Australia's National
Science Agency

A Review of the Health Effects of Avocados

Genevieve James-Martin
Paige Brooker
Gilly Hendrie
Welma Stonehouse

10 March 2022

Prepared for Bite Communications (on behalf of Hort Innovation and Australian Avocado Growers)

Citation

James-Martin G, Brooker P, Hendrie G and Stonehouse W (2022) A Review of the Health Effects of Avocados. CSIRO, Australia.

Copyright

Important disclaimer

Contents

Figures

Tables

Abbreviations

apoB	Apolipoprotein B
BGL	Blood glucose level
BMI	Body mass index
BP	Blood pressure
CI	Confidence interval
CVD	Cardiovascular disease
GRADE	Grading of Recommendations Assessment, Development and Evaluation
HDL-C	High-density lipoprotein cholesterol
HbA1c	Glycated haemoglobin
HOMA-IR	Homeostatic model of insulin resistance
LDL-C	Low-density lipoprotein cholesterol
MD	Mean difference
MPOD	Macular pigment optical density
MUFA	Monounsaturated fatty acid
Ox-LDL	Oxidised low-density lipoprotein
PUFA	Polyunsaturated fatty acid
SCFA	Short chain fatty acid
SD	Standard deviation
SE	Standard error
TC	Total cholesterol
TG	Triglyceride
VAT	Visceral adipose tissue
WC	Waist circumference
RCT	Randomised controlled trial
T2DM	Type 2 diabetes

Acknowledgments

This project has been funded by Hort Innovation, using the avocado research and development levy and contributions from the Australian Government. Hort Innovation is the grower-owned, not-for-profit research and development corporation for Australian horticulture.

The authors thank Darren Jones, of CSIRO Library Services, for developing and executing the literature search strategy.

Executive summary

Background to the problem

Cardiovascular disease is one of the most widespread non-communicable diseases globally and remains the leading cause of death in Australia. Dietary intake, obesity, high blood pressure and elevated LDL-cholesterol (the "bad" cholesterol) levels are important modifiable risk factors for many diseases including cardiovascular disease. Initiatives to improve dietary intake will help to improve risk factors of disease among the Australian population.

The consumption of dietary fats is an important part of dietary guidance for the prevention and management of cardiovascular disease. The balance of saturated fat and unsaturated fats in the average Australian diet does not align with the recommendations for cardiovascular disease. Reducing saturated fat intake and increasing unsaturated fats, including monounsaturated fatty acids and polyunsaturated fatty acids, is recommended. Avocados are an example of a food that is considered a rich source of unsaturated fats, and they also contain a range of other nutrients and phytochemicals beneficial for cardiovascular health.

Avocados are nutrient dense foods in themselves, and research also suggests that people who eat avocados seem to have better diets overall. However, the consumption of avocados is relatively low among Australians. Among a nationally representative sample (in 2011-12), only 16% of the population reported to consume avocados with an estimated average of 16 grams per day, equating to less than 3 grams per person across the whole population.

Avocado consumption has the potential to influence the risk of cardiovascular disease. To date, two systematic reviews with meta-analyses have been conducted on the consumption of avocado and blood lipids, and their findings were mixed. Since the publication of the latest of these two reviews in 2018, additional studies have been published which provide data on cardiometabolic outcomes and additional health outcomes not previously reviewed. Therefore, the purpose of the present study was to review all the available scientific evidence on the effects of avocado consumption on cardiometabolic health and other emerging health outcomes. As part of this, a meta-regression was conducted to explore the optimal serving size and frequency of avocado consumption that is associated with positive health outcomes, and any indications of adverse effects of higher consumption.

Approach to the review

Part I of the review examined the effects of avocado intake on risk factors for cardiometabolic health by conducting a systematic review and meta-analyses. The consumption of avocado was compared to control diets containing no avocado, lower avocado, or an alternative dietary fat source. Studies included adults who were healthy or at increased risk or diagnosed with cardiovascular disease or type 2 diabetes. A secondary aim of this systematic review was to determine the optimal serving size of avocado and frequency of consumption that may generate positive cardiometabolic outcomes.

Part II of the review provided an overview of avocado trials and observational studies focusing on outcomes other than risk factors for cardiometabolic health. Given this area of research is emerging a scoping review was conducted, instead of a systematic review.

Findings: Avocados and cardiovascular health

There were 10 different studies, reported across 12 research publications, which examined the impacts of avocado on cardiometabolic health including (i) blood lipids, (ii) body composition, (iii) other markers of cardiometabolic health such as blood glucose and blood pressure. This evidence suggests that daily consumption of avocado, compared to no or lower avocado intake, resulted in a minor reduction in total cholesterol but did not affect low-density lipoprotein-cholesterol (LDL-C), high-density lipoprotein-cholesterol (HDL-C) or triglycerides in free living, mixed populations including healthy weight adults, those with obesity, type 2 diabetics (T2DM), and dyslipidaemic adults. The reduction in total cholesterol was largely influenced by those studies in adults with hypercholesterolaemia (high total cholesterol or LDL-C) at baseline Furthermore, in this population, avocado intake also resulted in a reduction in LDL cholesterol compared to control diets. However, for reasons associated with the studies design, our confidence in the effect was low and more studies of better quality are needed to be more confident about the benefits of consuming avocados on cardiovascular health.

