

Fasting Compared With Nonfasting Triglycerides and Risk of Cardiovascular Events in Women

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IN CONTRAST TO TOTAL CHOLESTEROL, low-density lipoprotein cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C), which are well-established independent risk factors for cardiovascular disease,¹ the importance of triglycerides remains controversial. In part this controversy reflects the fact that, due to the inverse correlation of triglyceride levels with those of HDL-C, adjustment for HDL-C attenuates the relationship between triglycerides and cardiovascular disease. A recent meta-analysis suggested that the adjusted risk ratio for coronary heart disease among individuals in the highest third of triglyceride levels compared with those in the lowest third decreases from approximately 2.0 to 1.5 after accounting for HDL-C levels.²

A second aspect of the controversy stems from the manner in which triglyceride levels are typically measured. Current national guidelines recommend that blood for lipid profiles be drawn after an 8- to 12-hour fast.¹ Because plasma triglyceride levels can increase substantially postprandially, fasting levels ostensibly avoid the variability associated with meals and provide a

See also pp 299 and 336.

Context The association of triglycerides with incident cardiovascular disease remains controversial. Although triglyceride levels are typically obtained in the fasting state, postprandial hypertriglyceridemia may play an important role in atherosclerosis.

Objective To determine the association of triglyceride levels (fasting vs nonfasting) and risk of future cardiovascular events.

Design, Setting, and Participants Prospective study of 26 509 initially healthy US women (20 118 fasting and 6391 nonfasting) participating in the Women's Health Study, enrolled between November 1992 and July 1995 and undergoing follow-up for a median of 11.4 years. Triglyceride levels were measured in blood samples obtained at time of enrollment.

Main Outcome Measure Hazard ratios for incident cardiovascular events (nonfatal myocardial infarction, nonfatal ischemic stroke, coronary revascularization, or cardiovascular death).

Results At baseline, triglyceride levels in fasting as well as nonfasting women correlated with traditional cardiac risk factors and markers of insulin resistance. During a median follow-up of 11.4 years, 1001 participants experienced an incident cardiovascular event (including 276 nonfatal myocardial infarctions, 265 ischemic strokes, 628 coronary revascularizations, and 163 cardiovascular deaths), for an overall rate of 3.46 cardiovascular events per 1000 person-years of follow-up. After adjusting for age, blood pressure, smoking, and use of hormone therapy, both fasting and nonfasting triglyceride levels predicted cardiovascular events. Among fasting participants, further adjustment for levels of total and high-density lipoprotein cholesterol and measures of insulin resistance weakened this association (fully adjusted hazard ratio [95% confidence interval] for increasing tertiles of triglyceride levels: 1 [reference], 1.21 [0.96-1.52], and 1.09 [0.85-1.41] [$P = .90$ for trend]). In contrast, nonfasting triglyceride levels maintained a strong independent relationship with cardiovascular events in fully adjusted models (hazard ratio [95% confidence interval] for increasing tertiles of levels: 1 [reference], 1.44 [0.90-2.29], and 1.98 [1.21-3.25] [$P = .006$ for trend]). In secondary analyses stratified by time since participants' last meal, triglyceride levels measured 2 to 4 hours postprandially had the strongest association with cardiovascular events (fully adjusted hazard ratio [95% confidence interval] for highest vs lowest tertiles of levels, 4.48 [1.98-10.15] [$P < .001$ for trend]), and this association progressively decreased with longer periods of fasting.

Conclusions In this cohort of initially healthy women, nonfasting triglyceride levels were associated with incident cardiovascular events, independent of traditional cardiac risk factors, levels of other lipids, and markers of insulin resistance; by contrast, fasting triglyceride levels showed little independent relationship.

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more stable estimate for risk assessment. However, postprandial lipids may play an important role in the pathogenesis of cardiovascular disease because postprandial triglyceride-rich remnant lipoproteins can penetrate the endothelial cell layer and reside in the subendothelial space, where they can contribute to the formation of foam cells, a hallmark of early atherosclerosis.³⁻⁵ Elevated postprandial levels of triglycerides, via higher peak concentrations or delayed clearance, also might represent an abnormal response to an oral fat load that reflects insulin resistance,⁶⁻⁸ a condition associated with a host of metabolic abnormalities that predispose an individual to cardiovascular disease.^{9,10} To clarify the importance of the prandial state when measuring triglyceride levels, we evaluated the association of fasting and nonfasting levels with incident cardiovascular events in a large prospective cohort of initially healthy women, independent of traditional cardiac risk factors, levels of other lipids, and markers of insulin resistance.

