GHb Is a Better Predictor of Cardiovascular Disease Than Fasting or Postchallenge Plasma Glucose in Women Without Diabetes

The Rancho Bernardo Study

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OBJECTIVE — To examine the relation between GHb, fasting plasma glucose (FPG), postchallenge plasma glucose (PCPG), and mortality from cardiovascular disease (CVD) and ischemic heart disease (IHD) in older adults.

RESEARCH DESIGN AND METHODS — A community-based study of 1,239 nondiabetic older adults followed for an average of 8 years, from baseline (1984–1987) to 1993.

RESULTS — GHb, but not FPG or PCPG, was significantly related to CVD and IHD mortality in women but not men. The age-adjusted relative hazard for those in the highest quintile of GHb (≥6.7%) compared with women with lower levels was 2.37 for fatal CVD (95% CI = 1.30–4.31, $P = 0.005$) and 2.43 for IHD (95% CI = 1.12–5.25, $P = 0.024$). This association persisted after adjustment for all covariates (age, systolic blood pressure, BMI, LDL, HDL, triglycerides, cigarette smoking, antihypertensive medication use, and estrogen use). GHb was significantly associated with LDL and HDL levels in women, but the association between GHb and CVD or IHD persisted after adjustment for these lipoproteins.

CONCLUSIONS — We conclude that GHb is a better predictor of CVD and IHD mortality than FPG or PCPG in women without diabetes; no single measure of glycemia was predictive in men. The reason for the sex difference is unexplained.

Diabetes is a strong risk factor for cardiovascular disease (CVD) (1), but the evidence that hyperglycemia is a risk factor for CVD or ischemic heart disease (IHD) in individuals without diabetes is contradictory. Thus, the Pooling Project, which examined data from 15 studies, found no consistent evidence for a graded or threshold association of fasting plasma glucose (FPG) or postchallenge plasma glucose (PCPG) in nearly 40,000 middle-aged men without known diabetes who were followed for 4–15 years (2).

In more recent prospective studies, evidence for an association between nondiabetic hyperglycemia and IHD in men remains inconsistent. For example, blood glucose levels were not associated with the 8-year risk of CHD in nondiabetic Puerto Rican men followed for 8 years (3) or in nondiabetic men from Chicago who were followed for 9 years (4). The Bedford Survey (5) also found no independent association of glycemia with IHD risk in men with impaired glucose tolerance, but three other prospective studies of nondiabetic men in the U.K. found a rather extreme threshold effect (6–8). Fuller et al. (6), for example, found that postchallenge glycemia was associated with IHD in nondiabetic men only when the blood glucose concentration was in the top 5% of the distribution. Less evidence for a threshold effect was found in the Tecumseh Study (9), in which there was an increased risk of IHD in nondiabetic men whose glucose levels were in the top three quintiles of distribution, and in the Honolulu Heart Study (10), in which PCPG levels were linearly associated with CHD risk in nondiabetic men of Japanese ancestry. The Paris Prospective Study (11) found a threshold effect for the top quintile of PCPG, but this disappeared after multivariate adjustment.

The few prospective studies of asymptomatic hyperglycemia and CHD that included women also have yielded contradictory results. In the Bedford Survey (5), there was a significantly increased risk of IHD in women with impaired glucose tolerance. Similarly, there was a significant independent risk of IHD in women in the Chicago Heart Association Detection Project cohort (4). In contrast, fasting hyperglycemia showed a threshold association in women from the Rancho Bernardo cohort (12), and glycemia was not associated with CHD in Tecumseh women (9).

Previous prospective studies of the association between glycemia and CVD have relied on FPG, PCPG, or casual glucose levels. These methods are imprecise because of large intraindividual variation, such that single measures of blood glucose poorly characterize usual glycemia for individuals (13–19). In contrast, GHb provides an integrated measure of the level of glycemia over the past 2–3

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AGE, advanced glycation end product; CVD, cardiovascular disease; FPG, fasting plasma glucose; IHD, ischemic heart disease; OGTT, oral glucose tolerance test; PCPG, postchallenge plasma glucose; RH, relative hazard.

months (20–22) and might therefore be a better predictor of subsequent CVD.

The present study was designed to determine whether GHb predicts mortality due to all causes, CVD, or IHD better than FPG or PCPG levels. Results are based on a prospective study of 1,239 nondiabetic older adults from the Rancho Bernardo cohort who were followed for an average of 8 years.

