Smoking and apolipoprotein levels: A meta-analysis of published data

Alba Romero Kauss, Meagan Antunes, Guillaume de La Bourdonnaye, Sandrine Pouly, Matthew Hankins, Annie Heremans, Angela van der Plas *

PMI R&D, Philip Morris Products S.A., Quai Jeanrenaud 5, CH-2000 Neuchâtel, Switzerland

Abstract

Background: Apolipoproteins are major components of lipoproteins such as high-density lipoprotein (HDL) and very-low-density lipoprotein and are considered nontraditional markers in the risk assessment for cardiovascular disease. The goal of this review was to quantify the effects of smoking and smoking cessation on serum levels of apolipoproteins AI, AII, and B and the ratio of apolipoproteins B and AI.

Methods: PubMed and Scopus were searched up to June 2021 to identify publications that reported the levels of apolipoproteins AI, AII, and B and the apolipoprotein B/AI ratio in smokers and nonsmokers as well as articles reporting the effect of smoking cessation on the same endpoints. Meta-analyses were performed when a sufficient number (n ≥ 3) of articles evaluating the same outcome were available.

Results: Forty-nine studies had assessed apolipoprotein levels in smokers and nonsmokers. The meta-analyses comparing the levels of apolipoproteins AI and AII showed decreased levels in smokers relative to nonsmokers. On the other hand, the apolipoprotein B levels and apolipoprotein B/AI ratio were increased in smokers relative to nonsmokers. Insufficient publications were available on which to perform meta-analyses on the effects of smoking cessation on apolipoprotein levels.

Conclusions: Smoking is associated with reduced levels of apolipoproteins AI and AII (in line with reduced levels of HLD-cholesterol) and increased apolipoprotein B levels and apolipoprotein B/AI ratio, thereby confirming the negative impact of smoking on lipid metabolism, which contributes to increased cardiovascular risk. More data are needed to elucidate the effects of smoking cessation on these cardiovascular risk endpoints.

1. Introduction

Cigarette smoking has been implicated as a major risk factor in chronic pulmonary disease, carcinogenesis and atherosclerotic arterial disease leading to cardiovascular disease (CVD) [1–3]. The mechanisms through which smoking increases the risk of atherosclerosis and CVD include oxidative damage and the alteration of lipid levels [4,5]. For instance, it has been shown that cigarette smoking decreases high-density lipoprotein-cholesterol (HDL-C) [6].

Even though low-density lipoprotein-cholesterol (LDL-C) is recognized as the primary lipid risk factor for CVD [7], other lipid measurements have been associated with an increased or decreased risk of CVD, such as apolipoproteins, which are the protein components of lipoproteins [8]. Two major types of apolipoproteins have been described: apolipoprotein A (I and II) and apolipoprotein B. Apolipoprotein AI, the major constituent of HDL, comprises roughly 70% of the HDL protein mass [9]. It mediates the efflux of cholesterol from the peripheral cell membranes and activates lecithin cholesterol acetyltransferase, which is an important enzyme in the reverse transport of cholesterol from peripheral tissues to the liver [10]. Apolipoprotein AII is the second most abundant protein in HDL, accounting for about 20% of total HDL [11]. Apolipoprotein B is an essential component of LDL and very-low-density lipoprotein (VLDL-C). It facilitates cholesterol delivery to peripheral tissues and promotes LDL-C accumulation by acting as a ligand for LDL-C receptors [1]. Apolipoprotein B is the major protein not only of LDL but also of all atherogenic particles, e.g. VLDL and intermediate-density lipoprotein (IDL). Because only one apolipoprotein B protein is present per particle [12], the total apolipoprotein B value reflects the total number of potentially atherogenic particles [13]. Due to the close association between apolipoproteins and serum lipids, their measurement has been proposed as an important factor in predicting the risk of cardiovascular diseases [8,10]. Additionally, population-based studies have suggested that the apolipoprotein B/AI ratio could be used as an indicator of the balance between atherogenic and atheroprotective...
cholesterol transport, as an increase in this ratio predicts cardiovascular risk more accurately and strongly than the use of either apolipoprotein B or apolipoprotein AI individually, or any of the other cholesterol indexes [14–16].

Studies assessing the association of smoking status and smoking cessation with lipid parameters are abundant in the literature. A review of the association of HDL-C levels and smoking status was performed by Maeda et al. [17], demonstrating increased levels of HDL-C after smoking cessation. To date, no meta-analyses have been published comparing the effects of smoking status or smoking cessation on the levels of apolipoproteins, which is the objective of the present review.

2. Methods

Searches were performed through PubMed and Scopus databases to identify publications that evaluated the relationship between smoking or smoking cessation and plasma levels of apolipoproteins, including AI, AII, and B, and the ratio of apolipoprotein B to apolipoprotein AI in humans. The last search was performed on June 25, 2021. The following query was used in PubMed: ('apolipoproteins'[MeSH Terms] OR 'apo-lipoproteins'[All Fields] OR 'apolipoprotein'[All Fields]) AND ('smoking'[MeSH Terms] OR 'smoking'[All Fields] OR 'tobacco'[MeSH Terms] OR 'tobacco'[All Fields] OR 'tobacco products'[MeSH Terms] OR 'tobacco'[All Fields] OR 'tobacco products'[All Fields]) OR cessation [All Fields] OR quitting [All Fields]). In Scopus, the following query was used: apolipoprotein AND (smoking OR tobacco OR cessation OR quitting). The reference lists of the publications obtained through the original search were checked for additional articles.

2.1. Study selection

Case-control, cohort, or interventional studies in healthy adult subjects reporting any measurements of apolipoproteins AI, AII, and B levels and/or apolipoprotein B/AI ratio by smoking exposure (smokers vs. nonsmokers and baseline vs. time after smoking cessation) were included if they provided the following: mean or median values by group, standard deviation (SD) or standard error (SE), sample size per group, or had enough information to allow for the calculation of mean and SD. Review articles, case reports, editorials, and duplicates of publications of data that were part of a more recent publication were not included.

2.2. Data extraction

The following information was extracted from each study independently by three researchers: first author’s name, year of publication, country where study was performed, study design and population characteristics, number of participants per exposure group, mean, SD or SE, and smoking status definition. Apolipoprotein levels were converted from mg/dL to g/L as needed.

2.3. Statistical analysis

All analyses were implemented using the ‘meta’ [18] and ‘dmetar’ [19] packages in R 4.0.5 [20].

To quantify the effects of smoking and smoking cessation on apolipoprotein levels, pooled mean differences between smokers and nonsmokers (when assessing effects of smoking on apolipoproteins) or differences between baseline and follow-up measures after smoking cessation (when assessing the effects of quitting smoking on apolipoprotein levels) and 95% confidence intervals (95% CI) were calculated using the fixed-effects model in the ‘metamean’ function [18]. The ‘metamean’ function uses the inverse variance method for pooling, giving studies with small variance relatively higher weight and studies with larger variance relatively smaller weight [18]. Meta-analyses were carried out using the ‘metacont’ function when at least three measures of a single estimate were available. The degree of heterogeneity between the study results was assessed by the I² statistic. Funnel plot symmetry and Egger’s regression test were used to evaluate publication bias [21]. Statistical significance was assessed at α = 0.05.

To explore possible sources of heterogeneity, the meta-analyses were performed using the random-effects model and sensitivity analyses were conducted using the ‘Influence/Analysis’ function, to eliminate studies that contributed the most to heterogeneity. This function conducts an influence analysis using the ‘Leave-One-Out’ paradigm and produces data for four influence diagnostics (Baujat plot, Viechtbauer-Cheung influence characteristics plot, and forest plots for the leave-one-out analysis, sorted by effect size and I²). The Baujat plot and forest plot sorted by I² were used to identify the studies with high heterogeneity contribution and low influence on the overall results. These studies represent outliers and were removed to reduce the amount of between-study heterogeneity. Additionally, subgroup analyses were performed based on smoking status, geographical region, study design, period of publication, and sex.

The reason for using different models in the meta-analyses is that, while the fixed-effects model assumes that the estimated effects from the component studies in a meta-analysis come from a single homogeneous population, the random-effects model seeks to account for the fact that the study effect estimates are often more variable than assumed in the fixed-effects model [22].

3. Results

3.1. Identified studies

A Prisma flow diagram detailing the retrieval process of articles from the different sources used can be found in Fig. 1. For the analyses of smoking status and its association to apolipoprotein levels, a total of 66 publications that assessed the effect of smoking status on any of the following parameters were identified: 39 studies on apolipoprotein AI [23–61], ten studies on apolipoprotein AII [23,24,26,32,34,43,47,53,55], 40 studies on apolipoprotein B [23,24,27–30,32,34,35,37–42,44–46,48–69], and 31 studies on the ratio of apolipoprotein B to AI [23,24,27–30,32,34,35,37–42,44–46,48–58,62,70]. These publications compared the levels of the aforementioned lipid parameters in smokers vs. nonsmokers. Out of the 66 publications, 49 studies were included in the analyses and their characteristics can be found in Table 1. Additionally, the forest plots can be found in Appendix 1. The other 17 studies were not included because they involved diseased populations [71,72], had incomplete information [73–81], reported inaccurate or inconsistent measurement units [82,83], the populations studied included children [84], the publication was a meta-analysis [85], or because the nonsmoking group included smokers [86,87]. Table 2.

Using influence diagnostics, four studies (Casasnovas et al. [27], Pasupathi et al. [60], Sirisali et al. [59], and Sharma et al. [67]) were identified as having a large impact on between-study heterogeneity. Thus, sensitivity analyses were conducted excluding these studies.

For the analysis of smoking cessation impact on apolipoprotein levels, eight studies were retrieved. In total, five studies had complete information, with follow-up times of six weeks [88], eight weeks [89], 12 weeks [90,91], and 90 days [92]. Of the other studies, one included patients that had followed up from two to six weeks after smoking cessation [93], one reported results from smoking reduction [94] and one was a duplicate of another publication [95]. Due to the lack of a sufficient number of studies, no meta-analysis was performed to evaluate the effects of smoking cessation on apolipoprotein levels. The characteristics of the retrieved studies can be found in Tables 3 and 4.

