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Estimation of cardiovascular risk based on total cholesterol versus total cholesterol/high-density lipoprotein within different ethnic groups: The HELIUS study

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Abstract

Aims: European guidelines recommend estimating cardiovascular disease risk using the Systematic COronary Risk Evaluation (SCORE) algorithm. Two versions of SCORE are available: one based on the total cholesterol/high-density lipoprotein cholesterol ratio, and one based on total cholesterol alone. Cardiovascular risk classification between the two algorithms may differ, particularly among ethnic minority groups with a lipid profile different from the ethnic majority groups among whom the SCORE algorithms were validated. Thus in this study we determined whether discrepancies in cardiovascular risk classification between the two SCORE algorithms are more common in ethnic minority groups relative to the Dutch.

Methods: Using HELIUS study data (Amsterdam, The Netherlands), we obtained data from 7572 participants without self-reported prior cardiovascular disease of Dutch, South-Asian Surinamese, African Surinamese, Ghanaian, Turkish and Moroccan ethnic origin. For both SCORE algorithms, cardiovascular risk was estimated and used to categorise participants as low (<1%), medium (1–5%), high (5–10%) or very high (≥10%) risk. Odds of differential cardiovascular risk classification were determined by logistic regression analyses.

Results: The percentage of participants classified differently between the algorithms ranged from 8.7% to 12.4% among ethnic minority men versus 11.4% among Dutch men, and from 1.9% to 5.5% among ethnic minority women versus 6.2% among Dutch women. Relative to the Dutch, only Turkish and Moroccan women showed significantly different (lower) odds of differential cardiovascular risk classification.

Conclusion: We found no indication that discrepancies in cardiovascular risk classification between the two SCORE algorithms are consistently more common in ethnic minority groups than among ethnic majority groups.

Keywords

SCORE, cholesterol, primary prevention, cardiovascular risk estimation, ethnic groups, HELIUS study

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Introduction

According to most cardiovascular disease (CVD) prevention guidelines, pharmacological intervention to reduce cardiovascular risk is indicated only among those considered to be at high risk of CVD.[1,2] This cardiovascular risk status is often determined by estimated 10-year cardiovascular risk.[1,2] European guidelines recommend to estimate 10-year cardiovascular risk using the Systematic COronary Risk Evaluation (SCORE) algorithm, which was developed in 2003 by

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Conroy et al.[3] using prospective data from several European cohorts.[2] This algorithm estimates the 10-year risk of fatal CVD based on age, sex, systolic blood pressure (SBP), smoking status and the total cholesterol (TC)/high-density lipoprotein (HDL) cholesterol ratio.[2,3]

European guidelines recommend the use of this TC-based SCORE algorithm over another version of the SCORE algorithm that was developed based on TC/HDL-cholesterol.[2] This recommendation is based on the finding that estimated cardiovascular risk did not differ strongly between the two algorithms.[2–4] Only 6.5% of the participants showed a discrepancy of 1% or more, and although the predictive accuracy of the TC/HDL-cholesterol algorithm was slightly higher than that of the TC algorithm, the simplicity of the TC algorithm was preferred.[3,4] Although a recent analysis from the Copenhagen study confirms that the TC/HDL-cholesterol algorithm may not be superior to the TC algorithm, HDL-cholesterol may be important among those with a risk level just below the threshold for treatment, as a low HDL-cholesterol level may qualify these individuals for such treatment.[2,5]

The comparison of the TC/HDL-cholesterol and the TC algorithm may have important implications especially in populations that may show a high discrepancy between the two algorithms, as the choice for one or the other algorithm may directly influence preventive treatment decisions. Discrepancies may occur among certain ethnic minority groups because of a different association between TC-cholesterol and TC/HDL-cholesterol compared with the populations included in validation studies.[6–10] For example, in The Netherlands, certain ethnic minority groups (e.g. Turkish, Moroccan) show lower TC levels, but higher TC/HDL-cholesterol levels relative to the Dutch, resulting in a potentially different pattern of discrepancies between the SCORE algorithms.[6,7,10] Thus, in our study, we aim to assess the occurrence of differential cardiovascular risk classification between the two SCORE algorithms, and determine whether this differs between ethnic groups.