RESEARCH DESIGN AND METHODS

— Between 1972 and 1974, 82% of the adult white residents in a middle- to upper-middle-class community, Rancho Bernardo, California, participated in a survey of heart disease risk factors (23). From 1984 to 1987, 81% of the survivors aged 40 or older at the time of the initial visit participated in a follow-up visit designed to study diabetes (24). At this visit, standard questionnaires were used to ask about each participant's demographic, behavioral, and medical history. Questions included history of diabetes, heart disease, cigarette smoking, and treatment for hypertension or diabetes. Medication use was validated by examination of pills and prescriptions brought to the clinic for that purpose.

Height and weight were measured with the subject wearing light clothing and no shoes. BMI was used as an estimate of obesity. Blood pressure was measured in seated subjects by a certified technician, using the Hypertension Detection Follow-up Program protocol (25) and a standard mercury sphygmomanometer. Plasma cholesterol and triglyceride levels were measured by enzymatic assay in a Centers for Disease Control certified laboratory. HDL was measured by heparin and manganese extraction, using procedures outlined in the Lipid Research Clinics Manual of Laboratory Operations (26). A standard 12-lead resting electrocardiogram was analyzed by the Minnesota Coding Center, using the Minnesota Code Protocol (27).

A standard 75-g oral glucose tolerance test (OGTT) was performed in the morning after a requested 12-h fast. Blood samples were taken before and 2 h after the glucose challenge. Fasting and 2-h PCPG levels were measured using the glucose oxidase method in a diabetes research laboratory. World Health Organization (28) criteria for epidemiological studies were used to define NIDDM (FPG ≥140 mg/dl, a 2-h PCPG ≥200 mg/dl, a history of diabetes diagnosed by a physician, or use of a medication for diabetes).

All participants seen after January 1985 (11 months into the study) had nonlabile $GHbA_1$ levels measured by high-performance liquid chromatography using an automated analyzer in a commercial laboratory (Smith Kline, Van Nuys, CA); fasting whole blood samples were collected into heparinized tubes, stored at room temperature, and transported within 24 h to the laboratory for analysis. (Using this method, GHb is stable for up to 4 days.) Labile GHb was removed, and the erythrocytes were incubated for 1 h with potassium biphthalate at 37°C. The cells were diluted with a hemolyzing reagent (1:150 dilution), eluted through a cation exchange resin composed of methacrylic acid methacrylate ester with phosphate buffers (29), and separated into three fractions: A_{1a}, A_{1b}, and A_{1c}. GHb represents the sum of all three fractions.

All of the cohort have been followed for survival status from baseline (1984–1987) to 1993, an average of 8 years, with death certificates obtained for all decedents. The cause of death was coded by a certified nosologist, using the ninth revision of the International Classification of Diseases, Adapted (ICD-9) (30). CVD was defined as the underlying cause of death with ICD-9 codes 401–438, and IHD was defined with ICD-9 codes 410–414.

Participants with diabetes by history or by glucose tolerance test criteria at the 1984–1987 visit were excluded from this analysis to study the relation of glycemia, not diabetes, to CVD. Subjects who had fasted for <12 h were also excluded (n = 29), as were two subjects with unexplained extreme GHb values.

Distribution of heart disease risk factors was assessed by analysis of covariance using quintiles of GHb, FPG, and PCPG and by partial correlation using continuous variables. Cox proportional hazard models were used to determine the independent association of each glucose parameter with all-cause, CVD, and IHD mortality after adjustment for confounding variables. This model assesses the length of time to each event, giving more weight to premature mortality. All analyses were performed separately for men and for women. Statistical analysis was done using the SAS statistical package (31). No adjustments were made for multiple comparisons; rather, exact P values are shown.

RESULTS

— There were 549 men and 690 women who were aged 55 years or older, had fasted for at least 12 h before the OGTT and GHb measurement, and were free of diabetes in 1984–1987. The

Table 1—*Distribution of quintiles of GHb, FPG, and PCPG in 549 nondiabetic men and 690 nondiabetic women: Rancho Bernardo, CA, 1984–1987*

Quintiles	Men	Women
GHb (%)		
1st (low)	5.0 ± 0.3 (3.6–5.3)	5.1 ± 0.4 (3.4–5.5)
2nd	5.6 ± 0.1 (5.4–5.7)	5.8 ± 0.1 (5.6–5.9)
3rd	6.0 ± 0.1 (5.8–6.1)	6.1 ± 0.1 (6.0–6.2)
4th	6.4 ± 0.1 (6.2–6.5)	6.4 ± 0.1 (6.3–6.6)
5th (high)	7.0 ± 0.4 (6.6–8.6)	7.1 ± 0.4 (6.7–8.9)
FPG (mg/dl)		
1st (low)	85.2 ± 7.1 (51–91)	81.6 ± 7.9 (44–87)
2nd	94.4 ± 1.3 (92–96)	90.2 ± 1.7 (88–92)
3rd	98.9 ± 1.4 (97–101)	95.1 ± 1.1 (93–97)
4th	104.2 ± 1.6 (102–107)	100.5 ± 2.0 (98–104)
5th (high)	115.5 ± 6.9 (108–139)	111.5 ± 6.4 (105–133)
PCPG (mg/dl)		
1st (low)	75.1 ± 13.0 (33–90)	84.8 ± 12.6 (43–99)
2nd	100.2 ± 5.2 (91–108)	108.8 ± 5.3 (100–117)
3rd	116.5 ± 4.9 (109–125)	125.4 ± 5.0 (118–133)
4th	136.4 ± 7.2 (126–150)	144.7 ± 6.7 (134–158)
5th (high)	172.5 ± 13.8 (151–199)	175.3 ± 11.7 (159–199)