The consumption of avocados appears not to have an impact on body weight, even when consumed in relatively high amounts. Avocados can be thought of as high in kilojoules and fat, however across the studies in this review no negative effects on body weight were observed. There was limited evidence examining the other health markers of interest such as blood glucose levels and blood pressure, and these studies had inconsistent findings, so no conclusions could be made on the effect of avocados on these outcomes.

Findings: Avocados and other health outcomes

The scoping review identified five areas where the effects of avocado consumption have been investigated: (i) gut microbiome; (ii) cognitive function; (iii) eye health; (iv) inflammation; and (v) skin health. There were only five different studies, reported across six research publications, so the evidence is still considered emerging. The limited evidence available suggests that avocado consumption may improve the gut microbiome by changing the microbial diversity and abundances. The consumption of avocados may also improve specific domains of cognitive function such as recall. The effects of avocado consumption on eye health were mixed and there was no evidence of an effect of avocado consumption on markers of inflammation. One study in the review examined skin firmness and found positive changes in forehead firmness of those who consumed avocado relative to those who did not, however there were positive changes in other markers of skin health over time in both groups, suggesting there may be other variables contributing to this change.

Generally, across all the outcomes, but particularly for outcomes other than for cardiovascular disease, the number of studies available is limited, and the design of these studies and how the outcomes are measured is mixed. This means there is not an adequate and consistent body of evidence at this stage to support a beneficial effect of avocados on these other health related outcomes. More rigorously controlled studies using consistent methods to measure outcomes are needed to better understand the unique role of avocado in health.

Findings: Optimal serving for health

The amount of avocado provided in studies ranged from about 100 g to 330 g. There was some evidence of a dose response relationship between avocado intake and total cholesterol, LDL-C, HDL-C and triglycerides. However, this relationship was only present when the study that provided participants with 330 grams per day was included. This amount was much more than most of the studies and seemed to skew the results. This specific study was also questioned for its quality and accuracy of data reporting, so these findings need to be interpreted with caution.

There was not enough evidence to determine if there are positive health benefits from increasing the average serve size from 50 grams to 75 grams, however no negative effects on blood lipids or body weight were observed either when study participants consumed avocado in amounts that exceeded this. So overall, it seems unlikely that there would be any adverse health outcomes associated with increasing the recommended serving size from 50 grams to 75 grams.

Considerations and future directions

The strengths of the review include the broad strategy and the systematic approach to the search. The systematic review and meta-analyses were conducted using a best practice approach, with study quality assessed and sensitivity examined where possible.

To better understand the effects of avocados on health there is a need for more well-designed studies with larger and more diverse samples of participants. These studies should also be longer in duration, and test different doses of avocados, with careful consideration given to the health attributes of the study population given that the effect may be different for those at a higher level of risk such as those with elevated blood lipids at baseline.

Following more research to establish the health benefits of avocado, consideration of these findings in the context of current eating habits is needed. The current (2011-12) estimated average consumption of avocado is less than 20 grams per day among those Australians who consume it, so a 3-4 fold increase in consumption would be needed to align intakes with current or proposed serving recommendations. Additional work is needed to support Australians to change their eating habits to include more avocados in their daily diets.

Introduction

Cardiovascular disease is one of the most widespread non-communicable diseases, affecting 523 million people worldwide in 2019 [1]. In Australia, despite a decline over the last 50 years, coronary heart disease remains the leading cause of death, accounting for 1 in 5 deaths [2] and is the major contributor to total disease burden [3]. Dietary intake, obesity, high blood pressure and elevated LDL-cholesterol levels are important modifiable risk factors for many diseases. Globally, cardiovascular diseases and diabetes are among the diseases most commonly attributed to poor diet and excess weight [4]. In Australia, more than 40% of the burden of cardiovascular disease and 34% of burden of endocrine disease was attributable to dietary risk factors [3]. With two-thirds of Australian adults considered overweight or obese and most consuming a diet that is not in line with guidelines for health, there is need to change behaviour and improve risk factors among the population.

Consumption of dietary fats is an important part of dietary guidance for the prevention and management of non-communicable diseases, in particular cardiovascular and metabolic diseases [5]. Substitution of saturated fatty acids with unsaturated fatty acids including monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) is a recommended strategy for primary and secondary prevention for cardiovascular disease as this has been shown to decrease low-density lipoprotein cholesterol (LDL-C), the primary target for reducing cardiovascular disease risk [6] and increase high-density lipoprotein cholesterol (HDL-C) cholesterol [5]. Specific targets for dietary fats have been recommended including the consumption of less than 10% of total energy as saturated fats and 4-10% of total energy as omega-6 polyunsaturated fat [7, 8]. These recommendations are translated and communicated to the public through national food based dietary guidelines documentation. For example, the Australian Dietary Guidelines recommend to "*Limit intake of foods containing saturated fat…*" and "*Replace high fat foods which contain predominantly saturated fats such as butter, cream, cooking margarine, coconut and palm oil with foods which contain predominantly polyunsaturated and monounsaturated fats such as oils, spreads, nut butters/pastes and avocado"* [9].