3.2. Effects of smoking status on apolipoprotein AI levels

The pooled mean levels of apolipoprotein AI were 1.48 ± 0.009 g/L.
in smokers and 1.54 ± 0.007 g/L in nonsmokers. The meta-analysis for apolipoprotein AI levels between smokers and nonsmokers was performed using data from 39 studies including 59 estimates [23–61]. The fixed-effects model mean difference was −0.038 (95% CI: −0.041, −0.035 g/L, I² = 90%). Because of the large heterogeneity, sensitivity analyses were conducted, removing the studies that were driving the most heterogeneity. Data from 36 studies, including 55 estimates, were included in the sensitivity analysis. The fixed-effects model mean difference was −0.036 (95% CI: −0.039, −0.033 g/L, I² = 71%). To address the large heterogeneity further, exploratory subgroup analyses were performed. The results of these analyses can be found in Table 5. Stratification by region explained up to 28% points of the I² value when considering only studies run in the Americas (mean difference = −0.062, 95% CI: −0.073, −0.051 g/L, I² = 43%), while most of the heterogeneity originated from Asian and African studies (I² = 73%). Stratification by study design explained up to one percentage point (mean difference = −0.034, 95% CI: −0.039, −0.029 g/L, I² = 70%) in the cohort subgroup. The stratification by period of publication explained up to 59% points (mean difference = −0.010, 95% CI: −0.013, 0.003 g/L, I² = 12%) in the 2000s subgroup and the stratification by smoking definition explained up to 20% points (mean difference = −0.074, 95% CI: −0.134, −0.014 g/L, I² = 51%) in the CPD subgroup. The stratification by sex explained up to 30% points (mean difference = −0.040, 95% CI: −0.044, −0.036 g/L, I² = 41%) in the women-only subgroup.

Applying the random-effects model to acknowledge the high inter-study heterogeneity resulted in a mean difference of −0.073 (95% CI: −0.088, −0.058 g/L) for all studies and −0.042 (95% CI: −0.053, −0.032 g/L) for studies included in the sensitivity analysis. Visual inspection of the funnel plots (Fig. 2) revealed some asymmetries, with several large studies showing a significant negative effect. Funnel plot asymmetry was confirmed using Egger’s regression test (intercept = −0.62, 95% CI: −1.17, −0.07, t = −2.21, p = 0.03) and may be an indication of publication bias.

3.3. Effects of smoking status on apolipoprotein AI levels

The pooled mean levels of apolipoprotein AI were 0.32 ± 0.007 g/L in smokers and 0.30 ± 0.004 g/L in nonsmokers. The meta-analysis of the effects of smoking on apolipoprotein AI levels included ten studies and 16 estimates [23,24,26,32,34,36,43,47,53,55] and found no differences in levels of apolipoprotein AI between smokers and nonsmokers (mean difference = −0.003; 95% CI: −0.007, 0.002 g/L, I² = 66%). Considerable heterogeneity was found among studies; however, the studies identified through the sensitivity analysis did not report estimates for apolipoprotein AI levels. Therefore, subgroup analyses were performed using data from all studies. The detailed results of these analyses can be found in Table 6. The stratification by study design explained up to 25% points of the I² value (mean difference = 0.002, 95% CI: −0.003, 0.007 g/L, I² = 41% in the cross-sectional studies subgroup). The stratified analysis by period of publication explained 14% points (mean difference = −0.014, 95% CI: −0.023, −0.005 g/L, I² = 52%) in the 1990s subgroup. The major source of heterogeneity seemed to come from studies in the US (mean difference = −0.038, 95% CI: −0.027, 0.011 g/L, I² = 73%) and studies involving only women (mean difference = 0.014, 95% CI: −0.007, 0.008 g/L, I² = 74%). Applying the random-effects model estimated a mean difference of −0.004 (95% CI: −0.014, 0.004 g/L). The evaluation of the funnel plot (Fig. 3) did not show evidence of publication bias and the results of Egger’s regression test were not significant (intercept = 0.20, 95% CI: −1.32, 1.73, t = 0.26, p = 0.80).

3.4. Effects of smoking status on apolipoprotein B levels

The pooled mean levels of apolipoprotein B were 1.06 ± 0.009 g/L in smokers and 0.98 ± 0.007 g/L in nonsmokers. The meta-analysis of apolipoprotein B levels in smokers versus nonsmokers included 40 studies and 63 estimates [23,24,27–30,32,34,35,37–42,44–46,48–58, 62,70]. Increased levels of apolipoprotein B were found in smokers using the fixed-effects model (mean difference = −0.056, 95% CI: 0.053, 0.059 g/L, I² = 95%). The heterogeneity was high; therefore, sensitivity and subgroup analyses were performed. The sensitivity analysis included data from 36 studies and 59 estimates. The fixed-effects mean difference was 0.053 g/L (95% CI: 0.050, 0.055 g/L, I² = 81%). The results of the subgroup analysis can be found in Table 7. The stratified analyses found that four percentage points of the I² value could be explained by including only cohort studies (mean difference = −0.057, 95% CI: 0.053, 0.061 g/L, I² = 77%), 14% points could be explained by limiting the studies to those performed in Europe (mean difference =
Table 1

Study characteristics of selected studies of smoking status and apolipoprotein levels.

| Study | Reference          | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Smokers Mean ± SD | Nonsmokers Mean ± SD | Mean difference Δ (95% CI) | Adjustment |
|-------|--------------------|---------|--------------|--------------------|--------------------|------------------|-------------|----------|-------------------|-----------------------|--------------------------------|------------|
| 1     | Adachi et al. [23] | Japan   | Cross-Sectional | 169 healthy men with an average age of 54.7 ± 8.5 years | Stratified by CPD | 1.1  | Apolipoprotein AII | Men 20 + CPD | 1.38 ± 0.27 | 0.87 ± 0.19 | 0.41 | -0.01 [-0.11, 0.09] | None |
|       |                    |         |              |                    |                    | 1.2  | Apolipoprotein AI | Men 20 + CPD | 1.38 ± 0.27 | 0.87 ± 0.19 | 0.41 | -0.01 [-0.11, 0.09] | None |
|       |                    |         |              |                    |                    | 1.3  | Apolipoprotein B | Men 20 + CPD | 0.34 ± 0.08 | 0.18 ± 0.04 | 0.30 | -0.01 [-0.08, 0.06] | None |
|       |                    |         |              |                    |                    | 1.4  | Ratio Apo B/Apo AI | Men 20 + CPD | 0.34 ± 0.08 | 0.18 ± 0.04 | 0.30 | -0.01 [-0.08, 0.06] | None |
| 2     | Ashavaid et al. [24] | India | Cross-Sectional | 470 men and 281 women aged 25–65 years | None | 2.1  | Apolipoprotein AII | All Men | 1.22 ± 0.18 | 0.98 ± 0.14 | 0.24 | -0.08 [-0.30, 0.14] | None |
|       |                    |         |              |                    |                    | 2.2  | Apolipoprotein AI | All Men | 1.22 ± 0.18 | 0.98 ± 0.14 | 0.24 | -0.08 [-0.30, 0.14] | None |
|       |                    |         |              |                    |                    | 2.3  | Apolipoprotein B | All Men | 0.28 ± 0.04 | 0.21 ± 0.06 | 0.07 | -0.03 [-0.06, 0.0] | None |
|       |                    |         |              |                    |                    | 2.4  | Ratio Apo B/Apo AI | All Men | 0.28 ± 0.04 | 0.21 ± 0.06 | 0.07 | -0.03 [-0.06, 0.0] | None |
|       |                    |         |              |                    |                    | 2.5  | Apolipoprotein AII | All Women | 1.24 ± 0.22 | 0.98 ± 0.14 | 0.26 | -0.07 [-0.16, 0.04] | None |
|       |                    |         |              |                    |                    | 2.6  | Apolipoprotein AI | All Women | 1.24 ± 0.22 | 0.98 ± 0.14 | 0.26 | -0.07 [-0.16, 0.04] | None |
|       |                    |         |              |                    |                    | 2.7  | Apolipoprotein B | All Women | 0.91 ± 0.21 | 0.76 ± 0.17 | 0.15 | -0.06 [-0.16, 0.04] | None |
|       |                    |         |              |                    |                    | 2.8  | Ratio Apo B/Apo AI | All Women | 0.91 ± 0.21 | 0.76 ± 0.17 | 0.15 | -0.06 [-0.16, 0.04] | None |
| 3     | Assmann et al. [25] | Germany | Cohort | 3509 men and 1648 women, adults with no information on age. | Stratified by CPD | 3.1  | Apolipoprotein AII | Men 20 + CPD | 1.38 ± 0.25 | 1.09 ± 0.20 | -0.04 [-0.07, 0.01] | None |
|       |                    |         |              |                    |                    | 3.2  | Apolipoprotein AI | Men 20 + CPD | 1.38 ± 0.25 | 1.09 ± 0.20 | -0.04 [-0.07, 0.01] | None |
| 4     | Berg et al. [26]    | Sweden | Cross-Sectional | 97 men aged 40–42 years | Stratified by CPD | 4.1  | Apolipoprotein AII | Men 10 + CPD | 1.24 ± 0.30 | 0.34 ± 0.04 | -0.15 [-0.24, 0.06] | None |
|       |                    |         |              |                    |                    | 4.2  | Apolipoprotein AI | Men 10 + CPD | 1.24 ± 0.30 | 0.34 ± 0.04 | -0.15 [-0.24, 0.06] | None |
Table 1 (continued)

| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Adjustment |
|-------|-----------|---------|-------------|--------------------|--------------------|-----------------|-------------|----------|------------|
|       |           |         |             |                    |                    | Apolipoprotein  |            |          |            |
| 5     | Casasnovas et al. [27] | Spain | Cohort | 572 young men with an average age of 19.9 ± 0.9 years | None | 5.1 | Apolipoprotein AI | All Men | 0.17 | -0.01 [−0.03, 0.01] |
|       |           |         |             |                    |                    | Apolipoprotein AI | Men 10+ CPD | 0.29 ± 0.04 |          |            |
| 6     | Chen et al. [28] | China | Cohort | 84 men aged 40-80 years | 10+ CPD for at least 10 years | 6.1 | Apolipoprotein AI | All Men | 1.39 ± 0.139 | 0.05 [−0.01, 0.11] |
|       |           |         |             |                    |                    | Apolipoprotein AI | All Men | 1.39 ± 0.139 | 0.05 [−0.01, 0.11] |
|       |           |         |             |                    |                    | Apolipoprotein B | All Men | 0.23 | -0.01 [-0.01, -0.01] |
|       |           |         |             |                    |                    | Ratio Apo B/Apo AI | All Men | 0.54 ± 0.111 |          |            |
| 7     | Chu et al. [29] | Taiwan | Cross-Sectional | 781 men aged 18-24 years | Stratified by CPD | 7.1 | Apolipoprotein AI | All Men | 1.45 ± 0.145 | -0.03 [−0.19, 0.13] |
|       |           |         |             |                    |                    | Apolipoprotein AI | All Men | 1.45 ± 0.145 | -0.03 [−0.19, 0.13] |
|       |           |         |             |                    |                    | Apolipoprotein B | All Men | 0.23 | -0.01 [-0.01, -0.01] |
|       |           |         |             |                    |                    | Ratio Apo B/Apo AI | All Men | 0.78 ± 0.28 |          |            |
|       |           |         |             |                    |                    | Age and body weight |            |          |            |
| 8     | Cuesta et al. [30] | Spain | Cross-Sectional | 391 men aged 20-65 years | Stratified by CPD | 8.1 | Apolipoprotein AI | Men 20-29 | 1.64 ± 0.37 | -0.06 [−0.20, 0.08] |
|       |           |         |             |                    |                    | Apolipoprotein AI | Men 30+ CPD | 1.68 ± 0.39 | -0.08 [-0.24, -0.08] |
|       |           |         |             |                    |                    | Apolipoprotein B | Men 30-39 | 0.88 ± 0.24 |          |            |
|       |           |         |             |                    |                    | Ratio Apo B/Apo AI | Men 30-39 | 1.02 ± 0.28 | -0.10 [−0.29, 0.09] |
|       |           |         |             |                    |                    | Men 40-49 | 0.60 ± 0.05 | 0.16 (0.05, 0.17) |

