levels were found to be statistically different, there is overlap between cases and controls for both markers. CCL18 and CHIT1 have been shown to be useful biomarkers in Gaucher disease and for the assessment of therapeutic efficacy. Additional studies have also demonstrated that CHIT1 is elevated in atherosclerosis and periprosthetic loosening. Our data confirm elevated serum CHIT1 activity in osteolysis patients. These results also suggest that macrophages in periprosthetic osteolysis display an M2 like phenotype, consistent with findings that a pro-inflammatory macrophage response may only be transient in nature.

Table 1

<table>
<thead>
<tr>
<th>Gene Analyzed</th>
<th>Osteolysis Patients</th>
<th>Osteoarthritis Controls</th>
<th>Fold Difference</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CHIT-1</td>
<td>21.24 (±28.34)</td>
<td>0.32 (±0.64)</td>
<td>66.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CCL18</td>
<td>7.33 (±5.64)</td>
<td>2.52 (±3.16)</td>
<td>2.9</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

REFERENCES: