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Longitudinal Documentation of Serum Cartilage Oligomeric Matrix Protein and Patient-Reported Outcomes in Collegiate Soccer Athletes over the Course of an Athletic Season

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Longitudinal documentation of serum cartilage oligomeric matrix protein and patient reported outcomes in collegiate soccer athletes over the course of an athletic season

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Abstract

Background—Serum cartilage oligomeric matrix protein (sCOMP) is a biomarker for cartilage degradation. Patient reported outcomes (PRO) are used to document post-injury recovery and may be used to prospectively identify changes in the course of a season. It is unknown what effect intense, continuous physical activity has on sCOMP levels and PRO values in athletes over the duration of a soccer season.

Hypothesis/Purpose—The purpose of this study was to longitudinally document sCOMP levels, and to determine if changes in PROs occur in collegiate soccer athletes during a season. The hypotheses tested were that sCOMP levels and PRO scores would remain stable over the duration of the spring soccer season.

Study Design—Case series; Level of evidence, 4.

Methods—29 NCAA Division-I soccer athletes (18 males, 11 females, age:19.6±1.2 years, height:177.8±7.4cm, mass:73.8±10.2kg) participated in three [pre-(T1), mid-(T2), and post-season(T3)] data collection sessions. Subjects were included if they participated in the spring soccer season and were free of severe knee injury at the time of data collection. At each session subjects completed PROs (Lysholm, IKDC) before serum collection.
Results—For sCOMP(ng/mL) there was a significant effect for time with significant increases at T$_2$ (1723.5±257.9, p<0.0001) and T$_3$ (1624.7±231.6, p=0.002) when compared with T$_1$ (1482.9±217.9). For each of the PROs there was a significant effect for time with increases from T$_1$-T$_2$ and T$_2$-T$_3$.

Conclusions—These data indicate sCOMP levels increased as athletes reported an increased level of function over time. However, the differences in sCOMP levels did not reach the calculated MDC value and the differences in PRO scores did not reach previously calculated MDC values. It is unclear if these increases in sCOMP levels were due to an increase in cartilage matrix breakdown or turnover. Even though these elevations may not be clinically meaningful, this biomarker may have the potential to be used for future research studies investigating the effects of exercise on overall joint health in longitudinal studies. In addition, these results indicate fluctuations in sCOMP occur during a competitive season and must be taken into consideration for future biomarker studies.

Key Terms
articular cartilage; biomarkers; soccer; cartilage matrix

INTRODUCTION

Primary osteoarthritis (OA) is characterized by irreversible joint destruction such as cartilage degradation, osteophyte formation, and joint space narrowing, and causes pain, loss of function, activity limitations, and participation restrictions in the millions of individuals it affects each year. It has been theorized that continuous and intense physical activity can cause the development of primary OA in elite athletes. In addition, there has been speculation regarding how articular cartilage responds to increases in activity level and determination of the levels related to articular cartilage damage due to intense physical activity is unclear. It has been hypothesized that sports that include rapid acceleration and deceleration moments, continuous training where the joints sustain high impact, and athletes that compete at elite levels for an extended period of time are at an increased risk of developing OA. However, it is unknown as to the exact amount, types, and intensity of exercise that is detrimental to the articular cartilage in the human joint. At this time there are limited tools available to prospectively investigate the relationship between early stages of degenerative joint disease, such as articular cartilage degradation, and athletic participation.

Described as an instrument to measure the progress of a disease or the effect of a treatment on disease progression, biomarkers may serve as a tool to elucidate the effects of exercise on articular cartilage and the eventual development of primary OA. A biomarker for cartilage degradation, known as serum cartilage oligomeric matrix protein (sCOMP), has been shown to be elevated after intense exercise. Serum COMP is a non-collagenous protein identified in synovium, ligamentous tissue, tendon, meniscus, and primarily articular cartilage. Serum COMP levels are elevated after intense exercise and are elevated in subjects who participated in less strenuous forms of physical activity. This indicates that exercise stresses the articular cartilage. However, the balance of cartilage turnover caused by exercise and that which may cause long term ramifications is unknown.

