Supplemental materials

Supplemental Results

Do cytokines add discrimination to models using traditional predictors?

The c-statistic (area under the Receiver Operating Characteristic curve) for the logistic regression model predicting the composite outcome using clinical and demographic characteristics only (including age, race, sex, smoking, alcohol use, ACEi/ARB treatment, DM, HTN and eGFR) was 0.807 (95% confidence interval 79 - 0.82). When TNF-α, IL-6, serum albumin, and fibrinogen were included, the new model had c=.836 (95% CI 0.82 - 0.85), a significant improvement ($p=0.013$). Thus, the cytokines add discrimination to the model using traditional clinical variables. We also examined sensitivity and specificity of both models. In particular, we examined the specificity at the point in the risk score distribution where sensitivity was closest to 0.80. We found that in the model without cytokines, when sensitivity was 0.80, specificity was 0.656. In the model with cytokines, when sensitivity was 0.805, specificity was 0.70. Thus, these cytokines add some predictive accuracy and discrimination to the model that includes only traditional clinical predictors. If we then add proteinuria (UACR) to this model, the new model has c=0.88 (95% CI 0.87 - 0.90), which is significantly better than the model with traditional predictors + cytokines ($p<0.0001$). With sensitivity of 0.80, specificity is also 0.80. Thus, including all the possible predictors produces a model with the best discrimination.

Supplemental Tables

Supplemental Table 1. Cut-points used to calculate quartiles

Variable	**Values**
hS-CRP (mg/L)	
Quartile 1	<1.05
Quartile 2	1.05 to <2.57
Quartile 3	2.57 to <6.51
Quartile 4	≥ 6.51
Fibrinogen (g/L)	
Quartile 1	<3.39
Quartile 2	3.39 to <4.04
Quartile 3	4.04 to <4.80
Quartile 4	≥ 4.80
IL-1β (pg/ml)	
Quartile 1 & 2	< 0.206
Quartile 3	0.206 to <1.286
Quartile 4	≥ 1.286
IL-1RA (pg/ml)	
Quartile 1	< 390
Quartile 2	390 to <715.7
Quartile 3	715.7 to < 1551.0
Quartile 4	≥ 1551.0
IL-6 (pg/ml)	
Quartile 1	< 1.167
Quartile 2	1.167 to <1.903
Quartile 3	1.903 to < 3.151
Quartile 4	≥ 3.151
TNF-α (pg/ml)	
Quartile 1	< 1.5
Quartile 2	1.5 to <2.2
Quartile 3	2.2 to <3.2
Quartile 4	≥ 3.2
TGF-β (pg/mL)	
Quartile 1	<6.47
Quartile 2	6.47 to <10.96
Quartile 3	10.96 to <17.86

Quartile 4	≥ 17.86
UACR	
Quartile 1	<8.651
Quartile 2	8.651 to < 51.936
Quartile 3	51.936 to <458.827
Quartile 4	≥ 458.827
Serum Albumin (g/dl)	
Quartile 1	< 3.7
Quartile 2	3.7 to < 4.0
Quartile 3	4.0 to <4.2
Quartile 4	≥ 4.2

hs-CRP, High sensitive C-Reactive Protein; Interleukin-1RA, interleukin-1 receptor antagonist; TNFα, Tumor necrosis factor α; TGF-β, transforming growth factor-β

Supplemental Table 2. Distribution of end-stage renal disease and Cox regression hazard ratios for cytokine quartiles

	N with ESRD (%)	**p-value**	**Model 1** Hazard ratio (95% Confidence interval)	**p-value**	**Model 2** Hazard ratio (95% Confidence interval)	**p-value**	**Model 3** Hazard ratio (95% Confidence interval)	**p-value**
Fibrinogen		<0.001		<0.001		<0.001		<0.001
Quartile-1	86 (9.9%)		Reference group		Reference group		Reference group	
Quartile-2	132 (15.0%)		1.55 (1.18-2.04)		1.09 (0.83-1.44)		1.04 (0.79-1.37)	
Quartile-3	187 (21.4%)		2.29 (1.77-2.95)		1.40 (1.08-1.82)		1.18 (0.91-1.53)	
Quartile-4	302 (37.3%)		4.86 (3.82-6.18)		2.08 (1.61-2.68)		1.51 (1.17-1.96)	
Interleukin-1β *		<0.001		<0.001		0.99		0.56
Quartile 1 & 2	294 (16.9%)		Reference group		Reference group		Reference group	
Quartile-3	189 (22.3%)		1.39 (1.15-1.66)		1.00 (0.83-1.21)		0.98 (0.81-1.18)	
Quartile-4	224 (26.6%)		1.83 (1.54-2.18)		0.99 (0.83-1.19)		0.91 (0.76-1.09)	
Interleukin-1RA		0.02		0.009		0.83		0.56
Quartile-1	157 (18.0%)		Reference group		Reference group		Reference group	
Quartile-2	161 (19.1%)		1.06 (0.85-1.32)		0.95 (0.76-1.19)		0.95 (0.76-1.18)	
Quartile-3	194 (22.4%)		1.26 (1.02-1.56)		0.92 (0.74-1.14)		0.92 (0.74-1.14)	
Quartile-4	195 (23.1%)		1.37 (1.11-1.69)		0.91 (0.73-1.13)		0.86 (0.69-1.06)	
Interleukin-6		<0.001		<0.001		0.008		0.087
Quartile-1	103 (11.5%)		Reference group		Reference group		Reference group	
Quartile-2	174 (19.8%)		1.87 (1.46-2.38)		1.29 (1.01-1.65)		1.28 (1.00-1.64)	
Quartile-3	207 (24.2%)		2.44 (1.92-3.09)		1.43 (1.12-1.84)		1.35 (1.06-1.73)	
Quartile-4	223 (27.8%)		2.94 (2.33-3.71)		1.50 (1.18-1.91)		1.33 (1.04-1.70)	
TNF-α		<0.001		<0.001		<0.001		<0.001
Quartile-1	57 (7.0%)		Reference group		Reference group		Reference group	

Quartile-2	127 (14.8%)		2.28 (1.67-3.12)		1.19 (0.87-1.63)		0.98 (0.72-1.34)	
Quartile-3	224 (25.9%)		4.49 (3.36-6.01)		1.60 (1.18-2.16)		1.33 (0.99-1.80)	
Quartile-4	299 (33.7%)		6.51 (4.90-8.65)		1.92 (1.43-2.58)		1.47 (1.09-1.97)	
Serum Albumin		<0.001		<0.001		<0.001		<0.001
Quartile-1	315 (41.9%)		5.47 (4.42-6.77)		3.91 (3.14-4.88)		1.75 (1.37-2.22)	
Quartile-2	173 (20.8%)		2.20 (1.73-2.78)		1.88 (1.48-2.39)		1.33 (1.04-1.69)	
Quartile-3	94 (14.5%)		1.52 (1.15-1.99)		1.49 (1.13-1.96)		1.24 (0.94-1.63)	
Quartile-4	115 (10.0%)		Reference group		Reference group		Reference group	

Only biomarkers showing significant association with outcomes in univariate analysis were examined in multivariate analysis.

Interleukin-1RA, interleukin-1 receptor antagonist; TNFα, Tumor necrosis factor α; TGF-β, transforming growth factor-β