Methods

The HELIUS (Healthy Life in an Urban Setting) study is a large-scale cohort study on health and healthcare utilisation among different ethnic groups living in Amsterdam, The Netherlands. The aims and design of the HELIUS study have been published previously.[11,12] In brief, data collection took place from 2011 to 2015. Participants between 18 and 70 years of age living in Amsterdam were randomly sampled by way of the municipality register, after stratification by ethnicity. A total of 90,019 subjects received a written invitation. Approximately 55% were contacted, either by regular mail or after an additional home visit by an ethnically matched interviewer. Of those, 24,789 agreed to participate.[12] Baseline data were obtained among participants of Dutch, South-Asian Surinamese, African Surinamese, Ghanaian, Turkish and Moroccan ethnic origin. The study protocols were approved by the AMC Ethical Review Board, and all participants provided written informed consent.

Ethnicity

Participants' ethnicity was defined according to the country of birth of the participant as well as that of his/her parents.[13] A participant was considered as of non-Dutch ethnic origin if he/she was born abroad and has at least one parent born abroad (first generation), or he/she was born in The Netherlands but both his/her parents were born abroad (second generation). Of the Surinamese immigrants in The Netherlands, approximately 80% are either of African or South-Asian origin. After data collection, Surinamese subgroups were classified according to self-reported ethnic origin. For the Dutch sample, we invited people who were born in The Netherlands and whose parents were born in The Netherlands.

Cardiovascular risk

All participants were asked to bring their prescribed medications to the research location, which were categorised using the anatomical therapeutic chemical (ATC) classification system.[14] Blood pressure (BP) lowering medication included diuretics, beta-blockers, calcium antagonists and agents acting on the renin–angiotensin–aldosterone system and centrally acting antihypertensive drugs (ATC codes C02, C03, C07, C08, C09). Lipid-lowering medication was classified as ATC code C10. Glucose-lowering medication was classified as ATC code A10.

The 10-year risk of cardiovascular mortality was estimated using the TC-based SCORE algorithm and by using the TC/HDL-cholesterol-based SCORE algorithm for low-risk countries, based on age, sex, SBP, smoking status and either TC or TC/HDL-cholesterol ratio (for the TC algorithm and HDL-cholesterol algorithm, respectively).[2] The SCORE algorithms were kindly provided to HELIUS by van Dis et al.[15] Smoking status and the occurrence of prior CVD were assessed by questionnaire. SBP was measured using a validated automated digital BP device (WatchBP Home; Microlife AG) on the left arm in a seated position after the person had been seated for at least 5 minutes. BP measurements were performed in duplicate and the mean of the two measurements was used in the analyses. Participants were considered to have diabetes if they reported a diabetes diagnosis in

the questionnaire, were using glucose-lowering medication and/or when they had an increased (≥7.0 mmol/l) fasting glucose. Fasting blood samples were drawn after an overnight fast. Glucose (mmol/l) was determined using an enzymatic spectrophotometric (UV) method, using hexokinase as the primary enzyme (Roche Diagnostics, Japan). TC and HDL-cholesterol and triglycerides were determined by enzymatic colorimetric spectrophotometry (Roche Diagnostics, Japan).

Study population

Of the 24,789 HELIUS participants, baseline data from both physical examination and questionnaire were available for 22,165 participants. Of these participants, 9594 were potentially eligible for cardiovascular risk estimation according to current European CVD prevention guidelines (i.e. participants without prior CVD or diabetes and between 40 and 65 years of age).[2] Of the potentially eligible participants, we excluded those with a Javanese Surinamese ($n = 131$), 'other/unknown Surinamese' ($n = 134$) or 'unknown/other' ethnicity ($n = 22$) due to low statistical power. Next we excluded those receiving BP-lowering or lipid-lowering medication ($n = 1683$). We then excluded participants based on missing data regarding prior CVD, diabetes, or components of SCORE risk factors ($n = 51$). Finally, we excluded one participant with an unlikely low cholesterol value (i.e. a TC of 1.960 and HDL-cholesterol of 0.080 mmol/l). This resulted in a study population of 7572 participants.

