Serum retinol, the acute phase response, and the apparent misclassification of vitamin A status in the third National Health and Nutrition Examination Survey\textsuperscript{1–3}

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ABSTRACT
Background: Serum retinol decreases transiently during the acute phase response and can thus result in misclassification of vitamin A status.
Objective: Our objective was to determine the prevalence of acute phase response activation in a representative sample of the US population, identify the factors associated with this activation, and determine whether persons with an active acute phase response have lower serum retinol concentrations.
Design: Data from the third National Health and Nutrition Examination Survey (NHANES III) were analyzed. A serum C-reactive protein (CRP) concentration $\geq 10$ mg/L indicated an active acute phase response.
Results: Mean serum retinol was lowest in subjects aged $<10$ y and increased with age. Concentrations were higher in males than in females aged 20–59 y. The prevalence of a CRP concentration $\geq 10$ mg/L was lowest in subjects aged $<20$ y ($\leq 4\%$) and increased with age to a maximum of nearly 15%. An elevated CRP concentration was 2.4-fold greater in females than in males aged 20–69 y. Serum retinol was lower in subjects with elevated CRP concentrations.
Conclusions: Serum retinol increases with age and males have higher mean values than do females aged 20–59 y. The prevalence of a CRP concentration $\geq 10$ mg/L also increases with age, is 2-fold greater in females than in males aged 20–69 y, and is associated with common inflammatory conditions. Thus, inflammation appeared to contribute to the misclassification of vitamin A status in the NHANES III population, and serum CRP is useful in identifying subjects who may be misclassified. Am J Clin Nutr 2000;72:1170–8.

KEY WORDS Retinol, vitamin A, acute phase response, C-reactive protein, CRP, third National Health and Nutrition Examination Survey, NHANES III, infection, inflammation

INTRODUCTION
Serum retinol concentrations are normally maintained within a narrow range in individuals with adequate liver vitamin A stores. When liver stores are depleted, serum retinol concentrations decrease. Thus, serum retinol is a useful indicator of vitamin A status and can be used to identify subjects with low or depleted liver vitamin A stores (1, 2). However, serum retinol concentrations decrease transiently during the acute phase response to infection (3–7). Such decreases do not reflect changes in liver vitamin A stores and, thus, can interfere with the use of serum retinol as an indicator of vitamin A status. Similar changes also occur with other micronutrients, including iron and zinc (8). In populations in whom infections are highly prevalent, such as young children living in poor areas of developing countries, this phenomenon can be a source of false-positive results in surveys designed to determine the prevalence of micronutrient deficiencies (9).

The decrease in serum retinol seen during infection is an integral part of the acute phase response, which is induced not only by infection but also by tissue-damaging trauma such as surgery or by chronic inflammatory conditions such as arthritis (10–12). During the acute phase response the synthesis of negative acute phase proteins, such as retinol binding protein (RBP), decreases (13). This contributes to the lower serum concentration of these proteins, although other factors also contribute (4, 14). A hallmark of the acute phase response is the rapid and substantial increase in synthesis of positive acute phase proteins, which are involved in controlling infection and resolving tissue damage caused by inflammation (11). For example, serum C-reactive protein (CRP) concentrations increase within a few hours (15) and reach peak concentrations as much as 1000-fold above baseline within a few days (12). The half-life of CRP in serum is 6 h; thus, serum CRP concentrations decrease to normal within days after the proinflammatory stimulus has been eliminated (16).

Serum retinol has been used to assess the vitamin A status of the US population in the National Health and Nutrition Examination Surveys (NHANES) and the Hispanic HANES...
The effect of the acute phase response on serum retinol has not been evaluated in these surveys. Because serum CRP concentrations were measured in the third NHANES (NHANES III) survey, we evaluated the effect of elevated serum CRP concentrations (≥10 mg/L) (12) on serum retinol concentrations and the percentage of subjects with concentrations below the cutoff value used to identify those who are likely to have marginal liver vitamin A stores (≤1.05 μmol/L) (1, 2, 20). In addition, we evaluated the prevalence of elevated CRP concentrations in this population and examined the association of infectious and inflammatory conditions with the risk of having elevated serum CRP concentrations.

SUBJECTS AND METHODS

NHANES III data

NHANES III was a representative survey of the US population and was conducted in 2 phases from 1988 to 1994 (21). Each phase included a random sample of the noninstitutionalized US population living in households. The total sample size from which subjects were recruited was ≈40,000 persons aged >2 mo and was clustered in 81 counties. Some groups were oversampled (eg, children aged <5 y and adults aged >60 y). Initial contact was made at selected households to identify subjects who were then recruited into the study. Information was collected in the household during a subsequent appointment from ≈34,000 subjects. After these household interviews were conducted, subjects were asked to come to a mobile examination center for a physical examination and blood draw; ≈31,000 subjects complied. Serum retinol and CRP concentrations were available from 10,617 males and 11,729 females aged ≥4 y.