Data are means ± SD (range).

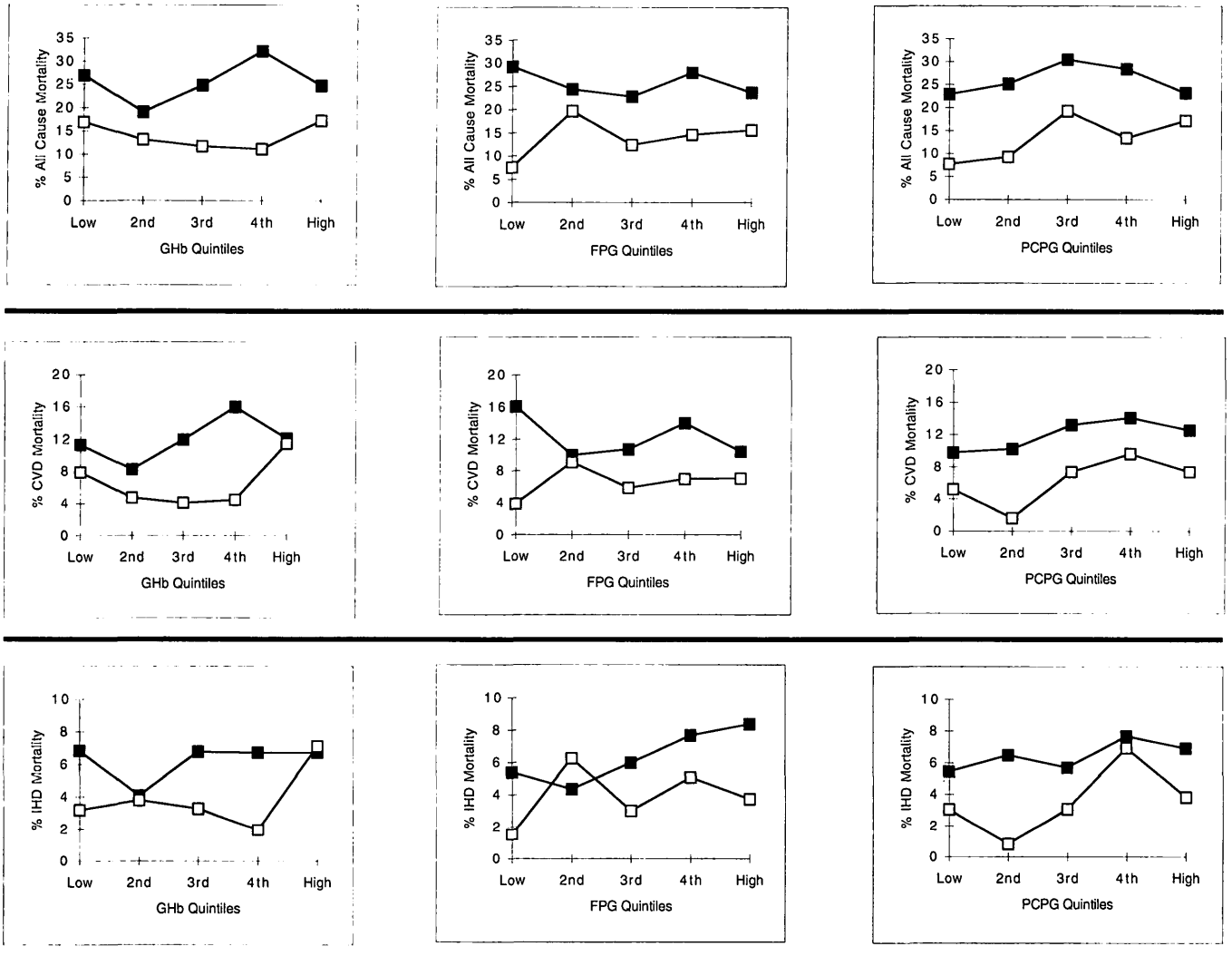

Figure 1—*Age-adjusted mortality rates by GHb, FPG, and PCPG quintiles.* ■ *men,* □ *women.*

mean age of both men and women was 70 years (range 55–90 years for men and 55–92 years for women).

