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Original Article

High-sensitive factor I and C-reactive protein based biomarkers for coronary artery disease

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Abstract: An analysis of high-sensitive factor I and C-reactive proteins as biomarkers for coronary artery disease has been performed from 19 anticipated cohort studies that included 21,567 participants having no information about coronary artery disease. Besides, the clinical implications of statin therapy initiated due to assessment of factor I and C-reactive proteins have also been modeled during studies. The measure of risk discrimination (C-index) was increased (by 0.0101) as per the prognostic model for coronary artery disease with respect to sex, smoking status, age, blood pressure, total cholesterol level along with diabetic history characteristic parameters. The C-index was further raised by 0.0045 and 0.0053 when factor I and C-reactive proteins based information were added, respectively which finally predicted 10-year risk categories as: high (> 20%), medium (10% to < 20%), and low (< 10%) risks. We found 2,254 persons (among 15,000 adults (age ≥ 45 years)) would initially be classified as being at medium risk for coronary artery disease when only conventional risk factors were used as calculated risk. Besides, persons with a predicted risk of more than 20% as well as for persons suffering from other risk factors (*i.e.* diabetes), statin therapy was initiated (irrespective of their decade old predicted risk). We conclude that under current treatment guidelines assessment of factor I and C-reactive proteins levels (as biomarker) in people at medium risk for coronary artery disease could prevent one additional coronary artery disease risk over a period a decade for every 390-500 people screened.

Keywords: Factor I protein, C-reactive protein, biomarkers, coronary artery disease

Introduction

The C-reactive protein is described as the first acute-phase protein and known it is capable to precipitate the somatic C-polysaccharide of *Streptococcus pneumonia*. Besides, it is presently known as an exquisitely sensitive systemic inflammation and tissue damage based biomarker and to date, more than 30 epidemiological studies have shown its strong association with cardiovascular heart disease [1, 2]. This molecule has specific characteristics as inflammatory stimuli as it binds to phosphocholine on microbes and extrinsic ligands bearing phosphocholine residues like native and modified plasma lipoproteins, phospholipids and related compounds, apoptotic cells and other damaged cell membranes [3].

We are living in the era of multivariable statistical derived models as "global risk assessment" scores to check who is cardiovascular risk at but these models have some limitations which have prompted us to the search of novel biomarkers for coronary artery disease. To achieve this goal C-reactive protein and factor I biomarkers have been explored. C-reactive protein has been chosen due to its high-sensitive assays which had received widespread interest and an accumulated large database as its potential role as a predictor of cardiovascular risk by several associations, Canadian Cardiovascular Society [4], Centers for Disease Control and Prevention and American Heart Association [5], European Society of Cardiology [6], National Academy of Clinical Biochemistry Laboratory Medicine Practice Guidelines [7], as well as American College of Cardiology Foundation-American Heart Association Task Force [8], recently. While usefulness of factor I as biomarker is still inconclusive and hence

inspired us to explore it as biomarker of inflammation for coronary artery disease.

On account of usage of factor I and C-reactive protein and need for biomarker of inflammation for coronary artery disease we analyzed records of 21,567 people without a background of coronary artery disease at baseline from 19 anticipated cohort studies. Thus, this approach was used to quantify the improvement in the prediction of a first coronary artery disease risk event by adding the assessment of circulating biomarkers of inflammation into the risk factors' assessment to explore the usage of factor I and C-reactive proteins as biomarkers for coronary artery disease.

Materials and methods

Study cohorts

For anticipated cohort studies, participants were chosen (during follow-up), specifically who did not have a background of baseline coronary artery disease were chosen (during follow-up). Participants were followed up for more than a year to record the data. When we obtained baseline level data of factor I, C-reactive protein or both only then we included them for studies. Additionally, complete information on sex, age, smoking status, systolic blood pressure, diabetic history, as well as the total and high-density lipoprotein cholesterol's levels had also been collected. Besides these, some other data related to albumin-levels, leukocyte-counts, socioeconomic factors, family history of coronary artery disease and low-density lipoprotein cholesterol levels were also available, but only as a subset of study participants. Definition of coronary artery disease was used in the entire studies on the basis of World Health Organization criteria. The study was designed by us and then approved by the ethics review committee of China-Japan Union Hospital of Jilin University, Changchun, Jilin, China.