NHANES III data were obtained on CD-ROM (series 11, no. 1A, July 1997 and no. 2A, April 1998) from the Data Dissemination Branch, National Center for Heath Statistics (21). The following variables (and variable names) were used in this analysis: serum retinol concentration (VAPS1), serum C-reactive protein concentration (CRP), age (HSAGE1), sex (HSSEX), race (DMARAC1), body mass index (BMI, in kg/m2; BMBMI), examining physician’s diagnosis of a possible active infection (PEP13C), self-report of acute infection in the past few days (PEP13C), self-report of respiratory infection in the past 3 wk (PEP4Q), self-report of respiratory infection in the past 3 wk (PEP4Q), self-report of cigarette smoking status (HAR3, subset of subjects aged ≥18 y), previous physician’s diagnosis of chronic bronchitis that the subject indicates is still present (HAC1F/2F and HYE1H/3H), previous physician’s diagnosis of asthma that the subject indicates is still present (HAC1E/2E and HYE1G/3G), previous physician’s diagnosis of hay fever that the subject indicates is still present (HAC1F/2F and HYE1H/3H), previous physician’s diagnosis of arthritis (HCA1A/B, rheumatoid arthritis, osteoarthritis, or arthritis of unknown type), diabetes (HAD1), heart attack (HAF10), osteoporosis (HAG11), and depression (PEP10). The presence of hypertension was assessed by measuring blood pressure (systolic: ≥160 mm Hg, PEPMNK1R; diastolic: ≥95 mm Hg, PEPMNK5R) (22, 23). Dental examination variables from subjects aged ≥1 y representing gingival bleeding (DEPUGN and DEPLGN) in association with the 2 surfaces of 7 upper and 7 lower teeth were used to create a gingivitis score: no gingivitis to mild gingivitis, bleeding at 0–5 sites, moderate-to-severe gingivitis, and bleeding at >5 sites. Subjects for whom 12 of the 28 sites could not be examined were excluded. A similar classification was used previously (24). A summary severity variable (0, no arthritis manifestations in examined joints; 1, 1 joint involved; 2, 2 joints involved; 3, 3 joints involved; and 4, 4 joints involved) was created for symptomatic arthritis in subjects aged ≥60 y by using physical examination variables that assess arthritis manifestations at the wrist (PEP4A), knuckles (PEP4B), great toe (PEP10), and knees (PEP10B). Serum cotinine was used as a marker to determine smoking status; subjects with a concentration >10 μg/L were considered to be current smokers (25). An inverse serum rheumatoid factor antibody titer ≥40 was used to identify subjects aged >60 y with rheumatoid arthritis (26–28). A positive serum Helicobacter pylori antibody titer was used to identify subjects with past or current H. pylori infection. This assay was performed only on surplus sera from children and adolescents aged 6–19 y.

Data analysis

Data were analyzed by using SAS (version 6.12; SAS Institute Inc, Cary, NC). Weights obtained at the medical examination center and at the subjects’ homes were used to determine mean serum retinol concentrations and prevalence estimates for elevated serum CRP that are representative of the US population. Weights were not used in other analyses because representative population estimates were not needed. The primary analyses were designed to examine the associations of the main outcome variables—serum retinol and CRP concentrations—with infectious and inflammatory conditions after adjustment for demographic variables, primarily age and sex. Descriptive statistics were computed for all of the demographic and dependent variables, including means, medians, and SDs for continuous data and frequency distributions for each of the categorical variables. Initial analyses were based on frequency tables of the outcomes and risk factors; chi-square tests, Student’s t tests, and logistic regression models were used to gauge the degree of association. Age-adjusted odds ratios (ORs) were calculated by using a logistic regression model. Results of the exploratory analyses were used to make informed decisions about which risk factors to include as potential covariates in the final analyses. The major hypotheses were evaluated by using logistic regression models with binary outcomes. Stepwise regression techniques were applied to assist in the selection of variables for the final multivariate model. Main effects, which were adjusted for age, were included in each model. Additionally, interaction terms were investigated when identified. The final multivariate models were created separately for subjects aged <17 y compared with those aged ≥17 y and separately for the 2 sexes. These age groupings were selected because questions related to chronic disease variables were typically asked only of subjects aged ≥17 y. The 0.05 level of significance was used for all statistical tests.

RESULTS

Serum retinol, age, and sex

Serum retinol concentrations increased significantly with age and reached a plateau in males by the fifth decade of life (aged 40–49 y) (Figure 1). Serum retinol in females increased during the first 2 decades, remained constant between 20 and 49 y of age, then increased by 60–69 y of age to values equivalent to those for men. Serum retinol concentrations were significantly higher in males than in females between 20 and 59 y of age, but
not in younger or older subjects (Figure 1). Essentially the same curves were seen when only subjects with CRP values <10 mg/L were analyzed (data not shown), indicating that the differences between the sexes were not related to differences in the prevalence of elevated CRP concentrations.