For men and women, respectively, the GHb values (means ± SD) were 6.10 ± 0.73 and 5.96 ± 0.73%; the FPG levels were 99.9 ± 11.2 and 96.2 ± 11.0 mg/100 ml; and the PCPG levels were 120 ± 34.5 and 128 ± 32.4 mg/100 ml. The sex-specific distribution of GHb, FPG, and PCPG by quintile is shown in Table 1. GHb and PCPG increased with age (*P* value for linear trend = 0.0001 in men and 0.002 in women for GHb, and *P* = 0.0001 in men and women for PCPG). Therefore, all subsequent analyses were age-adjusted.

As shown in Fig. 1, no significant associations were seen in men or women between GHb, FPG, or PCPG levels and age-adjusted all-cause mortality. In men, GHb, FPG, and PCPG levels were unrelated to CVD or IHD death; FPG and PCPG were also unrelated to CVD or IHD deaths in women. In contrast, both CVD and IHD mortality were increased in women in the highest quintile of GHb (≥6.7%) compared with the lower four combined (*P* = 0.012 and 0.041, respectively).

The age-adjusted relative hazard (RH) for CVD mortality was 2.37 (95% CI = 1.30–4.31, *P* = 0.005) for women with GHb levels in the highest quintile (Table 2). In men, the upward trend was not statistically significant; the age-adjusted CVD RH of the top GHb quintile (≥6.6%) versus the four lower quintiles was 1.09 (95% CI = 0.63–1.93) (Table 2).

In women, there was also a 2.43-fold increase in age-adjusted risk of IHD mortality (95% CI = 1.12–5.25) for GHb levels in the highest quintile compared with the lower levels (Table 2). In men at the highest quintile of GHb, the age-adjusted RH for IHD was 1.25 (95% CI = 0.59–2.68) (Table 2).

To determine whether the sex difference was explained by a stronger association of GHb with heart disease risk factors in women than men, we examined the sex-specific association between age-adjusted heart disease risk factors by quintile of GHb and by partial correlations. The results were similar, and only the partial correlations are shown (Table 3). GHb was not associated with blood pressure, triglyceride levels, or BMI in either sex (all *P* values for linear trend ≥0.08). It was also unassociated with cigarette smoking (data not shown). In contrast, plasma LDL levels increased and HDL levels decreased in a statistically significant stepwise fashion with each increasing quintile of GHb only in women (*P* = 0.002 for LDL; *P* = 0.004 for HDL).

Table 2—*RHs (proportional hazards model) for all-cause, CVD, and IHD mortality (top versus lower four quintiles): Rancho Bernardo, CA, 1984–1993*

	Men		Women	
	RH	95% CI	RH	95% CI
All-cause mortality				
GHb				
Age-adjusted	1.03	0.69–1.53	1.46	0.93–2.28
Fully adjusted	0.92	0.61–1.39	1.49	0.94–2.36
FPG				
Age-adjusted	0.84	0.54–1.29	1.34	0.85–2.17
Fully adjusted	0.71	0.45–1.11	1.33	0.80–2.20
PCPG				
Age-adjusted	0.83	0.56–1.22	1.22	0.78–1.91
Fully adjusted	0.73	0.48–1.10	1.17	0.74–1.84
CVD mortality				
GHb				
Age-adjusted	1.09	0.63–1.93	2.37	1.30–4.31
Fully adjusted	1.10	0.61–1.97	2.61	1.40–4.88
FPG				
Age-adjusted	0.84	0.45–1.56	1.25	0.62–2.53
Fully adjusted	0.75	0.39–1.46	1.30	0.61–2.81
PCPG				
Age-adjusted	1.01	0.59–1.71	1.01	0.53–1.97
Fully adjusted	0.83	0.47–1.45	1.01	0.51–2.00
IHD mortality				
GHb				
Age-adjusted	1.25	0.59–2.68	2.43	1.12–5.25
Fully adjusted	1.25	0.56–2.78	2.59	1.14–5.85
FPG				
Age-adjusted	1.16	0.53–2.56	1.00	0.38–2.66
Fully adjusted	0.98	0.42–2.30	0.98	0.34–2.80
PCPG				
Age-adjusted	1.15	0.56–2.38	0.84	0.34–2.09
Fully adjusted	0.90	0.41–1.96	0.87	0.34–2.21

The fully adjusted model is adjusted for age, systolic blood pressure, BMI, plasma LDL, plasma HDL, plasma triglyceride, cigarette smoking, antihypertensive medication use, and, in women, estrogen use.