Several studies have investigated changes in sCOMP during exercise. For example, in healthy subjects who participated in moderate walking exercises, marathons, and ultra-marathons, there was an increase in sCOMP levels after activity. Serum COMP levels have been documented to return to baseline levels 30 minutes after a moderate walking exercise and a longer period of time for more intense exercise.
marathon, sCOMP levels returned to baseline 24–48 hours after the race, and for an ultra-marathon sCOMP levels returned to baseline 6 days after the race.\textsuperscript{23, 33} Therefore, elevated levels of sCOMP as a result of physical activity may indicate that this biomarker may be useful for establishing the relationships between exercise and articular cartilage changes.

It has been reported that elite level athletes who participate in high impact sports for an extended period of time may have more risk for developing primary OA.\textsuperscript{6, 25, 37} However, there are no prospective, longitudinal studies investigating the effects of continuous, intense physical activity over time on sCOMP. Therefore, to further investigate sCOMP as a biomarker for cartilage damage induced by exercise, it is important to prospectively study those who participate in high impact sports. Documentation of the stability of this marker over the duration of an athletic season, rather than one bout of exercise, is needed for further investigation of the effects of exercise on articular cartilage. In addition, patient reported outcome instruments (PROs) are used to document self-reported function and knee related symptoms. These can be used throughout the duration of the season to provide additional information that might explain changes in sCOMP levels such as decrease in function, or an increase in pain or knee related symptoms.

The primary purpose of this investigation was to document the stability of sCOMP in collegiate soccer athletes during a spring soccer season. In addition, we aimed to document patient reported outcomes assessing pain and function related to the knee joint using two separate PROs. The hypotheses tested were that sCOMP levels and PRO scores would remain stable over the duration of the spring soccer season. Finally, as a secondary analysis, we aimed to determine if a relationship would be observed between changes in sCOMP levels and PRO scores.

**MATERIALS and METHODS**

**Design**

A prospective case series study was employed to determine the stability of sCOMP over the duration of a spring soccer season.

**Population**

A volunteer sample of 29 Division I NCAA soccer athletes (18 males, 11 females, age: 19.6±1.2 years, height: 177.8±7.4cm, mass: 73.8±10.2kg) participated in this research study. Subjects were included if they were actively participating in the spring soccer season. Subjects were excluded if they were not participating in soccer related activities. Informed consent was obtained from the subjects participating in the study. This study was approved by the Institutional Review Board at the University of Kentucky.

**Procedure**

Subjects reported for data collection at three time points (pre- (T\textsubscript{1}), mid- (T\textsubscript{2}) and post-season (T\textsubscript{3})) over the duration of their spring soccer season (February - May). Upon arrival, each subject was asked to remain seated for 30 minutes. It is recommended that subjects remain seated before serum collection in order for serum levels to return to baseline after any moderate exercise such as walking to the data collection site.\textsuperscript{1, 31} During the 30 minutes of rest the subjects were asked to complete two PROs (International Knee Documentation Committee (IKDC) and Lysholm). Once the subjects had been seated for 30 minutes and both PROs were completed, a maximum of 10 mL of blood was collected from the antecubital fossa.
Enzyme-linked Immunosorbent Assay

The blood was immediately placed on ice and transported for separation. After clotting at room temperature, sera were separated and stored in a −20°C freezer and eventually transported into a −80°C freezer until assayed. Once all samples were collected, sCOMP concentrations were determined using a commercial enzyme-linked immunosorbent assay (Euro-Diagnostica, ALPCO, Salem, NH). Serum COMP values are expressed as ng/mL. The average intra-assay coefficient of variance (CV) of all controls was 1%, the average intra-assay CV of all samples was 1%, and the average inter-assay CV of all controls was 4%.