Model 1: Unadjusted

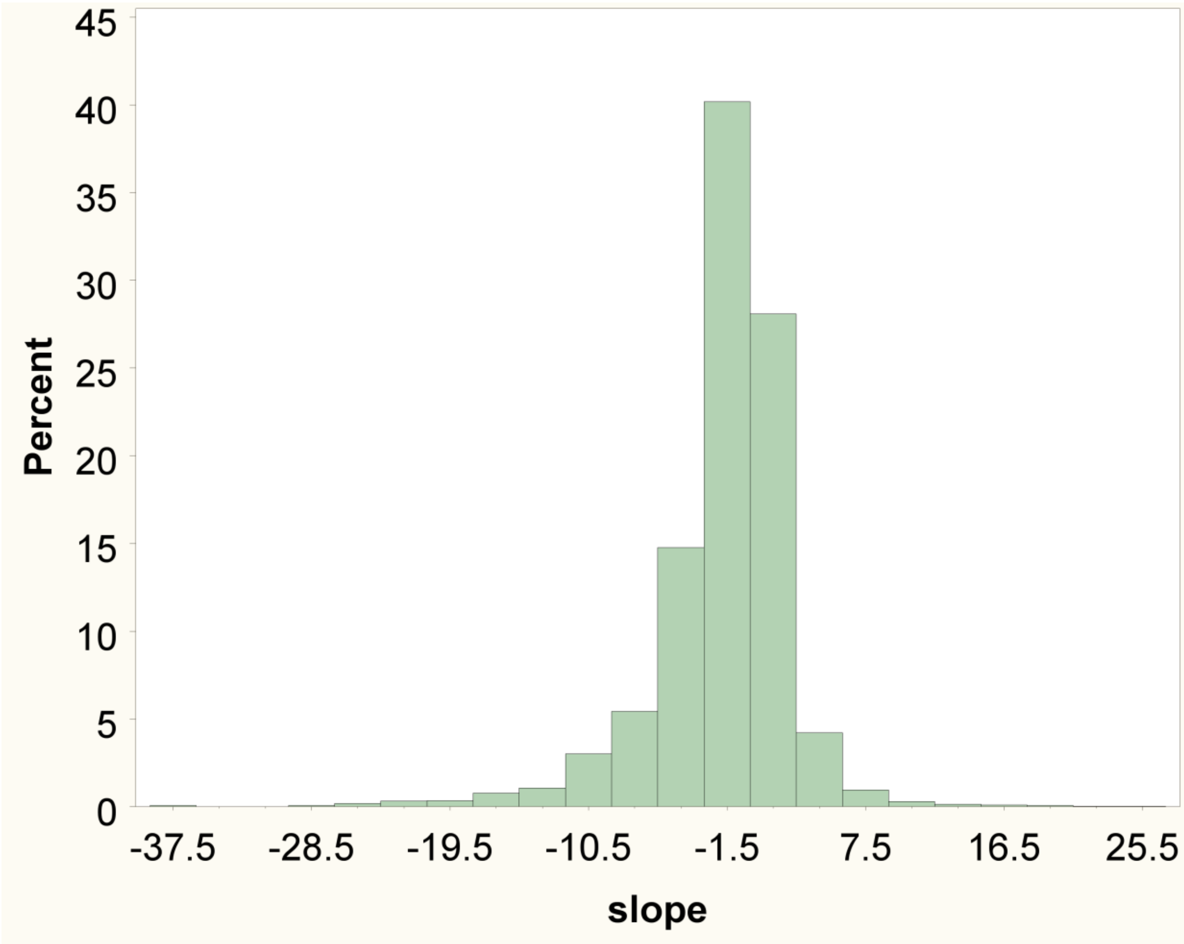
Model 2: Covariates adjusted for included baseline eGFR, age, sex, race, cholesterol, hypertension, diabetes, ACEi/ARB, BMI, alcohol use and smoking

Model 3: Covariates from model 2 and baseline UACR

*Due to the skewed distribution of IL-1β, quartiles 1 & 2 are combined for this variable.

Supplemental Figures

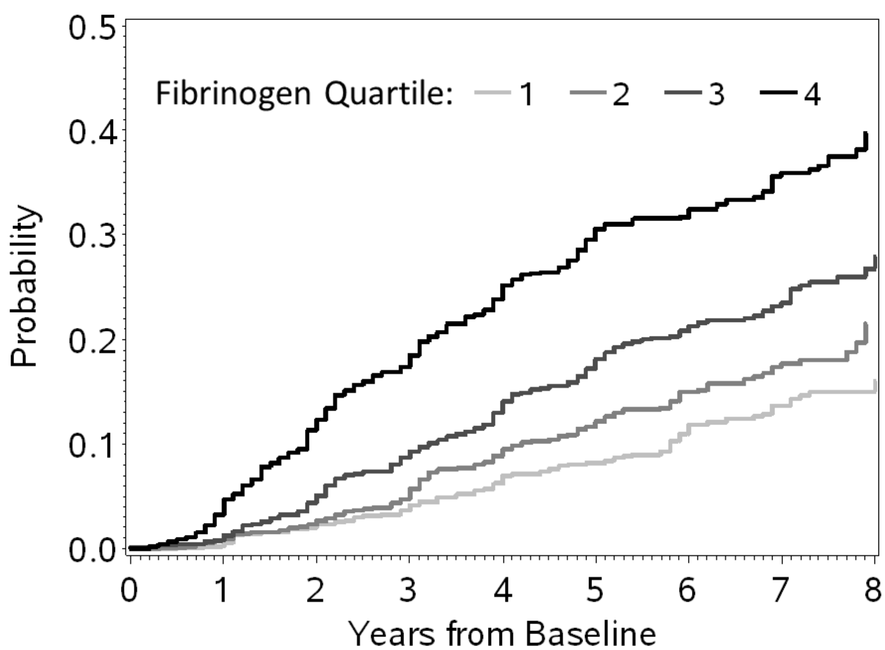
Supplementary Figure 1: Frequency histogram for slope of eGFR change by year (ml/min/1.73m^2)