Nevertheless, in proportional hazards models adjusted for these significant variables (age, LDL, and HDL), the RH of GHb with CVD or IHD mortality was essentially unchanged (RH = 2.48, 95% CI = 1.4–4.5, $P = 0.003$ for CVD mortality, and RH = 2.56, 95% CI = 1.2–5.6, $P = 0.02$ for IHD mortality, in women).

Although FPG and PCPG levels were not predictive of CVD death in women, they were more correlated with risk factors other than GHb (Table 3). Proportional hazards models adjusting for all covariates (age, systolic blood pressure, BMI, plasma LDL cholesterol, plasma HDL cholesterol, plasma triglyceride, cigarette smoking, antihypertensive medication use, and estrogen use in women) did not materially change the absent associations of these glucose measures with CVD or IHD, as shown in Table 2.

There was a highly significant stepwise trend for decreasing prevalence of postmenopausal estrogen use by increasing the quintile of GHb (not shown). When the association of GHb with future CVD or IHD mortality was examined in models adjusted for age and current estrogen use, the associations were not materially changed (RH = 2.35, 95% CI = 1.29–4.28, $P = 0.005$ for CVD mortality, and RH = 2.40, 95% CI = 1.11–5.2, $P = 0.03$ for IHD mortality). In another analysis that excluded the 208 women currently using estrogen therapy, the positive association of GHb with CVD and IHD death persisted (RH = 3.0, 95% CI = 1.5–5.8, $P = 0.001$ for CVD, and RH = 3.3, 95% CI = 1.4–7.8, $P = 0.007$ for IHD) for GHb levels in the highest quintile when compared with lower GHb levels. Again, there was no significant association between FPG or PCPG levels and CVD or IHD mortality.

To determine whether antihypertensive medications, most commonly thiazides, contributed to these associations, separate proportional hazards analyses were done, adjusting for age and blood pressure medication; the results were essentially unchanged. In analyses that excluded the 143 men and 202 women who used antihypertensives medication(s), the positive relation between GHb and CVD in women persisted (RH = 2.80, 95% CI = 1.20–6.20, $P = 0.01$) in the highest GHb quintile compared with the lower levels. The positive relation between GHb and IHD mortality in untreated women (RH = 2.10, 95% CI = 0.66–7.00) was not significant, probably because of the small number of events.

To determine whether the absent association in men was due to survivor bias, such that only the healthiest men survived, analyses were repeated after stratification by age. There were no significant associations between GHb and mortality among the 266 men <70 years of age or the 283 men age 70 years or older. In addition, RHs were similar in each age-group. Among women, RHs for CVD and IHD remained elevated in each age-group but were significant only among the 363 women 70 years or older (RH = 2.4, 95% CI = 1.1–5.3 for IHD, and RH 2.3, 95% CI = 1.3–4.3 for CVD).

CONCLUSIONS — In this prospective study of adults without diabetes, GHb, but not FPG or PCPG, predicted future CVD and IHD mortality, but only in women. To our knowledge, the only other community-based study of GHb and CVD is a cross-sectional study from Framingham in which GHb was found to be associated with prevalent CVD, again only in women (32).

The reason for the sex difference observed here and cross-sectionally in Framingham is not known (32). One possible explanation is survival bias; men are more likely to have died of CVD before the OGTT clinic visit than women, even at these older ages. This is compatible with the observation that fasting hyperglycemia below the diabetic range (FPG <140 mg/100 ml) significantly predicted 9-year CVD, IHD, and all-cause mortality in

Table 3—*Partial correlations (continuous) for risk factors and GHb, FPG, and PCPG: Rancho Bernardo, CA, 1984–1987*

	Men		Women	
Risk factors	*r*	*P* value	*r*	*P* value
GHb				
Age	0.166	0.0001	0.117	0.002
Systolic blood pressure	−0.052	0.22	0.011	0.78
Diastolic blood pressure	−0.040	0.35	−0.008	0.84
LDL cholesterol	0.016	0.70	0.119	0.002
HDL cholesterol	0.003	0.94	−0.110	0.004
Triglyceride (log)	0.044	0.31	0.081	0.27
BMI	0.011	0.80	0.067	0.08
FPG				
Age	−0.072	0.09	−0.036	0.35
Systolic blood pressure	0.019	0.65	0.103	0.007
Diastolic blood pressure	−0.052	0.23	0.651	0.09
LDL cholesterol	−0.013	0.76	0.102	0.008
HDL cholesterol	−0.048	0.27	−0.148	0.0001
Triglyceride (log)	0.097	0.02	0.153	0.0001
BMI	0.127	0.003	0.169	0.0001
PCPG				
Age	0.244	0.0001	0.216	0.0001
Systolic blood pressure	0.099	0.02	0.149	0.0001
Diastolic blood pressure	0.044	0.31	0.044	0.25
LDL cholesterol	−0.003	0.94	0.024	0.53
HDL cholesterol	−0.194	0.0001	−0.162	0.0001
Triglyceride (log)	0.190	0.0001	0.223	0.0001
BMI	0.056	0.19	0.106	0.006

P value for partial correlation.