Statistical analyses

Descriptive variables were reported as means with standard deviations, except for smoking status, which was reported as the percentage of current smokers. Because the relation between TC and TC/HDL-cholesterol can differ by sex, men and women were analysed separately.[6] We first determined the agreement between the two SCORE algorithms by developing Bland–Altman plots.[16] To that end, we calculated the difference between the SCORE algorithms as TC/HDL-cholesterol-based estimated cardiovascular risk minus TC-based estimated cardiovascular risk for each participant, and plotted this difference against the average estimated cardiovascular risk between the two SCORE algorithms.[16] For comparability between ethnic groups, sex-specific cut-off values for x-axes and y-axes of Bland–Altman plots were used.

Next, we determined the extent of differential cardiovascular risk classification between the two SCORE algorithms. For each algorithm separately, we first determined the estimated cardiovascular risk and categorised participants into cardiovascular risk categories recommended by European guidelines, i.e. low risk (estimated cardiovascular risk <1%), moderate risk (1–5%) high risk (5–10%) or very high risk (≥10%).[2] For each category of TC/HDL-cholesterol-based cardiovascular risk, we determined the proportion of participants in each TC-based risk category. Furthermore, using binary logistic regression analysis, we determined whether the crude (model 1) or age-adjusted (model 2) odds of being classified differently by the two algorithms differed between ethnic groups. Furthermore, to determine whether ethnic differences in differential cardiovascular risk classification are due to ethnic differences in absolute estimated cardiovascular risk (i.e. because the estimated cardiovascular risk is closer to cardiovascular risk category thresholds), we additionally adjusted for TC/HDL-cholesterol-based estimated cardiovascular risk in a final model (model 3).

Finally, in previous longitudinal studies regarding the predictive value of the SCORE algorithms, authors reported either the Spearman's rank correlation coefficient between the two algorithms or the percentage of participants showing discrepancy in estimated cardiovascular risk of 1% or more.[3–5] To compare our study population with the populations used in these previous studies, we also determined these measures. Moreover, after determining among which participants the difference between estimated cardiovascular risk was equal to or higher than 1%, we explored whether this prevalence differed between ethnic groups by binary logistic regression analyses, adjusted for age and cardiovascular risk as estimated by the TC/HDL-cholesterol SCORE algorithm.

Results

Age did not differ strongly between ethnic groups (Table 1). Compared with the Dutch, we observed similar or higher rates of smoking among all ethnic minority groups except for Ghanaians and Moroccan women, higher SBP among South-Asian Surinamese, African Surinamese and Ghanaian participants, and similar or lower TC and HDL-cholesterol, except among Ghanaians who had higher HDL-cholesterol. TC/HDL-cholesterol relative to the Dutch showed more ethnic heterogeneity, with higher TC/HDL-cholesterol among South-Asian Surinamese, Turkish and Moroccan participants, but similar or lower TC/HDL-cholesterol among African Surinamese and Ghanaian participants.

Bland–Altman plots showed a high agreement at low average estimated cardiovascular risk (Figures 1 and 2). At higher estimated cardiovascular risk, the agreement between the SCORE algorithms was lower, especially among Dutch men, Dutch women and African Surinamese women. Furthermore, we observed a

Table 1. General characteristics by ethnicity.