Statistical analysis

The main outcome of the studies was a first coronary artery disease event. The $\log_e$-transformed values of leukocyte count and C-reactive protein were used during entire analyses. The risk prediction models were based on a Cox proportional hazards model as per reported literature [9] according to study, sex, and (where applicable) randomized trial arm, but log hazard ratios (common coefficients) were estimated across studies. Briefly, for each stratum k = 1, 2, •••, K (for K distinct combinations of study, sex, and trial arm), with $i = 1, 2, \bullet\bullet\bullet, n_k$ individuals in stratum K and the i^{th} individual having baseline covariate values $X_i = (x_{i1}, x_{i2}, \bullet\bullet\bullet x_{ip})$, the probability of surviving beyond t years after baseline without a coronary artery disease event was modeled as

$$S(t \mid X_i, k) = S_{0k}(t)^{\exp(\beta X_i')}$$

However, the probability of a coronary artery disease event within t years after baseline was simply one minus the survival probability, such as

$$\Pr(T \leq t \mid \mathbf{X}_i, k) = 1 - S(t \mid \mathbf{X}_i, k) = 1 - S_{0k}(t)^{\exp(\beta X_i')}$$

We censored the outcomes for patients who died from non-coronary artery disease event causes, so the above probability refers to the risk of a coronary artery disease event in the absence of deaths from other causes. In both the equations the evolution of risk over time was modeled independently for each stratum (i.e. study, sex, and trial arm), which is represented by the non-parametric baseline survival $S_{0k}(t)$.

Prognostic models were compared with the assessment of discrimination using the C-index [10] and the D measure [11] as per reported work to quantify the extent of the model up to which it can predict the disease events [12]. On the other hand for an arbitrary selection of participants, the C-index is the probability that, the individual with the shorter survival time has the higher value of the linear predictor $\beta X_i'$. The overall measure is calculated as

$$C = \frac{C_o + 0.5_u}{C_o + d + u}$$

where Co, u, and d are the number of concordant, undecided and discordant pairs respectively. In present case we allowed a constrained pair-wise comparisons *i.e.* only pairing of participants within the same strata. In other words we can say that the concordance/discordance counts did not include comparison of males to females nor participants from different studies.

Table 1. Baseline characteristics and hazard ratios for first coronary artery disease

Characteristics	Assessment of factor I protein		Assessment of C-Reactive protein	
	Participants/total* (N = 737/15,781)	Hazard ratio (95% CI)†	Participants/total* (N = 810/14,565)	Hazard ratio (95% CI)†
Sex (male; no.)	8363	Not Available#	6991	Not Available#
Age (mean ± SD; yr)	60.9 ± 8.3	1.80 (1.75-1.84)	61.3 ± 8.9	1.89 (1.87-1.93)
Smoking Status (Current; no.)	3787	1.72 (1.65-1.79)	3204	1.65 (1.61-1.69)
Systolic B.P. (mean ± SD; mm Hg)	138 ± 17	1.29 (1.26-1.31)	137 ± 18	1.27 (1.25-1.29)
Diabetic History (no.)	947	1.90 (1.81-2.02)	873	1.73 (1.61-1.84)
Total Cholesterol (mean ± SD; mmol/L)	5.78 ± 1.11	1.21 (1.18-1.23)	5.82 ± 1.09	1.19 (1.16-1.22)
HDL Cholesterol (mean ± SD; mmol/L)	1.34 ± 0.34	0.84 (0.81-0.86)	1.34 ± 0.39	0.87 (0.85-0.90)
Factor I (g/L)	3.11 ± 0.73	1.16 (1.14-1.18)	-	-
Log_e(C-Reactive Protein) (mg/L)	-	-	0.60 ± 1.07	1.22 (1.17-1.23)
Anticipated Cohort Studies$	BRUN; BWHHS; COPEN; EAS; FINRISK97; HISAYAMA; IKNS; LEADER; MESA; NHANESIII; NSHS; PROCAM; SHHEC; SHS		AFTCAPS; ARIC; BRUN; BWHHS; EAS; FINE-FIN; FINE-IT; FINRISK97; KIHD; NHANESIII; TARFS; WOSCOPS	

*Data on factor I are from 14 studies in which 737 participants had a first-ever coronary artery disease outcome during follow-up and data on C-reactive protein are from 12 studies in which 810 participants had a first-ever coronary artery disease outcome during follow-up. †The hazard ratios were calculated per 1-SD increment in the measured level or as compared with the relevant reference category. Hazard ratios were adjusted for sex, age, smoking status, systolic blood pressure, diabetic history, and levels of total and high-density lipoprotein (HDL) cholesterol and factor I or C-reactive protein. #Because these models were stratified by sex. SD: Standard Deviation; yr: Year; $AFTCAPS: Air Force/Texas Coronary Atherosclerosis Prevention Study [15]; ARIC: Atherosclerosis Risk in Communities Study [16]; BRUN: Bruneck Study [17]; BWHHS: British Women's Heart and Health Study [18]; COPEN: Copenhagen City Heart Study [19]; EAS: Edinburgh Artery Study [20]; FINE-FIN: Finland, Italy and Netherlands Elderly Study-Finland cohort [21]; FINE-IT: Finland, Italy and Netherlands Elderly Study-Italian cohort [21]; FINRISK97: Finrisk Cohort 1997 [22]; HISAYAMA: Hisayama Study [23]; IKNS: Ikawa, Kyowa, and Noichi Study [24]; LEADER: Lower Extremity Arterial Disease Event Reduction Trial [25]; MESA: Multi-Ethnic Study of Atherosclerosi [26]; NHANESIII: Third National Health and Nutrition Examination Survey [27]; NSHS: Nova Scotia Health Survey [28]; PROCAM: Prospective Cardiovascular Munster Study [29]; SHHEC: Scottish Heart Health Extended Cohort [30]; SHS: Strong Heart Study [31]; TARFS: Turkish Adult Risk Factor Study [32]; WOSCOPS: West of Scotland Coronary Prevention Study [33].

