C-Reactive Protein, Insulin Resistance, and Metabolic Syndrome in a Population With a High Burden of Subclinical Infection

Insights from the Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) study

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OBJECTIVE — To explore relationships between C-reactive protein (CRP), subclinical infection, insulin resistance, and metabolic syndrome.

RESEARCH DESIGN AND METHODS — Data from 1,174 Eskimos, aged ≥18 years, from the Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) study were analyzed; 40 participants with diabetes were eliminated. Baseline assessment included interviews, physical exam, and blood and urine sampling. Metabolic syndrome was assessed using Adult Treatment Panel III criteria. CRP and antibodies to common pathogens were measured.

RESULTS — Although CRP was related in univariate analyses to insulin resistance and metabolic syndrome, relations were attenuated or eliminated after adjustment for relevant covariates. CRP was not higher among those with impaired fasting glucose (IFG), and pathogen burden was not related to insulin resistance, metabolic syndrome, or IFG.

CONCLUSIONS — Pathogen burden and inflammation do not seem to be related to insulin resistance, metabolic syndrome, or IFG in this population. The inflammatory process may reflect insulin resistance or its correlates but most likely is not causative.

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Insulin resistance, metabolic syndrome, and diabetes are associated with inflammation (1–3). It is unclear, however, whether the inflammatory process causes insulin resistance and accelerates progression to diabetes or whether insulin resistance and diabetes increase inflammation. While Alaskan Eskimos have high rates of cardiovascular disease (4) and subclinical infection (5), diabetes, insulin resistance, and metabolic syndrome occur less often than in U.S. whites (6). Therefore, exploration of the relationships between subclinical infection, inflammatory markers, insulin resistance, and diabetes in this population may illuminate possible mechanisms.

RESEARCH DESIGN AND METHODS

— The Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) study population includes Eskimos residing in the Norton Sound region of Alaska. Of this population, 1,214 family members, aged ≥18 years, were recruited (October 2000–April 2004) (7). Questionnaires provided demographic, health habits, and medical history data. Sitting blood pressure was measured, and the mean of the second and third measurements was used for analysis. Fasting blood measures included glucose, insulin, fibrinogen, high-sensitivity C-reactive protein (CRP), and homocysteine (HCY) (7). Dietary intake was assessed using a food-frequency questionnaire validated for Alaska Natives (8).

IgG, IgA, and IgM antibodies to *C. pneumoniae* and IgG antibodies to other pathogens were determined using commercially available enzyme-linked immunosorbent assay (ELISA) kits (5). Obesity was defined as BMI ≥30 kg/m^2, metabolic syndrome was defined by Adult Treatment Panel III criteria, and insulin resistance was defined by homeostasis model assessment (HOMA) (9). Diabetes and impaired fasting glucose (IFG) were defined by American Diabetes Association criteria. Participants wore a Digi-walker pedometer for 7 days.

Excluded from this study were 10 men and 30 women with diabetes. Participant characteristics were compared using the Kruskal-Wallis test, Pearson's χ^2 test, Wilcoxon's rank-sum test, or Fisher's exact test. ANOVA was used to compare least-square means of insulin resistance or IFG, as estimated by HOMA of insulin resistance (HOMA-IR), for each quartile of CRP and HCY and number of subclinical infections. Logistic regression was used to compute adjusted odds ratios of metabolic syndrome across CRP or HCY quartiles (using the first quartile as the reference) and levels of subclinical infection (using levels 1 and 2 as the reference). For each analysis, data were first adjusted for age and sex and then analyzed in models including BMI, alcohol use, smoking status, physical activity, and

Table 1—*Relationships among HOMA-IR, metabolic syndrome, IFG, CRP, and pathogen burden, the GOCADAN study*

	HOMA-IR		Metabolic syndrome		IFG	
	Model 1*	Model 2†	Model 1*	Model 2†	Model 1*	Model 2†
CRP quartile						
1	1.58 ± 1.04	1.62 ± 1.05	1 (referent)	1 (referent)	1 (referent)	1 (referent)
2	1.75 ± 1.04	1.63 ± 1.06	1.91 (0.90–4.07)	1.90 (0.56–6.43)	1.59 (0.81–3.11)	2.01 (0.57–7.11)
3	1.91 ± 1.04	1.66 ± 1.06	4.58 (2.29–9.14)	3.51 (1.14–10.85)	2.55 (1.37–4.76)	2.81 (0.86–9.12)
4	2.04 ± 1.04	1.73 ± 1.06	6.47 (3.30–12.72)	2.82 (0.92–8.67)	2.80 (1.51–5.17)	2.43 (0.75–7.84)
P	<0.0001	0.341	<0.0001	0.137	0.003	0.379
Pathogen burden						
1	1.90 ± 1.11	1.59 ± 1.13	1 (referent)	1 (referent)	—	—
2	1.97 ± 1.06	1.81 ± 1.07	0.87 (0.29–2.60)	0.64 (0.11–3.71)	1 (referent)‡	1 (referent)‡
3	1.79 ± 1.03	1.83 ± 1.04	0.83 (0.30–2.25)	0.69 (0.15–3.23)	0.78 (0.44–1.40)	0.94 (0.32–2.72)
4	1.84 ± 1.03	1.78 ± 1.04	0.98 (0.36–2.66)	0.70 (0.15–3.35)	0.88 (0.50–1.56)	1.08 (0.39–3.02)
5	1.74 ± 1.06	1.62 ± 1.07	0.73 (0.25–2.15)	0.44 (0.08–2.37)	0.70 (0.35–1.40)	0.89 (0.27–2.93)
P	0.562	0.368	0.831	0.831	0.7118	0.9643