	Dutch	South-Asian Surinamese	African Surinamese	Ghanaian	Turkish	Moroccan
Men						
N	893	362	672	357	518	497
Age (years)	52.03 (7.3)	49.10 (6.13)	51.90 (6.50)	50.05 (5.96)	48.37 (5.43)	49.38 (6.37)
Smoking (%)	21.6	38.7	47.8	8.4	39.8	25.2
SBP (mmHg)	129.6 (15.9)	130.1 (15.8)	133.6 (17.4)	138.7 (18.0)	127.0 (14.0)	127.6 (14.0)
Total cholesterol (mmol/l)	5.46 (0.96)	5.45 (0.92)	5.05 (0.98)	5.18 (1.01)	5.27 (0.89)	4.98 (0.83)
HDL-cholesterol (mmol/l)	1.42 (0.39)	1.18 (0.29)	1.40 (0.39)	1.54 (0.42)	1.14 (0.29)	1.16 (0.28)
TC/HDL-cholesterol	4.12 (1.30)	4.85 (1.32)	3.85 (1.22)	3.56 (1.09)	4.88 (1.44)	4.49 (1.21)
Women						
N	1110	542	904	485	547	685
Age (years)	52.04 (7.13)	49.97 (6.23)	50.67 (6.35)	47.67 (5.37)	47.40 (5.34)	48.73 (6.31)
Smoking (%)	21.1	18.3	23.7	2.3	26.0	2.5
SBP[a] (mmHg)	120.8 (15.7)	125.8 (18.0)	129.6 (17.8)	135.8 (18.8)	122.2 (15.2)	121.8 (15.1)
Total cholesterol (mmol/l)	5.50 (1.04)	5.38 (0.94)	5.14 (0.95)	5.12 (0.96)	5.22 (0.93)	4.99 (0.86)
HDL-cholesterol (mmol/l)	1.81 (0.44)	1.51 (0.39)	1.66 (0.44)	1.74 (4.23)	1.45 (0.36)	1.45 (0.34)
TC/HDL-cholesterol	3.22 (1.07)	3.77 (1.10)	3.29 (1.01)	3.07 (0.80)	3.81 (1.12)	3.59 (0.95)

N is presented as absolute number, smoking is presented as percentage, and other data are presented as mean (standard deviation) or percentages. SBP: systolic blood pressure; HDL: high-density lipoprotein; TC/HDL: total cholesterol/high-density lipoprotein cholesterol ratio.