However, the D measure was by first transforming each participant's linear predictor βXi' from the fitted Cox proportional hazards model to give standard normal order rank statistics (using Blom's approximation). During present analyses the standard normal rank order statistics were formed within studies to avoid potential influences from between study differences in covariate distributions. Division of the rank statistics with a factor of $\sqrt{\frac{\pi}{8}}$ gave z_i and then a second stratified Cox proportional hazards model is then fitted to these values. Thus, D is the regression coefficient of *z* from this second model. Thus we used two-stage method to examine the between-study heterogeneity through C-index and D measure calculations. As per number of coronary artery disease outcomes' contribution our studies were weighted. The I^2 statistic approach [13] was used to quantify the heterogeneity lied in the risk-discrimination and in this way it has measured all the changes. However, the well known chi-square tests were used to determine the discrimination measurements (across subgroups).

Prognostic models were also compared with the use of risk reclassification assessment [14]. Reclassification tables were constructed as per data from studies containing both the fatal and the nonfatal coronary artery disease outcomes which had examined the participants among three predicted decades coronary artery disease categories ("high" > 20%; "medium" 10% to < 20%; and "low" < 10%) when factor I and C-reactive proteins were included to the model of conventional risk factors. A reduction of coronary artery disease by 20% with statin treatment was assumed for clinical modeling. All *P* values were two-sided. The latest version of Stata software was utilized for the entire analyses.

Results

Characteristics of the study participants

We started the analysis with 19 studies involving 21,567 participants. However, the mean (± SD) age at baseline was 62 ± 8 years. Overall, there were 61% men out of the all the participants. Information on factor I levels was explored for total a 15,781 number of participants from 14 cohorts studies (**Table 1**), among whom there were first 737 coronary artery disease events. Information on C-reactive protein

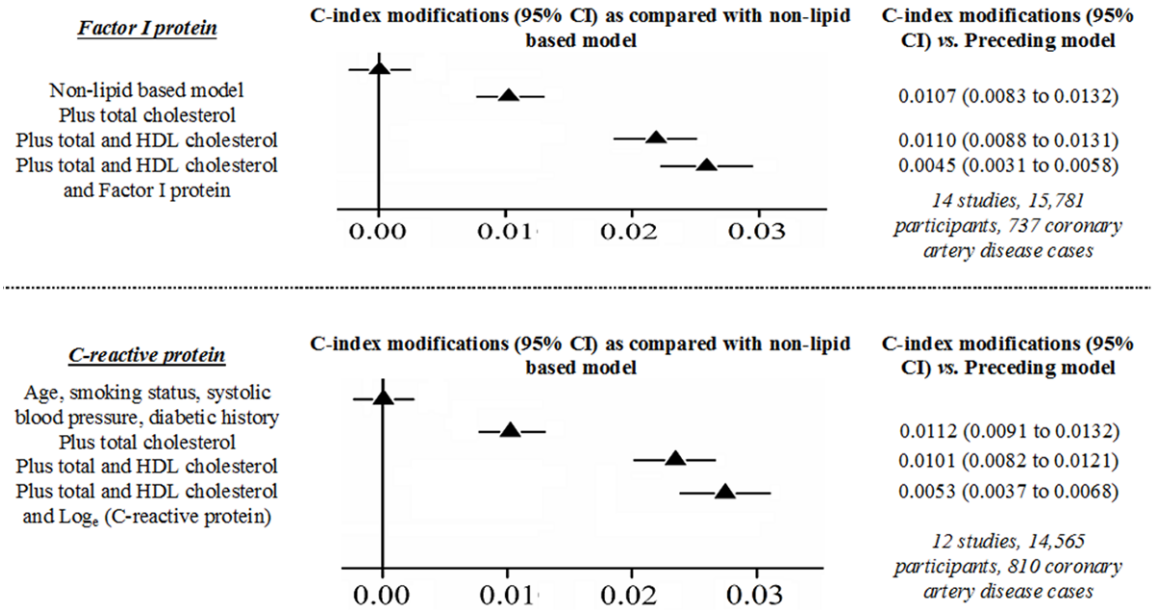

Figure 1. Modifications in C-index for coronary artery disease risk prediction after addition of lipids markers and biomarkers (factor I protein or C-reactive protein) to a non-lipid based model. The non-lipid based model denotes age, smoking status, systolic blood pressure, diabetic history (stratified as per sex). CI denotes confidence interval and HDL denotes high-density lipoprotein.