Data are least-square means ± SEM or odds ratio (95% CI). N = 1,174 participants included in the analysis. *Adjusted for age and sex. †Adjusted for age, sex, BMI, current smoking, current drinking, physical activity, n-3 fatty acid intake, and pathogen burden (in the CRP model). ‡Categories 1 and 2 combined as reference category.

fatty acid intake. In all computations (ANOVA and logistic regression), the transformed variables for HOMA-IR and CRP were used. Participants with CRP >10 mg/l (n = 21 [1.7%]) were excluded from CRP analyses. Because participants were members of extended families, kinship was accounted for in additional analyses.

RESULTS — Of the 1,174 participants (55% women), mean BMI was 27.5 kg/m^2, 31.5% were overweight, and 29.1% were obese. Metabolic syndrome was present in 14.3% (11% men, 17% women). Insulin was low (median 8.1 μU/ml). Mean CRP was 1.6 mg/l (median 0.9 mg/l), and mean HCY was 7.3 μmol/l. Measures for five common pathogens averaged 3.4 per participant, and CRP was positively correlated with pathogen burden (P = 0.02). BMI, percent women, IFG (in women), CRP, glucose, insulin, and waist circumference were higher and current smoking, drinking, and physical activity were lower with increasing tertiles of HOMA-IR or in metabolic syndrome (all P < 0.001).

With increasing quartiles of CRP in the simple models (Table 1), there was a significant increasing trend in HOMA-IR (P < 0.0001) and in probabilities of metabolic syndrome and IFG (P < 0.0001 and 0.003, respectively). However, in the multivariate models, relations between HOMA-IR, metabolic syndrome, IFG, and CRP were not significant (P = 0.341, 0.137, and 0.379, respectively). Relationships of pathogen burden with HOMA-IR, metabolic syndrome, and IFG were not significant in the simple or multivariate models. For HCY, neither model was significant.

CONCLUSIONS — GOCADAN provides a unique setting for exploring relationships between inflammation, insulin resistance, and metabolic syndrome. Obesity is not pervasive, diabetes rates are low, rates of insulin resistance and other metabolic syndrome components vary, and chronic inflammation is prevalent in the population. Although CRP in univariate comparisons was higher in those with insulin resistance and metabolic syndrome, pathogen burden was not. After adjustment for confounders, no consistent relationships were observed between HOMA-IR, metabolic syndrome, or IFG and CRP or subclinical pathogen burden.

Low-grade chronic inflammation has been shown in vitro to promote insulin resistance (10). Although cross-sectional studies have demonstrated relationships between CRP and measures of insulin resistance or metabolic syndrome (11–13), longitudinal studies have varied, with CRP predictive of diabetes in some studies (14,15) but not others (3,13). In the Insulin Resistance and Atherosclerosis Study (IRAS), Monitoring Trends and Determinants in Cardiovascular Disease (MONICA) study, Augsburg study, and Atherosclerosis Risk in Communities (ARIC) study, odds of diabetes with increasing CRP became nonsignificant after adjustment for BMI or other covariates (3,13,14), suggesting that elevations in inflammatory markers seen in insulin resistance, metabolic syndrome, and diabetes are not causative but a consequence of these abnormalities. Some (15) argue that inclusion of BMI in the models represents an overadjustment, proposing that inflammation precedes obesity; however, until a causal relationship has been established, it seems prudent to focus on the complete models.

The lack of a relationship between chronic inflammation and insulin resistance and/or metabolic syndrome in this population is supported by the observation that, despite chronic subclinical infection, this population has low incidence of insulin resistance and diabetes. As in other populations, diabetes is associated with the female sex, obesity, and greater insulin resistance (6); CRP is positively associated with BMI and is higher with diabetes, smoking, and increasing pathogen burden (data not shown); thus, CRP appears to be an indicator of chronic inflammation. The reasons for the high subclinical pathogen burden are unclear; they may be related to confinement in close quarters in winter and lack of adequate medical care.