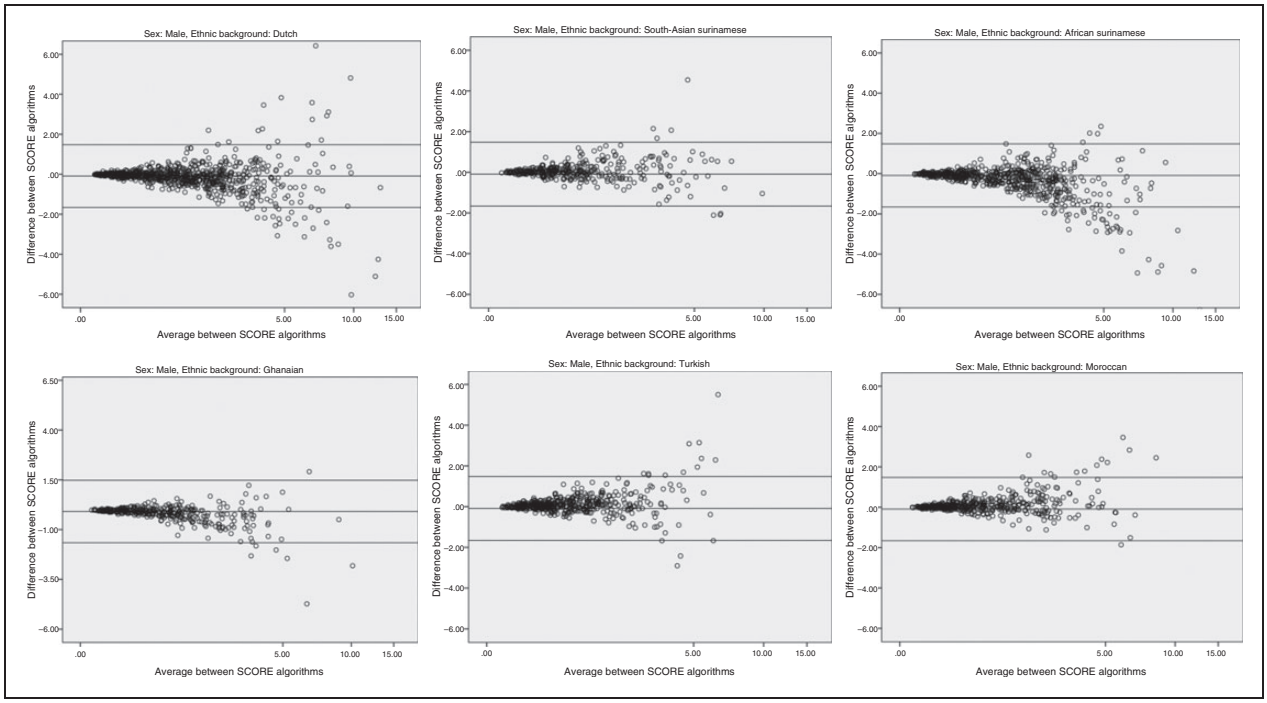

Figure 1. The difference (a) between Systematic COronary Risk Evaluation (SCORE) algorithms plotted against the average (b) between the SCORE algorithms among men, by ethnicity. (a) Calculated as total cholesterol (TC)/high-density lipoprotein (HDL)-cholesterol-based estimated cardiovascular risk minus TC-based estimated cardiovascular risk; (b) calculated as the average between the TC/HDL-cholesterol based estimated cardiovascular risk and the TC-based estimated cardiovascular risk.