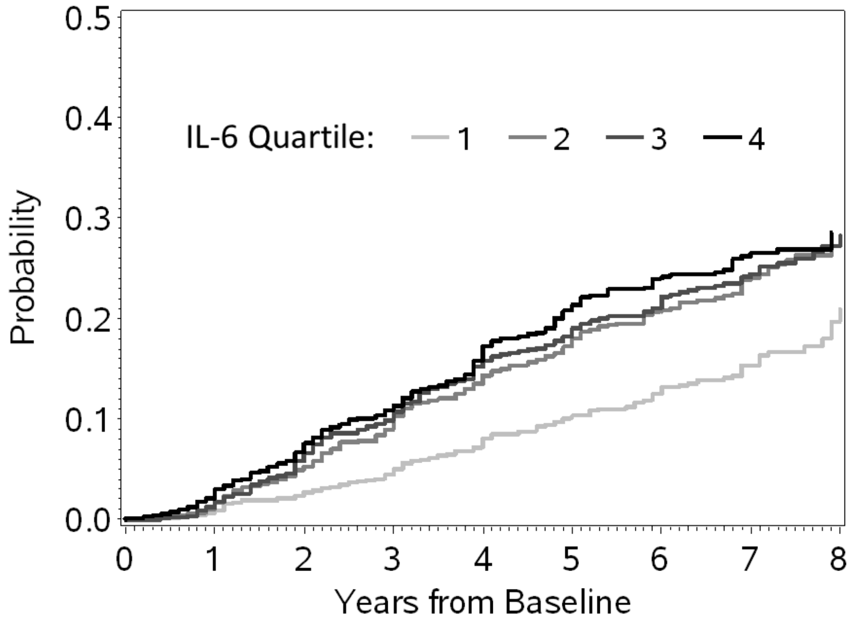
<u>Supplemental Figure 2</u>. Adjusted cumulative incidence functions for the composite endpoint stratified by cytokine quartile, with death as a competing endpoint. Adjusted for baseline eGFR, UACR, race, HTN, alcohol use, smoking, ACE/ARB use, age, BMI, DM, sex, total cholesterol. Gray's test for equivalence of cumulative incidence functions was used to test for differences between quartiles.

A) Fibrinogen: Chi-square 145.63, df=3, p<.0001
B) Interleukin-6: Chi-square 34.63, df=3, p<.0001
C) Tumor necrosis factor-α: Chi-square 154.52, df=3, p<0.001
D) Serum Albumin: Chi-square 197.33, df=3, p<0.001