levels was available for 14,565 participants from 12 anticipated cohorts (**Table 1**), among whom there were first 810 coronary artery disease events. Information on both factor I and C-reactive proteins levels was available for 8,410 participants. **Table 1** is shown here to express the baseline characteristics of participants in the factor I and C-reactive proteins analyses, along with the most important factor viz. the adjusted hazard ratios of first coronary artery disease events. Detailed data related to anticipated cohort studies are not shown to avoid mere over-compilation of data.

Factor I and C-reactive proteins as biomarkers: Incremental values in C-index

To the prognostic model for first coronary artery disease events included data on sex, age, smoking status, systolic blood pressure, diabetic history, along with total and high-density lipoprotein cholesterol's level, the addition of HDL cholesterol led to increase in C-index by 0.0101 (**Figure 1**; **Table 2**). And, addition of factor I and C-reactive proteins biomarkers of inflammation to this model enhanced the C-index by 0.0045 for factor I and by 0.0053 for C-reactive protein (**Figure 1**; **Table 3**). Thus factor I and C-reactive proteins biomarkers yielded 0.91% and 1.61% respective net reclassification improvement for the predicted decade risk categories of high (> 20%), medium (10% to < 20%), and low (< 10%) ($P < 0.02$; data not shown). The corresponding values for the integrated discrimination index were 0.0029 and 0.0038 (data not shown). We can easily observe in **Figure 1** that the combined predictive value of total and HDL cholesterol was higher than that of either factor I or C-reactive proteins biomarkers.

In analyses that included measures of family history of coronary artery disease, body-mass index, or both risk factors (**Table 4**) both the biomarkers have shown similar effect, especially, in those analyses which had excluded extreme factor I and C-reactive protein values and also in those analyses which had excluded participants known to be taking blood pressure or lipid levels lowering medications at study entry (data not shown).

Interestingly, in those analyses which had excluded one risk factor at a time from conventional risk factors plus measures of factor I and C-reactive protein values based model and thus we observed broadly a similar pattern when compared to those analyses which had

Table 2. Modifications in C-index and net reclassification improvement for coronary artery disease risk prediction from addition of lipids markers and factor I protein (or C-reactive protein) to a non-lipid based model

	C-index (95% CI)	C-index modifications (95% CI)	*P* value for C-index modification differences	Net reclassification improvement (95% CI)
Factor I protein	*14 studies, 15,781 participants, 737 coronary artery disease cases*			*9 studies, 7,051 participants, 424 coronary artery disease cases*
Non-lipid based model	0.6022 (0.5981, 0.6067)	Reference model		Reference model
Plus total cholesterol	0.6127 (0.6081, 0.6172)	0.0107 (0.0084, 0.0130)	Reference	1.45 (0.72, 2.17)
Plus total and HDL cholesterol	0.6239 (0.6195, 0.6285)	0.0217 (0.0198, 0.0236)	< 0.0001	2.64 (1.73, 3.51)
Plus total and HDL cholesterol and Factor I protein	0.6284 (0.6239, 0.6330)	0.0262 (0.0243, 0.0281)	< 0.0001	3.40 (2.44, 4.36)
C-reactive protein	*12 studies, 14,565 participants, 810 coronary artery disease cases*			*6 studies, 6,344 participants, 408 coronary artery disease cases*
Non-lipid based model	0.6009 (0.5965, 0.6055)	Reference model		Reference model
Plus total cholesterol	0.6121 (0.5976, 0.6166)	0.0112 (0.0093, 0.0131)	Reference	1.22 (0.53, 1.93)
Plus total and HDL cholesterol	0.6221 (0.6177, 0.6268)	0.0213 (0.0194, 0.0232)	< 0.0001	1.89 (0.91, 2.78)
Plus total and HDL cholesterol and C-reactive protein	0.6275 (0.6230, 0.6319)	0.0266 (0.0247, 0.0285)	< 0.0001	3.41 (2.42, 4.42)

The non-lipid based model denotes age, smoking status, systolic blood pressure, diabetic history (stratified as per sex). CI denotes confidence interval and HDL denotes high-density lipoprotein.