METHODS

Study Participants

The study cohort was derived from participants in the Women's Health Study, a previously completed randomized controlled trial of aspirin and vitamin E in the primary prevention of cardiovascular disease and cancer in women aged 45 years and older. Details of the study design and primary outcomes have been reported.¹¹⁻¹³ The study protocol was approved by the institutional review board of Brigham and Women's Hospital, Boston, Massachusetts. All participants provided written informed consent.

All participants provided baseline demographic data and health histories and were asked to provide a blood sample at enrollment. Participants were requested, but not required, to have the sample drawn in the morning before eating, and reported the number of hours since their last meal before the blood draw. Of the full cohort of 39 876 women, 27 939 provided a baseline

blood sample for analysis. Participants whose last meal was 8 or more hours prior to their blood draw comprised the fasting cohort (n=20 118), and those who had eaten within 8 hours of their blood draw comprised the nonfasting cohort (n=6391); those with unknown time since last meal (n=1430) were excluded from the analysis.

Laboratory Methods

Blood samples were collected at enrollment in tubes containing EDTA and were shipped cold overnight to a core laboratory certified by the US Centers for Disease Control and Prevention and the National Heart, Lung, and Blood Institute Lipid Standardization Program. There, samples were centrifuged and the plasma was stored in liquid nitrogen until analysis.

Triglyceride levels were measured enzymatically, with correction for endogenous glycerol,¹⁴ using a Hitachi 917 analyzer and reagents and calibrators from Roche Diagnostics (Indianapolis, Indiana). Triglycerides at concentrations of 84.0 and 201.8 mg/dL (to convert to millimoles per liter, multiply by 0.0113) were determined in the laboratory with a day-to-day reproducibility of 1.8% (SD, 1.6 mg/dL) and 1.7% (SD, 3.5 mg/dL), respectively. Levels of total cholesterol and HDL-C were measured enzymatically on a Hitachi 911 autoanalyzer (Roche Diagnostics, Basel, Switzerland), and levels of LDL-C were determined directly (Genzyme, Cambridge, Massachusetts). These assays are approved for clinical use by the US Food and Drug Administration.

Outcomes

Incident cardiovascular events were a composite of confirmed nonfatal myocardial infarction, nonfatal ischemic stroke, coronary revascularization, or death due to cardiovascular causes. Myocardial infarction was defined by World Health Organization criteria of characteristic symptoms accompanied by elevated levels of cardiac enzymes or by diagnostic electrocardiographic changes. Stroke was defined as a new neurologic deficit of sudden onset that persisted for

at least 24 hours; computed tomography scans or magnetic resonance images were available for most events and were used to distinguish ischemic from hemorrhagic strokes. Coronary revascularization included percutaneous coronary interventions and coronary artery bypass graft surgery. All events were adjudicated by an end points committee. In participants with more than 1 cardiovascular event, only the first was used in these analyses. Follow-up morbidity and mortality data were available for 97.2% and 99.4%, respectively, of the Women's Health Study participants.

Statistical Analyses

All analyses were performed separately for fasting and nonfasting participants. For the main analyses, triglyceride levels were categorized into tertiles for consistency with a recent meta-analysis² and for stability of effect estimates by maintaining adequate numbers of events in reference groups. Because distributions differed between fasting and nonfasting participants, separate tertile cut points were defined for each cohort. To address whether our results might differ on the basis of cut points used, secondary analyses were performed dividing the study population into quintiles and treating triglyceride levels as a natural logarithm-transformed continuous variable.

Differences between baseline characteristics of participants within each triglyceride category were analyzed using the Cochran-Armitage test for trend for proportions and analysis of variance for continuous measures. Nonparametric Spearman rank correlations were computed for triglycerides with other biomarkers due to the right-skewed distribution.

Cox proportional hazard models were used to compute hazard ratios (HRs) and 95% confidence intervals (CIs) for each tertile or quintile of triglyceride level, using the lowest category as the reference group. Linear tests for trend were performed using the median triglyceride value within each tertile or quintile as an ordinal vari-

able. All analyses were adjusted for random treatment assignment to aspirin, vitamin E, and beta carotene. Models were initially adjusted for potential confounders by traditional nonlipid cardiac risk factors (age, blood pressure, smoking status, and use of hormone therapy). To determine the role of triglycerides independent of other lipid markers, models were then adjusted for levels of total cholesterol and HDL-C. Final multivariable models were additionally adjusted for history of diabetes mellitus, body mass index, and natural logarithm-transformed levels of high-sensitivity C-reactive protein as metabolic risk factors related to insulin resistance with the potential to affect lipid levels.^{8,15} Proportionality of hazards was confirmed by Wald χ^2 testing of the interaction between natural logarithm of triglyceride levels and natural logarithm of follow-up time.