(continued on next page)
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Adjustments |
|-------|-----------|---------|--------------|--------------------|-------------------|-----------------|------------|----------|-------------|
|       |           |         |              |                    |                   |                 |            |          |             |
| 9     | Dallal et al. [31] | USA      | Cross-Sectional | 146 women and 92 men of Chinese origin, aged 60–96 years. (no female smokers) | None | 9.1 | Apolipoprotein AI | All Men | 1.18 ± 0.32 | 1.18 ± 0.35 | 0.00 [−0.22, 0.22] | None |
| 10    | Dallongeville et al. [32] | France   | Cohort | 180 men aged 45–65 years | 5 + CPD | 10.1, 10.2, 10.3, 10.4 | Apolipoprotein AI | All Men | 1.50 ± 0.28 | 1.54 ± 0.31 | -0.04 [−0.13, 0.05] | None |
| 11    | Dedonder-Decoopman et al. [33] | France | Cross-Sectional | 206 men and 271 women, no information on age. | Stratified by CPD | 11.1, 11.2 | Apolipoprotein AI | Men 15 + CPD | 1.39 ± 0.26 | 1.46 ± 0.24 | -0.13 [−0.22, -0.04] | None |
| 12    | de Parscau & Fielding [34] | USA     | Cross-Sectional | 20 men, 10 smokers and 10 nonsmokers. All healthy aged 20–40 years | 20 + CPD | 12.1, 12.2 | Apolipoprotein AI | All Men | 1.09 ± 0.28 | 1.29 ± 0.17 | -0.20 [−0.37, -0.03] | Weight |

(continued on next page)
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Smokers Mean ± SD | Nonsmokers Mean ± SD | Mean difference Δ (95% CI) | Adjustment |
|-------|-----------|---------|--------------|--------------------|--------------------|-----------------|-------------|----------|-------------------|------------------------|----------------------------|------------|
| 12.3  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 12.4  |           |         |              |                    |                    | 1.55 ± 0.24     | All Men     | 0.59 ± 0.03 | 0.41 ± 0.00 | -0.18 [0.01, 0.08] | 0.35 (0.23, 0.47) | None         |
| 13.1  | Dirican et al. [35] | UK  | Cross-Sectional | All men, 27 cigarette smokers aged 41.9 ± 6.7 years and 31 nonsmokers aged 41.5 ± 6.9 years | 10 + CPD for at least 7 years | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 13.2  |           |         |              |                    |                    | 1.55 ± 0.24     | All Men     | 0.59 ± 0.03 | 0.41 ± 0.00 | -0.18 [0.01, 0.08] | 0.35 (0.23, 0.47) | None         |
| 13.3  |           |         |              |                    |                    | 1.55 ± 0.24     | All Men     | 0.59 ± 0.03 | 0.41 ± 0.00 | -0.18 [0.01, 0.08] | 0.35 (0.23, 0.47) | None         |
| 14.1  | Donahue et al. [36] | USA  | Cross-Sectional | 172 men and women aged 20–24 years | None | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 14.2  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 14.3  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 14.4  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 14.5  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 14.6  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 15.1  | Dullaart et al. [37] | Netherlands | Cross-Sectional | 42 men aged 21–60 years | 5 + CPD | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 15.2  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 15.3  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.1  | Foulon et al. [38] | France  | Cross-Sectional | 251 women and 72 men aged 20–29 years | None | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.2  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.3  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.4  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.5  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |
| 16.6  |           |         |              |                    |                    | 1.49 ± 0.25     | All Men     | 0.76 ± 0.19 | 0.59 ± 0.03 | 0.17 (0.01, 0.35) | 0.27 (0.23, 0.31) | None         |

(continued on next page)
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Mean difference | Adjustment |
|-------|------------|---------|--------------|--------------------|--------------------|-----------------|-------------|----------|----------------|------------|
| 17    | Frey et al. [39] | USA | Cohort | 4196 never smokers and 3176 current smokers aged 35–75 years. Men and women | None | 17.1 | Apolipoprotein AI | All Men | 0.92 ± 0.21 | 0.03 [-0.04, 0.10] | Study |
|       |            |       |              |                    |                   | 17.2 | Apolipoprotein B | All Women | 0.74 ± 0.19 | 0.13 [0.11, 0.15] |
|       |            |       |              |                    |                   | 17.3 | Ratio Apo B/Apo AI | All | 0.77 ± 0.05 | 0.04 (0.03, 0.05) |
| 18    | Frondelius et al. [40] | Sweden | Cohort | Men and women aged 44–74 years. Including 7080 current smokers and 9668 never smokers. | None | 18.1 | Apolipoprotein AI | All Men | 1.45 ± 0.25 | -0.06 [-0.07, -0.05] | Age and screening date. |
|       |            |       |              |                    |                   | 18.2 | Apolipoprotein B | All Women | 1.12 ± 0.26 | 0.09 (0.08, 0.10) |
|       |            |       |              |                    |                   | 18.3 | Ratio Apo B/Apo AI | All | 0.80 ± 0.23 | 0.09 [0.08,0.10] |
|       |            |       |              |                    |                   | 18.4 | Apolipoprotein AI | All Men | 1.98 ± 0.48 | -0.02 [-0.03, -0.01] |
|       |            |       |              |                    |                   | 18.5 | Apolipoprotein B | All Women | 1.12 ± 0.13 | -0.04 [-0.05, -0.04] |
|       |            |       |              |                    |                   | 18.6 | Ratio Apo B/Apo AI | All | 0.75 ± 0.13 | 0.06 (0.05, 0.07) |
| 19    | Gomo et al. [41] | Zimbabwe | Cross-Sectional | 589 men aged 20–65 years | Stratified by CPD | 19.1 | Apolipoprotein AI | All Men | 2.01 ± 0.45 | -0.03 [-0.15, -0.09] | None |
|       |            |       |              |                    |                   | 19.2 | Apolipoprotein B | Men 20+ CPD | 2.04 ± 0.54 | -0.45 [-0.61, -0.29] |
|       |            |       |              |                    |                   | 19.3 | Ratio Apo B/Apo AI | Men 20+ CPD | 0.99 ± 0.34 | -0.10 [-0.18, -0.02] |
|       |            |       |              |                    |                   | 19.4 | Apolipoprotein AI | All Men | 1.98 ± 0.48 | 0.03 [0.12, 0.18] |
|       |            |       |              |                    |                   | 19.5 | Apolipoprotein B | Men 20+ CPD | 0.99 ± 0.44 | 0.03 [0.12, 0.18] |
|       |            |       |              |                    |                   | 19.6 | Ratio Apo B/Apo AI | Men 20+ CPD | 0.49 ± 0.04 | 0.03 [0.12, 0.18] |

(continued on next page)
Table 1 (continued)

| Study | Reference | Country         | Study design | Study participants                                                                 | Smoking definition | Smokers Estimate number | Nonsmokers Estimate number | Mean difference Δ (95% CI) | Adjustment                      |
|-------|-----------|-----------------|--------------|------------------------------------------------------------------------------------|--------------------|-------------------------|---------------------------|--------------------------------|--------------------------------|
| 20    | Haarbo et al. [42] | Denmark | Cohort       | 148 women aged 45–55 with natural menopause 6 months to 3 years previously          | None               | 20.1 Apolipoprotein AI 1.43 ± 0.17 | All Women 1.53 ± 0.26     | -0.10 [-0.17, -0.03]       | Abdominal fat%                  |
|       |           |                 |              |                                                                                    |                    | 20.2 Apolipoprotein B 0.17 | All Women 0.98 ± 0.25     |                               |                                |
|       |           |                 |              |                                                                                    |                    | 20.3 Apolipoprotein Ratio Apo B/Apo AI 1.03 ± 0.23 | All Women 0.66 ± 0.19     | 0.05 [-0.03, 0.13]          |                                |
|       |           |                 |              |                                                                                    |                    |                         |                           | 0.07 (0.01, 0.13)           |                                |
| 21    | Haffner et al. [43] | USA      | RCT-Cohort   | 33 men and 17 women, aged 38-64                                                   | None               | 21.1 Apolipoprotein AI 1.17 ± 0.21 | All Men 1.37 ± 0.21       | -0.20 [-0.35, -0.05]        | None                           |
|       |           |                 |              |                                                                                    |                    | 21.2 Apolipoprotein B 0.21 | All Men 1.39 ± 0.24       |                               |                                |
|       |           |                 |              |                                                                                    |                    | 21.3 Apolipoprotein Ratio Apo B/Apo AI 1.29 ± 0.11 | All Men 0.34 ± 0.04       | -0.10 [-0.28, 0.08]         |                                |
|       |           |                 |              |                                                                                    |                    | 21.4 Apolipoprotein Ratio Apo B/Apo AI 0.29 ± 0.04 | All Men 0.32 ± 0.05       | -0.05 [-0.08, -0.02]        |                                |
|       |           |                 |              |                                                                                    |                    |                         |                           | -0.01 [-0.06, 0.04]         |                                |
| 22    | Haj Mouhammed et al. [70] | Tunisia | Cross-Sectional | 300 men and women, 138 nonsmokers aged 38.47 ± 21.91 years and 162 smokers aged 35.55 ± 16.03 years | None               | 22.1 Ratio Apo B/Apo AI 0.83 ± 0.04 | All 0.52 ± 0.15               | 0.31 (0.23, 0.39)             | Age, gender, BMI                |
| 23    | Hostmark et al. [61] | Norway | Cross-Sectional | 165 men aged 40-60 years                                                            | None               | 23.1 Apolipoprotein B 1.71 ± 0.42 | All Men 1.56 ± 0.39       | 0.15 [0.02, 0.28]            | None                           |
| 24    | Hugues et al. [44] | Singapore | Cross-Sectional | 418 men aged 30–69 years                                                            | None               | 24.1 Apolipoprotein AI 1.33 ± 0.13 | All Men 1.33 ± 0.18       | [-0.03, 0.03]                | Age, ethnic group and BMI    |
|       |           |                 |              |                                                                                    |                    | 24.2 Apolipoprotein Ratio Apo B/Apo AI 1.33 ± 0.13 | All Men 1.21 ± 0.27       | 0.03 [-0.02, 0.08]           |                                |
|       |           |                 |              |                                                                                    |                    | 24.3 Apolipoprotein Ratio Apo B/Apo AI 1.33 ± 0.13 | All Men 0.91 ± 0.06       | 0.02 (0.01, 0.03)           |                                |
| 25    | Kralova et al. [45] | Czech Republic | Cross-Sectional | 39 women, 21 nonsmokers aged 51.8 ± 2.4 years and 18 smokers aged 50.5 ± 3.2 years | 10 + CPD for at least 3 months | 25.1 Apolipoprotein AI 1.59 ± 0.29 | All Women 1.71 ± 0.27       | -0.12 [-0.30, 0.06]         | None                           |
|       |           |                 |              |                                                                                    |                    | 25.2 Apolipoprotein B 0.29 | All Women 1.02 ± 0.33       |                               |                                |
|       |           |                 |              |                                                                                    |                    | 25.3 Apolipoprotein Ratio Apo B/Apo AI 1.10 ± 0.39 | All Women 1.00 ± 0.17 | 0.08 [-0.15, 0.31]          |                                |
|       |           |                 |              |                                                                                    |                    |                         |                           | -0.31 [-0.41, -0.21]        |                                |