Patient Reported Outcomes

Subjects completed two PROs for this study, the IKDC and the Lysholm. These were used to assess function and symptoms related to the knee, and were completed by each subject at each time point. The IKDC is a valid and reliable PRO used to measure knee symptoms, level of function, and sports activity in patients after knee injuries.\(^{15, 21}\) Higher scores represent higher levels of function and fewer self-reported knee symptoms.\(^{15, 21}\) A change of ±9 points is required to indicate a clinically meaningful change in the patient’s knee symptoms.\(^{21}\) The IKDC has acceptable internal consistency and test-retest reliability.\(^{20}\)

The Lysholm knee scale measures eight condition specific domains for the knee.\(^{27, 38}\) These domains include limping, use of supporting device, stair climbing, squatting, walking, locking, instability, and pain.\(^{27, 38}\) The Lysholm is scored from 0 to 100, where a score of 95 to 100 is excellent, 89 to 94 is good, 65 to 83 is fair, and <65 is a poor.\(^{24}\) A minimal detectable change (MDC) for the Lysholm is ±8.9 points.\(^{3}\) For patients with chondral and ACL injuries, the Lysholm has acceptable internal consistency and test-retest reliability.\(^{3, 24}\)

Statistical Analysis

Descriptive statistics (mean ± standard deviation (SD)) for sCOMP and PROs for each time point are presented in the Table 1. The independent variables were time (T\(_1\), T\(_2\), T\(_3\)) and sex (male, female). The dependent variables were sCOMP values and scores on two PROs (Lysholm, IKDC). Separate linear mixed models analyses were used to determine sex by time interactions and differences in each of the dependent variables (sCOMP, IKDC, Lysholm) for each of the time points (T\(_1\), T\(_2\), T\(_3\)). Paired-sample t-tests were used to explain significant interactions or main effects. Pearson correlations were employed to determine if a relationship exists between changes in sCOMP levels and PRO scores for the 18 subjects with sCOMP at each of the time points. Changes for T\(_3\) minus T\(_2\), T\(_3\) minus T\(_1\), and T\(_2\) minus T\(_1\) were calculated. Hedge’s g effect sizes\(^{17}\) with 95% confidence intervals (CI) were calculated to examine the magnitude of change and associated variability over time for each of the dependent variables. Calculated effect sizes were interpreted as weak if they were less than 0.40, moderate if between 0.41 and 0.69, and strong if greater than 0.70.\(^{8}\) Alpha was set \textit{a priori} at \(p \leq 0.05\). Statistical analyses were performed using IBM SPSS software, version 19.0 (Armonk, NY), and SAS version 19.2 (Cary, NC).

RESULTS

All injuries documented during the spring soccer season are listed in Appendix A and surgeries that were treated or performed before the spring soccer season are listed in Appendix B. Subjects were not excluded in this study if they had a history of injury or surgery. All subjects who participated with a history of surgery were medically cleared to participate in soccer activities as determined by an orthopaedic surgeon and a primary care physician. No ongoing or resolving musculoskeletal injuries were recorded in subjects that were enrolled into the study at the time of physical examination. All subjects that were
treated for musculoskeletal injuries during the spring soccer season were included if they were actively participating in soccer activities during the data collection sessions.

Figure 1 depicts the total number of sCOMP samples collected and processed for each of the time points. Missing data points were attributed to a) the inability of the phlebotomist to collect a sample, b) sample processing error, c) removal from the team, or d) severe knee injury at time of data collection. Figure 1 also depicts the time lapse between each of the data collection sessions. If sCOMP samples were not available for analysis, the PRO scores for those subjects remained in the analysis for each of the time points.

Serum COMP Values

Descriptive statistics for each of the time points can be found in the Table. When all 29 subjects were included in the data analysis, there was no significant sex by time interaction for sCOMP (p=0.44) or significant effect for sex (p=0.09). There was a significant effect of time on sCOMP levels (p<0.001), with significant increases between T1-T2 (p<0.001), T1-T3 (p=0.002), but not for T2-T3 (p=0.08). Calculated Hedge’s g effect sizes and 95% CIs for sCOMP were 1.0 (CI: 0.4, 1.6) for T1 vs T2 and 0.6 (CI: 0.02, 1.2) for T1 vs T3. The effect sizes for T1 vs T2 was interpreted as strong and the effect size for T1 vs T3 was interpreted as moderate, and the associated 95% CI for both effect sizes do not encompass zero.