Table 3. C-index for coronary artery disease after addition of biomarkers (factor I or C-reactive proteins) to conventional risk factors

	Reference model		Modifications after addition of biomarkers			
	C-index (95% CI)	*I^2 (95% CI)*	*C-index modifications (95% CI)*	*Z value*	*p value*	*I^2 (95% CI)*
Factor I protein (14 studies, 15,781 participants, 737 coronary artery disease cases)						
Age	0.5530 (0.5485, 0.5576)	96 (95, 97)	0.0163 (0.0141, 0.0181)	13.9	< 0.0001	79 (69, 88)
Above plus smoking status	0.5705 (0.5661, 0.5751)	95 (94, 96)	0.0131 (0.0106, 0.0154)	10.5	< 0.0001	65 (44, 84)
Above plus SBP*	0.5931 (0.5885, 0.5976)	95 (94, 96)	0.0092 (0.0076, 0.0109)	9.1	< 0.0001	48 (27, 69)
Above plus diabetic history	0.6022 (0.5981, 0.6067)	95 (94, 96)	0.0065 (0.0045, 0.0086)	8.3	< 0.0001	40 (19, 61)
Above plus total cholesterol	0.6127 (0.6081, 0.6172)	95 (94, 96)	0.0059 (0.0046, 0.0074)	7.3	< 0.0001	39 (8, 68)
Above plus HDL cholesterol	0.6239 (0.6195, 0.6285)	95 (94, 96)	0.0045 (0.0036, 0.0054)	5.1	< 0.0001	18 (0, 39)
C-reactive protein (12 studies, 14,565 participants, 810 coronary artery disease cases)						
Age	0.5115 (0.5076, 0.5159)	95 (94, 96)	0.0161 (0.0144, 0.0176)	13.2	< 0.0001	68 (47, 89)
Above plus smoking status	0.5236 (0.5190, 0.5280)	95 (94, 96)	0.0146 (0.0131, 0.0162)	11.3	< 0.0001	47 (26, 68)
Above plus SBP*	0.5948 (0.5902, 0.5992)	95 (94, 96)	0.0104 (0.0090, 0.0119)	9.2	< 0.0001	32 (11, 51)
Above plus diabetic history	0.6009 (0.5965, 0.6055)	94 (93, 95)	0.0081 (0.0064, 0.0096)	8.4	< 0.0001	25 (5, 44)
Above plus total cholesterol	0.6121 (0.5976, 0.6166)	94 (93, 95)	0.0063 (0.0051, 0.0075)	8.1	< 0.0001	17 (0, 37)
Above plus HDL cholesterol	0.6221 (0.6177, 0.6268)	94 (93, 95)	0.0053 (0.0044, 0.0062)	6.5	< 0.0001	4 (0, 34)

*SBP, systolic blood pressure; CI denotes confidence interval; and HDL denotes high-density lipoprotein.

Table 4. Risk determination (in terms of C-index) for coronary artery disease after addition of biomarkers (factor I or C-reactive proteins) to conventional risk factors plus body mass index and family history of the disease

	Reference model		Modifications after addition of biomarkers			
	C-index (95% CI)	*I^2 (95% CI)*	*C-index modifications (95% CI)*	*Z value*	*p value*	*I^2 (95% CI)*
Factor I protein (7 studies, 7,927 participants, 433 coronary artery disease cases)						
Conventional risk factors[#]	0.6372 (0.6322, 0.6423)	95 (93, 96)	0.016 (0.011, 0.021)	3.9	< 0.0001	39 (29, 47)
Above plus body mass index	0.6371 (0.6322, 0.6423)	95 (93, 96)	0.0020 (0.0013, 0.0027)	4.2	< 0.0001	35 (14, 53)
Above plus family history of coronary artery disease	0.6387 (0.6339, 0.6441)	95 (93, 96)	0.0020 (0.0013, 0.0027)	4.2	< 0.0001	28 (0, 48)
C-reactive protein (9 studies, 8,669 participants, 529 coronary artery disease cases)						
Conventional risk factors[#]	0.6333 (0.6282, 0.6381)	95 (93, 96)	0.021 (0.014, 0.027)	3.5	< 0.0001	28 (0, 52)
Above plus body mass index	0.6332 (0.6281, 0.6380)	95 (93, 96)	0.0023 (0.0014, 0.0065)	4.2	< 0.0001	21 (0, 46)
Above plus family history of coronary artery disease	0.6345 (0.6295, 0.6394)	95 (93, 96)	0.0023 (0.0014, 0.0065)	4.2	< 0.0001	14 (0, 39)

[#]Conventional risk factors include age, smoking status, systolic blood pressure, Diabetic history, total cholesterol and HDL cholesterol (stratified by sex); CI denotes confidence interval.