(continued on next page)
| Study | Reference | Country          | Study design        | Study participants                                                                 | Smoking definition | Estimate number | Measurement                  | Subgroup | Nonsmokers | Mean difference Δ (95% CI) | Adjustment |
|-------|-----------|------------------|---------------------|------------------------------------------------------------------------------------|--------------------|-----------------|------------------------------|----------|------------|-----------------------------|------------|
| 26    | Liao et al. [46] | China           | Cross-Sectional     | 2505 men and women aged 35–93 years, 1254 of Jing nationality and 1251 of Mulao nationality | None               | 26.1            | Apolipoprotein A1             | Jing Minority | 1.30 ± 0.13 | 1.36 ± 0.03 | 0.06 [-0.03, 0.03] | None        |
|       |           |                  |                     |                                                                                   |                    | 26.2            | Apolipoprotein A1             | Jing Minority | 1.30 ± 0.13 | 1.36 ± 0.03 | 0.06 [-0.03, 0.03] | None        |
|       |           |                  |                     |                                                                                   |                    | 26.3            | Apolipoprotein B               | Jing Minority | 0.23 ± 0.06 | 1.00 ± 0.55 | 0.77 [-0.05, 0.01] | None        |
|       |           |                  |                     |                                                                                   |                    | 26.4            | Apolipoprotein B               | Mulao Minority | 0.96 ± 0.10 | 0.74 ± 0.21 | 0.22 [-0.01, 0.00] | None        |
|       |           |                  |                     |                                                                                   |                    | 26.5            | Ratio Apo B/Apo A1            | Jing Minority | 1.38 ± 0.41 | 1.06 ± 0.23 | 0.00 [-0.07, 0.07] | None        |
|       |           |                  |                     |                                                                                   |                    | 26.6            | Ratio Apo B/Apo A1            | Mulao Minority | 1.38 ± 0.41 | 1.06 ± 0.23 | 0.00 [-0.07, 0.07] | None        |
| 27    | Maeda et al. [47] | Japan          | Cohort              | 349 males and 451 females aged 30–69 years                                        | Everyday smokers | 27.1            | Apolipoprotein A1             | All Men        | 1.46 ± 0.17 | 1.70 ± 0.24 | -0.29 [-0.57, -0.01] | None        |
|       |           |                  |                     |                                                                                   |                    | 27.2            | Apolipoprotein A1             | All Men        | 1.48 ± 0.19 | 1.25 ± 0.28 | 0.23 [-0.19, 0.22] | None        |
|       |           |                  |                     |                                                                                   |                    | 27.3            | Apolipoprotein B               | All Women      | 0.36 ± 0.05 | 0.74 ± 0.04 | 0.38 [-0.20, 0.11] | None        |
|       |           |                  |                     |                                                                                   |                    | 27.4            | Ratio Apo B/Apo A1            | All Women      | 0.34 ± 0.05 | 0.34 ± 0.05 | 0.00 [-0.01, 0.00] | None        |
| 28    | Malczewska et al. [48] | Poland | Cross-Sectional     | 57 females aged 36–46 years                                                        | None               | 28.1            | Apolipoprotein A1             | All Women      | 1.70 ± 0.24 | 1.89 ± 0.19 | -0.19 [-0.35, -0.02] | None        |
|       |           |                  |                     |                                                                                   |                    | 28.2            | Apolipoprotein A1             | All Women      | 1.41 ± 0.14 | 1.25 ± 0.28 | 0.16 [-0.05, 0.05] | None        |
|       |           |                  |                     |                                                                                   |                    | 28.3            | Apolipoprotein B               | All Women      | 0.32 ± 0.05 | 0.74 ± 0.04 | 0.42 [-0.11, 0.22] | None        |
|       |           |                  |                     |                                                                                   |                    |                 | Ratio Apo B/Apo A1            | All Women      | 1.26 ± 0.16 | 0.89 ± 0.05 | 0.37 [-0.16, 0.20] | None        |
| 29    | Meenakshiduram et al. [49] | India | Cross-Sectional     | 274 men active smokers and 78 controls aged 40-59 years                            | Stratified by p/y | 29.1            | Apolipoprotein A1             | All Men        | 1.26 ± 0.19 | 1.89 ± 0.19 | -0.63 [-1.29, -0.01] | None        |
|       |           |                  |                     |                                                                                   |                    | 29.2            | Apolipoprotein A1             | All Men        | 1.15 ± 0.25 | 1.64 ± 0.21 | 0.49 [-0.09, 0.01] | None        |
|       |           |                  |                     |                                                                                   |                    | 29.3            | Apolipoprotein B               | All Men        | 0.25 ± 0.10 | 0.92 ± 0.05 | 0.67 [-0.06, 0.18] | None        |
|       |           |                  |                     |                                                                                   |                    |                 | Ratio Apo B/Apo A1            | All Men        | 1.27 ± 0.28 | 1.89 ± 0.19 | 0.62 [-0.05, 0.20] | None        |
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Nonsubgroup | Mean difference Δ (95% CI) | Adjustment |
|-------|-----------|---------|--------------|---------------------|-------------------|----------------|------------|----------|-------------|-----------------------------|------------|
| 30    | Molgaard & Olsson [50] | Sweden | Cohort | 140 men aged 45–69 years | Stratified by CPD | 30.1 | Apolipoprotein Al | All Men | 1.10 ± 0.12 | 1.34 ± 0.17 | -0.02 [-0.09, 0.05] | None |
|       |           |         |            |                     |                   | 30.2 | Apolipoprotein B | Men 15+ | 1.32 ± 0.12 | 1.33 ± 0.16 | -0.01 [-0.10, 0.08] | |
|       |           |         |            |                     |                   | 30.4 | Apolipoprotein CPD | All Men | 1.21 ± 0.27 | 1.23 ± 0.28 | -0.02 [-0.07, 0.19] | |
|       |           |         |            |                     |                   | 30.5 | Ratio Apo B/Apo | All Men | 0.86 ± 0.04 | 0.92 ± 0.06 | -0.06 [-0.12, 0.14] | |
|       |           |         |            |                     |                   | 30.6 | Apolipoprotein Al | All Men | -0.02 [0.09, 0.05] | -0.01 [0.10, 0.08] | |
|       |           |         |            |                     |                   |     |             | All Men | 0.07 [-0.05, 0.19] | 0.12 [0.04, 0.28] | |
| 31    | Paololetti et al. [51] | Italy | Cross-Sectional | 22 men aged 21–56 years | 20 + CPD | 31.1 | Apolipoprotein Al | All Men | 1.35 ± 0.17 | 0.87 ± 0.12 | 0.55 [-0.09, 0.19] | None |
|       |           |         |            |                     |                   | 31.2 | Apolipoprotein B | All Men | 1.40 ± 0.09 | 0.87 ± 0.12 | 0.52 [-0.08, 0.12] | |
|       |           |         |            |                     |                   |     |             | All Men | 1.28 ± 0.36 | 0.36 ± 0.06 | 0.02 [-0.07, 0.14] | |
|       |           |         |            |                     |                   |     |             | Men 15+ | 1.35 ± 0.36 | 0.36 ± 0.06 | 0.02 [-0.07, 0.14] | |
|       |           |         |            |                     |                   |     |             | All Men | 1.02 ± 0.09 | 0.09 ± 0.09 | 0.03 [-0.14, 0.11] | |
| 32    | Pasupathi et al. [60] | India | Cross-Sectional | 100 smokers aged 40.1 ± 10.3 years and 100 nonsmokers aged 43.3 ± 9.7 years. All men. | None | 32.1 | Apolipoprotein Al | All Men | 1.42 ± 0.12 | 1.00 ± 0.17 | -0.49 [-0.54, -0.44] | None |
|       |           |         |            |                     |                   | 32.2 | Apolipoprotein B | All Men | 0.93 ± 0.17 | 0.70 ± 0.03 | 0.23 [0.63, 0.71] | |
|       |           |         |            |                     |                   | 32.3 | Apolipoprotein CPD | All Men | 1.67 ± 0.15 | 1.81 ± 0.62 | 0.14 [0.04, 0.24] | |
|       |           |         |            |                     |                   |     |             | All Men | 1.17 ± 0.02 | 0.10 ± 0.03 | 0.07 [-0.05, 0.19] | |
| 33    | Periti et al. [65] | Italy | Cross-Sectional | 253 men aged 21–61 years | None | 33.1 | Apolipoprotein B | Men BMI 1 | 1.01 ± 0.21 | 1.20 ± 0.25 | -0.22 [-0.18, 0.14] | Analysis stratified by BMI and Age tertiles |
|       |           |         |            |                     |                   | 33.2 | Apolipoprotein Age 1 | Men BMI 1 | 1.20 ± 0.25 | 1.20 ± 0.25 | -0.00 [-0.26, 0.10] | |
|       |           |         |            |                     |                   | 33.3 | Apolipoprotein Age 2 | Men BMI 1 | 1.15 ± 0.27 | 1.31 ± 0.21 | -0.02 [-0.25, 0.21] | |
|       |           |         |            |                     |                   | 33.4 | Apolipoprotein Age 3 | Men BMI 1 | 1.12 ± 0.21 | 1.25 ± 0.16 | -0.04 [-0.05, 0.25] | |
|       |           |         |            |                     |                   | 33.5 | Apolipoprotein Age 1 | Men BMI 2 | 1.22 ± 0.32 | 1.22 ± 0.32 | 0.00 [0.05, 0.11] | |
|       |           |         |            |                     |                   | 33.6 | Apolipoprotein Age 2 | Men BMI 2 | 1.19 ± 0.15 | 0.15 ± 0.15 | 0.04 [-0.14, 0.20] | |
|       |           |         |            |                     |                   | 33.7 | Apolipoprotein Age 3 | Men BMI 2 | 1.19 ± 0.15 | 0.15 ± 0.15 | 0.04 [-0.14, 0.20] | |
|       |           |         |            |                     |                   | 33.8 | Apolipoprotein Age 1 | Men BMI 2 | 1.19 ± 0.15 | 0.15 ± 0.15 | 0.04 [-0.14, 0.20] | |
|       |           |         |            |                     |                   | 33.9 | Apolipoprotein Age 2 | Men BMI 2 | 1.19 ± 0.15 | 0.15 ± 0.15 | 0.04 [-0.14, 0.20] | |
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement Subgroup | Adjustment |
|-------|-----------|---------|--------------|--------------------|-------------------|-----------------|---------------------|------------|
|       |           |         |              |                    |                   |                 | Smokers Mean ± SD   | Nonsmokers Mean ± SD | Mean difference Δ (95% CI) |          |
| 34    | Rodenas et al. [69] | Spain | Cross-Sectional | 203 men aged 30–55 years | Stratified by CPD | 34.1 | Apolipoprotein B | Men 20 + CPD 1.21 ± 0.24 | Men 20 + CPD 1.15 ± 0.23 | 0.06 [- 0.05, 0.17] | None |
|       |           |         |              |                    |                   |                 |                      |                      |                       |          |
| 35    | Roggi et al. [52] | Italy | Cross-Sectional | 510 men and women aged 20–79 years. | Stratified by CPD | 35.1 | Apolipoprotein Al | Men 20 + CPD 1.53 ± 0.41 | Men 20 + CPD 1.66 ± 0.52 | 0.15 [- 0.42, 0.12] | None |
|       |           |         |              |                    |                   |                 |                      |                      |                       |          |
|       |           |         |              |                    |                   |                 |                      |                      |                       |          |
| 36    | Sakurabayashi et al. [53] | Japan | Cross-Sectional | 1018 men and 1167 women aged 20–69 years | Any cigarette use during the previous year | 36.1 | Apolipoprotein Al | All Men 1.38 ± 0.16 | All Men 1.43 ± 0.17 | 0.00 [- 0.04, 0.04] | None |
|       |           |         |              |                    |                   |                 |                      |                      |                       |          |
|       |           |         |              |                    |                   |                 |                      |                      |                       |          |