These age-related increases in serum retinol, as well as the higher values in middle-aged men, were seen independently in the 3 racial groups used in NHANES III: white, black, and other, which includes Asians, Pacific Islanders, and American Indians. Although no significant differences were seen among the 3 racial groups in any individual decade, when comparisons were made among males and females separately, the overall difference between blacks and whites was significant for both males and females (P < 0.001), as was true when whites were compared with other (P < 0.001). There were no significant differences between the blacks and others for either males or females. The weighted, mean serum retinol concentrations for multiple comparisons among these age and sex categories). Values were weighted to correct for sampling and nonresponse errors and represent the US population. The numbers of male and female subjects, respectively, in decades 1–9 were 1733 and 1682, 1859 and 2035, 1542 and 1746, 1377 and 1741, 1168 and 1295, 826 and 958, 1130 and 1105, 804 and 924, and 591 and 632, respectively.

CRP and serum retinol

A serum CRP concentration ≥10 mg/L is widely accepted as indicative of an active acute phase response (12), although lower cutoff values have sometimes been used (28, 29). In the NHANES III subjects, we found that mean serum retinol concentrations were significantly lower in subjects with serum CRP concentrations ≥10 mg/L than they were in subjects with normal CRP concentrations. This was true for males in all age categories (except those aged 80–89 y) and for females aged >30 y (except those aged 50–59 y) (Figure 2). The mean difference between subjects with and without elevated CRP concentrations was somewhat greater for males (0.26 ± 0.27 μmol/L, when compared by decade) than for females (0.17 ± 0.08 μmol/L; P = 0.024 by paired Student’s t test). This difference was most evident in those aged 20–59 y (Figure 2) and was not a result of males with CRP concentrations ≥10 mg/L having higher mean CRP concentrations than females because mean values for those with CRP concentrations ≥10 mg/L did not differ between the sexes in these decades (data not shown). This differential association of CRP with serum retinol by sex was confirmed by multiple regression analysis: when age (in y; β = 0.0137, P < 0.0001), sex (female = 1; β = −0.15421, P < 0.0001), and CRP concentrations (mg/L; β = −0.07367, P < 0.0001) were used to predict serum retinol concentrations, significant interaction terms were found for age × CRP (β = −0.00075116, P = 0.0003), age × sex (β = −0.00074976, P = 0.0103), and sex × CRP (β = 0.0384, P < 0.0001). The intercept for the equation was 1.43995 and the r² value was 0.2787. The significant sex × CRP interaction term confirmed that the association of CRP with serum retinol was different between males and females, although the fundamental nature of the association (elevated serum CRP was associated with lower serum retinol) was the same.

In the evaluation of vitamin A status in well-nourished populations, a cutoff value of ≤1.05 μmol/L has been used to identify subjects with inadequate vitamin A stores (1, 2, 20). Because subjects with elevated serum CRP concentrations have lower mean serum retinol concentrations than do those with normal vitamin A stores, this study suggested that CRP might be associated with vitamin A deficiency.
TABLE 1
Percentage of NHANES III subjects (number with elevated CRP/total number of subjects) with serum retinol concentrations ≤1.05 \( \mu \text{mol/L} \) in those with elevated (≥10 mg/L) and normal (<10 mg/L) serum CRP concentrations.

<table>
<thead>
<tr>
<th>Age group</th>
<th>Male CRP &lt; 10 mg/L</th>
<th>Male CRP ≥ 10 mg/L</th>
<th>Female CRP &lt; 10 mg/L</th>
<th>Female CRP ≥ 10 mg/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤10 y</td>
<td>23.6 (311/1320)</td>
<td>66.7 (18/27)</td>
<td>22.7 (280/1232)</td>
<td>68.8 (33/48)</td>
</tr>
<tr>
<td>10–19 y</td>
<td>5.1 (91/1795)</td>
<td>94.9 (134/352)</td>
<td>5.4 (106/1955)</td>
<td>94.6 (178/188)</td>
</tr>
<tr>
<td>≥20 y</td>
<td>0.73 (50/6886)</td>
<td>99.3 (23546)</td>
<td>2.4 (175/7374)</td>
<td>97.6 (70/1052)</td>
</tr>
</tbody>
</table>

1NHANES III, third National Health and Nutrition Examination Survey; CRP, C-reactive protein.

2Significantly different from subjects < 4 y of age.

3Significantly different from the same sex with CRP < 10 mg/L, \( P < 0.001 \).

4Significantly different from males with CRP < 10 mg/L, \( P < 0.001 \).