Rancho Bernardo men 12 years earlier (33), when the average age of the cohort was 63 years. It is also possible that more men than women had borderline hyperglycemia that worsened to diabetes (and would thus have been excluded from the present study) because men in Rancho Bernardo were significantly more overweight (12) and had more diabetes (34) than women at baseline. On the other hand, the sex effect may not be due to differential survival, since other prospective studies of younger cohorts also found sex differences. Thus, both the Bedford Survey (5) and the Chicago Heart Association Detection Project (4) found nondiabetic glycemia to be associated with IHD in women, but not in men. In addition, stratification by age in the present study (<70 or >70 years) yielded comparable results.

A stronger association of GHb measures with other heart disease risk factors in women than in men is also compatible with survival bias as an explanation for the sex difference. LDL and HDL cholesterol levels were found to be significantly associated with GHb levels only in women. However, the RHs remained little changed after adjustment for these lipoproteins, making it less likely that they account for the observed differences.

Antihypertensive medications may decrease CVD risk but can also cause hyperglycemia. If more men than women were taking thiazides, this might explain the sex difference. In Rancho Bernardo, however, the percentage of men taking antihypertensive medications (26.1%) was similar to the proportion in women (29.3%), and the results did not change after adjustment for or stratification by antihypertensive use. Analysis adjusted for or stratified by estrogen therapy also did not materially change the results.

The poor predictive power of FPG and PCPG reported here is consistent with the studies that found no association or only an extreme threshold effect, as summarized earlier. The reason for the failure to show a consistent association between CVD and FPG or PCPG may reflect the well-known large intraindividual variation and poor reproducibility of individual glucose tests, leading to substantial misclassification (13–19,35–38). With more standardized glucose tolerance test procedures, more recent studies have confirmed the poor reproducibility of FPG and PCPG levels in repeated-measure studies (39–43).

In contrast, GHb levels, which represent the integrated glucose values over the preceding months, show good reproducibility (20–22,44). For example, Dunn et al. (44) found that HbA_{1c} measured 7 days to 16 months apart had a coefficient of variation of 6.9% in 121 adults without diabetes. We theorize that GHb is a better predictor of CVD and IHD mortality than FPG or PCPG because individuals are better classified with regard to their usual level of glycemia. Therefore, GHb may also be better at identifying those subjects with diabetes who were misclassified as not having diabetes or those who subsequently developed diabetes before death. The association of GHb with CVD and IHD mortality shown in Fig. 1 suggests a threshold effect, such that only those in the highest quintile were at increased risk.

Although our original thesis was that any advantage of GHb over other plasma glucose levels reflected less misclassification, there is another possible reason for the stronger association. GHb is one of many body proteins that are irreversibly glycated in proportion to glucose concentration (45). These advanced glycation end products (AGEs) bind to lipoproteins and to AGE-specific receptors; the latter can induce production of interleukin-1 and tumor necrosis factor cytokines, which may injure the arterial wall. Thus, AGEs may contribute to the genesis of atherosclerosis (46), and GHb could be a better predictor of CVD than glucose, not because it is more representative of normal glucose but because it is more directly in the pathogenic pathway.

Whatever the mechanism, these results may have more immediate implications with regard to screening for hyperglycemia. Unlike OGTT or FPG, GHb does not require subjects to be seen in the morning after a 12-h fast. No glucose load is required, and only a single blood sample is necessary. These time and cost savings are only partially offset by the higher cost of the GHb laboratory test and the limited comparability of different meth-

ods of determining GHb levels (47). Therefore, the results of the present study suggest that GHb might be superior to FPG or PCPG levels as a risk factor for CVD. Additional studies of the mechanism, the reasons for the intriguing and unexplained sex differences, and the predictive power of GHb as a heart disease risk factor are warranted.