In addition, we performed a linear mixed model for the 18 athletes for which there were data for each of the time points. When the 18 subjects with all data points in the data analysis, there was no significant sex by time interaction for sCOMP (p=0.70). There was a significant effect for sex (p=0.03). There was a significant effect for time on sCOMP levels (p<0.001), with significant increases between T1-T2 (p<0.001), T1-T3 (p=0.005), but not for T2-T3 (p=0.14).

Finally, we calculated means (±SDs) for four different groups: 1) all 29 athletes who participated, 2) athletes with a documented history of injury or surgery, 3) athletes who were treated for an injury during the soccer season, and 4) athletes with no documented injury during the spring soccer season or surgery history (Figure 2). It must be noted that it is possible for athletes to be in group 2 and group 3 for this analysis.

sCOMP Minimal Detectable Change

We calculated minimal detectable change (MDC) values using intersession reliability via an intraclass correlation coefficient (ICC) mixed model (single measure), and standard error of measurement (SEM). A total of 18 subjects had sCOMP values for all three data time points and these values were used to calculate MDC value. The MDC value was calculated with a 95% level of confidence using the formula SEM * 1.96 * √3. Single measure intersession reliability over the three collection time points was ICC(2,1) =0.61 (SEM=136.9ng/mL). The MDC value for sCOMP over these three data points was 464.6ng/mL.

Patient Reported Outcomes

Descriptive statistics for each of the PROs can be found in Table 1. There was no significant sex by time interaction for either the Lysholm (p=0.52) or the IKDC (p=0.17). There was no significant effect for sex for either the Lysholm (p=0.09) or the IKDC (p=0.23). A significant effect for time was noted for the Lysholm (p=0.03), with significant increases between T1-T2 (p=0.03), and T2-T3 (p=0.06). A significant effect for time was noted for the IKDC (p=0.02), with significant increases between T1-T3 (p=0.02) and T2-T3 (p=0.04).
Calculated effect sizes for the IKDC were 0.1 (CI: −0.4, 0.6) for T1 vs T2 and 0.5 (CI:−0.1, 1.1) for T1 vs T3 with one effect size interpreted as weak and one effect size interpreted as moderate. For the Lysholm, the effect sizes were −0.1 (CI: −0.6, 0.5) for T1 vs T2 and 0.4 (CI: −0.2, 1.0) for T1 vs T3, with one effect size interpreted as weak and one effect size interpreted as moderate. However, caution must be used when interpreting these results as for each effect size, the 95% CIs did encompass zero.

**Relationship between changes in sCOMP levels and PRO scores**

There were no significant relationships between changes in sCOMP levels and PRO scores when T2 and T1 were compared. The relationships between change in sCOMP levels and IKDC and Lysholm scores were r=0.3 (p=0.2) and r=0.2 (p=0.4), respectively. There was a significant positive relationship between changes in sCOMP levels and IKDC scores (r=0.5, p=0.03), and sCOMP levels and Lysholm scores (r=0.5, p=0.05) between the T3 and T2 time points. Finally, there was no relationship between sCOMP levels and IKDC (r=0.1, p=0.7) or the Lysholm (r= 0.08, p=0.7) for changes calculated from T3 to T1.

**DISCUSSION**

The purpose of this study was to document the stability of sCOMP levels and PRO scores in collegiate soccer athletes at three time points during a competitive athletic season. Serum COMP levels and PRO scores increased over the duration of an athletic season. It was also demonstrated that the differences between the time points did not exceed the intersession MDC value of 464.6ng/mL reinforcing that the changes in our measures, while important to note, may not have been clinically different.

There are few published studies that report baseline levels for young, healthy athletes or patients.26, 36 Previously reported levels of sCOMP for six healthy athletes with no history of joint disease or joint injury ages 30±9 was 47μg/mL with a range from 10–109μg/mL.26 Additionally, sCOMP baseline levels using healthy blood donors ages 20–65, and healthy children ages 1–20, were reported to be 11.3μg/mL and 10.3μg/mL, respectively.36 The baseline levels for the cohort in this study, who had a history of injury or surgery was 1482.9 ng/mL. When interpreting sCOMP results, it is important to realize inter-study variation may be due to several other factors including the ELISA utilized, age, BMI, race, and previous surgical intervention.7, 22 We believe it is important for authors to report these demographics to allow for proper interpretation of sCOMP data, particularly in athletic populations who are likely to have had previous musculoskeletal injuries.

