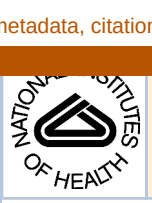



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# Cigarette smoking, disease severity, and autoantibody expression in African Americans with recent-onset rheumatoid arthritis

**Ted R. Mikuls, MD, MSPH**[1], **Laura B. Hughes, MD, MSPH**[2], **Andrew O. Westfall, MS**[2], **V. Michael Holers, MD**[3], **Lezlie Parrish, MPH**[3], **Desiree van der Heijde**[4], **Maaltje van Everdingen, MD, PhD**[4], **Graciela S. Alarcón, MD, MPH**[2], **Doyt L. Conn, MD**[5], **Beth Jonas, MD**[6], **Leigh F. Callahan, PhD**[6], **Edwin A. Smith, MD**[7], **Gary Gilkeson, MD**[7], **George Howard, DrPH**[2], **Larry W. Moreland, MD**[2,*], and **S. Louis Bridges Jr., MD, PhD**[2]

[1] University of Nebraska Medical Center, the Nebraska Arthritis Outcomes Research Center (NAORC), and Omaha VA Medical Center, Omaha, NE, USA [2] University of Alabama at Birmingham, Birmingham, AL, USA [3] University of Colorado Health Sciences Center, Denver, CO [4] Leiden, Netherlands [5] Emory University, Atlanta, GA, USA [6] University of North Carolina, Chapel Hill, NC, USA [7] Medical University of South Carolina, Charleston, SC, USA

## Abstract

**Objective—**To examine the association of smoking with clinical and serologic features in African Americans with recent-onset rheumatoid arthritis (RA) and to explore whether this association is dependent on the presence of the *HLA-DRB1* shared epitope (SE).

**Methods—**In African Americans with recent-onset RA (n = 300), we examined the association of cigarette smoking (current vs. past vs. never and pack-years of exposure) with anti-CCP antibody, rheumatoid factor (RF) (-IgM and -IgA), rheumatoid nodules, and baseline radiographic erosions using logistic and cumulative logistic regression (adjusting for SE status). We also examined for evidence of interaction between smoking status and SE for all outcomes.

**Results—**Although there was no association with RF-IgA seropositivity, current smokers were approximately twice as likely as never smokers to have higher IgA-RF concentrations (based on tertiles; OR = 1.74; 95% CI 1.05–2.88) and nodules (OR = 2.43; 95% CI 1.13–5.22). These associations were most pronounced in those with more than 20 pack-years of exposure. There was no association of smoking status or cumulative tobacco exposure with anti-CCP antibody, IgM-RF, or radiographic erosions. There was also no evidence of a biologic or statistical SE-smoking interaction for any of the outcomes examined.

**Conclusion—**This is the first study to systematically examine the association of cigarette smoking with RA-related features in African Americans. Cigarette smoking is associated with both subcutaneous nodules and higher serum concentrations of IgA-RF in African Americans with RA, associations that may have important implications for long-term outcomes in this population.

Corresponding Author and Request for Reprints: Ted R. Mikuls, MD, MSPH, Associate Professor, Department of Medicine, University of Nebraska Medical Center and Omaha VA Medical Center, 986270 Nebraska Medical Center, Omaha, NE, USA 68198-6270; phone (402) 559-7288; fax (402) 559-6788; email: tmikuls@unmc.edu.

*Current affiliation: Division of Rheumatology and Clinical Immunology, University of Pittsburgh, Pittsburgh, PA

### Keywords

rheumatoid arthritis; African Americans; cigarette smoking; rheumatoid factor; anti-CCP antibody

Since the first report by Vessey nearly twenty years ago [1], cigarette smoking has been shown in numerous investigations to be associated with rheumatoid arthritis (RA) susceptibility [2–10]. In a national study of disease-discordant monozygotic twins, smoking was associated with a twelve-fold increased risk of RA [11], an association that appears to be most pronounced in those with long-term exposure (> 20 years duration) [7,12,13]. The impact of smoking on RA risk also appears to be mediated by certain genetic risk factors with the *HLA-DRB1* shared epitope (SE) being the most notable. In a recent case-control study, there was no significant association of smoking with seropositive RA in cases without the SE. Ever having smoked, however, was associated with a more than six-fold increased risk of anti-CCP (cyclic citrullinated peptide) antibody-positive RA among those with a single SE-containing *HLA-DRB1* allele and a more than 20-fold increased risk in those with two SE-containing alleles [2].