All secondary analyses were performed using the final, fully adjusted multivariable model. The possibility that cardiovascular risk among participants choosing not to fast differed from those who fasted was tested by including a fasting indicator variable in the model. To

further explore the importance of the cut-off time for fasting participants being defined as at least 8 hours since their last meal, models were run with participants categorized by postprandial time (<2, 2-4, 4-8, 8-12, or ≥ 12 hours). Analyses were repeated using each individual component of the composite cardiovascular end point as the outcome. To ascertain the independence of triglycerides from HDL-C and to test for multiplicative interactions in predicting cardiovascular risk, models were run using prespecified clinical categories of HDL-C (< or ≥ 50 mg/dL [to convert to millimoles per liter, multiply by 0.0259]) and triglycerides (< or ≥ 150 mg/dL).

All *P* values were 2-tailed, and *P* < .05 was considered statistically significant. All statistical analyses were performed using SAS version 9.1 (SAS Institute Inc, Cary, North Carolina).

RESULTS

The median (interquartile range) triglyceride levels among fasting and nonfasting participants were 115 (81-169) mg/dL and 133 (93-196) mg/dL, respectively. Baseline characteristics of participants at study entry are de-

tailed in TABLE 1. Among both fasting and nonfasting participants, women with higher levels of triglycerides were significantly more likely to have other cardiac risk factors and markers for the metabolic syndrome. However, when comparing fasting with nonfasting participants, no substantive differences were seen for alcohol use, exercise frequency, total cholesterol, HDL-C, body mass index, high-sensitivity C-reactive protein, or glycated hemoglobin. Compared with the fasting group, those in the nonfasting group were slightly younger, less likely to have hypertension, and had lower levels of LDL-C. Fasting and nonfasting levels of triglycerides were significantly correlated with other measured biomarkers, and correlation coefficients were similar between the 2 groups (TABLE 2).

During a median 11.4 years of follow up, 1001 participants experienced a first cardiovascular event (including 276 nonfatal myocardial infarctions, 265 ischemic strokes, 628 coronary revascularizations, and 163 cardiovascular deaths), for an overall event rate of 3.46 per 1000 person-years of follow-up. Hazard ratios for cardiovas-

Table 1. Baseline Characteristics of Participants by Tertiles of Triglycerides, According to Fasting Status^a

Characteristic	Fasting				Nonfasting			
	All (n = 20 118)	Triglyceride Tertile, mg/dL			All (n = 6391)	Triglyceride Tertile, mg/dL		
		≤ 90 (n = 6590)	91-147 (n = 6802)	≥ 148 (n = 6726)		≤ 104 (n = 2084)	105-170 (n = 2174)	≥ 171 (n = 2133)
Age, mean (SD), y	54.5 (7.2)	53.1 (6.9)	54.8 (7.3)	55.6 (7.2)	53.3 (6.6)	52.2 (6.2)	53.5 (6.7)	54.2 (6.8)
Hypertension, No. (%)	5235 (26.0)	1070 (16.2)	1718 (25.3)	2447 (36.4)	1437 (22.5)	296 (14.2)	435 (20.0)	706 (33.1)
Current smoking, No. (%)	2362 (11.8)	663 (10.1)	815 (12.0)	884 (13.2)	713 (11.2)	179 (8.6)	238 (11.0)	296 (13.9)
Diabetes mellitus, No. (%)	518 (2.6)	65 (1.0)	115 (1.7)	338 (5.0)	198 (3.1)	35 (1.7)	38 (1.8)	125 (5.9)
Postmenopausal, No. (%)	11 179 (55.6)	2996 (45.5)	3907 (57.5)	4276 (63.7)	3241 (50.9)	879 (42.3)	1166 (53.8)	1196 (56.2)
Hormone use, No. (%)	8747 (43.6)	2235 (34.0)	3023 (44.5)	3489 (52.0)	2817 (44.2)	763 (36.7)	1041 (47.9)	1013 (47.7)
≥ 1 Alcoholic drink/d, No. (%)	2116 (10.5)	817 (12.4)	740 (10.9)	559 (8.3)	644 (10.1)	261 (12.5)	207 (9.5)	176 (8.3)
Exercise < once/wk, No. (%)	11 456 (57.0)	3387 (51.4)	3918 (57.6)	4151 (61.7)	3615 (56.6)	1069 (51.3)	1221 (56.2)	1325 (62.1)
Total cholesterol, mean (SD), mg/dL	213 (42)	193 (35)	212 (37)	232 (43)	210 (42)	191 (34)	208 (37)	229 (45)
LDL-C, mean (SD), mg/dL	126 (34)	113 (29)	128 (32)	136 (37)	120 (33)	108 (28)	122 (31)	130 (36)
HDL-C, mean (SD), mg/dL	54 (15)	60 (15)	54 (14)	47 (13)	54 (15)	60 (15)	54 (15)	46 (13)
Body mass index, mean (SD) ^b	25.9 (5.0)	24.2 (4.1)	25.9 (5.0)	27.6 (5.1)	25.9 (5.0)	24.0 (3.9)	25.7 (4.8)	27.9 (5.3)
Glycated hemoglobin, mean (SD), %	5.10 (0.60)	5.00 (0.41)	5.06 (0.48)	5.22 (0.81)	5.10 (0.63)	5.00 (0.45)	5.05 (0.47)	5.26 (0.85)
High-sensitivity CRP, median (IQR), mg/L	2.03 (0.82-4.39)	1.01 (0.45-2.32)	2.02 (0.89-4.21)	3.51 (1.78-6.24)	1.96 (0.78-4.33)	0.99 (0.43-2.24)	2.03 (0.84-4.02)	3.40 (1.61-6.48)