Concentrations, we evaluated the effect of elevated CRP concentrations on categorization using 1.05 \( \mu \text{mol/L} \) as the cutoff value for serum retinol. This evaluation was done initially by age (ie, in decades), but was then collapsed into 3 age groups. The percentage of subjects with serum retinol concentrations ≤1.05 \( \mu \text{mol/L} \) was highest in the youngest age group, intermediate in those aged 11–20 y, and lowest in subjects aged >20 y (Table 1). The percentage of subjects with serum retinol concentrations below this cutoff was significantly greater in those with elevated CRP concentrations in all age categories and in both sexes. When ORs were calculated and adjusted for age within these age categories (in y), subjects with elevated serum CRP concentrations were from 3.1- to 8.9-fold more likely to have a serum retinol concentration ≤1.05 \( \mu \text{mol/L} \) (Table 2).

Age, sex, and serum CRP

The percentage of subjects with elevated serum CRP concentrations increased from childhood through late middle age, with significantly more females than males having elevated values between 20 and 59 y of age (Figure 3). Fewer than 4% of subjects aged <20 y had elevated CRP values, but the prevalence among females increased in 20–29-y-olds to 7.6%, then increased more gradually, peaking at 14.4% in 60–69-y-olds. Among males, prevalences increased slowly to 14.0% in 80–89-y-olds. On average, prevalences or elevated CRP concentrations were 2.4-fold (range: 1.8–3.2-fold) greater in females than in males aged 20–59 y. Thus, women were more likely to have elevated CRP concentrations compared with those with normal (<10 mg/L) serum CRP concentrations. Subjects who reported a previous physician’s diagnosis of arthritis (no arthritis, rheumatoid arthritis, osteoarthritis, or “don’t know”) also had elevated serum CRP concentrations. Percentages of subjects with elevated CRP concentrations in these categories were 8.3% (1103/13313), 19.5% (135/692; \( P < 0.001 \) compared with the group with no arthritis), 13.6% (94/689; \( P < 0.001 \)), and 13.8% (295/2137; \( P < 0.001 \)). The prevalences for the groups with rheumatoid arthritis and osteoarthritis were significantly different from one another (\( P = 0.0043 \)). The prevalence of elevated CRP concentrations in subjects with any type of arthritis was 14.9% (524/3518), which was significantly greater than the

Infection and serum CRP

Several variables indicating active or recent infection were associated with serum CRP concentrations ≥10 mg/L, as shown in Table 3. A physician’s diagnosis of a possible active infection; a subject’s self-report of having a cough, cold, or other acute illness in the previous few days; and a “yes” response to the question “In the past 3 wk have you had any respiratory infections, such as the flu, pneumonia, bronchitis or a severe cold” were all associated with an increased risk of having elevated CRP concentrations. Subjects with gingivitis were also more likely to have elevated CRP concentrations, which is consistent with previous findings (24, 30). Although \( H. \text{pylori} \) infection is associated with gastric and duodenal inflammation, no association of past or present \( H. \text{pylori} \) infection (as indicated by a positive antibody titer) was seen with elevated CRP concentrations in these subjects.

Arthritis and serum CRP

Chronic, noninfectious inflammatory conditions may also activate the acute phase response and thus elevate serum CRP concentrations. Clinically active rheumatoid arthritis was shown to be associated with elevated CRP concentrations (31). Subjects aged >60 y in the NHANES III study population were examined for signs of arthritis. When subjects were scored by a physician for the presence of tenderness, swelling, or pain in the wrist, knuckle, great toe, and knee joints, the percentages with such signs in 0, 1, 2, 3, or 4 of these sites who also had elevated serum CRP concentrations were 10.7% (108/1013), 11.1% (292/2663), 13.0% (20/154), 22.4% (49/219), and 16.8% (16/95) \( (P < 0.001) \), respectively. When corrected for age, the OR for males was significantly >1.0, whereas the association for females was not significant (Table 3). Subjects who reported a previous physician’s diagnosis of arthritis (no arthritis, rheumatoid arthritis, osteoarthritis, or “don’t know”) also had elevated serum CRP concentrations. Percentages of subjects with elevated CRP concentrations in these categories were 8.3% (1103/13313), 19.5% (135/692; \( P < 0.001 \) compared with the group with no arthritis), 13.6% (94/689; \( P < 0.001 \)), and 13.8% (295/2137; \( P < 0.001 \)). The prevalences for the groups with rheumatoid arthritis and osteoarthritis were significantly different from one another (\( P = 0.0043 \)). The prevalence of elevated CRP concentrations in subjects with any type of arthritis was 14.9% (524/3518), which was significantly greater than the

TABLE 2
Odds ratios (adjusted for age in y) of having a serum retinol concentration ≤1.05 \( \mu \text{mol/L} \) in subjects with elevated serum CRP (≥10 mg/L) concentrations compared with those with normal (<10 mg/L) serum CRP concentrations in the NHANES III population.