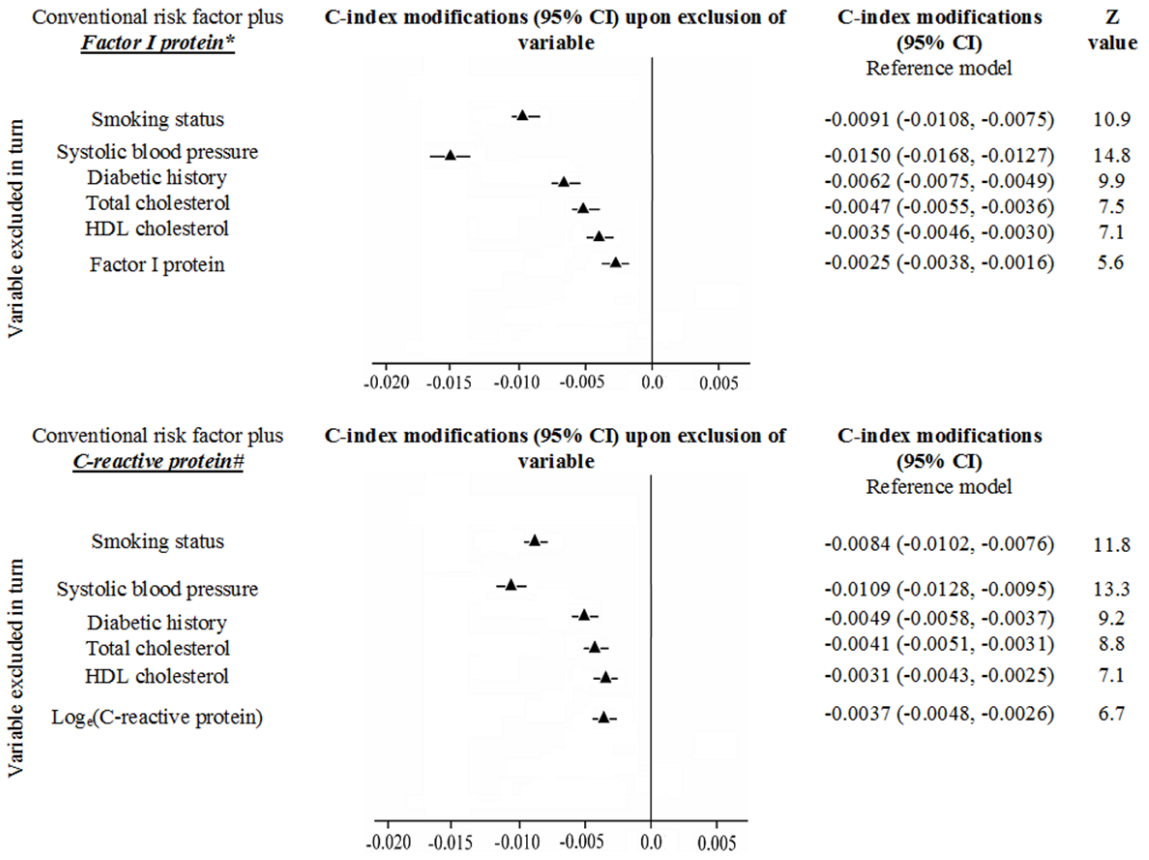

Figure 2. Modifications in C-index for coronary artery disease risk prediction after exclusion of individual risk factors from a full model containing conventional risk factors and biomarkers (factor I protein or C-reactive protein). CI denotes confidence interval and HDL denotes high-density lipoprotein. *14 studies, 737 coronary artery disease cases; #12 studies, 810 coronary artery disease cases.

excluded the values of factor I and C-reactive protein levels (**Figure 2**).

Factor I and C-reactive proteins as biomarkers: Assessment in clinically relevant subgroups

We observed that the factor I and C-reactive proteins have shown improved coronary artery disease risk discrimination only in men ($P < 0.001$ for the interaction) (**Figure 3**). Besides, C-reactive protein as biomarker was efficient in current smokers ($P < 0.001$ for the interaction) (**Figure 3**). However, there were no significant differences in coronary artery disease risk discrimination observed in other clinically relevant subgroups (**Figure 3**). Nevertheless, in further analyses of stratified participants as per their exclusion as smokers (or to their smoking status) we analyzed the persistence of the differences in the improvement of coronary artery disease risk discrimination with change in sex with the use of factor I and C-reactive proteins levels. The similar trend was observed (although information on such treatments was incomplete) in analyses in which hormonal treatment receiving women at baseline were not incorporated.

Reclassification from 8 studies that involved only both the genders as 4,510 men with 456 first coronary artery disease events and 6,795 women with 367 first coronary artery disease events with at least 10 years of follow-up to further explore sex-specific findings was assessed. In these studies, we observed, the net reclassification improvement along with C-reactive protein's measurement among men was 1.31% (95% confidence interval [CI], -0.17 to 2.82; $P = 0.08$). On the other hand the same characteristic value among the opposite gender was 0.41% (95% CI, -0.34 to 1.57; $P = 0.51$). According to sex, the effect of modification was not observed with measures of either systolic blood pressure or HDL cholesterol (data not shown). In the similar way, when analyses of factor I and C-reactive