Abbreviations: CRP, C-reactive protein; HDL-C, high-density lipoprotein cholesterol; IQR, interquartile range; LDL-C, low-density lipoprotein cholesterol. SI conversion factors: To convert triglycerides to mmol/L, multiply by 0.0113; total cholesterol, LDL-C, and HDL-C to mmol/L, multiply by 0.0259; and high-sensitivity CRP to nmol/L, multiply by 9.524.

^a*P* < .001 for trend for all comparisons across tertiles for fasting and for nonfasting participants.

^bCalculated as weight in kilograms divided by height in meters squared.

cular events among fasting and nonfasting participants are presented in TABLE 3 by tertiles of triglyceride levels. In univariable analyses of event rates and after adjusting for age, blood pressure, smoking, and use of hormone therapy (model 1), both fasting and nonfasting triglyceride levels were strongly associated with cardiovascular events. Among fasting participants, adjusting for total cholesterol and HDL-C (model 2) and for indicators of insulin resistance (diabetes mellitus, body mass index, and high-sensitivity

C-reactive protein) (model 3) substantially reduced the association between fasting triglyceride levels and cardiovascular events (fully adjusted HRs [95% CIs] for increasing tertiles of fasting triglyceride levels: 1 [reference], 1.21 [0.96-1.52], and 1.09 [0.85-1.41] [$P=.90$ for trend]). In contrast, nonfasting levels maintained a strong association with cardiovascular events in fully adjusted models, with HRs (95% CIs) for increasing tertiles of nonfasting levels of 1 (reference), 1.44 (0.90-2.29), and 1.98 (1.21-3.25) ($P=.006$ for trend). The proportionality assumption was valid for these models as there was no significant interaction between triglyceride levels and time.

To address the possibility that results were driven by the choice of cut points used in the 2 groups, analyses were repeated categorizing the population into quintiles rather than tertiles without substantive differences (TABLE 4). We additionally examined triglyceride levels as a natural logarithm-transformed continuous variable with similar results (Table 3); in fully-adjusted models, a 1-unit in-

crease in log(triglyceride level) (eg, corresponding to an increase in triglyceride level from 55 mg/dL to 150 mg/dL) was associated with an HR (95% CI) of 1.11 (0.92-1.34) for fasting participants ($P=.30$) and of 1.67 (1.18-2.35) for nonfasting participants ($P=.004$).

The inclusion of other potential confounders, including alcohol consumption, exercise, and level of glycated hemoglobin, had no substantive impact on these results. The use of LDL-C levels instead of total cholesterol levels in fully adjusted models likewise provided similar results. Because use of hormone therapy has been shown to increase triglyceride levels by approximately 20%,¹⁶ analyses were also run in the subgroup of hormone nonusers with similar results. Fasting status of participants was not associated with risk of cardiovascular events in univariate analysis or in fully adjusted models.