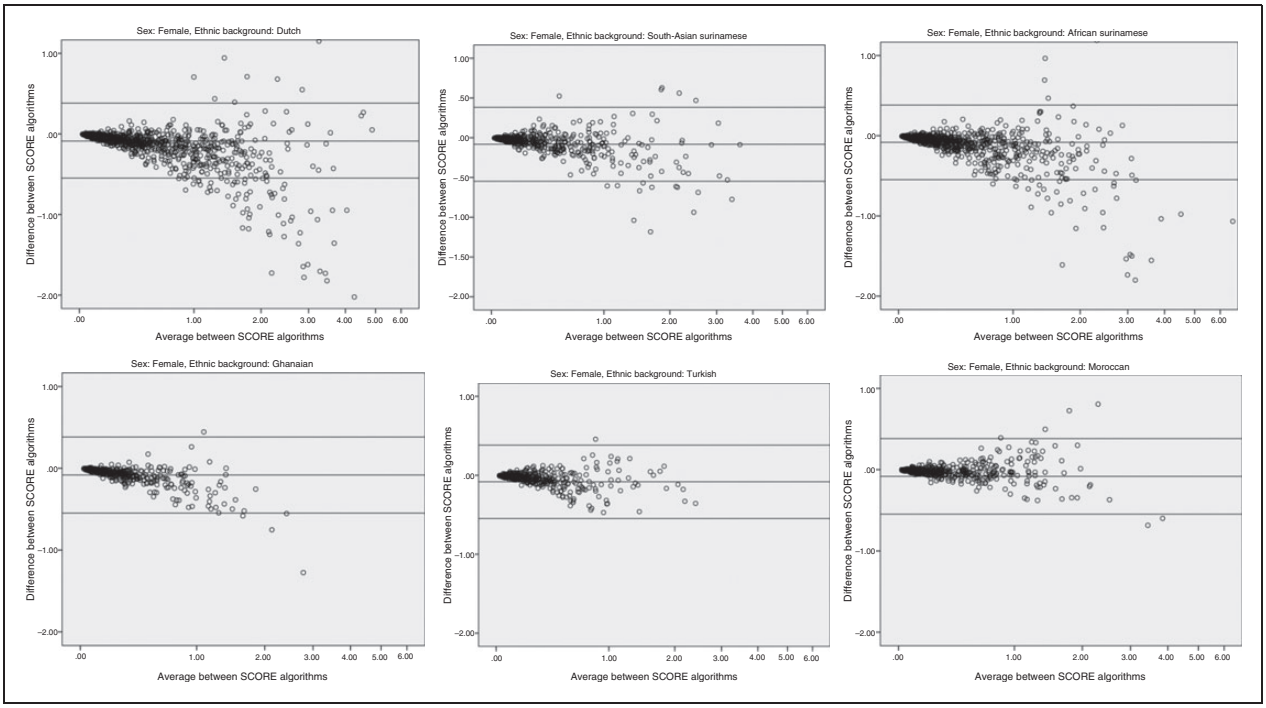

Figure 2. The difference (a) between Systematic COronary Risk Evaluation (SCORE) algorithms plotted against the average (b) between the SCORE algorithms among women, by ethnicity. (a) Calculated as total cholesterol (TC)/high-density lipoprotein (HDL)-cholesterol-based estimated cardiovascular risk minus TC-based estimated cardiovascular risk; (b) calculated as the average between the TC/HDL-cholesterol based estimated cardiovascular risk and the TC-based estimated cardiovascular risk.

slightly overall higher agreement between the SCORE algorithms among South-Asian Surinamese women and among Ghanaian, Turkish and Moroccan participants than the Dutch.

After applying classifications as recommended by European CVD prevention guidelines (i.e. low, medium, high, very high cardiovascular risk), we found that some discrepancies in CVD risk classification between the two algorithms did occur, in both directions (i.e. some participants were classified as higher risk, but other participants were classified as lower risk in one algorithm relative to the other, Table 2). For low and moderate cardiovascular risk, a high proportion of participants was classified similarly between the SCORE algorithms. In contrast, the proportion classified similarly between the algorithms was lower at high and very high risk (e.g. 61.9% of men that were classified as high risk by the TC/HDL-cholesterol SCORE algorithm were also classified as high risk by the TC SCORE algorithm). Moreover, in general, cardiovascular risk status in women was classified slightly lower by the TC/HDL-cholesterol algorithm than the TC algorithm. This pattern was not observed among men.

The occurrence of differential cardiovascular risk classification between the two algorithms ranged from 8.7% among Ghanaian men to 12.4% among South-Asian Surinamese men versus 11.4% among Dutch men, and did not differ significantly between ethnic minority groups and the Dutch (Table 3). Among women, the occurrence of differential cardiovascular risk classification ranged from 1.9% among Moroccans to 5.5% among African Surinamese versus 6.2% among the Dutch. Crude odds of differential cardiovascular risk classification were lower among Ghanaian, Turkish and Moroccan women relative to the Dutch. After adjustment for age, only Moroccan women showed significantly lower odds relative to the Dutch. In contrast, after adjustment for underlying estimated cardiovascular risk, both Turkish and Moroccan women showed significantly lower odds of differential cardiovascular risk classification relative to the Dutch.

Finally, for comparison with previous studies, we determined the Spearman correlation coefficient between the two algorithms, as well as the proportion of participants with a discrepancy between the SCORE algorithms of 1% or more (see Supplementary Tables 1 and 2). A high correlation between the algorithms was found in all subgroups, ranging from 0.961 to 0.990. Among men, the proportion of participants with a discrepancy of at least 1% ranged from 4.8% to 14.4% and was particularly high among Dutch and African Surinamese men. Among women, the proportion ranged from 0.00% to 2.34% and was particularly high among the Dutch.

Table 2. Distribution of TC based SCORE categories and TC/HDL-cholesterol-based SCORE categories, by sex.