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References

1. National Diabetes Data Group: *Diabetes in America*. 2nd ed. Washington, DC, U.S. Govt. Printing Office, 1995 (NIH publ. no. 95–1468)
2. Stamler R, Stamler J (Eds.): International Collaborative Group: Asymptomatic hyperglycemia and coronary heart disease: a series of papers by the International Collaborative Group based on studies in fifteen populations. *J Chron Dis* 32:683–837, 1979
3. Cruz-Vidal M, Garcia-Palmieri MR, Costas R, Sorlie PD, Havlik RJ: Abnormal blood glucose and coronary heart disease: the Puerto Rico heart health program. *Diabetes Care* 6:556–561, 1983
4. Pan WH, Cedres LB, Liu K, Dyer A, Schoenberger JA, Shekelle RB, Stamler R, Smith D, Collette P, Stamler J: Relationship of clinical diabetes and asymptomatic hyperglycemia to risk of coronary heart disease mortality in men and women. *Am J Epidemiol* 123:504–516, 1986
5. Jarrett RJ, McCartney P, Keen H: The Bedford Survey: ten year mortality rates in newly diagnosed diabetes, borderline diabetics and normoglycaemic controls and risk indices for coronary heart disease in borderline diabetics. *Diabetologia* 22:79–84, 1982
6. Fuller JH, Shipley MJ, Rose GA, Jarrett RJ, Keen H: Mortality from coronary heart disease and stroke in relation to degree of glycemia: the Whitehall Study. *Br Med J* 287:867–870, 1983
7. Yarnell JW, Pickering JE, Elwood PC, Baker IA, Bainton D, Dawkins C, Phillips DI: Does nondiabetic hyperglycemia predict future IHD? Evidence from the Caerphilly and Speedwell studies. *J Clin Epidemiol* 47:383–388, 1994
8. Perry IJ, Wannamethee SG, Whincup PH, Shaper AG: Asymptomatic hyperglycaemia and major ischaemic heart disease events in Britain. *J Epidemiol Community Health* 48:538–542, 1994
9. Butler WJ, Ostrander LD Jr, Carman WJ, Lamphiear DE: Mortality from coronary heart disease in the Tecumseh study: long-term effect of diabetes mellitus, glucose intolerance, and other risk factors. *Am J Epidemiol* 121:541–547, 1985
10. Donahue RP, Abbott RD, Reed DM, Yano K: Postchallenge glucose concentration and coronary heart disease in men of Japanese ancestry: Honolulu Heart Program. *Diabetes* 36:689–692, 1987
11. Eschwege E, Richard JL, Thibult N, Ducimetiere P, Warnet JM, Claude JR, Rosselin GE: Coronary heart disease mortality in relation to diabetes, blood glucose and plasma insulin levels: the Paris Prospective Study, ten years later. *Horm Metab Res* 15 (Suppl.):41–46, 1985
12. Scheidt-Nave C, Barrett-Connor E, Wingard DL, Cohn BC, Edelstein SL: Sex differences in fasting glycemia as a risk factor for ischemic heart disease death. *Am J Epidemiol* 133:565–576, 1990
13. Kosaka K, Mizuno Y, Kuzuya T: Reproducibility of oral glucose tolerance test and the rice-meal test in mild diabetics. *Diabetes* 15:901–904, 1966
14. Ganda OP, Day JL, Soelgner JS, Connon JJ, Gleason RE: Reproducibility and comparative analysis of repeated intravenous and oral glucose tolerance tests. *Diabetes* 27:715–725, 1978
15. Cummings ST, Fraser CG: Variability of capillary plasma glucose in healthy individuals in repeated 75 g oral glucose tolerance tests. *Ann Clin Biochem* 25:634–637, 1988
16. McDonald GW, Burnham CE, Lewis WF: Reproducibility of glucose intolerance in 101 non-diabetic women. *Publ Health Rep* 84:353, 1969
17. McDonald GW, Fisher GF, Nurnham CE: Reproducibility of the oral glucose tolerance test. *Diabetes* 14:473, 1965
18. West KM, Wulff JA, Reigerl DG, Fitzgerald DT: Oral carbohydrate tolerance tests. *Arch Intern Med* 113:641–648, 1964
19. Olefsky JM, Reaven GM: Insulin and glucose responses to identical oral glucose tolerance tests performed forty-eight hours apart. *Diabetes* 23:449–453, 1974
20. Bunn HF, Haney DM, Kamin S, Gabbay KH, Gallop PM: The biosynthesis of hemoglobin A1c: slow glycosylation of hemoglobin in vivo. *J Clin Invest* 57:1652–1659, 1976
21. Peterson CM (Ed.): Symposium on nonenzymatic glycosylation and browning reactions. *Diabetes* 31 (Suppl. 3):11–82, 1982
22. McDonald JM, Davis JE: Glycosylated hemoglobins and diabetes mellitus. *Hum Pathol* 10:279–291, 1979
23. Criqui MH, Barrett-Connor E, Austin M: Differences between respondents in a population-based cardiovascular disease study. *Am J Epidemiol* 108:367–372, 1978
24. Wingard DL, Sinsheimer P, Barrett-Connor EL, McPhillips JB: Community-based study of prevalence of NIDDM in older adults. *Diabetes Care* 13:3–8, 1990
25. Holbrook TL, Barrett-Connor E, Wingard DL: A prospective population-based study of alcohol use and non-insulin-dependent diabetes mellitus. *Am J Epidemiol* 132: 902–909, 1990
26. Lipid Research Clinics Program: *Lipid Research Clinics Manual of Laboratory Operations*. Vol 1. *Lipid and Lipoprotein Analysis*. Washington, DC: U.S. Dept. of Health, Education and Welfare, 1974 (DHEW publ. no. [NIH] 75–628)
27. Rose GA, Blackburn H, Gillum RF, Prineas RJ: *Cardiovascular Survey Methods*. 2nd ed. Geneva, World Health Org., 1982
28. World Health Organization: *WHO Expert Committee on Diabetes Mellitus. Second Report*. Geneva, World Health Org., 1980. (Tech. Rep. Ser., no. 646)
29. McPhillips JB, Barrett-Connor E: A comparison of glycosylated hemoglobin A1 measured by column and high performance liquid chromatography. (unpublished)
30. *International Classification of Diseases, Adapted (ICD-9)*. Alexandria, VA, Saint Anthony Publishing, 1992
31. *SAS User's Guide: Statistics. Version 5 Edition*. Cary, NC, SAS, 1985
32. Singer DE, Nathan DM, Anderson KM, Wilson PW, Evans JC: Association of HbA1c with prevalent cardiovascular disease in the original cohort of the Framingham Heart Study. *Diabetes* 41:202–208, 1992
33. Barrett-Connor E, Wingard DL, Criqui MH, Suarez L: Is borderline fasting hyperglycemia a risk factor for cardiovascular death? *J Chron Dis* 37:773–779, 1984
34. Barrett-Connor E, Cohn BA, Wingard DL, Edelstein SL: Why is diabetes mellitus a stronger risk factor for fatal ischemic heart disease in women than in men? The Rancho Bernardo Study. *Diabetes*, reprinted from *JAMA* 265:627–631, 1991
35. Siperstein MD: The glucose tolerance test: a pitfall in the diagnosis of diabetes mellitus. *Adv Intern Med* 20:297–323, 1975
36. Liu K, Stamler R, Cooper R, Shekelle RB, Schoenberger JA, Berkson DM, Lindberg HA, Marquardt J, Stevens E, Tokich T: Methodological problems in characterizing an individual's plasma glucose. *J Chron Dis* 35:475–485, 1982
37. Yudkin JS, Alberti KGMM, McLarty DG, Swai ABM: Impaired glucose tolerance: is it a risk factor for diabetes or a diagnostic ragbag? *Br Med J* 301:397–402, 1990
38. Unger RH: The standard two-hour glucose tolerance test in the diagnosis of diabetes mellitus in subjects without fasting