In analyses stratified by postprandial time, women who had eaten 2 to 4 hours prior to phlebotomy had the strongest association between triglyceride levels and cardiovascular events ($P<.001$ for trend across tertiles) (FIGURE 1). This relationship became

Table 2. Spearman Rank Correlations of Triglyceride Levels With Other Covariates, by Fasting Status^a

Covariate	Fasting	Nonfasting
Non-HDL-C	0.56	0.57
High-sensitivity CRP	0.43	0.41
Total cholesterol	0.41	0.41
HDL-C	-0.40	-0.43
Body mass index	0.33	0.37
LDL-C	0.30	0.28
Blood pressure category	0.24	0.25
Glycated hemoglobin	0.18	0.22
Age	0.18	0.15
Alcohol use	-0.12	-0.13
Exercise level	-0.09	-0.11

Abbreviations: CRP, C-reactive protein; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol.
^a $P < .001$ for all coefficients shown.

Table 3. Association of Triglyceride Levels With Incident Cardiovascular Disease According to Fasting Status

	Triglyceride Tertile			P Value for Trend	Per Unit Increase in Log(Triglyceride Level) ^a	P Value
	1	2	3			
Fasting (n = 20 118)						
Triglyceride level, mg/dL	≤90	91-147	≥148			
No. of participants	6590	6802	6726			
No. of events	126	262	398			
Event rate per 1000 person-y	1.74	3.52	5.48			
Model 1 ^b	1 [Reference]	1.63 (1.31-2.02)	2.23 (1.82-2.74)	<.001	1.94 (1.71-2.22)	<.001
Model 2 ^c	1 [Reference]	1.27 (1.02-1.59)	1.32 (1.03-1.68)	.09	1.34 (1.12-1.60)	.001
Model 3 ^d	1 [Reference]	1.21 (0.96-1.52)	1.09 (0.85-1.41)	.90	1.11 (0.92-1.34)	.30
Nonfasting (n = 6391)						
Triglyceride level, mg/dL	≤104	105-170	≥171			
No. of participants	2084	2174	2133			
No. of events	31	61	123			
Event rate per 1000 person-y	1.35	2.55	5.34			
Model 1 ^b	1 [Reference]	1.48 (0.95-2.29)	2.53 (1.69-3.79)	<.001	2.12 (1.66-2.70)	<.001
Model 2 ^c	1 [Reference]	1.31 (0.83-2.05)	1.94 (1.21-3.10)	.003	1.91 (1.37-2.67)	<.001
Model 3 ^d	1 [Reference]	1.44 (0.90-2.29)	1.98 (1.21-3.25)	.006	1.67 (1.18-2.35)	.004

SI conversion factor: To convert triglyceride values to mmol/L, multiply by 0.0113.

^aFor example, corresponding to an increase in triglyceride level from 55 mg/dL to 150 mg/dL.

^bAdjusted for age, blood pressure, smoking, and use of hormone therapy.

^cAdjusted for covariates in model 1 plus total and high-density lipoprotein cholesterol.

^dAdjusted for covariates in model 2 plus diabetes mellitus, body mass index, and high-sensitivity C-reactive protein.

more attenuated as more time elapsed after the participants' last meal. Triglyceride levels measured within 2 hours of a meal showed no association with cardiovascular risk (HR, 0.59 [95% CI, 0.15-2.28], *P* = .70 for trend). Similar results were obtained when levels were reanalyzed as a natural logarithm-transformed continuous variable.

Fully adjusted HRs for extreme tertiles of triglyceride levels are presented in FIGURE 2 for the individual end points of nonfatal myocardial infarction, nonfatal ischemic stroke, coronary revascularization, and cardiovascular death. Results from the composite cardiovascular end point were consistent across the individual end points studied among both fasting and nonfasting women.

Because prior work has suggested that adjustment for HDL-C eliminates or substantially modifies the relationship of triglycerides with cardiovascular disease,¹⁷⁻¹⁹ we repeated our analyses stratified by HDL-C level (FIGURE 3). In participants with normal HDL-C levels (≥ 50 mg/dL), only elevated nonfasting levels of triglycerides were independently associated with events. While

high levels of triglycerides in both groups increased risk in women with HDL-C levels below the clinical cutoff of 50 mg/dL, the magnitude of this effect was greater in the nonfasting group. No significant multiplicative interactions were found for triglycerides with HDL-C among either fasting or nonfasting participants.