<table>
<thead>
<tr>
<th>Age group</th>
<th>Male Odds ratio 95% CI</th>
<th>Female Odds ratio 95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤10 y</td>
<td>6.62 2.91, 15.04</td>
<td>8.56 4.51, 16.25</td>
</tr>
<tr>
<td>10–19 y</td>
<td>8.91 4.24, 18.72</td>
<td>6.26 3.47, 11.30</td>
</tr>
<tr>
<td>≥20 y</td>
<td>5.41 3.21, 9.10</td>
<td>3.13 2.35, 4.17</td>
</tr>
</tbody>
</table>

1CRP, C-reactive protein; NHANES III, third National Health and Nutrition Examination Survey. Sample sizes are the same as in Table 1.

2Blood was not collected from subjects <4 y of age.
In agreement with these findings, a previous diagnosis of a 
vaccinated CRP concentrations in these subjects (Table 3). Heart disease 
However, hypertension was not significantly associated with ele-
induce the acute phase response, as was reported previously (34).

Serum CRP 
Hypertension, heart disease, body weight, diabetes, and 
and chronic bronchitis the risk was seen only in females. 

elevated serum CRP (Table 1).

Three respiratory conditions—emphysema, chronic bronchitis, 
and asthma—were also found to be associated with elevated serum 
CRP concentrations (Table 3). For emphysema the increased risk 
concentrations in females but not in males. Conversely, respira-
tory infection was associated with elevated CRP in males but not 
in females. BMI was positively associated with elevated CRP 
concentrations in both sexes. Gingivitis and smoking were not 
included in the initial stepwise analysis because missing data for 
these variables greatly reduced the sample size. However, when 
these variables were individually added to the final model 
(Table 4), smoking showed a significant positive association with 
elevated serum CRP concentrations in females (OR: 7.53; 95% 
CI: 1.58, 35.94; n = 795) but not in males, whereas gingivitis did 
not show a significant association in either sex (data not shown).

The association of infectious and chronic disease variables 
heart attack in the NHANES III subjects was associated with a 
significantly higher risk of elevated CRP concentrations (Table 3).

Two recent studies found a positive association between BMI 
and elevated serum CRP concentrations (28, 36). Using the 
NHANES III data set, Visser et al (28) reported that subjects aged 
> 17 y with a BMI > 30 had a significantly higher prevalence of 
serum CRP concentrations ≥ 10 mg/L than did other subjects, with 
a much stronger association in females. In subjects aged ≥ 17 y, we 
found that the prevalences of elevated CRP concentrations in 
subjects with BMIs < 18.5, 18.5–24.9, 25.0–29.9, and ≥ 30 were 2.6% 
(86/3355), 5.1% (430/8414), 7.9% (487/6155), and 17.9% 
(772/4317) (P < 0.001). The OR was significantly > 1.0 for 
females, but not for males, when adjusted for age (Table 3). This 
association was also significant in subjects aged < 17 y (Table 4).

We found that female but not male diabetic subjects were at 
increased risk of elevated CRP concentrations (Table 3). To deter-
de whether subjects with type 1 diabetes were at higher risk of 
elevated serum CRP concentrations than were diabetic subjects 
who did not use insulin, ORs were calculated among subjects with 
diabetes. When corrected for age, the OR for male insulin users 
(1.10; range: 0.63–1.94) was not significantly different from 1.0. 
In females, however, the OR was 2.03 (1.43–2.90), showing a 
significant risk of elevated CRP concentrations (P < 0.001).

Other inflammatory conditions and serum CRP

Not all types of chronic inflammation were associated with 
elevated serum CRP concentrations. Neither the presence of der-
matitis (as diagnosed by a physician during the NHANES III 
physical examination) nor the subject’s report of hay fever was 
associated with elevated CRP concentrations (Table 3).

Multiple logistic regression: infection, inflammation, and 
serum CRP

We used multiple logistic regression analysis to determine 
whether these infectious and chronic diseases were associated 
with elevated serum CRP concentrations. Information on several 
chronic diseases (diabetes, heart disease, hypertension, arthritis, 
and gout) was not collected for subjects aged < 17 y, so data from 
this group of subjects were analyzed separately. Variables that 
contributed significantly to prediction of the risk of elevated 
serum CRP in either males or females were first selected by using 
stepwise regression from among the following variables: physi-
can diagnosis of infection, self-report of acute infection, and 
self-report of respiratory infection, bronchitis, asthma, BMI, hay 
fever, or dermatitis. Age (in y) was forced to remain in the model. 
The selected variables were then included in a single model. In 
this final model (Table 4), a current or recent infection was asso-
ciated with an increased risk of elevated serum CRP. A diagnosis 
of asthma was also significantly associated with elevated CRP 
concentrations in females but not in males. Conversely, respira-
tory infection was associated with elevated CRP in males but not 
in females. BMI was positively associated with elevated CRP 
concentrations in both sexes. Gingivitis and smoking were not 
included in the initial stepwise analysis because missing data for 
these variables greatly reduced the sample size. However, when 
these variables were individually added to the final model 
(Table 4), smoking showed a significant positive association with 
elevated serum CRP concentrations in females (OR: 7.53; 95% 
CI: 1.58, 35.94; n = 795) but not in males, whereas gingivitis did 
not show a significant association in either sex (data not shown).