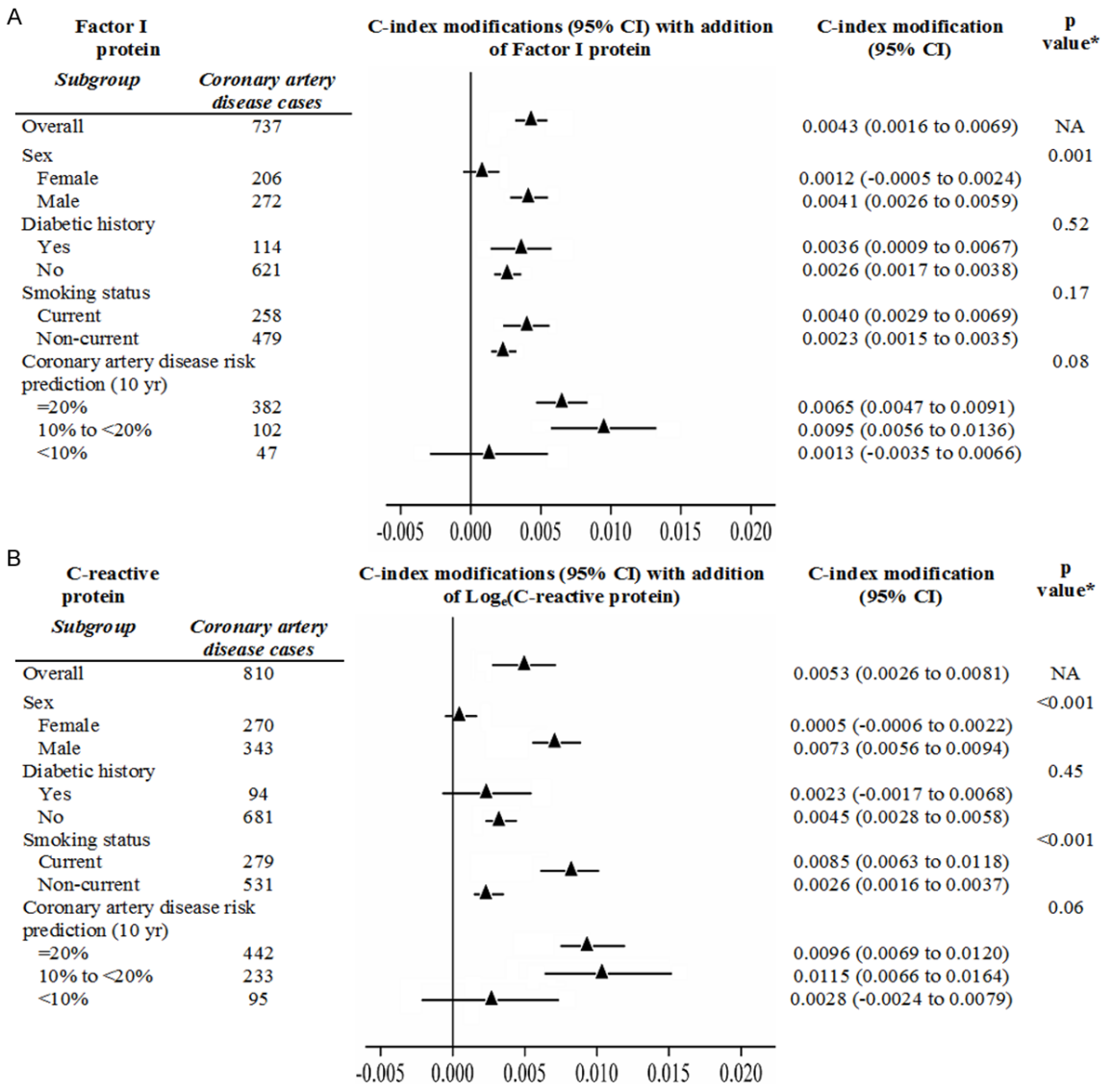

Figure 3. Modifications in C-index for coronary artery disease risk prediction with addition of information on biomarkers ((A) factor I protein or (B) C-reactive protein) to a model with conventional risk factors (based on the subgroups). CI denotes confidence interval. Only Studies with information on all subgroup levels were used to fulfill the comparison purpose and hence, subgroup total numbers do not always equal overall total numbers. *For heterogeneity.

proteins levels were conducted with the data that included participants of three age groups (45- < 55 years, 55- < 65 years, and ≥ 65 years; P = 0.59) we observed no significant effect of modification according to the change in age groups.