COMMENT

In this large-scale, prospective cohort of initially healthy US women, we observed that higher nonfasting triglyceride levels were strongly associated with an increased risk of future cardiovascular events, independent of baseline cardiac risk factors, levels of other lipids, and markers of insulin resistance. In contrast, fasting triglyceride levels showed little independent association with cardiovascular events. Associations were particularly strong among individuals who had their blood drawn 2 to 4 hours after a meal, and this relationship weakened as more time elapsed postprandially.

Several biological mechanisms provide a plausible explanation for the association between postprandial triglyceride levels and cardiovascular disease.

Following food consumption, triglycerides are transported from the small intestines via chylomicrons through the bloodstream. Lipolysis of the triglycerides within chylomicrons, catalyzed by lipoprotein lipase in tissues, transforms these particles into atherogenic, triglyceride-rich remnant lipoproteins. An elevated postprandial triglyceride level, reflecting either a higher peak level or a delay in clearance of triglyceride-rich particles, can lead to an accumulation of these atherogenic particles.²⁰ The data presented in Figure 1 correspond to the expected time course of postprandial triglyceride metabolism; in response to a meal, triglycerides and remnant lipoprotein concentrations both typically increase to their peaks by approximately 4 hours and decline thereafter.²¹ Taken together, our results support the broad hypothesis that atherosclerosis is, at least in part, a "postprandial phenomenon."^{20,22,23}

Along with the possibly direct atherogenic effects of postprandial lipids, several established cardiac risk factors are associated with triglyceride levels, as seen in Tables 1 and 2. In particular, high levels are one manifestation of the constellation of metabolic dis-

Table 4. Association of Triglyceride Levels With Incident Cardiovascular Disease According to Fasting Status

	Triglyceride Quintile					<i>P</i> for Trend
	1	2	3	4	5	
Fasting (n = 20 118)						
Triglyceride level, mg/dL	≤ 73	74-98	99-132	133-184	≥ 185	
No. of participants	3915	3981	4133	4052	4037	
No. of events	61	87	115	155	241	
Event rate per 1000 person-y	1.41	1.98	2.55	3.52	5.55	
Model 1 ^a	1 [Reference]	1.49 (1.08-2.05)	2.01 (1.49-2.71)	1.93 (1.43-2.60)	3.02 (2.27-4.02)	<.001
Model 2 ^b	1 [Reference]	1.26 (0.91-1.73)	1.50 (1.10-2.04)	1.26 (0.91-1.73)	1.61 (1.16-2.25)	.02
Model 3 ^c	1 [Reference]	1.18 (0.85-1.64)	1.41 (1.03-1.93)	1.08 (0.78-1.51)	1.27 (0.90-1.78)	.60
Nonfasting (n = 6391)						
Triglyceride level, mg/dL	≤ 85	86-113	114-154	155-214	≥ 215	
No. of participants	1273	1233	1320	1273	1292	
No. of events	18	20	43	47	87	
Event rate per 1000 person-y	1.28	1.47	2.97	3.38	6.27	
Model 1 ^a	1 [Reference]	0.90 (0.47-1.72)	1.78 (1.02-3.10)	1.72 (0.99-2.98)	2.81 (1.68-4.73)	<.001
Model 2 ^b	1 [Reference]	0.83 (0.43-1.61)	1.57 (0.89-2.78)	1.41 (0.79-2.55)	2.09 (1.13-3.86)	.003
Model 3 ^c	1 [Reference]	0.89 (0.45-1.75)	1.63 (0.90-2.96)	1.60 (0.87-2.95)	1.99 (1.05-3.78)	.02

SI conversion factor: To convert triglycerides to mmol/L, multiply by 0.0113.

^aAdjusted for age, blood pressure, smoking, and use of hormone therapy.

^bAdjusted for covariates in model 1 plus total and high-density lipoprotein cholesterol.