Despite the advice of dietary guidelines, the most recent nationally representative dietary intake survey of Australians suggested that saturated fat (including trans fatty acids) contributed an average 12% of energy, while monounsaturated fat (MUFA) also contributed 12% and polyunsaturated fat (PUFA) contributed 4.7% of total energy [10]. Therefore, the balance of fatty acid intake of Australians is not consistent with the recommendations for the prevention and management of cardiovascular and metabolic disease. Consuming more food high in unsaturated fatty acids, preferably in place of those higher in saturated fatty acids would be recommended to reduce the risks associated with poor diet. Avocados are an example of a food that is considered a rich source of unsaturated fats, containing 16 g per 100 g fat, of which ~63% is MUFA and 13% PUFA [11]. Along with this they contain a range of other nutrients and phytochemicals with cardiometabolic health benefits [12] including fibre, potassium, vitamin E and polyphenols (antioxidants), vitamin K1, B-vitamins (folate, pantothenic acid, niacin) and xanthophyll carotenoids (lutein, and beta-carotene) and phytosterols [13, 14].

The consumption of avocados is relatively low among Australians, with a secondary analysis of the 2011–2012 National Nutrition and Physical Activity Survey reporting an average of 2.6 (95% CI: 2.4, 2.8) grams of avocado consumed per person per day overall and 16.1 (95% CI: 15.4, 16.8) grams per day in consumers of avocado, who made up 15.9% of respondents [15]. Although consumed in small amounts, avocado consumers seem to have overall better diets which may in part be due to important contribution to daily nutrient intake, and/or by replacing unhealthy foods. Analysis of both Australian [15] and US [16, 17] dietary intake data has shown that avocado consumers have higher intakes of favourable nutrients such as

dietary fibre, MUFA, PUFA, vitamin E, magnesium and potassium, along with higher intakes of fruit and vegetables and lower intakes of unhealthy “discretionary” foods. Furthermore, consumers of avocado have been shown to have lower body weight, body mass index and waist circumference than non-consumers [15, 16]. More recent consumption data may show an even greater impact of avocado intake on total nutrient intakes of the population as avocado consumption may have steadily increased in the past decade, if production volume supplied to the Australian market is considered an indicator of population consumption. In 2019-2020 per capita consumption was estimated as 3.88 kg [18].

Avocado consumption has the potential to influence the modifiable dietary risk factors for cardiovascular disease. To date, two systematic reviews with meta-analyses have been conducted on the consumption of avocado and blood lipids. One review [19] concluded that when the studies were pooled in a meta-analysis there were favourable improvements in blood lipids with significant reductions in total cholesterol (TC), LDL-C, and triglyceride, and a non-significant decrease in HDL-C observed. However, in this meta-analysis comparisons were only made between change in baseline and end values in the intervention group and did not account for the control interventions, potentially biasing the interpretations of the data. A more recent review by Mahmassani et al. [20] found no significant differences in serum TC, LDL-C and triglyceride concentrations, and a favourable increase in serum HDL-cholesterol when the avocado interventions were compared to control interventions. However, the authors warned caution in interpretation of these findings given the significant heterogeneity of studies and lack of larger, longer-term trials.

Since the publication of the review by Mahmassani in 2018, additional studies have been published which provide data on cardiometabolic outcomes and additional health outcomes not previously explored [21-24]. A recent narrative review on health effects of avocados [25] summarised the findings of some of these studies, however the review was not systematic and meta-analyses were not conducted. Therefore, the purpose of the present study was to review all the available scientific evidence on the effects of avocado consumption on cardiometabolic health and other emerging health outcomes. As part of this, a meta-regression was conducted to explore the optimal serving size and frequency of avocado consumption that is associated with positive health outcomes, and any indications of adverse effects of higher consumption.

Part I Systematic review of avocados and cardiometabolic health outcomes

1 Methods

1.1 Aim

A systematic review and meta-analyses were conducted to examine the effects of avocado intake on risk factors for cardiometabolic health. The effects of consuming diets containing avocado compared to control diets containing no avocado, lower avocado, or an alternative dietary fat source (MUFA or other source), were investigated in adults who were healthy or at increased risk or diagnosed with CVD or type 2 diabetes. Secondary aims were to determine the optimal serving size of avocado and frequency of consumption that may generate positive cardiometabolic outcomes. The systematic review and meta-analyses were conducted and reported using the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) [26].

1.2 Data sources and study eligibility

1.2.1 Literature search

A comprehensive literature search was conducted on 10 November 2021 across four scientific journal databases (PubMed, Web of Science (core collection), Scopus and ProQuest) combining terms that included the exposure (avocado) and the outcomes of interest (terms related to cardiometabolic health outcomes, see Table 1). The search included original literature published from January 1990 through October 2021. In addition, a search of Google Scholar retrieving the first 400 results as well as a search of the Clinical Trials Registries database [27] were conducted to ensure that relevant studies were captured. Appendix A provides the search strategy.

1.2.2 Study inclusion and exclusion criteria

Studies published from 1990 onwards and available in English were included if they met the in- and exclusion criteria summarised in Table 1.