	TC-SCORE			
	Low	Moderate	High	Very high
HDL-SCORE (men)				
Low	1450 (91.8)	130 (8.2)	0 (0.0)	0 (0.0)
Moderate	136 (8.5)	1412 (88.0)	57 (3.6)	0 (0.0)
High	0 (0.0)	28 (26.7)	65 (61.9)	12 (11.4)
Very high	1 (11.1)	1 (11.1)	2 (22.2)	5 (55.6)
HDL-SCORE (women)				
Low	3639 (95.8)	160 (4.2)	0 (0.0)	0 (0.0)
Moderate	19 (4.1)	447 (95.5)	2 (0.4)	0 (0.0)
High	0 (0.0)	1 (16.7)	4 (66.7)	1 (16.7)
Very high	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

Data are presented as total number of participants (total percentage per row).
TC: total cholesterol; SCORE: Systematic COronary Risk Evaluation which estimates the 10-year risk of cardiovascular disease (%) based on the occurrence of cardiovascular disease risk factors, including either TC or TC/HDL-cholesterol; TC/HDL: the ratio between TC and HDL-cholesterol ratio.
[l]ow: <1%; moderate: 1–5%; high: 5–10%; very high: >10%.

Table 3. Prevalence and odds ratio (95% confidence interval, ref Dutch) for differential cardiovascular risk classification[a] per ethnic group, by sex.

	Prevalence	Model 1 (crude)	Model 2 (age)	Model 3 (HDL-cholesterol-SCORE)
Men				
Dutch	11.4	Ref	Ref	Ref
SA Surinamese	12.4	1.10 (0.76; 1.60)	1.28 (0.88; 1.89)	1.18 (0.80; 1.72)
African Surinamese	10.6	0.92 (0.67; 1.26)	0.93 (0.67; 1.29)	0.92 (0.66; 1.28)
Ghanaian	8.7	0.74 (0.48; 1.13)	0.82 (0.54; 1.26)	0.86 (0.57; 1.32)
Turkish	12.4	1.09 (0.78; 1.53)	1.33 (0.94; 1.87)	1.26 (0.90; 1.77)
Moroccan	10.9	0.95 (0.67; 1.34)	1.08 (0.76; 1.54)	1.08 (0.76; 1.55)
Women				
Dutch	6.2	Ref	Ref	Ref
SA Surinamese	4.4	0.70 (0.43; 1.13)	1.11 (0.67; 1.82)	0.79 (0.49; 1.29)
African Surinamese	5.5	0.88 (0.61; 1.29)	1.25 (0.84; 1.86)	0.98 (0.66; 1.44)
Ghanaian	2.9	**0.45 (0.25; 0.81)**	1.24 (0.67; 2.30)	0.64 (0.35; 1.16)
Turkish	2.4	**0.37 (0.20; 0.67)**	1.03 (0.54; 1.95)	**0.52 (0.28; 0.97)**
Moroccan	1.9	**0.29 (0.16; 0.53)**	**0.53 (0.29; 0.98)**	**0.37 (0.20; 0.69)**

[a]Differential classification is defined as the total cholesterol (TC)-Systematic COronary Risk Evaluation (SCORE) category not corresponding to the TC/high-density lipoprotein (HDL)-cholesterol-SCORE category in either direction.
Bold type signifies statistically significant odds ratio.
SA: South-Asian.

Discussion

Key findings

There is a high overall agreement between the SCORE algorithms among all ethnic groups. This agreement tends to be lower at higher estimated cardiovascular risk, with a substantial occurrence of differential cardiovascular risk classification among participants classified as high or very high risk We found no indication that such differential classification of cardiovascular risk occurs more often among ethnic minority groups than in the Dutch. Among women, differential

cardiovascular risk classification may, in fact, occur less frequently in some ethnic minority groups than in the Dutch.

Evaluation of potential limitations

Some selection bias may have occurred. Non-response analyses showed no differences in socioeconomic status between responders and non-responders, suggesting that non-responders may not differ strongly from responders.[12] However, because data regarding cardiovascular risk factors among non-responders were not available, it was impossible to check whether the association between the two SCORE algorithms differed between responders and non-responders, for instance due to lower participation among those with health problems.[17,18]

In our study, lipid levels, fasting glucose and BP were measured on a single occasion, whereas clinical guidelines recommend a second measurement, especially when participants are close to or above a treatment threshold.[2,19] A repeated measurement might have influenced discrepancies between the two SCORE algorithms, by providing more accurate measurements of TC and TC/HDL-cholesterol. Consequently, we may have overestimated the occurrence of differential cardiovascular risk classification between the two SCORE algorithms as compared with what is common in daily clinical practice.