^cAdjusted for covariates in model 2 plus diabetes mellitus, body mass index, and high-sensitivity C-reactive protein.

turbances associated with insulin resistance and the metabolic syndrome. Elevated postprandial levels of triglycerides may represent an abnormal response to an oral fat load due to insulin resistance, a hypothesis supported by previous case-control studies in individuals with diabetes mellitus or the metabolic syndrome.^{24,25}

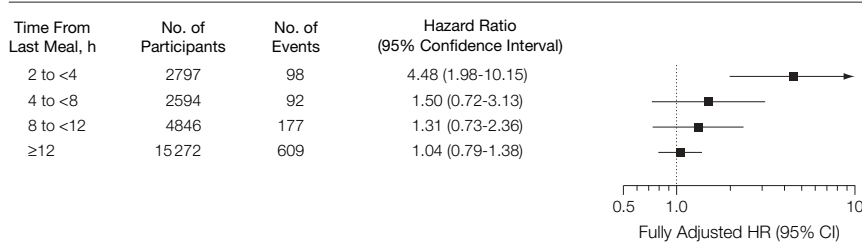
While the relationship between nonfasting triglyceride levels and cardiovascular events in our data persisted after adjustment for many markers of insulin resistance including diabetes mellitus, HDL-C level, blood pressure, body mass index, and high-sensitivity C-reactive protein level, we cannot fully exclude the possibility that

part of the observed effect for nonfasting levels of triglycerides may be due to residual confounding from unmeasured components of the metabolic syndrome. Conversely, our final model may provide a conservative estimate of the effect of triglycerides by adjusting for closely related metabolic parameters.

We believe that the current data demonstrating strong differences between nonfasting and fasting triglyceride levels in terms of vascular risk prediction may help to explain inconsistencies in previous triglyceride studies. The Friedewald equation for the calculation of LDL-C level using fasting levels of lipids,²⁶ coupled with the notion of greater reliability for triglyceride levels measured in the fasting state, have led to national guidelines recommending fasting lipid profiles to assess cardiovascular risk.¹ However, by emphasizing fasting measures, the overall association between plasma triglycerides and vascular risk may be systematically underestimated in the published literature. Our data supporting the importance of nonfasting levels of triglycerides are consistent with prior cross-sectional analyses correlating postprandial levels to extent of carotid atherosclerosis^{21,27} and with case-control studies demonstrating higher postprandial levels among individuals with coronary heart disease compared with healthy controls.²⁸⁻³² Moreover, some prospective studies also have previously demonstrated an independent association between nonfasting triglyceride levels and cardiovascular disease.³³⁻³⁶ However, the use of “casual” blood draws—ie, those for which fasting was not required—in these studies may have underestimated the association between nonfasting triglyceride levels and cardiovascular risk by combining participants in different postprandial states. As suggested by Figure 1, even stronger effects may have been observed had these studies systematically measured levels of triglycerides 2 to 4 hours after food consumption.

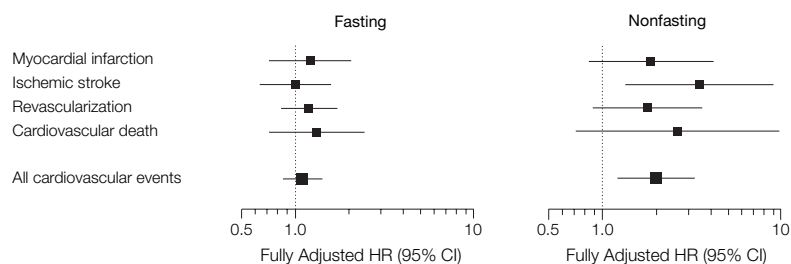
The results of this study suggest that postprandial triglyceride levels may be

Figure 1. Association of Triglyceride Levels With Future Cardiovascular Events, Stratified by Time Since Last Meal



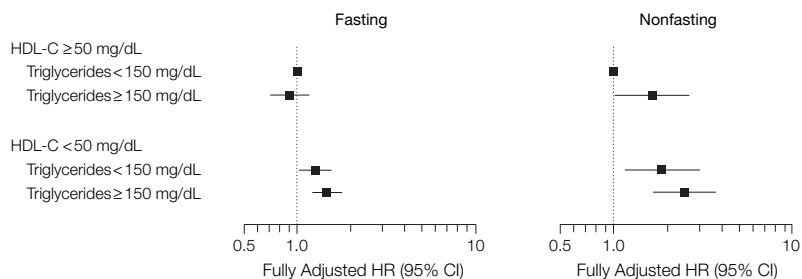
Hazard ratio (HR) and 95% confidence interval (CI) for highest vs lowest tertiles of triglyceride level (see Table 3 for values), adjusted for age, blood pressure, smoking, hormone use, levels of total and high-density lipoprotein cholesterol, diabetes mellitus, body mass index, and high-sensitivity C-reactive protein level.