Table 1. Inclusion and exclusion criteria for systematic review of avocado and cardiometabolic health effects

Population	Adults (aged ≥18 years) who were healthy (including those who were overweight and obese) or had metabolic syndrome, an increased risk of, or a diagnosis of CVD or type 2 diabetes
Intervention/Exposure	Avocado-enriched diet which used the avocado fruit (i.e. flesh)
Comparison	Diet containing no avocado (i.e. usual diet) or that were lower in avocado (defined as an amount of ≤50% of the intervention dose) or contained an alternative dietary fat source (MUFA or otherwise)
Outcomes	Lipids (LDL-C, HDL-C, TC, TG, TC:HDL-C ratio, non-HDL-C, apoB) Absolute/relative CVD risk Markers of oxidative stress (i.e oxidised LDL) Blood glucose, HbA1c, insulin levels, indices of insulin sensitivity (QUICKI) and insulin resistance (HOMA-IR), diabetes risk Systolic and diastolic blood pressure Anthropometric and body composition outcomes (weight, BMI, waist circumference, hip to waist ratio, adipose tissue, fat mass, fat free mass)
Study design	Intervention studies (i.e. parallel and cross-over RCTs, non-randomised controlled trials)

	Prospective cohort studies
Study duration	≥3 weeks
Exclusion criteria	Animal studies *In vitro* studies Acute studies Studies in adults with disease states other than those specified in the inclusion criteria Studies using components of avocado other than flesh (e.g. avocado oil, avocado extract, avocado seed) Studies where avocados were not the primary source of MUFA in the diet (i.e. provided <50% energy from MUFA)

Abbreviations: CVD, cardiovascular disease; MUFA, monounsaturated fatty acid; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TC, total cholesterol; TG, triglyceride; apoB, apolipoprotein B; HbA1c, glycated haemoglobin; QUICKI, quantitative insulin-sensitivity check index; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance; BMI, body mass index; RCT, randomised controlled trial.

1.2.3 Study selection

All duplicate publications were removed. Relevant studies were identified in duplicate by two independent reviewers (GJM, PB). Covidence software [28] was used to screen titles and abstracts followed by review of the full-text publications. Any disagreements were resolved by consensus between the two reviewers.

1.3 Data extraction

Data from each study was extracted by one of two investigators (GJM) and reviewed for errors and inconsistencies by the other investigator (PB). When necessary, uncertainties were resolved by discussion between the investigators. The following data were extracted: first author's family name; year of publication; geographic location; primary objective; funding source; study design (duration, randomisation procedures, blinding, treatment allocation concealment); participant information (sample size (numbers enrolled and completed), attrition and reasons, sex distribution, mean age, mean body mass index (BMI), co-conditions, inclusion and exclusion criteria); details regarding intervention and control treatments; mean dietary intakes (energy, macronutrients, cholesterol); methods used to assess outcomes; outcome results; and the conclusions reported by authors.

For each outcome, the mean and standard deviation (SD) at baseline, end of the intervention and change was extracted for the intervention and control arms. When multiple time points were reported only the end of intervention point was used.

The reported amount of avocado prescribed each day as part of the intervention was recorded. Where this was not provided as a weight, a gram amount was calculated based on the assumption that an average avocado weighs 150g (see Appendix B).

1.4 Statistical analysis

For each outcome, the mean change and standard deviation (SD) of change from baseline to endpoint for intervention groups were entered into Review Manager 5.4.1 [29]. If the SD was not provided, it was calculated from the standard error (SE) or 95% confidence interval (CI). One study reported outcome data as median and percentiles and this was converted to mean and SDs using the method described by Wan 2014 [30]. If only baseline and end data were reported, the mean change was calculated by deducting the baseline from the end value. When possible, the SD was then imputed from a mean correlation coefficient. Insufficient data were available to calculate mean correlation coefficients for any of the outcomes. Using the method of Higgins [31], a mean correlation coefficient can be calculated from other studies (>1) in the

meta-analysis that provided SDs for baseline, end and change values. For lipid outcomes, a correlation coefficient of 0.50 were used, as reported in Mahmassani et al. [20]. SDs were imputed for five studies [32-36]. However, for other outcomes, if change values were not reported, these studies were lost from the meta-analysis.

All outcomes were converted to the same unit of measure, i.e. lipid outcomes in mmol/L were converted to mg/dL using the following conversion factors: 38.67 for TC, LDL-C and HDL-C and 88.57 for triglycerides.

Meta-analysis was performed when ≥2 studies reported the relevant data on a single outcome.

When trials reported multiple comparisons relevant for inclusion in the meta-analysis, and one group was used more than once as a comparison group, the sample size of the specific group was divided by the number of times the group was used as comparison to avoid data duplication and provide appropriate weighting for the results.

The primary meta-analyses compared mean (SD) differences in total cholesterol, LDL-C, HDL-C, TC:HDL ratio triglycerides, BMI, body weight, body fat, visceral adipose tissue, blood glucose levels, insulin resistance and blood pressure between avocado and control interventions. Due to heterogeneity between studies and to avoid false positive results for subgroup analysis, a random-effects model was used to generate Forest plots with weighted mean differences and 95%CI. Heterogeneity between studies was indicated using a combination of Chi^2- ($p < 0.1$) and I^2 statistics (I^2 0%–40% = low; 30%–60% = moderate; 50%–90% = substantial and 75%–100% = considerable heterogeneity) and considering the variation of point estimates and overlap of CIs across studies [37].

Meta-regression was used to examine the effects of avocado dosage on effect size. Meta-regression was conducted using Comprehensive Meta-Analysis software [38] when there was 8 or more studies.