In addition to its association with disease risk, smoking also appears to be an important determinant of RA disease expression. Specifically, smoking has been associated with the presence of extra-articular disease manifestations [14] (including rheumatoid nodules), radiographic severity [15–17], and serum rheumatoid factor (RF) [15,16,18,19]. Reports examining associations of smoking with anti-CCP antibody in RA have been inconsistent with at least one study showing a positive association [20] while another showed no such association [21]. Studies to date examining the association of smoking with RA disease features have almost exclusively involved subjects of European ancestry. The lack of such studies among ethnic/racial minorities is relevant for three reasons: 1) the prevalence of smoking in African Americans appears to be increasing [22] while ‘quit rates’ in African Americans are lower compared to Caucasians [23], 2) disease characteristics related to smoking portend a poor outcome in RA [24–30], and 3) smoking appears to have a disproportionately negative impact on minority patients with select chronic conditions [31,32] The objective of this cross-sectional study was to examine the association of cigarette smoking with disease expression in a group of well characterized African Americans with recent-onset RA.

## Patients and Methods

### Study Population

RA cases (n = 300) were participants in the Consortium for the Longitudinal Evaluation of African-Americans with Early Rheumatoid Arthritis (CLEAR)[33–35]. Subjects were enrolled through one of four sites in the southeast U.S. (University of Alabama at Birmingham, Emory University, Medical University of South Carolina, and the University of North Carolina). The study was approved by the Institutional Review Board (IRB) at each participating center and all study subjects provided informed written consent prior to participation. Cases satisfied the American College of Rheumatology (ACR) RA classification criteria [36], had less than two years of disease duration, and self-reported African American race/ethnicity (there was no requirement for additional first- or second-degree relatives of African American ancestry). Smoking status (current, former, never) was collected at the time of enrollment and for ever smokers, pack-years of smoking served as a measure of cumulative exposure. All subjects underwent formal examination that included an assessment for the presence of subcutaneous nodules. Radiographs of the hands and wrists were obtained at baseline (defined as < 2 years duration, with a mean of 13.7 months disease duration) and scored using the van der Heijde modified Sharp score [37]. Validated scores were available for 192 of the 300 cases under study for whom radiographs were obtained. Subjects with available scores were classified as having

either erosive disease (Sharp erosion score > 0) or non-erosive disease (Sharp erosion score = 0) at baseline.

### Autoantibody Measurement

Autoantibody measurements, including anti-CCP antibody and RF isotypes, were performed as previously reported [33]. Anti-CCP (IgG) antibodies were measured in arbitrary units (U) per ml using a commercially available second generation enzyme linked immunosorbent assay (Diastat, Axis-Shield Diagnostics Ltd., Dundee, Scotland, UK) and were considered to be positive at a cut-off value ≥ 5 U/ml [33]. RF isotypes (IgA and IgM) were measured in international units (IU) per ml and were assessed using commercially available ELISA (INOVA Diagnostics Inc., San Diego, CA, USA). Positive tests were defined by levels of ≥ 10.8 IU/ml for IgA-RF and ≥ 9.5 IU/ml for IgM-RF [33].