(continued on next page)
Table 1 (continued)

| Study  | Reference          | Country     | Study design       | Study participants                                                                 | Smoking definition                        | Estimate number | Measurement                  | Subgroup                | Smokers Mean ± SD | Nonsmokers Mean ± SD | Mean difference Δ (95% CI) | Mean difference Δ (95% CI) | Adjustment |
|--------|--------------------|-------------|--------------------|------------------------------------------------------------------------------------|--------------------------------------------|-----------------|-------------------------------|------------------------|----------------------|----------------------|--------------------------------|--------------------------------|------------|
|        |                    |             |                    |                                                                                   |                                             |                 |                               |                        |                      |                      |                               |                               |            |
| 36.5   |                    |             |                    |                                                                                   |                                             |                 |                               |                        |                      |                      |                               |                               |            |
| 36.6   |                    |             |                    |                                                                                   |                                             |                 |                               |                        |                      |                      |                               |                               |            |
| 36.7   |                    |             |                    |                                                                                   |                                             |                 |                               |                        |                      |                      |                               |                               |            |
| 36.8   |                    |             |                    |                                                                                   |                                             |                 |                               |                        |                      |                      |                               |                               |            |
| 37     | Sanchez et al. [54] | Spain       | Cross-Sectional    | 41 men aged 19–49 years                                                           | None                                       | 37.1            | Apolipoprotein                | B                      | 1.47 ± 0.18         | 0.97 ± 0.16         | 0.00 [-0.01, 0.01]                             | -0.05 [-0.09, 0.01]           | None        | 0.31 ± 0.05         | 0.01 [-0.01, 0.02]                             |
| 38     | Saxena et al. [66]  | USA         | Cross-Sectional    | 9399 never smokers and 3663 current smokers aged 21 years or older. Men and women  | > 100 cigarettes in lifetime, currently smoking every day or some days | 38.1            | Apolipoprotein                | B                      | 0.99 ± 0.08         | 0.98 ± 0.30         | 0.01 [-0.27, 0.11]                             | 0.03 [0.09, 0.51]              | None        | 0.83 ± 0.07         | 0.46 [0.37, 0.55]                             |
| 39     | Sharma et al. [67]  | India       | Cross-Sectional    | 100 men aged 30–40 years                                                           | Stratified by CPD                          | 39.1            | Apolipoprotein                | B                      | 1.21 ± 0.14         | 0.77 ± 0.12         | 0.44 [0.38, 0.50]                             | -0.32 [-0.35, 0.03]            | None        | 1.32 ± 0.26         | -0.35 [-0.42, 0.28]                           |
| 40     | Siekmeyer et al. [55] | Germany    | Cross Sectional    | 68 men and women, 34 smokers with average age of 48 ± 7 years and 34 nonsmokers with average age 49.8 ± 5.8 years | None                                       | 40.1            | Apolipoprotein                | B                      | 1.39 ± 0.13         | 1.58 ± 0.36         | -0.19 [-0.35, 0.03]                             | 0.03 [-0.06, 0.12]             | None        | 0.45 ± 0.12         | 0.07 [-0.04, 0.18]                             |
| 41     | Sirisali et al. [59] | Thailand   | Cross Sectional    | 128 smokers and 67 nonsmokers, all male aged 35–65                                | Stratified by CPD                          | 41.1            | Apolipoprotein                | B                      | 1.00 ± 0.24         | 1.32 ± 0.26         | -0.32 [-0.39, 0.02]                             | -0.05 [-0.09, 0.01]            | None        | 1.03 ± 0.26         | 0.19 [0.09, 0.28]                             |

(continued on next page)
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Smokers Mean ± SD | Nonsmokers Mean ± SD | Mean difference Δ (95% CI) | Adjustment |
|-------|-----------|---------|--------------|-------------------|-------------------|-----------------|-------------|----------|-----------------|----------------------|------------------------|------------|
| 42    | Slagter et al. [56] | Netherlands | Cross-Sectional | 24389 men and 35078 women aged 18–80 years | Smoked during last month or for longer than a year and had not stopped | 42.1 | Apolipoprotein | All Men | 1.43 ± 0.22 | 1.02 ± 0.27 | -0.41 [-0.45, -0.36] | Age |
|       |           |         |              |                   |                   | 42.2 | Apolipoprotein | All Men | 1.04 ± 0.22 | 0.92 ± 0.27 | -0.05 [-0.10, 0.00] |       |
|       |           |         |              |                   |                   | 42.4 | Apolipoprotein | All Women | 0.87 ± 0.23 | 0.94 ± 0.23 | 0.07 [-0.03, 0.17] |       |
|       |           |         |              |                   |                   | 42.5 | Ratio Apo B/Apo AI | All Men | 1.57 ± 0.28 | 0.66 ± 0.04 | -0.57 [-0.64, -0.49] |       |
|       |           |         |              |                   |                   | 42.6 | Ratio Apo B/Apo AI | All Women | 0.54 ± 0.03 | 0.54 ± 0.03 | 0.00 [-0.02, 0.02] |       |

(continued on next page)
| Study | Reference | Country | Study design | Study participants | Smoking definition | Estimate number | Measurement | Subgroup | Nonsmokers | Mean difference | Adjustment |
|-------|-----------|---------|--------------|--------------------|-------------------|------------------|-------------|----------|------------|----------------|------------|
| 46    | Wang et al. [113] | USA | Cross-Sectional | 100 men and women subjects aged 25–84 | None | 46.1 | Apolipoprotein | All Men | 0.86 ± 0.04 | 1.69 ± 0.55 | -0.31 [-0.51, -0.11] | None |
| 47    | Yasue et al. [63] | Japan | Cohort | 438 men and women average age of 61.8 ± 11.0 years (smokers) and 68.2 ± 10.7 years (never smokers) | 10 CPD + for the 10 years or more | 47.1 | Apolipoprotein B | All | 1.02 ± 0.24 | 0.95 ± 0.21 | 0.07 [0.02, 0.12] | None |
| 48    | Zagozdzon et al. [62] | Poland | Cross-Sectional | 1168 men and 1245 women aged 18–79 years | At least one cigarette per day | 48.1 | Apolipoprotein B | All | 0.96 ± 0.28 | 0.92 ± 0.26 | 0.04 [0.01, 0.07] | None |
| 49    | Zaratin et al. [58] | Brazil | Cross-Sectional | 29 men aged 19–35 years | 10 + CPD for more than a year | 49.1 | Apolipoprotein B | All | 1.34 ± 0.27 | 1.25 ± 0.19 | 0.09 [-0.08, 0.26] | None |

CPD = cigarettes per day; OC = oral contraceptives; RCT = randomized controlled trial; p/y = pack/year.
characteristic of studies assessing levels of apolipoprotein levels after smoking cessation.

### Table 2

| Compound          | Analysis | Studies | Estimates | Mean difference, smokers – nonsmokers (95% CI) |
|-------------------|----------|---------|-----------|-----------------------------------------------|
|                   |          |         |           | Fixed effects<sup>a</sup>  | I<sup>2</sup> (%) | Random effects<sup>a</sup> |
| Apolipoprotein A g/L | All Studies | 39      | 59        | -0.036 [-0.041, -0.035]  | 90                  | -0.073 [-0.088, -0.058]  |
| Apolipoprotein B g/L | All Studies | 10      | 16        | -0.003 [-0.007, 0.002]  | 66                  | -0.004 [-0.014, 0.005]  |
| Ratio Apo B/Apo AI | All Studies | 40      | 63        | 0.056 [0.053, 0.059]     | 95                  | 0.063 [0.045, 0.081]     |

* The estimates used can be found in Table 1.

### Table 3

| Compound          | Analysis | Studies | Estimates | Mean difference, smokers – nonsmokers (95% CI) |
|-------------------|----------|---------|-----------|-----------------------------------------------|
|                   |          |         |           | Fixed effects<sup>a</sup>  | I<sup>2</sup> (%) | Random effects<sup>a</sup> |
| Apolipoprotein A g/L | Sensitivity | 36      | 55        | -0.036 [-0.039, -0.033]  | 71                  | -0.042 [-0.053, -0.032]  |
| Apolipoprotein B g/L | Sensitivity | 36      | 59        | 0.053 [0.050, 0.055]     | 81                  | 0.030 [0.018, 0.041]     |
| Ratio Apo B/Apo AI | Sensitivity | 28      | 42        | 0.050 [0.049, 0.050]     | 97                  | 0.059 [0.052, 0.066]     |

* The estimates used can be found in Table 1.