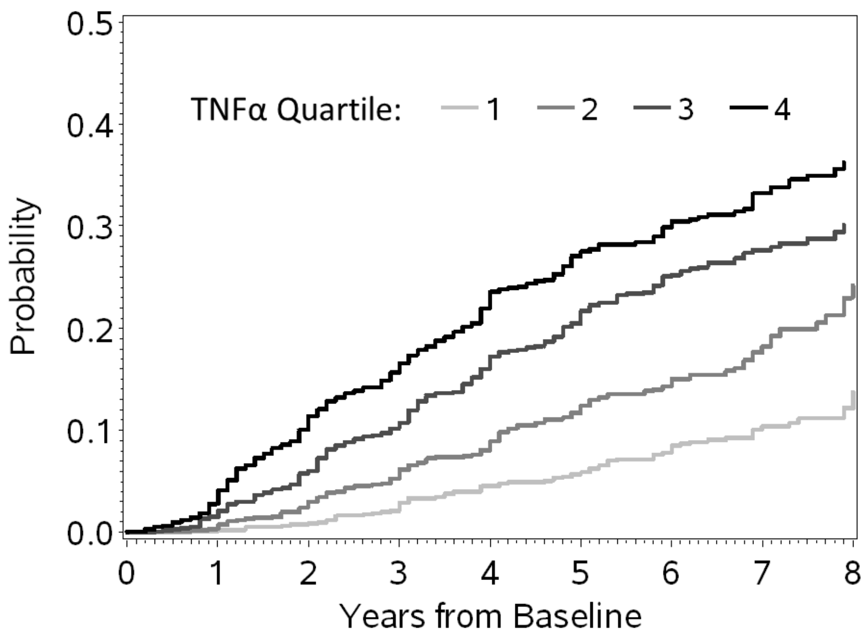
A) Fibrinogen



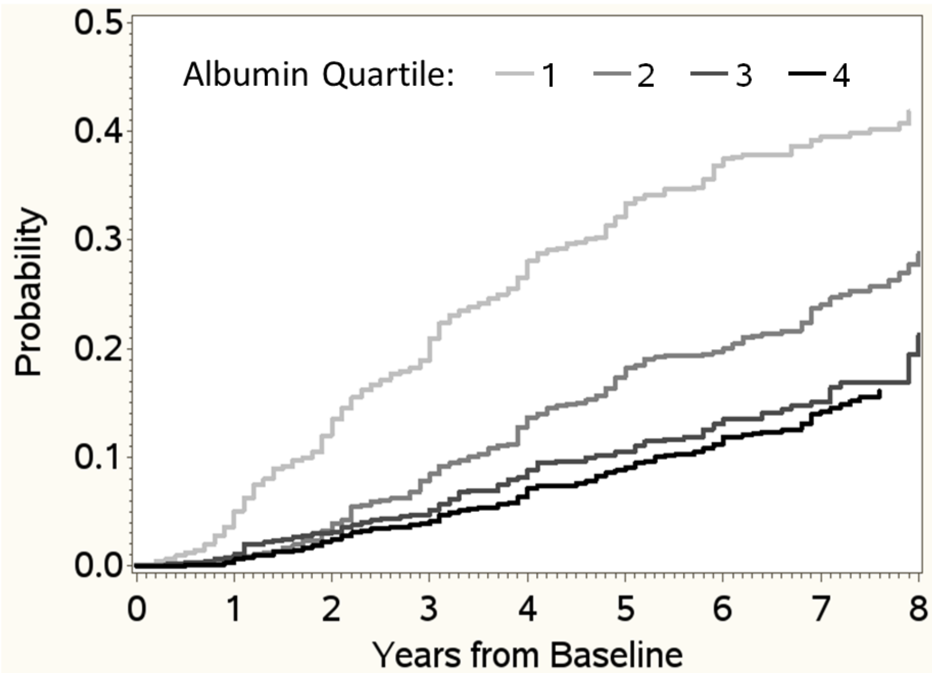
B) Interleukin-6



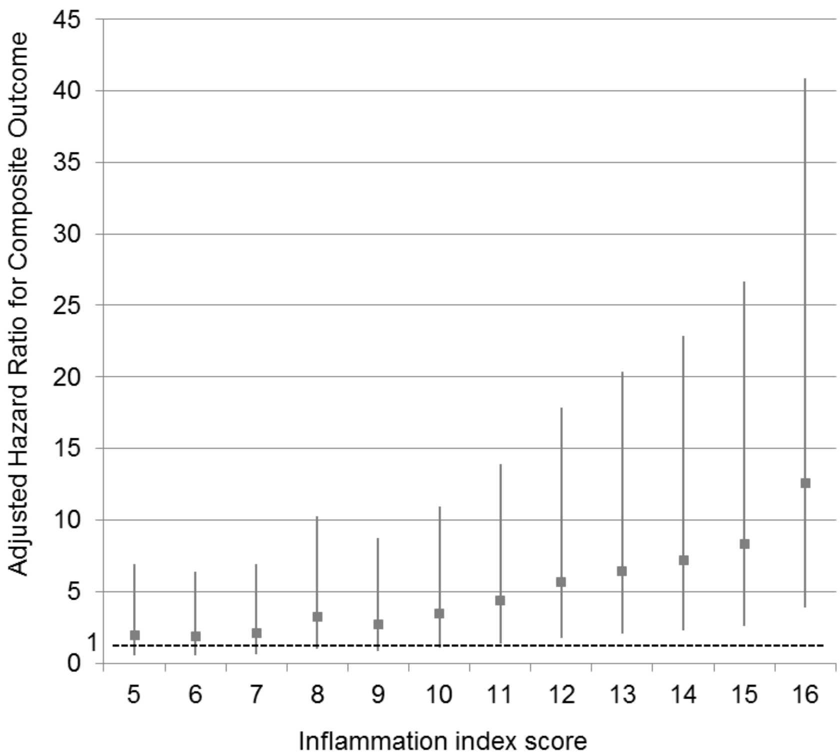
C) Tumor necrosis factor α



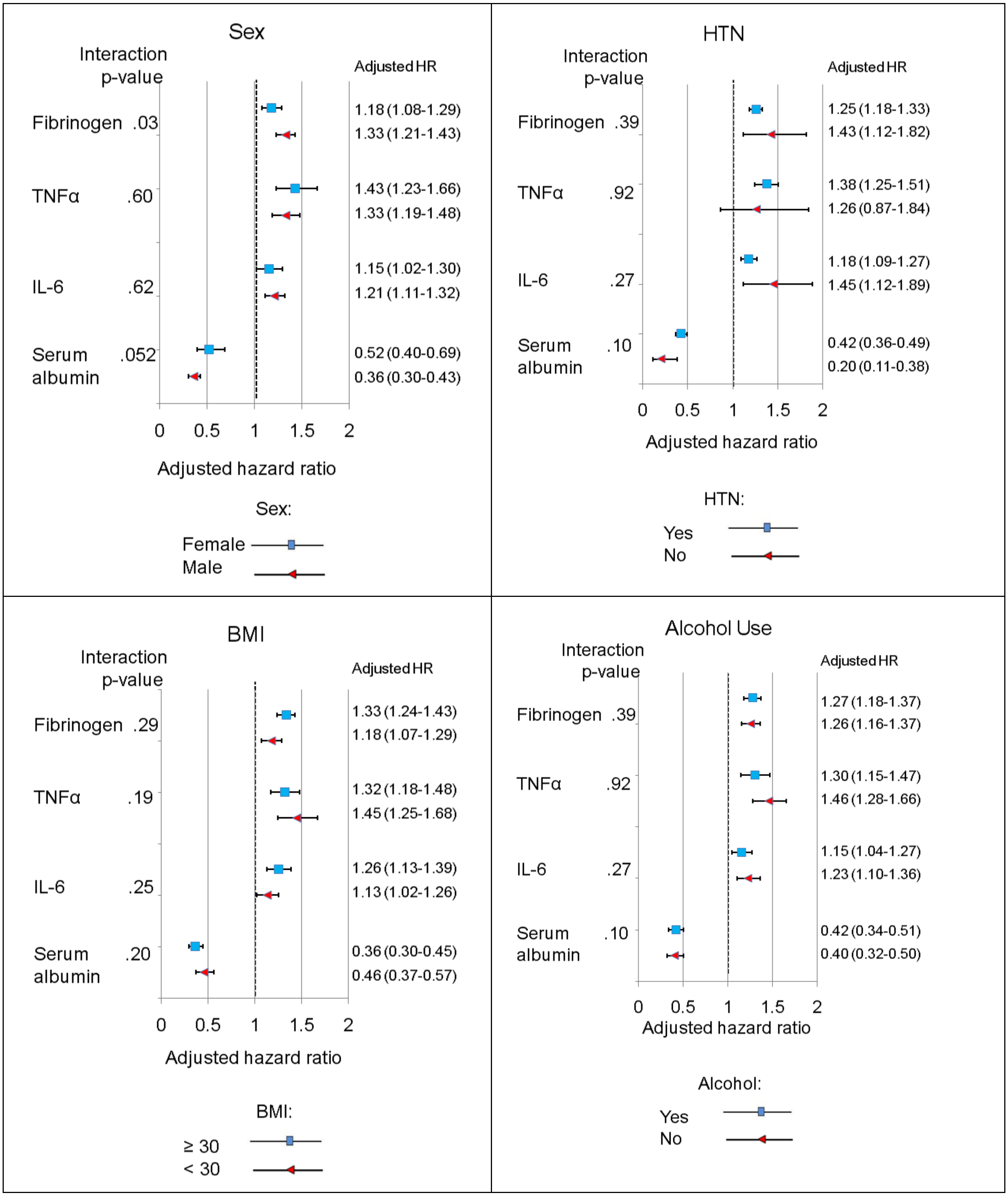
D) Serum Albumin

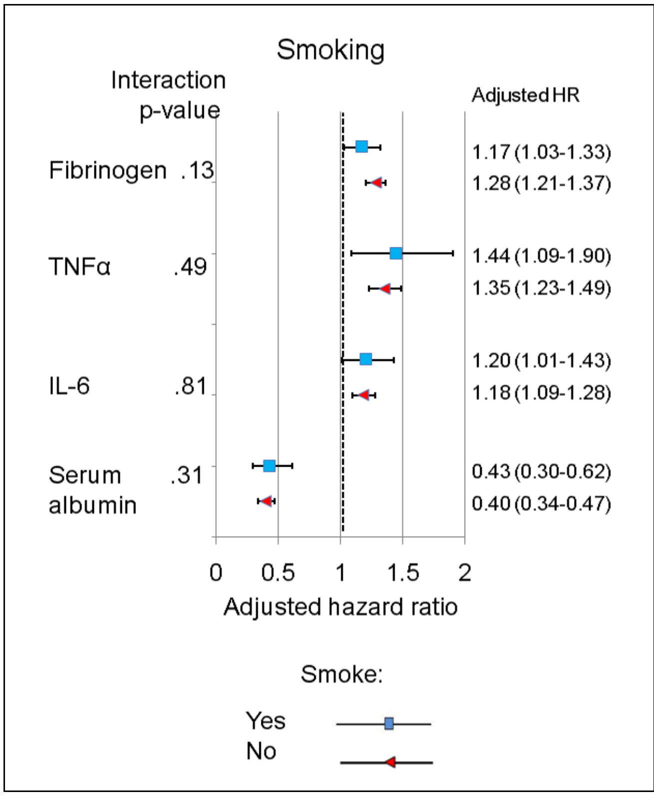


Supplementary Figure 3: Adjusted Hazard Ratios for inflammation score predicting composite outcome using Cox regression. The inflammation score was calculated by adding baseline quartiles of fibrinogen, IL-6, TNFα, and serum albumin (reverse scored). Error bars show the 95% confidence interval. Each score is compared with the lowest possible score of 4. Adjusted for baseline eGFR, SBP, DBP, age, race, sex, DM, alcohol use, smoking, total cholesterol, ACEi/ARB treatment. The hazard ratio for the composite outcome increased with increasing inflammation index score, and became significant at an inflammation index score of 10 (where HR was 3.45 [95% CI 1.09-10.97], p=0.036). When the inflammation index was treated as a continuous variable, it had HR 1.19 [95% CI 1.16-1.22], p<.0001.



Supplementary Figure 4. Interactions of Cytokines with Sex, Alcohol Abuse, Smoking, HTN, and BMI on the composite outcome.




Smoking
Interaction p-value
Adjusted HR
Fibrinogen .13
1.17 (1.03-1.33)
1.28 (1.21-1.37)
TNFα .49
1.44 (1.09-1.90)
1.35 (1.23-1.49)
IL-6 .81
1.20 (1.01-1.43)
1.18 (1.09-1.28)
Serum albumin .31
0.43 (0.30-0.62)
0.40 (0.34-0.47)
0
0.5
1
1.5
2
Adjusted hazard ratio
Smoke:
Yes
No