### HLA-DRB1 Genotyping

High resolution *HLA-DRB1* genotyping was performed as recently described with results showing a higher frequency of SE-containing alleles in African American cases compared to controls [38]. In brief, genotyping was completed through DNA sequencing of exon 2 using the alleleSEQR *HLA-DRB1* reagent kit and protocol (Atria Genetics, South San Francisco, CA). After PCR amplification, sequencing was performed on an ABI 377 automated sequencer (PE Applied Biosystems, Foster City, CA). An additional sequence reaction was performed to analyze codon 86 sequences, thus allowing for the resolution of ambiguous results for some of the exon 2 sequences. The sequences were analyzed using Assign SBT3 Software (Conexio Genomics, Australia), enabling assignment of genotypes based on recent library file of *HLA-DRB1* alleles. This method detects all SE positive alleles. When necessary, group-specific amplifications were performed and sequence analysis conducted to differentiate among alleles within a specific group.

### Statistical Analysis

Dichotomous outcomes including autoantibody status (positive vs. negative), the presence of nodules, and the presence of radiographic erosions were compared by smoking status using the Chi-square test in unadjusted analyses and subsequently by using multivariable logistic regression and adjusting for the presence of SE. In an additional analysis, we examined the association of smoking status with anti-CCP antibody positivity after stratifying subjects by SE status. The odds of having higher autoantibody concentrations or higher baseline erosion scores were compared among groups defined by baseline smoking status using cumulative logistic regression models (using the cumulative logit link function to generate a cumulative odds ratio (OR) [39]) adjusting for SE status; for this analysis outcome variables were categorized into tertiles. The OR and 95% CI reflect the cumulative odds of having an outcome value in the highest tertile vs. the low/medium tertiles *or* an outcome value in either the high/medium tertiles vs. the lowest tertile. In secondary analyses, we also examined the association of cumulative exposure categories (never, < 10, 10–20, and > 20 pack-years) with the outcomes of interest. Of 300 study participants, fourteen subjects were excluded from multivariable analyses due to missing values for SE (n = 10) or smoking status (n = 4). Additional subjects with missing data for nodules (n = 2) and erosions (n = 94) were excluded from multivariable analyses examining the association of smoking with these outcomes. There were an additional six subjects with a history of smoking but missing cumulative exposure data excluded from analyses examining the association of smoking pack-years with the outcomes of interest.

Gene-environment interactions (SE-smoking) were examined in two ways as reviewed elsewhere [40]. Based on recent reports of disease susceptibility [2,6,41], we assessed biologic (additive) interaction between ever smoking (current or past smoking combined) and SE status by examining for evidence of departure from additivity of effects as the interaction criteria as

described by Rothman et al [42]. Using this approach [40,43], we calculated the attributable proportion due to interaction (AP; where an AP = 0 corresponds to no interaction and an AP =1.0 corresponds to 'complete' interaction) and a corresponding 95% confidence interval using the method of Hosemer and Lemeshow [44]. To assess for evidence of statistical (or multiplicative) interaction, we modeled the SE-smoking product terms, using a p-value of < 0.05 as the threshold for significance. All analyses were conducted using SAS v9.1 (SAS Inc., Cary, NC).

## Results

Subject characteristics are summarized in Table 1.

African Americans study subjects were comprised predominantly of women (83%) and had a mean age of approximately 51 years. Almost half of subjects were ever smokers, 42% were SE positive, 27% had baseline radiographic erosions, and 14% had rheumatoid nodules. Only 23 subjects (8%) carried two SE containing alleles. Seropositivity was observed in 62% of subjects for anti-CCP antibody, 67% for IgA-RF, and 70% for IgM-RF.

The associations of smoking status with autoantibody status (positive vs. negative) and the presence of both nodules and erosions are summarized in Table 2.

In univariate or SE-adjusted analyses, there was no association of smoking status with seropositivity for anti-CCP antibody, IgA-RF, or IgM-RF nor was there an association of smoking with the presence of radiographic erosions. While there was no risk associated with past smoking, current smokers were more than twice as likely as never smokers (OR = 2.43; 95% CI 1.13–5.22) to have rheumatoid nodules after adjusting for SE status. In analysis stratified by SE status, there were also no significant associations of smoking status with anti-CCP antibody positivity (Table 2).