The association of infectious and chronic disease variables 

prevalence in the no-arthritis control group (P < 0.001). ORs 
calculated by using the latter variable indicated a significantly 
increased risk of elevated serum CRP concentrations in both 
males and females (Table 3). In addition, serum rheumatoid 
factor antibody was measured as an indicator of rheumatoid ar-
titis in subjects aged ≥ 60 y. Those with an inverse titer ≥ 40 
were at increased risk of elevated serum CRP concentrations 
(Table 3). Subjects reporting a diagnosis of gout, an inflamma-
tory joint disease caused by deposition of uric acid within joints 
(32), also had an increased risk of elevated CRP concentrations.

Smoking, chronic respiratory disease, and serum CRP

Smoking has been shown to be associated with elevated 
serum CRP concentrations, presumably because of the inflam-
mation caused by damage to lung tissue (33). When using self-
reported smoking to categorize subjects, we found no difference 
in the percentage of subjects with elevated serum CRP concen-
trations (data not shown). However, when a serum cotinine con-
centration > 10 μg/L was used as a marker of active smoking, 
both male and female smokers had a significantly greater risk of 
elevated serum CRP (Table 1).

Three respiratory conditions—emphysema, chronic bronchitis, 
and asthma—were also found to be associated with elevated serum 
CRP concentrations (Table 3). For emphysema the increased risk 
of elevated serum CRP was seen only in males, whereas for asthma and chronic bronchitis the risk was seen only in females.

Hypertension, heart disease, body weight, diabetes, and 
serum CRP

Hypertension may cause damage to blood vessels and thus 
induce the acute phase response, as was reported previously (34). 
However, hypertension was not significantly associated with ele-
vated CRP concentrations in these subjects (Table 3). Heart disease 
was also shown to be associated with elevated CRP concentrations 
(35). In agreement with these findings, a previous diagnosis of a

FIGURE 3. Mean (± SD) weighted percentages of male (○) and 
female (■) subjects with elevated serum C-reactive protein (CRP) 
concentrations (≥ 15 mg/L) in the third National Health and Nutrition Ex-
amination Survey population plotted by age in decades (decade 1: 4–9 y; 
decades 2–9: 10–19 y, 20–29 y, and so on). *Significantly different from 
males, P < 0.005 (Bonferroni adjustment for multiple comparisons; n = 9 
comparisons). Values were weighted to correct for sampling and nonre-
sponse errors and represent the US population. The numbers of subjects 
given in the legend to Figure 2.
with elevated serum CRP concentrations was also examined for subjects aged ≥17 y (Table 5). Variables for the final model were again selected by using stepwise regression from among the following variables: physician’s diagnosis of infection, self-report of acute infection, self-report of respiratory infection, self-report of a physician’s diagnosis of arthritis, smoking status, emphysema, bronchitis, asthma, hypertension, heart disease, BMI category, diabetes, gout, hay fever, and dermatitis. In the stepwise analysis, hypertension was significantly associated with the risk of elevated serum CRP concentrations for males but was not significant when included in the model shown in Table 5. Thus, hypertension was dropped from the final model. Variables that were significantly associated with elevated serum CRP again included the measures of infectious disease. In addition, chronic disease variables that retained their significant association with elevated serum CRP included arthritis, gout, chronic bronchitis, BMI, and diabetes. When gingivitis and smoking were individually added to the model, gingivitis was significantly associated with elevated CRP in males (OR: 1.91; 95% CI: 1.31, 2.78; n = 3935), but smoking was not significantly associated with elevated CRP in either sex (data not shown).

**DISCUSSION**

Serum retinol concentrations increased gradually with age in both males and females. This increase may reflect larger vitamin A stores in older subjects, but this association has not been shown directly. Women had lower serum retinol concentrations than did men during early adulthood and middle age, which may reflect lower liver stores in women but could also be due to differences in retinol metabolism. Serum RBP concentrations fluctuate during the menstrual cycle and increase with estrogen therapy (37), and may explain the lower concentrations in women.