1.4.1 Subgroup and sensitivity analysis

To explore potential reasons for differences in results between studies subgroup analysis were performed for baseline health status (hypercholesterolaemia (baseline LDL-C >115 mg/dL [6]) vs. normocholesterolaemia (baseline LDL-C <115 mg/dL); and type 2 diabetic vs. non-diabetic status), intervention duration (<8 weeks vs. ≥8 weeks, as suggested by European Food Safety Authority as an appropriate timeframe to test sustainability of blood lipids [39]), sex and dietary comparison (alternative dietary fat vs. non-dietary fat intervention e.g. high CHO diet). Sensitivity analysis was performed to assess the impact of each study on the overall result by leaving out one study at a time. Sensitivity analysis considered the following factors that could have affected the overall results: study quality (trials with high apparent bias), trials with imputed data, type of control intervention (trials using low avocado, low-fat and/or high carbohydrate diets), energy-restricted (weight loss) vs. eucaloric diets, ad libitum vs. energy-controlled diets, cross-over vs. parallel study design.

1.4.2 Publication bias

Publication bias was examined using funnel plots in which the SE of the studies were plotted against their corresponding effect sizes. Funnel plots plot the effect size against the study size. When this is done for ≥ 10 studies, this should ideally appear as an inverted funnel (or a triangle) with larger studies gathered around the top of the funnel as these generally have smaller standard errors, while smaller studies spread more widely at the bottom of the plot as they have larger SEs and more variable effect estimates. Publication bias may be suspected if there is asymmetry in the funnel, in particular, an absence of small studies which report a negative effect.

1.5 Quality assessment and grading the evidence

A critical appraisal of the quality of individual studies was undertaken in duplicate by two investigators (GJM, PB) using the Cochrane Risk of Bias Tool 2.0 for intervention studies [40] and the Newcastle Ottawa Scale for Cohort Studies . Disagreements were resolved by consensus between the two investigators and where needed discussion with a third investigator (WS). Studies assessed as “high risk of bias” were considered lower quality. No studies were excluded based on quality rating, but sensitivity analysis was conducted to assess the impact of these studies on the overall result.

Where possible, the overall quality of the body of evidence was rated using the Grading of Recommendations Assessment, Development and Evaluation (GRADE) guidelines considering the risk of bias, inconsistency, indirectness, imprecision, publication bias and assigning a quality level (high, moderate, low and very low) that reflects confidence that the estimates of the overall effects are correct [41]. Where there are insufficient studies to assess the quality of evidence using GRADE (ideally >10 to allow for assessment of publication bias using funnel plots), general strength of evidence categories (promising evidence, emerging evidence and limited evidence) were applied based on those developed by Bell [42] which consider the size of the evidence body, consistency of findings and quality of studies.

2 Results

Figure 1 outlines the number of studies that were assessed at each stage of the screening process. A total of 12 publications, reporting on 10 unique studies, were included in the review. Nine studies were randomised, controlled trials (RCT) and one was a prospective, cohort study.