Our results were similar when examining the association of cumulative smoking exposure with the dichotomous outcomes of interest. There were no associations of smoking pack-years with autoantibody seropositivity or the presence of radiographic erosions (Table 2). However, compared to never smokers, RA cases with high cumulative exposure (> 20 pack-years) were significantly more likely to have rheumatoid nodules (SE-adjusted OR = 2.65; 95% CI 1.10–6.37) while cases with intermediate levels of exposure (10–20 pack-years) showed a trend towards higher risk (OR = 2.01; 95% CI 0.71–5.75).

There was no association of smoking status with higher serum concentrations of anti-CCP antibody or IgM-RF based on tertiles, nor was there an association with radiographic erosion scores (Table 3). After adjusting for SE and compared to never smokers, current smokers were approximately two-times more likely to have higher concentrations of IgA-RF (OR = 1.74; 95% CI 1.05–2.88). The risk of higher IgA-RF concentrations appeared to be most pronounced in those with more than 20 pack-years of cumulative exposure (SE-adjusted OR = 2.79; 95% CI 1.50–5.17) while those with low and intermediate levels of cumulative exposure displayed no such risk (Table 3).

There was no evidence of statistical (multiplicative) interaction between smoking status (never, past, current) and SE for any of the outcomes examined (either dichotomous or ordered categorical outcomes) with p-values > 0.10 for all smoking-SE product terms (data not shown). Likewise, there was no evidence of significant biologic (additive) interaction between ever smoking and SE in our models examining dichotomous outcomes. In each circumstance, the 95% CI for the attributable proportion (AP) due to interaction included zero. Results from the assessments of possible biologic interactions are summarized in Table 4.

## Discussion

To our knowledge, this is the first study to examine the associations of cigarette smoking with RA-related disease characteristics in African Americans. In this population, current smoking is positively associated with the presence of both rheumatoid nodules and higher serum concentrations of IgA-RF, associations that appear to be independent of underlying SE status and most pronounced in those with higher levels of cumulative tobacco exposure. In contrast, our results suggest that smoking is not a major determinant of IgM-RF or anti-CCP antibody in African Americans with RA, either in terms of seropositivity or higher serum concentrations. In contrast to subjects of European ancestry with both early [16] and established disease [15,17], smoking does not appear to be strongly associated with the presence or extent of baseline radiographic erosions among African Americans with recent-onset RA.

The association of cigarette smoking with rheumatoid nodules and higher IgA-RF concentrations may have important implications in this population. IgA-RF, for instance, has been shown in RA patients of European/Caucasian descent to be an independent predictor of severe joint damage in established disease and has been associated with both poor functional status and extraarticular disease including pulmonary involvement [45–48]. In one study, 80% of RA patients with elevated IgA-RF concentrations had one or more extraarticular disease manifestations compared to only 21% of those with increased concentrations of IgM-RF but normal IgA-RF [48], highly relevant given the association of extraarticular findings with disease-related mortality [49]. Rheumatoid nodules, the most common extraarticular disease manifestation, have been associated with a more than four-fold increased risk of mortality in men with RA from the U.K. Norfolk Arthritis Registry [50].

It remains unknown whether smoking is a risk factor in African Americans for the development of RA (disease characterized by the presence of higher IgA-RF values) or whether smoking or its byproducts stimulate specific autoantibody expression. In a cross-sectional study of subjects of European ancestry without RA, both IgA-RF (34.4%) and IgM-RF (34.1%) were significantly more likely to be positive in smokers compared to non-smokers (21.9%), suggesting that smoking may have a direct effect on humoral immunity irrespective of disease status [51]. Padyukov et al found that smoking increases the risk of developing RF-positive RA but not seronegative RA, a risk that was confined to those positive for SE [6]. Although Padyukov and colleagues did not examine specific RF isotypes or anti-CCP antibody, their results support the existence of SE-smoking interaction in the risk of RF-positive disease. In a more recent study, Klareskog et al found a similar association of smoking for the development of anti-CCP antibody positive disease, a risk also limited to SE-positive subjects [2]. Based on subanalyses in subjects discordant for anti-CCP antibody and RF, their results also suggest that the development of anticitrulline immunity is the ‘primary’ pathogenic event in smokers carrying SE while the development of RF more likely represents a ‘secondary’ phenomenon.