hyperglycemia. *Ann Intern Med* 47:135–137, 1957
39. Riccardi G, Vaccaro O, Rivellese A, Pignalosa S, Tutino L, Mancini M: Reproducibility of the new diagnostic criteria for impaired glucose tolerance. *Am J Epidemiol* 121:422–429, 1985
40. Forrest RD, Jackson CA, Yudkin JS: The abbreviated glucose tolerance test in screening for diabetes: the Islington diabetes survey. *Diabetic Med* 5:557–561, 1988
41. Eriksson KF, Lindgarde F: Impaired glucose tolerance in a middle-aged male urban population: new approach for identifying high-risk cases. *Diabetologia* 33: 526–351, 1990
42. Balkau B, Eschwege E: Repeatability of the oral glucose tolerance test for the diagnosis of impaired glucose tolerance and diabetes mellitus. *Diabetologia* 34:201–203, 1991
43. Feskens EJM, Bowles CH, Kromhout D: Intra- and interindividual variability of glucose tolerance in an elderly population. *J Clin Epidemiol* 44:947–953, 1991
44. Dunn PJ, Cole RA, Soeldner MD, Gleason RE: Reproducibility of hemoglobin A1c and sensitivity to various degrees of glucose intolerance. *Ann Intern Med* 91:390–396, 1979
45. Brownlee M: Lilly Lecture 1993: glycosylation and diabetic complications. *Diabetes* 43:836–841, 1994
46. Gowen M (Ed.): *Cytokines and Bone Metabolism*. Boca Raton, FL, CRC Press, 1992
47. Benjamin RJ, Sacks DB: Glycated protein update: implications of recent studies, including the Diabetes Control and Complications Trial. *Clin Chem* 40:683–687, 1994