Interpretation of key findings

Earlier studies found ethnic differences in TC, HDL-cholesterol and TC/HDL-cholesterol.[7–10] We also found ethnic differences in these measures but, in contrast to our initial hypothesis, these differences were too small to result in a higher occurrence of differential cardiovascular risk classification. Replication of our findings in other countries is necessary, in part because ethnic-specific lipid profiles among migrants may differ from those who remained in the country of origin.[20]

Studies comparing the SCORE algorithms have reported strong associations between the two algorithms, which were generally similar to the associations found in our study.[3–5] For example, Mortensen et al.[5] reported that, in the Copenhagen study, the Spearman's correlation coefficient for the correlation between the SCORE algorithms was 0.93, which was comparable with the Spearman's correlation coefficients found among all our ethnic groups (i.e. ranging from 0.93 to 0.99). In addition, the percentage of participants showing a discrepancy of 1% or more in our study (4.2%) was comparable with that in the original validation studies. For example, Coomey et al.[4] reported that this was 6.5%.

In accordance with European CVD prevention guidelines, we estimated fatal CVD risk rather than total (fatal plus non-fatal CVD) risk.[2] However, some guidelines recommend to estimate total CVD risk rather than fatal CVD risk as primary prevention should also aim to prevent all burden of mortality and morbidity due to atherosclerotic disease and non-fatal CVD as the case fatality rate of CVD is declining.[21–23] We do not expect that ethnic differences in differential CVD risk classification based on estimated total CVD risk will differ from estimated fatal CVD risk, as both algorithms incorporate lipid profile in the same manner.

For our study, we used the Dutch version of the SCORE algorithm, which is based on the SCORE algorithms that were originally developed in 2003.[3] An update of this algorithm may be necessary.[24] Although an updated SCORE for the Dutch population (published in 2010) did not outperform the original SCORE algorithms, more recent data suggest that the current SCORE risk chart seriously underestimates cardiovascular risk in The Netherlands.[15,25] It would be of interest again to compare cardiovascular risk classification between the two algorithms among multiple ethnic groups, once these updated SCORE risk charts become available.

The necessity of validating SCORE risk estimation among ethnic minority groups has been stressed by CVD prevention guidelines and individual academics.[2,4,6,26,27] We found that the estimated CVD risk may differ between the two algorithms regardless of ethnic background. Considering that it is unclear which SCORE algorithm may provide more accurate CVD risk estimations, we recommend to validate and compare both versions of the SCORE algorithm in multi-ethnic settings, once the required longitudinal data to do so become available.[5,28] Alternatively, these studies may also compare the SCORE algorithms with more recent and more complex cardiovascular risk algorithms (e.g. HeartScore, which incorporates HDL-cholesterol on a continuous basis rather than as a ratio to TC, or algorithms incorporating genetic risk factors) to enable accurate evaluation of the necessity of complex cardiovascular risk algorithms among ethnic minority groups.[2,29,30]

Conclusion

In general, differential cardiovascular risk classification may occur among approximately 10% of men and 5% of women, but substantially higher proportions are found at the higher end of the cardiovascular risk spectrum. Differential risk classification does not occur more frequently among ethnic minority groups compared with the Dutch. Thus although the decision regarding which algorithm should be used in daily clinical practice may affect which individuals are eligible

for treatment, there is no indication that this generally affects ethnic minority groups more than ethnic majority groups in Europe under the presumption of equal predictive value between the two algorithms.

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Author contribution

WP and IGvV contributed to the conception of the work; all authors contributed to the design of the work. MBS and RJP contributed to the acquisition of the work; all authors contributed to the analysis and interpretation of the work. WP drafted the manuscript, MBS, RJP, AEK and IGvV critically revised the manuscript. All authors gave final approval and agree to be accountable for all aspects of the work ensuring integrity and accuracy.

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