In contrast to recent reports examining disease susceptibility [2,6,41] in Caucasians and the effect of smoking on autoantibody expression [20], we found no evidence to support the role of a SE-smoking interaction in RA disease expression among African Americans. However, our results are consistent with recent findings from three large North American Caucasian RA cohorts, showing no major SE-smoking interaction for anti-CCP antibody expression [52]. In a report of 407 Dutch subjects with recent-onset disease, a history of tobacco exposure was associated with an increased odds of anti-CCP antibody positivity in SE positive RA subjects but not in those negative for SE [20]. In contrast to our study, biologic interaction between SE and smoking in the study of Linn-Rasker et al [20] was defined as being present because the odds of having anti-CCP antibody for subjects having both tobacco exposure and SE (OR = 5.27; 95% CI 2.37–11.80) was higher than the summed odds ratios of subjects with only tobacco exposure (OR = 1.07; 95% CI 0.43–2.65) or SE alone (OR = 2.49; 95% CI 1.18–5.31). In post-hoc

analyses of these data, there was no evidence of statistical interaction (p-value of interaction term = 0.2) while the attributable proportion (AP) due to interaction was estimated to be 0.5 [53,54]. Even with the use of the less stringent definition [20], our results still provide no conclusive evidence of an interaction between smoking and SE for the development of anti-CCP antibody, RF (either IgM or IgA isotype), or radiographic erosions in African Americans with RA (although suggesting the presence of a borderline SE-smoking interaction for the presence of nodules).

Although our study includes the largest African American RA cohort systematically examined to date, it is likely that our analysis was not adequately powered to detect this gene-environment interaction. Because these subjects were recruited primarily from academic rheumatology practices, it is possible that these results are not generalizable to other African American populations with RA. Given the lower prevalence of *HLA-DRB1* alleles in African American cases relative to Caucasians with RA [55], we were not able to adequately examine the impact of SE 'dose' nor were we able to fully evaluate the risk conferred by specific *HLA-DRB1* alleles. This limitation is noteworthy because different SE alleles appear to differ in their interaction with smoking and predisposition to autoantibodies in patients of European ancestry [56]. It is possible that discrepant results observed across different populations (African American vs. Caucasian and/or North American vs. European) may be due to other 'unmeasured' risk factors involved in RA pathogenesis that are more common to select ethnic/racial groups (i.e. additional genes and/or environmental triggers)

In summary, the data presented in this report show that cigarette smoking is associated with select disease characteristics in African Americans with recent onset RA, features that include higher serum concentrations of IgA-RF and the presence of subcutaneous nodules. As reported in other studies of disease susceptibility, the associated risk of smoking appears to be greatest among subjects with the highest levels of cumulative exposure. Further studies that include a larger number of well-characterized cases and controls with extended follow-up will be needed to better define the complex relationship of smoking with pre-disposing risk alleles in RA susceptibility and disease expression in African Americans.

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**Table 1**

Subject characteristics of African Americans with recent onset rheumatoid arthritis (n = 300)

| Characteristic* | Mean (SD) or Number (%) |
|---|---|
| Female sex | 246 (82%) |
| Age, years | 50.6(13.5) |
| Disease duration at baseline visit, months | 12.8 (7.2) |
| Smoking Status | |
| Never | 142 (48%) |
| Past | 66 (22%) |
| Current | 88 (30%) |
| Ever smoked, < 10 pack-years | 63 (22%) |
| Ever smoked, 10–20 pack-years | 34(12%) |
| Ever smoked, > 20 pack-years | 51 (18%) |
| Shared epitope positive | 123 (42%) |
| Anti-CCP antibody positive | 186 (62%) |
| RF-IgA positive | 202 (67%) |
| RF-IgM | 211 (70%) |
| Baseline Radiographic erosions | 51 (27%) |
| Nodules | 41 (14%) |