This is the first study to report intersession MDC values for sCOMP levels in a physically active population over the duration of an athletic season. Based on our findings, an MDC value of greater than 464.6ng/mL would be necessary to imply a clinical change versus being considered normal variability. In addition, inter- and intraday MDC values for a physically active cohort with no history of lower extremity surgery have been calculated in a previous investigation.18 These results indicated an interday value of 292 ng/mL and an intraday value of 320 ng/mL are required to identify changes that exceed the variability of the marker.18 It appears that the MDC value of 464.6 ng/mL for these collegiate level athletes is higher than the MDC values that were calculated for a physically active cohort with no history of lower extremity surgery.18 These differences may be real, as subjects with a history of surgery were included in the present study but not in the previous investigation.18 In addition, the differences in MDC values may also exist because of the extended time frame of data collection. The MDC value calculated for this study spanned several weeks of time, where the previous investigation were calculated for within and between day differences.18 Recent literature has hypothesized the use of sCOMP as a biomarker to elucidate the effects of acute articular cartilage injury and the development of
preclinical posttraumatic osteoarthritis. The reported MDC values may be beneficial in future investigations and provide a comparison to determine meaningful changes associated with this measure.

This is the first study to report changes in sCOMP levels over the duration of an athletic season. Previous investigations have reported changes in sCOMP levels following single bouts of activity. The results of this study indicate a statistical difference in the increase in sCOMP levels between T1 and T2 (p=0.001), and T1 and T3 (p=0.002). In addition, there was a trend for a statistically significant decrease in sCOMP levels from T2 and T3 (p=0.08). However, based on the calculated MDC value of 464.6 ng/mL, differences of 241 ng/mL for T1-T2, 142 ng/mL for T1-T3 and 99 ng/mL for T2-T3 are not clinically significant as they do not exceed the variability associated with the measure.

Additionally, we performed a secondary analysis of only those subjects for which we had serum samples at all three time points. We identified statistically significant differences between each of the time points, similar to the analysis of all subjects (n=29). However, when the calculated MDC value is employed, none of the differences between these time points were clinically meaningful, T1-T2= 205 ng/mL, T1-T3= 137 ng/mL, and T2-T3= 68 ng/mL. Furthermore, when the athletes were grouped according to whether they were treated for an injury during the spring soccer season (Appendix A), had a previous history of injury or surgery (Appendix B), or those subjects without injury during the spring soccer season or history or surgery, we noticed similar patterns of change between each time point. Specifically, upon visual analysis there was a change when baseline values were compared with mid season and postseason however, the mean and standard deviations between groups overlap signifying there likely was no difference between groups (Figure 2). While the individuals with an injury that were treated during the spring season did not decrease as much as the other groups at T3, this can be attributed to the time of injury from serum collection. It may be that the athletes that were included in the ‘treated for an injury during the spring soccer season’ group sustained injuries in the last half of the season and this caused their levels to remain elevated.

At this time it is not known whether changes after single bouts of exercise are either indicative of an increase in cartilage matrix turnover or cartilage damage. The current findings, while not clinically significant, demonstrate that there are increases in sCOMP levels over the duration of an athletic season, likely attributed to the total participation or intensity of exercise. However, at this time we are unable to speculate whether these fluctuations were related to an increase in cartilage matrix turnover or matrix degradation. In addition, we are unable to determine the longitudinal effects of these fluctuations on overall articular cartilage health. Finally, we hypothesize there was a trend for decreasing sCOMP levels from T2-T3 due to a decrease in the amount of participation during the final four weeks of the season. Total time of participation data is not available to support this speculation; however, we do know that as the spring soccer season came to an end, the athletes participated in fewer practice sessions and games. Based on these results, future research studies are needed to investigate the influence participation on sCOMP levels in a physically active cohort.