Figure 2. Association of Triglyceride Levels With Individual Cardiovascular End Points, According to Fasting Status



Hazard ratio (HR) and 95% confidence interval (CI) for highest vs lowest tertiles of triglyceride level (see Table 3 for values), adjusted for age, blood pressure, smoking, hormone use, levels of total and high-density lipoprotein cholesterol, diabetes mellitus, body mass index, and high-sensitivity C-reactive protein level.

Figure 3. Associations of High vs Low Triglyceride Levels With Future Cardiovascular Events, Stratified by HDL-C Level



Hazard ratio (HR) and 95% confidence interval (CI) adjusted for age, blood pressure, smoking, hormone use, total cholesterol level, diabetes mellitus, body mass index, and high-sensitivity C-reactive protein level, using triglyceride levels <150 mg/dL (to convert to mmol/L, multiply by 0.0113) and high-density lipoprotein cholesterol (HDL-C) levels ≥50 mg/dL (to convert to mmol/L, multiply by 0.0259) as the reference group.

superior to fasting levels for assessment of cardiovascular risk, a hypothesis consistent with the concept of atherosclerosis as a postprandial disorder. Strengths of the study include its large sample size, prospective design, extended follow-up time with validated outcomes, and measurement of both fasting and nonfasting levels of triglycerides within the same cohort.

Nonetheless, limitations of this study also merit consideration. First, participants were not randomly assigned to fasting or nonfasting status. However, as shown in Table 1, baseline characteristics of fasting and nonfasting participants were similar for many major risk factors, and despite the slightly higher age, LDL-C level, and prevalence of hypertension among fasting women, fasting status itself was not associated with cardiovascular events. Second, despite the consistency of data in Figure 1 with respect to postprandial time and peak triglyceride levels, the last meal prior to phlebotomy was not given in a standardized manner. However, this potential limitation would likely lead to misclassification bias and therefore result, if anything, in an underestimation of the true effect. Third, given the variability of triglyceride levels, the single measurement of levels at study enrollment without repeated sampling could lead to regression dilution bias,^{2,36} particularly in the nonfasting state. Such an effect, however, also would bias the results toward a null finding rather than a strongly positive one. And fourth, our data are limited to women; thus, further studies are needed before generalizing the conclusions to men.

The use of nonfasting levels of triglycerides in risk assessment provides several potential advantages to clinical practice. Much of the 24-hour day is spent in the nonfasting state, especially considering the fact that triglycerides may take up to 12 hours to return to fasting levels after a meal.³⁷ If postprandial triglycerides are biologically active in atherogenesis, measurement of fasting levels may provide an inadequate representation of vascular

risk. In addition, despite the known strong correlation between fasting and postprandial levels of triglycerides,^{29,32,38} our data suggest that postprandial levels are a more robust indicator of cardiovascular risk, perhaps because the greater variability of postprandial levels captures important information about an individual's metabolism. At a practical level, the use of nonfasting triglyceride levels and the availability of assays to directly measure LDL-C levels could allow patients to have blood for a lipid profile drawn without the need to return to the laboratory after an overnight fast. Additionally, the apparent value seen in this study of triglyceride levels measured 2 to 4 hours after food consumption suggests that a "triglyceride tolerance test" using a standardized meal, analogous to a glucose tolerance test, warrants further evaluation as a potential indicator of a metabolic state predisposing individuals to higher risk for cardiovascular events.

Our observations may have implications for the design and conduct of clinical trials evaluating triglyceride-lowering medications. To date, almost all clinical trials of pharmaceutical agents targeting triglyceride levels have relied on fasting levels as inclusion criteria. However, if levels measured in the fasting state are not the best marker for the atherogenicity associated with hypertriglyceridemia, then it is possible that these trials might have targeted the wrong patient populations. By contrast, previous studies have demonstrated the benefits of several classes of drugs on postprandial elevations in triglyceride levels.³⁹⁻⁴³ Thus, based on the data presented here, future end point reduction trials of triglyceride-lowering agents might consider participant inclusion on the basis of nonfasting rather than fasting triglyceride levels.

Author Contributions: Dr Bansal and Dr Ridker had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Bansal, Mora, Ridker.

Acquisition of data: Buring, Rifai, Ridker.

Analysis and interpretation of data: Bansal, Mora, Sacks, Ridker.

Drafting of the manuscript: Bansal, Ridker.

Critical revision of the manuscript for important intellectual content: Bansal, Buring, Rifai, Mora, Sacks, Ridker.

Statistical analysis: Bansal, Mora.

Obtained funding: Buring, Ridker.

Administrative, technical, or material support: Rifai, Ridker.

Study supervision: Ridker.

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