In the NHANES III subjects, a serum CRP concentration ≥10 mg/L was used as the diagnostic cutpoint for CRP measurements. There was a higher prevalence of elevated CRP concentrations >10 mg/L (3–7). In our study, the difference in CRP concentrations had higher values than did women. This difference was not due to the fact that men with elevated serum concentrations were more likely to have marginal vitamin A stores, when a serum retinol concentration ≤1.05 μmol/L was used as the diagnostic cutpoint.
TABLE 4
Multiple logistic regression model to predict the risk of elevated serum CRP concentrations (≥10 mg/L) in subjects <17 y of age from the NHANES III population.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Male (n = 1610)</th>
<th>Female (n = 1718)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (in y)</td>
<td>Odds ratio</td>
<td>95% CI</td>
</tr>
<tr>
<td>18</td>
<td>0.95</td>
<td>0.83, 1.10</td>
</tr>
<tr>
<td>Physician diagnosis of possible active infection</td>
<td>2.58</td>
<td>0.96, 8.64</td>
</tr>
<tr>
<td>Self-report of acute infection in past few days</td>
<td>2.17(^2)</td>
<td>1.04, 4.53</td>
</tr>
<tr>
<td>Self-report of respiratory infection in past 3 wk</td>
<td>3.38(^2)</td>
<td>1.38, 8.29</td>
</tr>
<tr>
<td>Self-report of asthma diagnosis</td>
<td>0.84</td>
<td>0.27, 2.58</td>
</tr>
<tr>
<td>BMI (&lt;18.5, 18.5–24.99, 25–29.99, ≥30)(^3)</td>
<td>2.23(^2)</td>
<td>1.54, 3.23</td>
</tr>
</tbody>
</table>

\(^1\) CRP, C-reactive protein; NHANES III, National Health and Nutrition Examination Survey.

\(^2\) Significantly different from 1.00, P < 0.05.

\(^3\) Odds ratio indicates risk associated with progression from one BMI (in kg/m\(^2\)) category to the next.

TABLE 5
Multiple logistic regression model to predict the risk of elevated serum CRP concentrations (≥10 mg/L) in subjects ≥17 y of age from the NHANES III population.

<table>
<thead>
<tr>
<th>Variable remaining in equation</th>
<th>Males (n = 6991)</th>
<th>Females (n = 7714)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (in decades)</td>
<td>Odds ratio</td>
<td>95% CI</td>
</tr>
<tr>
<td>18</td>
<td>1.30(^2)</td>
<td>1.23, 1.37</td>
</tr>
<tr>
<td>Physician diagnosis of possible active infection</td>
<td>2.78(^2)</td>
<td>1.84, 4.21</td>
</tr>
<tr>
<td>Self-report of acute infection in past few days</td>
<td>2.24(^2)</td>
<td>1.80, 2.78</td>
</tr>
<tr>
<td>Self-report of arthritis diagnosis (≥17 y of age)</td>
<td>1.16</td>
<td>0.90, 1.48</td>
</tr>
<tr>
<td>Self-report of gout diagnosis</td>
<td>1.97(^2)</td>
<td>1.38, 2.82</td>
</tr>
<tr>
<td>Self-report of chronic bronchitis diagnosis</td>
<td>1.07</td>
<td>0.65, 1.78</td>
</tr>
<tr>
<td>BMI (&lt;18.5, 18.5–24.99, 25–29.99, ≥30)(^3)</td>
<td>1.18(^2)</td>
<td>1.03, 1.34</td>
</tr>
<tr>
<td>Self-report of diabetes diagnosis(^1)</td>
<td>1.01</td>
<td>0.72, 1.42</td>
</tr>
</tbody>
</table>

\(^1\) CRP, C-reactive protein; NHANES III, National Health and Nutrition Examination Survey.

\(^2\) Significantly different from 1.00, P < 0.05.

\(^3\) Odds ratio indicates risk associated with progression from one BMI (in kg/m\(^2\)) category to the next.

criterion. Although it is possible that these subjects did have marginal vitamin A stores, it is more likely that serum retinol concentrations decreased as a result of the acute phase response. This argument is buttressed by the fact that many studies have shown that low serum retinol concentrations observed during an acute infection rebound to (presumed) preinfection concentrations after recovery (3). Although similar observations have not been made during chronic inflammatory diseases, the data linking decreases in serum retinol to an active acute phase response are consistent and it is likely that such conditions complicate the assessment of vitamin A status, as do acute infections.

Women aged 20–59 y have a higher prevalence of serum CRP concentrations ≥10 mg/L than do men. It is possible that this difference is of endocrine origin. Healthy women (CRP < 10 mg/L) are known to have slightly higher serum CRP concentrations (∼1 mg/L) during the midcycle and luteal phases of the menstrual cycle than during the follicular phase (38), but the magnitude of this difference is not sufficient to explain the difference in prevalence of elevated serum CRP concentrations observed between men and women in the present study. It is also known that women have higher prevalences of certain chronic and autoimmune diseases than do men (39). This underlying difference in immune function may be related to the different prevalences of elevated serum CRP observed in the present study.