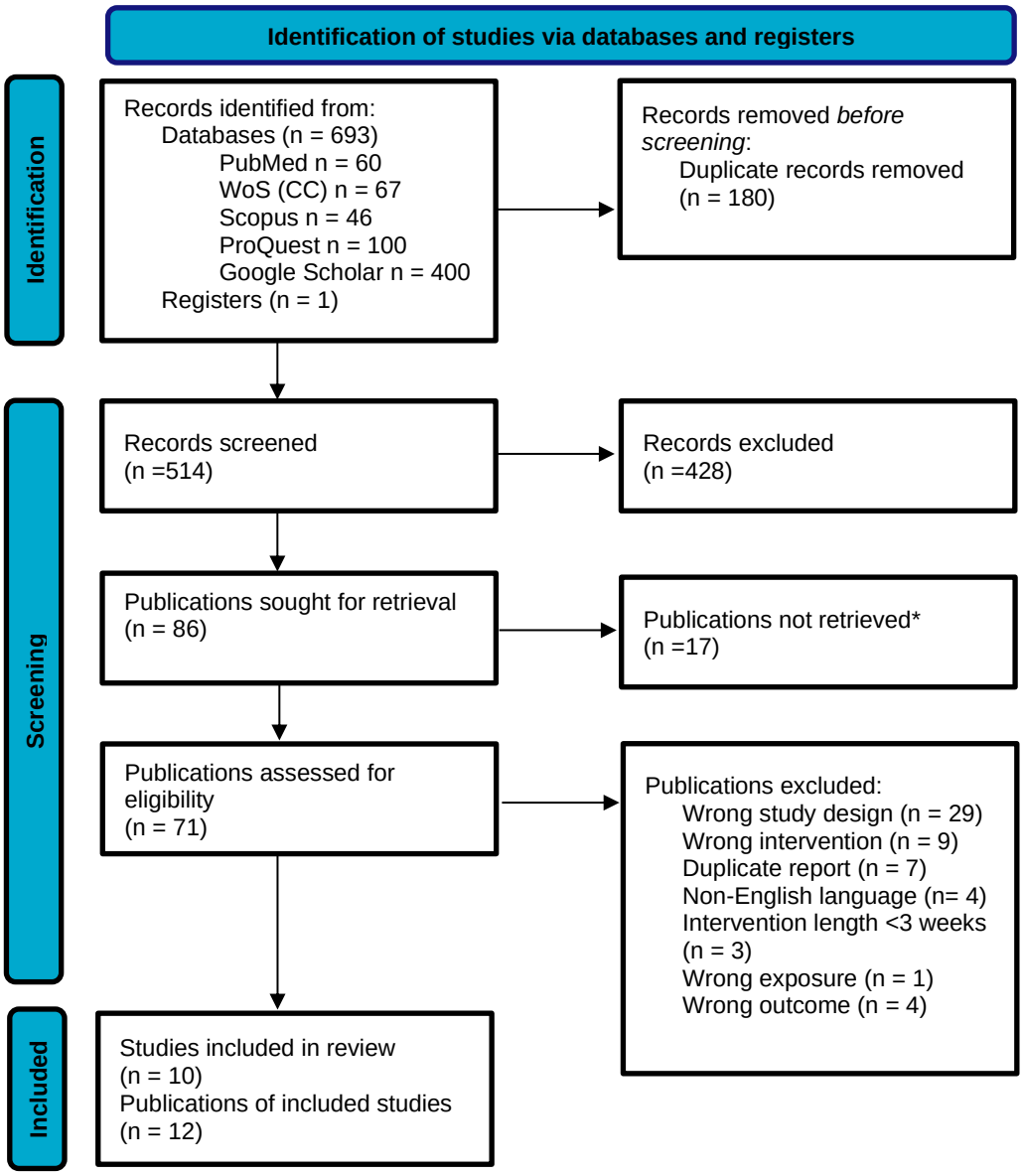

Figure 1. Study flow diagram showing a summary of the literature search and publication selection using the PRISMA format [26].

*Publications not retrieved included abstract or citation only records. Abbreviations: WoS (CC), Web of Science (Core Collection)

2.1 Description of studies included for review

2.1.1 Observational studies

One observational (prospective cohort) study was identified for inclusion [22]. This was an analysis of a subset (n = 55,407) of participants from the Adventist Health Study-2, which included adults (38% males, mean age 55.9 years (SD 13.7)) from the US and Canada. Assessment of exposure and outcome was collected from self-reported data with avocado intake measured at baseline through a food frequency questionnaire (FFQ) and weight and height self-reported at baseline and in two follow-up questionnaires (4 to 11 years after baseline). Due to religious beliefs, subjects in this cohort have unique health characteristics, with 38% of the sample following a vegetarian or vegan diet. Participants were categorised as consumers (low consumers >0 g to <32 g avocado per day, high consumers ≥32 g per day) or non-consumers of avocado. Two statistical methods were used to assess the relationship between avocado consumption and body weight, with results presented for the effect of avocado intake on weight change over time and the odds ratio for becoming overweight/obese.

2.1.2 Intervention studies

Participants

Nine intervention studies including a total of 340 participants were included in the review. Table 2 describes the study design and participant characteristics. Three studies had samples that included only women [32-34], while the remainder were mixed sex. Four studies [21, 23, 36, 43] were conducted in samples of people who were overweight and/or obese. Two studies recruited non-insulin dependent diabetics (T2DM) with hypertriglyceridemia [33, 34], two studies recruited hypercholesterolemic participants, based on high TC [32] or LDL-C [36] concentrations.

Study design

Four studies were randomised, cross-over design [32-34, 36] and four were randomised parallel [21, 23, 35, 43] with one study using a cluster randomised parallel design based on families [24]. Five studies tested interventions of less than 8 weeks (range 3-6 weeks) [32-34, 36, 43] and four assessed intervention of greater than 8 weeks (range 12-24 weeks) [21, 23, 24, 35].

Diets

The dietary comparisons and prescriptions assessed in studies varied. Two studies [21, 43] tested the health impacts of avocado in the context of an energy restricted diet (i.e. weigh loss), while the other studies were eucaloric in their energy prescription. All studies compared iso-caloric (i.e. energy matched) interventions, except for one study [24] which tested a higher versus lower allotment of avocados to families alongside the provision of nutrition education sessions. The majority of studies controlled the energy intake of participants through provision of structured dietary advice or eating plans, with the exception of two studies in which participants consumed an *ad libitum* diet [24, 35]. Due to the heterogenous nature of the dietary comparisons subgroup analysis was not conducted but was explored through sensitivity analysis.

Interventions

The dose of avocado provided in the intervention groups ranged from 99 g to 330 g per day. Only one study compared avocado intake with an alternative dietary source of MUFA fats (high oleic oils) [36], with the majority of studies using a control arm which excluded avocado and was lower in MUFA content than the intervention. Four studies did not report the MUFA content of the diets [21, 24, 35, 43]. In two studies, the intervention included a combination of MUFA sources (one avocado plus four teaspoons of olive oil),

however avocado remained the predominate MUFA source [33, 34]. All other studies had intervention comprised of avocado only.

Outcomes

Table 3 presents the number of studies which reported data for each out the outcomes and the number of studies for which data was able to be included in the meta-analysis. Meta-analysis was possible for 12 outcomes (total cholesterol, LDL-C, HDL-C, TC:HDL ratio, triglycerides, BMI, body weight, body fat, visceral adipose tissue, blood glucose levels, insulin resistance and blood pressure). No studies reported outcome data for insulin sensitivity, hip to waist ratio, diabetes risk or fat free mass.

Analysis

One study conducted an intention-to-treat analysis [24], while the remainder used per-protocol analysis of completers [21, 32, 35, 36, 43] or compliers [23, 33, 34].