### Table 4

| Study             | Country | Study design | Study participants | Treatment                                                                 | Findings |
|-------------------|---------|--------------|--------------------|--------------------------------------------------------------------------|----------|
| Eliasson et al. 1997 [89] | Finland | Cohort       | 17 non-obese healthy male smokers aged 49.3 ± 7.1 years. | Subjects did not receive any therapy and were not allowed to use nicotine replacement therapy. | After eight weeks of abstinence there were statistically significant increases in apolipoprotein A (from 1.23 ± 0.05 g/L to 1.39 ± 0.03 g/L). After 90 days of abstinence, apolipoprotein A and B levels increased (from 1.41 ± 0.19-1.46 ± 0.25 g/L and from 0.82 ± 0.20-0.83 ± 0.29 g/L, respectively), although neither difference was statistically significant. Switching to mTHS for 90 days resulted in an increase in apolipoprotein A levels from 1.40 ± 0.21-1.49 ± 0.24 g/L and a decrease in apolipoprotein B levels from 0.88 ± 0.29-0.85 ± 0.29 g/L. |
| Haziza et al. 2020 [92] | USA | Interventional | 160 healthy male and female smokers aged 37.7 ± 11.45 | Subjects were randomized to mTHS, mCC, or smoking abstinence for 5 days in confinement and 86 subsequent ambulatory days. Subjects in the smoking abstinence group were not allowed to use nicotine replacement therapy. | Serum apolipoprotein A levels significantly increased from baseline to 12 weeks in the group of successful quitters (1.52 ± 0.28 vs. 1.59 ± 0.27 g/L, p < 0.01). |
| Iwaoka et al. 2014 [90] | Japan | Interventional | 86 consecutive subjects 20 + years of age | Subjects received 12 weeks of cessation therapy with varencline. | Serum apolipoprotein A levels increased from baseline to 6 weeks in successful quitters (1.20 ± 0.15 vs. 1.24 ± 0.25 g/L and 0.51 ± 0.09 vs. 0.57 ± 0.1 g/L, respectively), although only the difference in apolipoprotein A II was statistically significant. |
| Masarei et al. 1991 [88] | Australia | Interventional | 64 male and female smokers aged 35.9 ± 2.3 years | Subjects received 6 regular counsellings at 2-weekly intervals and telephone support. | Serum apolipoprotein A and B levels increased from baseline to 6 weeks in successful quitters (1.20 ± 0.15 vs. 1.24 ± 0.25 g/L and 0.51 ± 0.09 vs. 0.57 ± 0.1 g/L, respectively), although only the difference in apolipoprotein A II was statistically significant. |
| Takata et al. 2014 [91] | Japan | Interventional | 32 smokers 27–64 years of age | Varencline or transdermal patch as part of a 12 week smoking cessation program. | Serum apolipoprotein A levels increased from baseline to 12 weeks in the success group (1.16 ± 0.21 vs. 1.21 ± 0.13 g/L, p < 0.23), although the difference was not statistically significant. |

The pooled mean ratios of apolipoprotein B to A were 0.62 ± 0.002 in smokers and 0.65 ± 0.001 in nonsmokers. The meta-analysis assessing the effects of smoking on the apolipoprotein B: A ratio included 31 studies with 46 estimates [23,24,27–30,32,34,35,37–42,44–46,48–58,62,70]. The fixed-effects model showed an increased ratio in smokers (mean difference = 0.047, 95% CI: 0.047, 0.048 g/L, I² = 99%). The heterogeneity was high; thus, sensitivity and subgroup analyses were
performed. The sensitivity analysis included data from 28 studies and 42 estimates; however, the results did not show much reduction in heterogeneity (mean difference = 0.050, 95% CI: 0.049, 0.050 g/L, \( I^2 = 97\% \)). Heterogeneity did not vary much across subgroups. Considering only studies from the 2000s resulted in the lowest heterogeneity; however, this revealed an opposite effect (mean difference = -0.048, 95% CI: -0.077, -0.018).

### Table 5

**Subgroup analyses of smoking and apolipoprotein AI levels (g/L).**

| Stratification                  | Studies | Effect estimates | Mean differences in apolipoprotein AI levels (g/L) (smokers - nonsmokers) | Fixed effects [95% CI] | \( I^2 \) (%) | Random effects [95% CI] |
|---------------------------------|---------|------------------|--------------------------------------------------------------------------|-------------------------|---------------|------------------------|
| **Region**                      |         |                  |                                                                          |                         |               |                        |
| Americas                        | 7       | 10               | -0.062 (43, -0.094)                                                     |                         |               |                        |
| Europe                          | 20      | 30               | -0.033 (64, -0.044)                                                     |                         |               |                        |
| Asia & Africa                   | 12      | 19               | -0.054 (96, -0.105)                                                     |                         |               |                        |
| **Study Design**                |         |                  |                                                                          |                         |               |                        |
| Cross                           | 28      | 44               | -0.040 (92, -0.098)                                                     |                         |               |                        |
| Sectional                       |         |                  |                                                                          |                         |               |                        |
| Cohort                          | 11      | 15               | -0.036 (76, -0.038)                                                     |                         |               |                        |
| **Period of Publication**       |         |                  |                                                                          |                         |               |                        |
| 1970–1980 s                     | 9       | 17               | -0.066 (62, -0.103)                                                     |                         |               |                        |
| 1990 s                          | 17      | 24               | -0.061 (87, -0.088)                                                     |                         |               |                        |
| 2000 s                          | 4       | 6                | -0.065 (98, -0.078)                                                     |                         |               |                        |
| 2010 s                          | 9       | 12               | -0.035 (84, -0.036)                                                     |                         |               |                        |
| **Smoking Definition\(^a\)**   |         |                  |                                                                          |                         |               |                        |
| No Definition/ Other            | 24      | 35               | -0.036 (93, -0.060)                                                     |                         |               |                        |
| > 5 CPD                         | 2       | 2                | NA                                                                        | NA                      |               |                        |
| > 10 CPD                        | 6       | 6                | -0.122 (56, -0.095)                                                     |                         |               |                        |
| > 15 CPD                        | 2       | 3                | -0.074 (51, -0.082)                                                     |                         |               |                        |
| > 20 CPD                        | 9       | 13               | -0.077 (87, -0.118)                                                     |                         |               |                        |
| **Sex\(^b\)**                  |         |                  |                                                                          |                         |               |                        |
| Men                             | 31      | 39               | -0.033 (93, -0.091)                                                     |                         |               |                        |
| Women                           | 14      | 15               | -0.040 (41, -0.039)                                                     |                         |               |                        |
| Combined                        | 5       | 5                | -0.052 (86, -0.049)                                                     |                         |               |                        |
| Total                           | 39\(^a\) | 59               | -0.038 (90, -0.073)                                                     |                         |               |                        |

\(^a\): sum of studies does not add up to 39, as some studies provided multiple stratum-specific estimates.

\(^b\): sum of studies does not add up to 10, as some studies provided multiple stratum-specific estimates.

### Table 6

**Subgroup analyses of smoking and apolipoprotein AI levels (g/L).**

| Stratification                  | Studies | Effect estimates | Mean differences in apolipoprotein AI levels (g/L) (smokers - nonsmokers) | Fixed effects [95% CI] | \( I^2 \) (%) | Random effects [95% CI] |
|---------------------------------|---------|------------------|--------------------------------------------------------------------------|-------------------------|---------------|------------------------|
| **Region**                      |         |                  |                                                                          |                         |               |                        |
| Americas                        | 3       | 6                | -0.008 (73, 0.012)                                                      |                         |               |                        |
| Europe                          | 3       | 3                | -0.002 (45, 0.004)                                                      |                         |               |                        |
| Asia & Africa                   | 4       | 7                | -0.003 (72, -0.007)                                                    |                         |               |                        |
| **Study Design**                |         |                  |                                                                          |                         |               |                        |
| Cross                           | 7       | 11               | 0.002 (41, 0.002)                                                       |                         |               |                        |
| Sectional                       |         |                  |                                                                          |                         |               |                        |
| Cohort                          | 3       | 5                | -0.019 (65, -0.017)                                                    |                         |               |                        |
| **Period of Publication**       |         |                  |                                                                          |                         |               |                        |
| 1970–1980 s                     | 4       | 7                | -0.009 (68, 0.004)                                                      |                         |               |                        |
| 1990 s                          | 4       | 5                | -0.014 (52, -0.008)                                                    |                         |               |                        |
| 2000 s                          | 2       | 4                | 0.002 (60, -0.001)                                                     |                         |               |                        |
| **Sex\(^b\)**                  |         |                  |                                                                          |                         |               |                        |
| Men                             | 9       | 9                | -0.005 (66, -0.005)                                                    |                         |               |                        |
| Women                           | 5       | 6                | 0.0002 (74, -0.0003)                                                   |                         |               |                        |
| Combined                        | 1       | 1                | NA                                                                        | NA                      |               |                        |
| Total                           | 10\(^a\) | 16               | -0.004 (66, -0.004)                                                    |                         |               |                        |

\(^a\): sum of studies does not add up to 10, as some studies provided multiple stratum-specific estimates.

\(^b\): sum of studies does not add up to 10, as some studies provided multiple stratum-specific estimates.
95% CI: −0.060, −0.036 g/L, $I^2=77$). Detailed results of the subgroup analyses can be found in Table 8. Despite the high heterogeneity, the random-effects model confirmed an increased ratio, with a mean difference of 0.078 g/L (95% CI: 0.068, 0.087 g/L) for all studies and 0.059 g/L (95% CI: 0.052, 0.066 g/L) for studies in the sensitivity analysis. The evaluation of the funnel plot showed (Fig. 5) no evidence of publication bias and the results of Egger’s regression test were not significant (intercept = 0.47, 95% CI: −1.65, 2.6, $t = 0.42$, $p = 0.66$).

4. Discussion

The meta-analyses performed comparing levels of apolipoprotein levels in smokers versus nonsmokers found that apolipoprotein AI and AII levels were higher in nonsmokers than in smokers. On the other hand, apolipoprotein B levels and apolipoprotein B:AI ratios were higher in smokers compared to nonsmokers. These findings describe an overall worse lipid profile in smokers compared to nonsmokers. A limited number of publications was found where the influence of smoking cessation on apolipoprotein levels or the ratio of apolipoprotein B to AI was assessed. For this reason, no meta-analysis was performed on the effects of smoking cessation and these endpoints. Nevertheless, the retrieved publications individually suggested that it is likely apolipoprotein AI and AII levels improve after quitting smoking.