*Denominator as noted with exception of missing values for shared epitope status (n = 10), age (n = 2), smoking status (n = 4), pack-years of smoking (n = 6) among ever smokers, disease duration (n = 2), nodules (n = 5), and radiographic erosions (n = 108 radiographs unscored at time of analysis); CCP = cyclic citullinated peptide

**Table 2**

Associations of smoking status (current vs. past vs. never) and categories of cumulative smoking exposure (pack-years) with autoantibody status (positive vs. negative), nodules (present vs. absent), and erosive disease (present vs. absent) among African Americans with recent-onset rheumatoid arthritis; all logistic regression models adjusted for presence of shared epitope (SE)[*†]

| | Anti-CCP Positivity | | RF-IgM Positivity | | RF-IgA Positivity | | Radiographic Erosions | | Subcutaneous Nodules | |
|---|---|---|---|---|---|---|---|---|---|---|
| | % | OR (95% CI) | % | OR (95% CI) | % | OR (95% CI) | % | OR (95% CI) | % | OR (95% CI) |
| **Smoking Status** | | **(n = 286)** | | **(n = 286)** | | **(n = 286)** | | **(n = 192)** | | **(n = 284)** |
| Never | 64% | Referent | 70% | Referent | 67% | Referent | 25% | Referent | 10% | Referent |
| Past | 63% | 0.99 (0.53–1.85) | 71% | 1.05 (0.54–2.02) | 69% | 1.11 (0.59–2.10) | 28% | 1.17 (0.52–2.60) | 9% | 0.90 (0.33–2.47) |
| Current | 60% | 0.88 (0.50–1.55) | 70% | 1.05 (0.57–1.91) | 67% | 1.00 (0.56–1.79) | 27% | 1.11 (0.52–2.34) | 21% | 2.43 (1.13–5.22) |
| **Pack- Years[‡]** | | **(n =280)** | | **(n =280)** | | **(n =280)** | | **(n =189)** | | **(n =278)** |
| Never | 64% | Referent | 70% | Referent | 67% | Referent | 25% | Referent | 10% | Referent |
| < 10 | 62% | 0.89 (0.47–1.69) | 70% | 0.97 (0.49–1.90) | 67% | 0.96 (0.50–1.84) | 23% | 0.89 (0.38–2.10) | 12% | 1.13 (0.43–2.97) |
| 10–20 | 58% | 0.83 (0.38–1.82) | 64% | 0.79 (0.35–1.77) | 61% | 0.78 (0.35–1.71) | 24% | 0.94 (0.33–2.66) | 18% | 2.01 (0.71–5.75) |
| >20 | 64% | 1.11 (0.56–1.36) | 78% | 1.65 (0.76–3.58) | 74% | 1.46 (0.70–3.02) | 32% | 1.44 (0.60–3.43) | 22% | 2.65 (1.10–6.37) |

[*] Univariate p-values (Chi-square test) for smoking staus: p = 0.75 for anti-CCP antibody; p = 0.95 for RF-IgM; p = 0.88 for RF-IgA; p = 0.93 for erosions; p = 0.02 for nodules; Univariate p-values for categories of pack-years (Mantel-Haenszel Chi-square test): p = 0.73 for anti-CCP antibody; p = 0.71 for RF-IgM; p = 0.74 for RF-IgA; p = 0.52 for erosions; p = 0.03 for nodules; in analyses stratified by SE status (0 vs. 1 or 2 copies), there was no significant association of current (OR = 0.7; 95% CI 0.3–1.4) or former smoking (OR = 0.9; 95% CI 0.4–1.9) (referent to never smoking) with anti-CCP antibody status in SE negative subjects; likewise, there was no significant association of current (OR = 1.3; 95% CI 0.5–3.4) or former smoking (OR = 1.3; 95% CI 0.5–3.5) with anti-CCP antibody status in SE positive subjects.