The current findings indicate a statistical difference between time points for the IKDC and Lysholm. While these differences are important, and documentation of change due to normal physical activity is important when employing PROs, these differences should be interpreted with caution, as they were not clinically significant based on previously reported MDC values. It has been reported that a change of ±9 points for the IKDC, and ±8.9 points for the Lysholm indicate a significant change in patient’s knee symptoms. For the IKDC, a 5.1 point difference between T1 (99.7±12.4) and T2 (94.8±6.0) and a 4 point
difference between T₂ (90.8±8.8) and T₃ (94.8±6.0) are not large enough to indicate a clinically significant change in a subject’s knee symptoms. For the Lysholm, a 2.4 point difference between scores of T₁ (92.4±8.4) and T₃ (96.3±7.1) and a 4 point difference between T₂ (92.3±10) and T₃ (96.3±7.1) are also not sufficiently large to indicate clinically significant changes in the subject’s knee symptoms. Furthermore, recent literature has reported normative data for the Lysholm knee score in patients with normal knees. The average Lysholm score was 94 (range 43–100) for uninjured participants. The current data are representative of this score, with the average for each of the time points ranging from 92 to 96. These PRO scores show that these athletes were functioning at a normal level and reported near normal joint health throughout the duration of the season.

LIMITATIONS

The present study is not without limitations. First, subjects that participated in the study were not excluded based on medical history. Subjects with a history of injury, including anterior cruciate ligament knee reconstruction (ACLR), were included in our study. Since the aim was to determine the stability of sCOMP over time, exclusion of patients with a previous history of severe knee injury or surgery was not required. Additionally, the injury and surgery history data were collected retrospectively. Medical records were reviewed for previous surgical or injury history. In addition, medical records were retrospectively reviewed to obtain data regarding injuries that were treated during the spring soccer season. Furthermore, both PROs employed in this study were specific to the knee joint. Serum COMP is not specific to cartilage degradation at the knee joint. Therefore, the use of PROs for other joints, such as the Foot and Ankle Ability Measurement (FAAM) or the Disabilities of the Arm, Shoulder and Hand (DASH) could have been employed to determine self-reported function for these joints. Finally, participation in activities outside of soccer practice and soccer associated activities such as lifting and running were not controlled for potential confounding effects.

CONCLUSION

The present findings indicate that sCOMP levels do change over duration of a soccer season; however, the difference between each of the separate time points was less than the calculated intersession MDC value of 464.6ng/mL. These results likely imply that the effects of intense, continuous physical activity resulted in an increase in cartilage turnover identified as increased levels of sCOMP; however, it is unclear whether this process had a negative influence on overall joint health. Future research investigations are warranted to further determine the relationship between these increased levels in sCOMP over the duration of an athletic season, cartilage degradation, and a possible connection with primary OA development in athletes who participate in continuous, intense physical activity.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References


Figure 1.
Flow chart depicting subject participation, data loss, and time between serum cartilage oligomeric matrix protein data collection sessions.
Figure 2.
Serum COMP values (±SD) for 1) all 29 athletes that participated, 2) athletes with a documented history of injury or surgery, 3) athletes who were treated for an injury during the soccer season, and 4) athletes with no documented injury during the spring soccer season or surgery history.
Table 1
Serum cartilage oligomeric matrix protein (sCOMP) levels and patient reported outcome scores (Lysholm and International Knee Documentation Committee (IKDC)) for each of the time points (pre-season ($T_1$), mid-season ($T_2$) and post-season ($T_3$)).

<table>
<thead>
<tr>
<th>Dependent Variable (Mean±SD)*</th>
<th>Pre-season ($T_1$)</th>
<th>Mid-season ($T_2$)</th>
<th>Post-season ($T_3$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>sCOMP (ng/mL)</td>
<td>1482.9±217.9a,b</td>
<td>1723.5±257.9</td>
<td>1624.7±231.6</td>
</tr>
<tr>
<td>Lysholm</td>
<td>93.1±8.1a</td>
<td>92.3±10a</td>
<td>96.3±7.1</td>
</tr>
<tr>
<td>IKDC</td>
<td>89.7±12.4a</td>
<td>90.8±8.8a</td>
<td>94.8±6.0</td>
</tr>
</tbody>
</table>

*Differences are significant at $p \leq 0.05$.  

*aSignificantly different from $T_3$.  

*bSignificantly different from $T_2$.  

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