Cigarette smoking can result in disease and disability and is the leading cause of preventable death [96]. Quitting greatly reduces the risk of developing smoking-related disease [97], but it has been proven difficult to achieve [98]. The FDA has published draft guidelines on modified risk tobacco products (MRTPs) [99]. These have led to the evaluation of risk reduction through the use of clinical risk markers [97], which should, in principle, be associated with smoking and be influenced by smoking cessation. A class of clinical risk endpoints usually assessed is lipids, but mostly with conventional measurements such as HDL-C and LDL-C [100]. Cigarette smoking is positively associated with increased levels of triglycerides and lower levels of (HDL-C) [6,101]. However, other than for HDL-C, no meta-analysis has been published on the effects of smoking and cessation on apolipoprotein levels.

Standard lipid analyses for cardiovascular risk assessment include measuring serum or plasma total cholesterol, triglycerides, and HDL-C after an overnight fast [102]. Data from the Framingham Offspring study indicate that LDL-C, small dense LDL-C, lipoprotein(a) or Lp(a), and HDL particle measurements add significant information about CVD risk to the standard lipid profile (summarized in [102]). Furthermore, it has been shown that cardiovascular risk assessment cannot rely solely on the determination of these measurements and that other lipid parameters can be more useful and more predictive of CVD [103,104]. For instance, the measurement of apolipoprotein AI in HDL particles by gel electrophoresis is important in assessing CVD risk and HDL functionality [102]. Evidence from epidemiologic data suggests that instead of measuring the cholesterol in LDL-C or HDL-C, measuring their respective apolipoproteins, apolipoprotein B and apolipoprotein AI, may improve coronary heart disease risk assessment, and in some observational and interventional studies, ratios of lipids and/or apolipoproteins have been better predictors of coronary heart disease risk than levels of any one lipid fraction [104–106]. According to our findings, smoking deteriorates lipid profiles by affecting the levels of all the apolipoprotein measurements.

### Table 7

| Stratification | Studies | Effect estimates | Mean differences in the ratio of apolipoprotein B (g/L) levels (smokers – nonsmokers) |
|---------------|---------|-----------------|---------------------------------------------------------------------------------|
|                |         |                 | Fixed effects [95% CI] | $I^2$ (%) | Random effects [95% CI] |
| **Region**     |         |                 |                       |           |                           |
| Americas       | 6       | 6               | 0.064 [0.055, 0.073]   | 89        | 0.062 [0.019, 0.105]     |
| Europe         | 20      | 37              | 0.055 [0.052, 0.059]   | 67        | 0.050 [0.038, 0.062]     |
| Asia & Africa  | 10      | 15              | -0.004 [0.007]        | 70        | -0.010 [-0.035, 0.015]   |
| **Study Design** |       |                 |                       |           |                           |
| Cross Sectional| 29      | 49              | 0.047 [0.043, 0.052]   | 81        | 0.023 [0.007, 0.039]     |
| Cohort         | 7       | 9               | 0.057 [0.053, 0.061]   | 77        | 0.066 [0.051, 0.081]     |
| **Period of**  |         |                 |                       |           |                           |
| **Publication**|         |                 |                       |           |                           |
| 1970–1980 s    | 6       | 9               | 0.054 [0.016, 0.092]   | 68        | 0.074 [0.004, 0.143]     |
| 1990 s         | 16      | 26              | 0.020 [0.003, 0.037]   | 67        | 0.038 [0.004, 0.073]     |
| 2000 s         | 5       | 7               | -0.003                | 83        | -0.018 [-0.058, 0.022]   |
| 2010 s         | 8       | 11              | 0.056 [0.053, 0.059]   | 87        | 0.053 [0.042, 0.064]     |
| 2020 s         | 1       | 2               | NA                    | NA        | NA                        |
| **Smoking**    |         |                 |                       |           |                           |
| **Definition** |         |                 |                       |           |                           |
| No Definition/| 24      | 40              | 0.053 [0.050, 0.056]   | 82        | 0.056 [0.037, 0.075]     |
| other          |         |                 |                       |           |                           |
| > 5 CPD        | 2       | 2               | NA                    | NA        | NA                        |
| > 10 CPD       | 4       | 4               | 0.006                 | 0         | 0.006                     |
| > 15 CPD       | 1       | 1               | NA                    | NA        | NA                        |
| > 20 CPD       | 8       | 11              | 0.007                 | 86        | 0.073                     |
| **Sex**        |         |                 |                       |           |                           |
| Men            | 28      | 42              | 0.054 [0.049, 0.058]   | 64        | 0.040 [0.025, 0.055]     |
| Women          | 10      | 10              | 0.051 [0.047, 0.055]   | 92        | 0.001 [-0.025, 0.027]    |
| Combined       | 5       | 6               | 0.057 [0.048, 0.066]   | 92        | 0.039                     |
| **Total**      | 36      | 59              | 0.053 [0.050, 0.055]   | 81        | 0.030 [0.018, 0.041]     |

* a: sum of studies does not add up to 37, as some studies provided multiple stratum-specific estimates.
Meta-analyses involve a two-step analysis approach, whereby in the first step, analyses of the data from the individual studies result in effect estimates and standard errors [107]. In the second step, these data are then combined. For this step, individual subject data are not generally available and effects are typically extracted from publications and then pooled [107]. Meta-analysis is a statistical method recommended by the Cochrane Collaboration [108] to combine individual results to make the best use of all available data and therefore increase the power of the analysis. Nonetheless, although meta-analyses are a robust method, they have limitations. These limitations are mostly related to the identification of studies, interstudy heterogeneity, and the availability of information [109]. For instance, in both the PubMed and Scopus searches and in the review of the reference lists, 66 publications were identified as assessing the influence of smoking status on apolipoprotein measurements, but only 49 had complete information that could be used in the analyses. Additionally, for the effects of smoking cessation, only five studies out of the eight retrieved contained useful information. Furthermore, substantial interstudy heterogeneity appeared in all analyses (I² values ranging from 66% in the apolipoprotein AII analysis to 99% in the apolipoprotein B/AI ratio analysis). The latter value is probably because ratios are usually not normally distributed, which could have added to the interstudy heterogeneity. It should be noted that the heterogeneity for apolipoprotein AI and B levels is mainly due to the size of the estimates, whereas for apolipoprotein AI levels, there is also heterogeneity in regard to the direction of the effect. The fact that the studies originated from diverse populations may have also contributed to the high interstudy heterogeneity, as many biological and environmental factors are known to influence apolipoprotein levels. For example, differences in lipoprotein metabolism [110], age [110,111], diet [110], BMI [112], and alcohol intake [110,112] have been shown to contribute to variation in serum apolipoprotein concentrations. For this reason, sensitivity analyses were conducted in which the studies that contributed most to the heterogeneity were removed. These studies included Casasnovas et al. [27], Pasupathi et al. [60], Sirisali et al. [59], and Sharma et al. [67]. The characteristics of the study populations in these studies may have differed from those in the other publications, resulting in increased heterogeneity. Participants in the study by Casasnovas et al. [27], for instance, had an average age of 19.9 ± 0.9 years, which is much lower than other studies. The results of the sensitivity analyses were consistent with the fixed analyses, and the interstudy heterogeneity was reduced by 19% and 14% for apolipoproteins AI and B, respectively, and by 2% for the apolipoprotein B/AI ratio. No sensitivity analysis was conducted for apolipoprotein AII as the excluded studies did not report data for this measure. Subgroup analyses and a random-effects model were implemented to account for the interstudy heterogeneity, both of which showed consistent results to the fixed analyses.

5. Conclusions

The meta-analyses show increased levels of apolipoproteins AI and AII in nonsmokers relative to smokers, while apolipoprotein B levels and apolipoprotein B/AI ratio were increased in smokers relative to nonsmokers. Based on the findings of the meta-analyses, smoking appears to negatively alter the lipid profile of current smokers relative to nonsmokers. These data need to be interpreted with caution, however, due to the high heterogeneity encountered in the analyses. Concerning smoking cessation and apolipoprotein levels, more data is needed to evaluate whether smoking cessation improves the levels of these proteins.

Table 8

| Stratification          | Studies | Effect estimates | Mean differences in apolipoprotein B/AI ratio levels (g/L) (smokers – nonsmokers) |
|-------------------------|---------|-----------------|----------------------------------------------------------------------------------|
|                         |         | Fixed effects   | I² (%)                           | Random effects [95% CI]     |
| **Region**              |         | [95% CI]        |                                  | [95% CI]                     |
| Americas                | 3       | 0.044 [0.035, 0.052] | 100 0.099 [-0.038, 0.235] |
| Europe                  | 16      | 0.051 [0.050, 0.051] | 95 0.068 [0.062, 0.074] |
| Asia & Africa           | 9       | 0.015 [0.011, 0.019] | 97 0.041 [0.013, 0.067] |
| **Study Design**        |         |                 |                                  |                               |
| Cross Sectional         | 22      | 0.049 [0.049, 0.050] | 98 0.056 [0.048, 0.064] |
| Cohort                  | 6       | 0.057 [0.054, 0.060] | 90 0.071 [0.057, 0.085] |
| **Period of Publication** |       |                 |                                  |                               |
| 1970–1980 s            | 4       | 0.030 [0.023, 0.037] | 99 0.102 [0.029, 0.174] |
| 1990 s                  | 13      | 0.047 [0.043, 0.051] | 96 0.073 [0.054, 0.093] |
| 2000 s                  | 3       | -0.048 [-0.060, -0.036] | 77 -0.033 [-0.066, -0.001] |
| 2010 s                  | 8       | 0.050 [0.050, 0.051] | 97 0.059 [0.052, 0.066] |
| **Smoking**             |         |                 |                                  |                               |
| No Definition/other     | 18      | 0.050 [0.049, 0.051] | 97 0.056 [0.048, 0.063] |
| > 5 CPD                | 2       | NA NA NA | NA NA |
| > 10 CPD               | 4       | -0.042 [-0.055, -0.029] | 96 -0.051 [-0.149, -0.046] |
| > 15 CPD               | 1       | NA NA NA | NA NA |
| > 20 CPD               | 7       | 0.061 [0.055, 0.067] | 98 0.085 [0.039, 0.130] |
| **Sex**                |         |                 |                                  |                               |
| Men                     | 22      | 0.049 [0.048, 0.050] | 98 0.073 [0.058, 0.088] |
| Women                   | 9       | 0.050 [0.049, 0.051] | 94 0.031 [0.016, 0.045] |
| Combined                | 5       | 0.076 [0.067, 0.085] | 97 0.095 [0.039, 0.151] |
| **Total**              | 28      | 0.050 [0.049, 0.050] | 97 0.059 [0.052, 0.066] |

* : sum of studies does not add up to 31, as some studies provided multiple stratum-specific estimates.
markers of cardiovascular risk.