Estimation of factor I and C-reactive proteins as biomarkers' potential for coronary artery disease prevention

To investigate the potential of factor I and C-reactive proteins as targeted biomarkers in people whose risk of a coronary artery disease risk event was 10% to < 20% over the subsequent 10 years we modeled this work. In a population of 15,000 adults (of age ≥ 45 years) 2,254 persons were classified as a predicted risk feature of coronary artery disease of 10% to < 20% over a period of 10 years (when only conventional risk factors were used for risk calculation). According to the Adult Treatment Panel III guidelines [34] allocation to statin treatment was conducted and found that 1,979 participants at "medium" risk were ineligible for statin treatment at that time. In addition to this, the assessment of factor I in these participants (1,979) reclassified 95 participants (4.8%) to a

predicted risk of ≥ 20%. Interestingly, approximately 20 out of 95 participants were expected to have a coronary artery disease risk within 10 years; correspondingly, additional assessment of C-reactive protein in the same participants had reclassified 113 participants (5.7%) to a predicted risk of ≥ 20%. Besides, approximately 25 out of these 113 participants were expected to have a coronary artery disease risk within a decade. When we assumed that a predicted risk of ≥ 20% reclassified person would begin statin therapy then as per Adult Treatment Panel III guidelines, such targeted factor I we found that about 4 (*i.e.* 0.20 × 20) additional coronary artery disease risk over a 10-year period could be prevented; the corresponding assessment of C-reactive protein led us to a result where about 5 (*i.e.* 0.20 × 25) additional coronary artery disease risk over a period of 10 years could be prevented.

We may say that targeted assessment of factor I in people at "medium" risk for a coronary artery disease risk could prevent an additional event over a decade for every 494 (1,979 ÷ 4) screened participants which has also resulted in about 23 additional participants began with statin therapy (*i.e.* 4.8% × 494). Similarly the corresponding would be screened number with targeted C-reactive protein assessment were about 396 (1,979 ÷ 5).

Discussion

In the present studies the effects of addition of factor I or C-reactive protein to the standard risk factors were evaluated to predict the risk of a first coronary artery disease risk in the analyses comprising of individual participant data from 19 anticipated cohort studies. Thus to establish the utility of factor I or C-reactive proteins as biomarkers of inflammation in coronary artery disease we then modeled a scenario in which factor I or C-reactive protein was assessed in participants having "medium" risk (a predicted risk of 10% to < 20% over a decade) after initial screening by using mere factors of conventional risk. As per the recommendations of the Adult Treatment Panel-III criteria [34], when measurement of a biomarker of inflammation coupled with initiation of statin therapy, we achieved a result data which suggest that one extra coronary artery disease risk would be prevented in the decade for approximately every 495 people were being assessed with factor I protein levels or approximately every 396 people in which C-reactive protein levels were assessed, on account of the initiation of statin therapy in about 23 additional participants. Our studies also suggested that analyses in which both factor I and C-reactive proteins levels were included had similar outcome in comparison to those in which either of the protein biomarker was used.

We assumed in our main model that the assessment of factor I or C-reactive protein would provide similar predictions of the coronary artery disease risk across population subgroups. Surprisingly, we found from an exploratory analysis that these biomarkers improve risk discrimination characteristic only in one gender (men but no significant improvement was observed in women). This is the limitation of our work that we could not find a proper explanation of this kind of result (sex-differentiated outcome of biomarkers). This area should be explored further to find out a good explanation for the same. Our results also confirm that we have developed such risk prediction scores which have tendency to combine the results of fatal and nonfatal coronary artery disease.

This study has its strengths and potential limitations. We involved analyses of population of several countries along with China and found harmonious outcomes among most of the measures based on risk discrimination and reclassification. We would like to acknowledge that even we used a standard decade-time frame along with conventional clinical risk categories, but these types of reclassification analyses are very fragile and highly dependent on how we chose the risk categories and landmark time.

We also observed that the differences due to studies in levels of factor I or C-reactive protein biomarkers of inflammation contributed relatively very little to the heterogenic characteristic feature. The reason behind this would be mostly to differing age ranges across cohorts. We assumed that all eligible people would receive statin and hence it might be possible that we overestimated the potential benefits of statin in our models. Besides this, we had incomplete information on statin use, and hence estimation of individual risk factors' or risk models' effect on results might be influenced.

At this point we would like to issue some points that measurement of factor I or C-reactive protein biomarkers of inflammation (used in present studies) must be checked at the comparative level using other emerging biomarkers (i.e. imaging biomarkers) with respect to their practicability, clinical benefit and cost-effectiveness.

In conclusion, after analyzing individual records of 19 cohort studies in the due process of investigation of the value of addition of factor I or C-reactive protein levels to conventional models to predict coronary artery disease risk among people of no background of coronary artery disease. We also estimated that initial screening having conventional risk factors, under current treatment guidelines, has led the additional assessment of factor I or C-reactive protein levels in people at "medium" risk for a coronary artery disease risk to prevent an additional event over a period of decade for every 390- 500 screened participants and hence established the utility of factor I and C-reactive proteins as biomarker of inflammation for coronary artery disease.

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Disclosure of conflict of interest

None.

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