The acute phase response was activated in both male and female NHANES III subjects with either acute (eg, cold) or chronic (eg, gingivitis) infections. One exception to this pattern was the case of recent respiratory infections in subjects aged <17 y, in whom the risk of elevated serum CRP was limited to males. Interestingly, in this same age group, the risk of elevated CRP in those with asthma was limited to females.

Smoking and chronic lung disease—emphysema, chronic bronchitis, and asthma—were associated with an increased risk of elevated serum CRP in the present study. Smoking was a significant risk factor in both sexes, whereas an association of emphysema with elevated serum CRP was observed in males only. Conversely, asthma and chronic bronchitis were risk factors in females only. Although the reasons for these sex differences were not defined, there are ample precedents for a significant association of sex with lung function and respiratory disease (39).

Both rheumatoid arthritis and osteoarthritis were positively associated with elevated serum CRP. This association was seen in both males and females when several markers of disease were assessed. Arthritis occurs primarily in older adults and its high prevalence in the US population (40% of those aged ≥65 y) (40) suggests that it may be an important cause of elevated CRP concentrations in older adults.

Heart disease, elevated BMI, and diabetes were all associated with the risk of elevated serum CRP concentrations in the NHANES III subjects, although hypertension, which was shown previously to be associated with elevated CRP concentrations (34), was not. Report of a previous heart attack was a significant predictor of elevated CRP concentrations in both males and females and presumably represents the inflammatory events.
associated with plaque formation in the coronary arteries (35). This association may also be the reason for the increased risk of elevated serum CRP concentrations observed in female but not in male diabetic subjects because diabetes in women increases their risk of developing premenopausal coronary artery disease to levels observed in men (41). Our observation that an elevated CRP concentration is associated with increased BMI recapitulates earlier findings based on NHANES III data as well as on other population-based data (28, 36). Our data differ from those of earlier studies because we extend to subjects aged <17 y the observation that higher BMIs are associated with an increased risk of elevated CRP concentrations.

It is likely that serum retinol decreases during the acute phase response to infection or chronic inflammation via the same mechanisms, although whether this response has some adaptive benefit for the host is unclear. In mild inflammation, the principal mechanism is probably a reduced transcription of messenger RNA for RBP, resulting in decreased release of RBP from the liver into the blood (13, 42), although changes in distribution between intravascular and extravascular compartments may occur as a result of changes in retinol metabolism or increased vascular permeability, which occurs during inflammation (10). During severe infections, retinol loss in the urine may also be associated with lower serum retinol concentrations (3). Thus, the decrease in serum retinol seems to be largely the result of decreased mobilization rather than of increased uptake from the blood. It is interesting that serum iron concentrations decrease during the acute phase response because iron is actively removed from the blood into phagocytic cells and the synthesis of the iron storage protein ferritin is positively regulated during the acute phase response via the same mechanisms used to increase the synthesis of CRP and other positive acute phase proteins (3). The presumed benefit to the host in this case is sequestration of iron away from pathogenic microorganisms, which also require iron for growth. No such benefit to the host has been postulated for decreased serum retinol concentrations. The argument that the liver shifts resources to rapid synthesis of positive acute phase proteins needed for host defense, and thus diminishes synthesis of less critical proteins (eg, RBP and albumin), seems the best, current explanation of this phenomenon.

These data indicate that an active acute phase response interferes with the assessment of vitamin A status in the NHANES III population and indicate that it is important to use serum CRP, or another indicator of the acute phase response (5, 10), to account for this phenomenon. Serum CRP seems to be a good choice because it increases rapidly to very high concentrations and then falls quickly in a time frame similar to that seen for the changes in serum RBP during the acute phase response. Although systematic data of this type are scarce, Banks et al (16) showed that cancer patients (most with normal concentrations of acute phase proteins) treated with interleukin responded with a nadir of 50% of normal within 2–4 d, with serum concentrations returning to normal by day 10. Serum CRP and amyloid A in the same subjects increased 100-fold by day 2 and were back to normal by day 15. Serum α₁-acid glycoprotein increased less dramatically and more slowly but also decreased by day 15; however, other positive acute phase proteins did not follow the pattern of changes in serum RBP so closely. For example, α₁-antichymotrypsin increased 2-fold by day 8 but had not decreased to normal concentrations in half of the patients by day 20. These data show that all acute phase proteins do not identify equally the interference by the acute phase response with assessment of vitamin A status. Serum CRP seems to be a reasonable choice (in light of the comparative data that are now available) for evaluating vitamin A status. The same may not be true of CRP as an indicator of changes in the serum concentration of other micronutrients.

REFERENCES