This study is limited by its cross-sectional design. Also, the high prevalence of subclinical infection may obscure relationships between obesity and insulin resistance on secretion of inflammatory cytokines. Finally, inflammatory mediators other than CRP that were not measured may be mediators.

In summary, while analyses of unique populations often lead to unexpected

findings, they can help to further understanding of complex disorders. Our study provides evidence that the inflammatory process may reflect diabetes or insulin resistance but is most likely not their cause and suggests that subclinical infection should be considered in further explorations of the inflammatory process in people with insulin resistance or metabolic syndrome.

References

1. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM: C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. *JAMA* 286:327–334, 2001
2. Spranger J, Kroke A, Möhlig M, Hoffmann K, Bergmann MM, Ristow M, Boeing H, Pfeiffer AF: Inflammatory cytokines and the risk to develop type 2 diabetes: results of the prospective population-based European Prospective Investigation into Cancer and Nutrition (EPIC)-Potsdam Study. *Diabetes* 52:812–817, 2003
3. Festa A, D'Agostino R Jr, Tracy RP, Haffner SM: Elevated levels of acute-phase proteins and plasminogen activator inhibitor-1 predict the development of type 2 diabetes: the Insulin Resistance Atherosclerosis Study. *Diabetes* 51:1131–1137, 2002
4. Lanier AP, Ehrsam G, Sardidge J: Alaska Native mortality 1989–1998. Anchorage, Alaska, Office of Alaska Native Health Research, Division of Community Health Services, Alaska Native Tribal Health Consortium, 2002
5. Zhu J, Katz RJ, Quyyumi AA, Canos DA, Rott D, Csako G, Zalles-Ganley A, Ogunmakinwa J, Wasserman AG, Epstein SE: Association of serum antibodies to heat-shock protein 65 with coronary calcification levels: suggestion of pathogen-triggered autoimmunity in early atherosclerosis. *Circulation* 109:36–41, 2004
6. Carter EA, MacCluer JW, Dyke B, Howard BV, Devereux RB, Ebbesson SO, Resnick HE: Diabetes mellitus and impaired fasting glucose in Alaska Eskimos: the Genetics of Coronary Artery Disease in Alaska Natives (GOCADAN) study. *Diabetologia* 49:29–35, 2006
7. Howard BV, Devereux RB, Cole SA, Davidson M, Dyke B, Ebbesson SO, Epstein SE, Robinson DR, Jarvis B, Kaufman DJ, Laston S, MacCluer JW, Okin PM, Roman MJ, Romenesko T, Ruotolo G, Swenson M, Wenger CR, Williams-Blangero S, Zhu J, Saccheus C, Fabsitz RR, Robbins DC: A genetic and epidemiologic study of cardiovascular disease in Alaska natives (GOCADAN): design and methods. *Int J Circumpolar Health* 64:206–221, 2005
8. Nobmann ED, Ponce R, Mattil C, Devereux R, Dyke B, Ebbesson SO, Laston S, MacCluer J, Robbins D, Romenesko T, Ruotolo G, Wenger CR, Howard BV: Dietary intakes vary with age among Eskimo adults of northwest Alaska in the GOCADAN study, 2000–2003. *J Nutr* 135:856–862, 2005
9. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC: Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 28:412–419, 1985
10. Shoelson SE, Herrero L, Naaz A: Obesity, inflammation, and insulin resistance. *Gastroenterology* 132:2169-2180, 2007
11. Duncan BB, Schmidt MI: The epidemiology of low-grade chronic systemic inflammation and type 2 diabetes. *Diabetes Technol Ther* 8:7–17, 2006
12. Ford ES: Body mass index, diabetes, and C-reactive protein among U.S. adults. *Diabetes Care* 22:1971–1977, 1999
13. Tutor CG: *C-reactive protein and diabetes in three American Indian populations: The Strong Heart Study*. Master thesis. Norman, Oklahoma, The University of Oklahoma, 2001.
14. Freeman DJ, Norrie J, Caslake MJ, Gaw A, Ford I, Lowe GD, O'Reilly DS, Packard CJ, Sattar N: C-reactive protein is an independent predictor of risk for the development of diabetes in the West of Scotland Coronary Prevention Study. *Diabetes* 51:1596–1600, 2002
15. Duncan BB, Schmidt MI, Pankow JS, Ballantyne CM, Couper D, Vigo A, Hoogeveen R, Folsom AR, Heiss G: Low-grade systemic inflammation and the development of type 2 diabetes: the Atherosclerosis Risk in Communities study. *Diabetes* 52:1799–1805, 2003