Compliance

Seven studies measured compliance to the dietary intervention [23, 24, 33-36, 43]. Three of these did not report compliance, however they implemented a compliance threshold of >80% for inclusion in per protocol analysis [23, 33, 34]. The remaining four studies reported compliance with the intervention which ranged from 83% [24] to 98% [35].

Quality assessment of individual studies

Six of the nine intervention studies were considered to have “some concerns” of bias [24, 33-36, 43] (Figure 2-4) and the remaining three intervention studies [21, 23, 32] were considered to have “high risk” of bias. The domains which were typically of concern related to the randomisation process and the selection of reported results. Regarding randomisation, six studies [21, 33, 34, 36, 43] did not adequately describe the process of randomisation and/or allocation concealment. For the domain of “selection of reported results”, none of the studies reported having a pre-specified analysis plan, and the study by Kahn et al. presented results stratified by sex for some outcomes. This appeared to be the result of post-hoc analysis, resulting in an overall “high” risk of bias. The studies by Henning et al. (2019) and Colquhoun et al. (1992) were also graded “high risk” of bias due to lack of reporting on compliance with the intervention.

The prospective cohort study [22] was scored 5 out of 9, using the Newcastle Ottawa Scale [44], indicating that it was not high quality. Points were deducted for a lack of description of those lost to follow up, using self-reported exposure and outcome data and using a selected group of individuals (Seventh-day Adventists) which are not representative of the wider population.

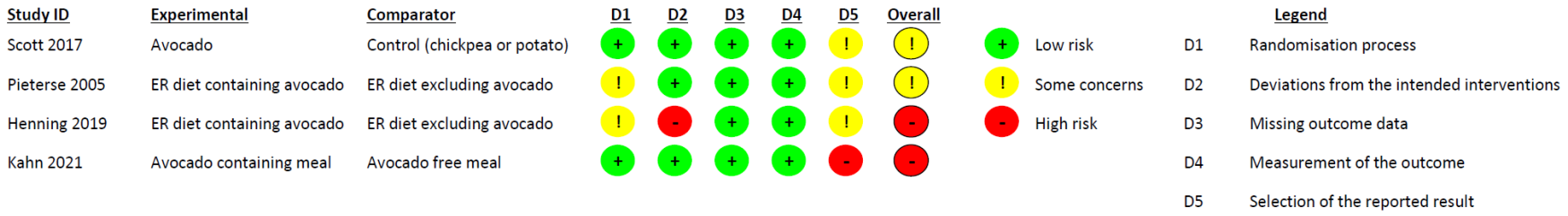

Figure 2. Quality assessment of parallel trials included for review using the Cochrane Risk of Bias 2.0 for randomised trials.

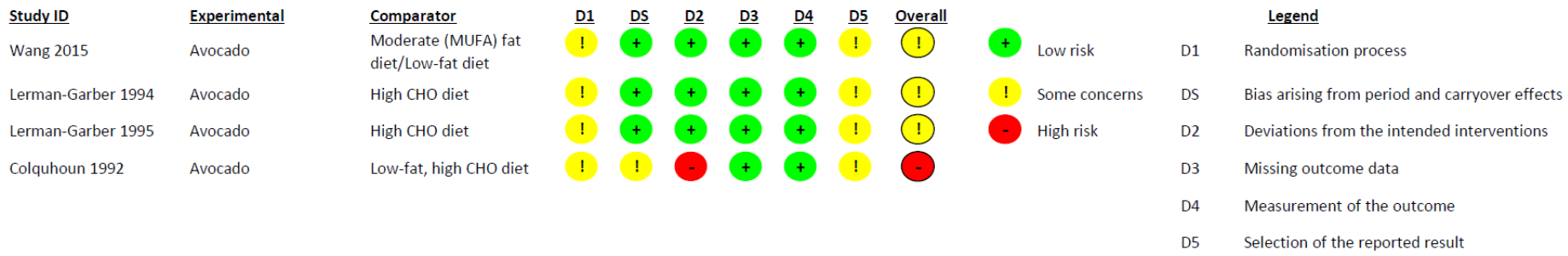

Figure 3. Quality assessment of cross-over trials included for review using the Cochrane Risk of Bias 2.0 for cross-over trials.

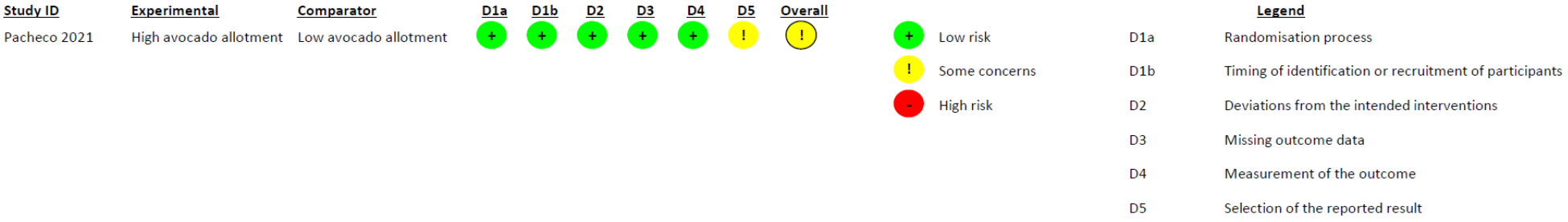

Figure 4. Quality assessment of cluster-randomised trials using the Cochrane Risk of Bias 2.0 for cluster-randomised trials.