Funding

Philip Morris International is the sole source of funding and sponsor of this research.

CRediT authorship contribution statement

Alba Romero Kauss: Data curation, Formal analysis, Writing – original draft. Meagan Antunes: Data curation, Analysis, Validation, Writing – review & editing, Writing – original draft. Sandrine Pouly: Conceptualization, Data validation. Matthew Hankins: Review of the original draft. Annie Heremans: Resources and review of the original draft. Angela van der Plas: Conceptualization, Writing – original draft.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: All authors are employed by Philip Morris International.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.toxrep.2022.05.009.

References

[1] A. Haapanen, et al., Carotid arteriosclerosis in identical twins discordant for cigarette smoking, Circulation 80 (1) (1989) 10–16.
[2] J.B. Kostis, D. Turkевич, J. Sharp, Association between leucocyte count and the presence and extent of coronary arteriosclerosis as determined by coronary arteriography, Am. J. Cardiol. 53 (8) (1984) 997–999.
[3] A.J. Alberg, D.R. Shopland, K.M. Cummings, The 2014 Surgeon General’s report: commemorating the 50th Anniversary of the 1964 report of the advisory committee to the US surgeon general and updating the evidence on the health consequences of cigarette smoking, Am. J. Epidemiol. 179 (4) (2014) 403–412.
[4] K.J. Rempher, Cardiovascular sequelae of tobacco smoking, Crit. Care Nurs. Clin. North Am. 18 (1) (2006) 13–20.
[5] B. Frei, et al., Gas phase oxidants of cigarette smoke induce lipid peroxidation and changes in Lipoprotein properties in human blood plasma, Protect. Effect. Ascorb. Acid. Biochem. J. 277 (Pt 1) (1991) 133–138.
[6] B.R. Winkelmann, K. von Holt, M. Unverderborn, Smoking and atherosclerotic cardiovascular disease: part IV: genetic markers associated with smoking, Biomark Med. 4 (2) (2010) 321–333.
[7] G. De Backer, et al., European guidelines on cardiovascular disease prevention in clinical practice. Third joint task force of european and other societies on cardiovascular disease prevention in clinical practice, Eur. Heart J. 24 (17) (2003) 1601–1610.
[8] A.I. Qureshi, et al., Apolipoproteins A-I and B and the likelihood of non-fatal stroke and myocardial infarction – data from the third national health and nutrition examination survey, Med Sci Monit 8 (5) (2002) CR313–CR316.
[9] W.S. Davidson, A.M. Gotto Jr., et al., Plasma very density lipoprotein contains a single molecule of apolipoprotein B, J. Lipid Res. 29 (11) (1988) 1461–1473.
[10] I. Goldenberg, et al., Current smoking, smoking cessation, and the risk of sudden cardiac death in patients with coronary artery disease, Arch. Intern. Med. 163 (19) (2003) 2301–2305.
[11] J.P. Segrest, et al., The amphilic alpha helix: a multifunctional structural motif in plasma apolipoproteins, Adv. Protein Chem. 45 (1994) 303–369.
[12] J. Elvov, et al., Plasma very low density lipoproteins contain a single molecule of apolipoprotein B, J. Lipid Res. 29 (11) (1988) 1461–1473.
[13] J. Kastelein, et al., Lipids, apolipoproteins, and their ratios in relation to cardiovascular events with statin treatment, Circulation 117 (23) (2008) 3002–3009.
[14] S. Yusuf, et al., Effect of potentially modifiable risk factors associated with myocardial infarction in 52 countries (the INTERHEART study): case-control study, Lancet 364 (9438) (2004) 937–952.
[15] A.M. Gotto Jr., et al., Relation between baseline and on-treatment lipid parameters and first acute major coronary events in the Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAP/TexCAPS), Circulation 101 (5) (2000) 577–584.
[16] J.J. Kastelein, et al., Lipids, apolipoproteins, and their ratios in relation to cardiovascular events with statin treatment, Circulation 117 (23) (2008) 3002–3009.
[17] K. Maeda, Y. Noguchi, T. Fukui, The effects of cessation from cigarette smoking on the lipid and lipoprotein profiles: a meta-analysis, Prev. Med. 37 (4) (2003) 283–290.
[18] S. Balduzzi, G. Rucker, G. Schwarzer, How to perform a meta-analysis with R: a practical tutorial, Evidence-Based Mental Health 22 (2019) 153–160.
[19] M. Harrer, et al., dmetar: companion R package for the guide doing meta-analysis in R, R package version 0 (2019) 09000.
[20] R Core Team, R: A Language and Environment for Statistical Computing, R Foundation for Statistical Computing, Vienna, Austria, 2021.
[21] P. Macaskill, S.D. Walter, L. Irwig, A comparison of methods to detect publication bias in meta-analysis, Stat Med 20 (4) (2001) 641–654.
[22] G. Schwarzer J.R., G. Rucker, , Fixed Effect and Random Effects Meta-Analysis, in Meta-Analysis with R (2015) 252.
[23] H.H. Adachi R., N. Yoshida, H. Tabito, H. Yoshina, The influence of environmental factors on serum apolipoproteins, Jpn. Athero. Soc. 20 (2–3) (1992) 117–125.
[24] T.F. Ashavai, et al., Lipid, lipoprotein, apolipoprotein and lipoprotein(a) levels: reference intervals in a healthy Indian population, J. Atheroscler Thromb. 12 (5) (2005) 251–259.
[25] G. Assmann, H. Schulte, H. Schriewer, The effects of cigarette smoking on serum lipids, Lipids, apolipoproteins, and their ratios in relation to cardiovascular disease: part IV: genetic markers associated with smoking, Biomark Med. 4 (2) (2010) 69–74.
[26] J.A. Casasnovas, et al., Tobacco, physical exercise and lipid profile, Eur. Heart J. 13 (4) (1992) 440–445.
[27] H.I. Chen, et al., The effects of cigarette smoking and smoking cessation on high-density lipoprotein functions: implications for coronary artery disease, Ann. Clin. Biochem. 56 (1) (2019) 100–111.
[28] N.F. Chu, et al., Relationship between smoking status and cardiovascular disease risk factors in young adult males in Taiwan, J. Cardiovasc Risk 3 (2) (1996) 205–208.
[29] C. Cuesta, et al., Effects of age and cigarette smoking on serum concentrations of lipids and apolipoproteins in a male military population, Atherosclerosis 80 (1) (1989) 33–39.
[30] G.E. Dallal, et al., Ascorbic acid, HDL cholesterol, and apolipoprotein A-I in an elderly Chinese population in Boston, J. Am. Coll. Nutr. 8 (1) (1989) 69–74.
[31] J. Dallongeville, et al., Cigarette smoking is associated with differences in nutritional habits and related to lipoprotein alterations independently of food and alcohol intake, Eur. J. Clin. Nutr. 50 (10) (1996) 647–654.
[32] E. Dedonder-Decoopman, et al., Plasma levels of VLDL- + LDL-cholesterol, HDL-cholesterol, triglycerides and apoproteins B and A-I in a healthy population–influence of several risk factors, Atherosclerosis 37 (4) (1980) 559–568.
[33] L. de Parrau, C.J. Fielding, Abnormal plasma cholesterol metabolism in cigarette smokers, Metabolism 35 (11) (1986) 1070–1073.
[34] M. Dirican, et al., Effects of smoking on serum lipid and lipoprotein concentrations and lecithin: cholesterol acyltransferase activity, J. Med. Invest. 46 (3–4) (1999) 169–173.
[35] R.P. Donahue, et al., Apolipoproteins AI, AI and B in young adults: associations with CHD risk factors. The Beaver County experience, J. Chronic Dis. 39 (10) (1986) 823–830.
[99] FDA (Food and Drug Administration), Guidance for industry - Modified risk tobacco product applications - Draft Guidance. 2012.

[100] B.K. Zedler, et al., Biomarkers of exposure and potential harm in adult smokers of 3–7 mg tar yield (Federal Trade Commission) cigarettes and in adult nonsmokers, Biomarkers 11 (3) (2006) 201–220.

[101] A.D. Gepner, et al., Effects of smoking and smoking cessation on lipids and lipoproteins: outcomes from a randomized clinical trial, Am. Heart J. 161 (1) (2011) 145–151.

[102] Schaefer, E.J., et al., The Measurement of Lipids, Lipoproteins, Apolipoproteins, Fatty Acids, and Sterols, and Next Generation Sequencing for the Diagnosis and Treatment of Lipid Disorders, in Endotext, K.R. Feingold, et al., Editors. 2000: South Dartmouth (MA).

[103] G. Assmann, Pro and con: high-density lipoprotein, triglycerides, and other lipid subfractions are the future of lipid management, Am. J. Cardiol. 87 (5A) (2001) 2B–7B.

[104] C.M. Ballantyne, R.C. Hoogeveen, Role of lipid and lipoprotein profiles in risk assessment and therapy, Am. Heart J. 146 (2) (2003) 227–233.

[105] Y.C. Hwang, et al., Prediction of future cardiovascular disease with an equation to estimate apolipoprotein B in patients with high cardiovascular risk: an analysis from the TNT and IDEAL study, Lipids Health Dis. 16 (1) (2017) 158.

[106] M.S. Islam, et al., Association between Serum Apolipoprotein-B and Acute Ischaemic Stroke, Mymensingh Med. J. 27 (2) (2018) 229–236.

[107] S.E. Seide, C. Rover, T. Friele, Likelihood-based random-effects meta-analysis with few studies: empirical and simulation studies, BMC Med. Res. Methodol. 19 (1) (2019) 16.

[108] Cochrane Handbook for Systematic Reviews of Interventions J.P.H.a.S. Green, Editor. 2011.

[109] E. Walker, A.V. Hernandez, M.W. Kattan, Meta-analysis: its strengths and limitations, Cleve Clin. J. Med. 75 (6) (2008) 431–439.

[110] K. Evans, M.F. Laker, Intra-individual factors affecting lipid, lipoprotein and apolipoprotein measurement: a review, Ann. Clin. Biochem. 32 (3) (1995) 261–280.

[111] E.J. Schaefer, et al., Effects of age, gender, and menopausal status on plasma low density lipoprotein cholesterol and apolipoprotein B levels in the framingham offspring study, J. Lipid Res. 35 (5) (1994) 799–802.

[112] E.J. Schaefer, et al., Factors associated with low and elevated plasma high density lipoprotein cholesterol and apolipoprotein A-I levels in the Framingham Offspring Study, J. Lipid Res. 35 (1994) 871–882.

[113] Q. Wang, et al., Serum apolipoprotein A-I quantification by LC-MS with a SILAC internal standard reveals reduced levels in smokers, Bioanalysis 7 (22) (2015) 2895–2911.