[†] CCP = cyclic citrullinated peptide; RF = rheumatoid factor

[‡] Excludes 10 subjects with missing data for SE and 4 subjects missing smoking data; an additional 6 subjects a known smoking history (3 with available erosion scores) were excluded from analysis due to missing values for pack-years of smoking.

## Table 3

Associations of smoking status (current vs. past vs. never) and categories of cumulative smoking exposure (pack-years) with higher tertiles of autoantibody concentrations and modified Sharp erosion scores among African Americans with recent-onset rheumatoid arthritis; cumulative logistic regression models (using cumulative logit link function to generate a cumulative OR and 95% CI) adjusted for presence of shared epitope (SE)*

| | Anti-CCP Antibody OR (95% CI) | RF-IgM OR (95% CI) | RF-IgA OR (95% CI) | Erosion Score OR (95% CI) |
|---|---|---|---|---|
| **Smoking Status** | **(n =286)** | **(n =286)** | **(n =286)** | **(n = 192)** |
| Never | Referent | Referent | Referent | Referent |
| Past | 1.13 (0.65–1.94) | 1.15(0.67–1.98) | 1.51 (0.88–2.61) | 1.24 (0.56–2.71) |
| Current | 1.11 (0.67–1.84) | 1.27 (0.77–2.09) | 1.74 (1.05–2.88) | 1.05 (0.50–2.21) |
| **Pack-Yearst†** | **(n =280)** | **(n =280)** | **(n =280)** | **(n =189)** |
| Never | Referent | Referent | Referent | Referent |
| < 10 | 1.21 (0.69–2.12) | 1.05 (0.60–1.83) | 1.25 (0.71–2.18) | 0.92 (0.40–2.14) |
| 10–20 | 0.87 (0.43–1.77) | 1.05 (0.52–2.11) | 1.30 (0.64–2.62) | 0.98 (0.35–2.71) |
| > 20 | 1.27 (0.70–2.78) | 1.65 (0.90–3.02) | 2.79 (1.50–5.17) | 1.18 (0.61–2.26) |

*CCP = cyclic citrullinated peptide; RF = rheumatoid factor; outcome variables categorized into tertiles; OR (and 95% CI) reflects the cumulative odds of having an outcome value in the highest tertile vs. the low/medium tertiles *or* an outcome value in either the high/medium tertiles vs. the lowest tertile.

†Six subjects a known smoking history (3 with available erosion scores) were excluded from analysis due to missing values for pack-years of smoking

**Table 4**

Assessment of biologic (additive) interaction between ever smoking and HLA-DRB1 shared epitope (SE) status among African Americans with recent-onset rheumatoid arthritis (RA); assessed using departure from additivity of effects as the interaction criteria*

| | Never Smoker/SE negative | Never smoker/SE positive OR (95% CI) | Ever smoker/SE negative OR (95% CI) | Ever smoker/SE positive OR (95% CI) | Attributable Proportion due to interaction (95% CI) |
|---|---|---|---|---|---|
| Anti-CCP Positivity (n = 286) | Referent | 1.74 (0.85–3.55) | 0.76 (0.41–1.41) | 2.23 (1.07–4.67) | 0.33 (−0.30–0.96) |
| RF-IgM Positivity (n = 286) | Referent | 1.85 (0.87–3.95) | 0.96 (0.51–1.83) | 2.25 (1.02–4.94) | 0.44 (−0.57–0.95) |
| RF-IgA Positivity (n = 286) | Referent | 1.51 (0.73–3.13) | 1.02 (0.54–1.93) | 1.64 (0.79–3.43) | 0.07 (−0.77–0.91) |
| Erosive Status (n = 192) | Referent | 0.91 (0.34–2.41) | 0.97 (0.40–2.33) | 1.24 (0.50–3.08) | 0.29 (−0.69–1.00) |
| Nodules (n = 284) | Referent | 0.94 (0.31–2.89) | 1.23 (0.47–3.24) | 2.30 (0.89–5.98) | 0.49 (−0.17–1.00) |

*No evidence of interaction when attributable proportion (AP) = 0; CCP = cyclic citrullinated peptide; RF = rheumatoid factor

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