Table 2. Study design and participant characteristics of studies included in the systematic literature review

First author, year (ref) (country)	Study design (intervention duration)	N Enrolled (N analysed)	Male, %	Ethnicity	BMI, kg/m² (SD)	Weight status	Age range Mean years (SD) [range]	Co-conditions	Diet comparison	Avocado g/day	MUFA (% E)		Energy prescription	Isocaloric interventions	Ad libitum diet
											Int	Ctrl			
Intervention studies															
Colquhoun et al. 1992 [32] Australia	Randomised, cross-over (3 wk)	15 (15)	0	NR	NR	NR	48.5 (6.4) [37-58]	Hypercholesterolaemia	Avocado diet vs. low-fat diet	330[3]	20[5]	14[5]	Eucaloric	Yes	No
Henning et al. 2019 [21] USA	Randomised, controlled, parallel (12 wk)	63 (51)	Int: 17 Ctrl: 26	NR	Int: 30.1 (3.2) Ctrl: 30.0 (3.7)	Overweight and obese	Int: 42.5 (12.7) [NR] Ctrl: 36.4 (10.8) [NR]	Nil reported	Hypocaloric diet with avocado vs. hypocaloric diet without avocado	150[2]	NR	NR	Negative (i.e. energy restricted)	Yes	No
Kahn et al. 2021[6] [23] USA	Randomised, controlled, parallel (12 wk)	163 (105)	39	80% Caucasian	32.6 (6.1)	Overweight and obese	34.5 (5.9) [NR]	Nil reported	Avocado containing meal vs. avocado free meal	175(M)/ 140 (F)	16[5]	12[5]	Eucaloric	Yes	No
Lerman-Garber et al. 1994 [34] Mexico	Randomised, cross-over (4 wk)	16 (12)	0	NR	28 (4)	NR	56 (8) [NR]	T2DM and hypertriglyceridemia	High-MUFA diet[1] vs. high CHO diet	150[2]	24[4]	7[4]	Eucaloric	Yes	No
Lerman-Garber et al. 1995 [33] Mexico	Randomised, cross-over (6 wk)	20 (13)	0	NR	25.2 (2.3)	NR	60 (7) [NR)	T2DM and hypertriglyceridemia	High-MUFA diet[1] vs. high CHO diet	150[2]	24[4]	7[4]	Eucaloric	Yes	No

Pacheco et al. 2021 [24] **USA**	Cluster-randomised, controlled, parallel (24 wk)	72 (72)	hAVO: 0 lAVO: 13	Self-identified Hispanic/Latino heritage	30.4 (6.4)	Nil reported	45.5 (9.9) [NR]	Nil reported	High avocado allotment plus nutrition intervention vs. low avocado allotment plus nutrition intervention	99[2]	NR	NR	Eucaloric	No	Yes
Pieterse et al. 2005 [43] **South Africa**	Randomised, controlled, parallel (6 wk)	61 (55)	21	NR	31.9 (3.9)	Overweight and obese	40.8 (8.9) [21-57]	Mixed population for lipid and blood pressure status	Energy-restricted diet containing avocado vs. energy restricted diet excluding avocado	200	NR	NR	Negative (i.e. energy restricted)	Yes	No
Scott et al. 2017 [35] **USA**	Randomised, controlled, parallel (24 wk)	48 (40)	63	NR	Exp: 24.1 (3.1) Ctrl: 24.2 (2.4)	NR	Int: 63.3 (11.1) [NR] Ctrl: 62.5 (9.2) [NR]	Nil reported	Avocado vs. control (chickpea and/or potato)	135	NR	NR	Eucaloric	Yes	Yes
Wang et al. 2015[7] [36] **USA**	Randomised, cross-over (5 wk)	45 (40)	60	Predominantly Caucasian and non-Hispanic (n=41)	28.2 (2.4)	Overweight	45 (13.3) [NR]	Hypercholesterolaemia	Avocado diet vs. lower fat diet vs. moderate fat diet	136	17[4]	11[4]	Eucaloric	Yes	No
Observational studies															
Heskey et al 2019 [22] **USA/Canada**	Prospective, cohort study	(NA) (55,407)	37	22% black, 78% non-black	Non-consumer 27.3 (4.8) Low intake	NA	55.9 (13.7) [NR]	NR	Consumer (Low avocado consumer (>0-<32	High: 37.8[5]	NR	NR	NA	NA	NA

	(4 to 11 year follow up)				26.1 (4.5) High intake 24.8 (4.4)				g/day), High consumer (≥32 g/day) vs non consumer	Low: 2.3[5]					

[1] High MUFA intervention was comprised of one avocado plus 4 teaspoons of olive oil [2]Calculated value based on average weight of an avocado 150 g [14], [3]Calculated values based on reported 20-35% energy from avocado [32]. [4] Planned [5] Measured, [6] Additional data sources from Hannon 2020 [45] [7] Additional data sourced from Wang 2020 [46]
Abbreviations: SD, standard deviation; BMI, body mass index; g/day, grams per day; Int, intervention group; USA, United States of America; wk, weeks; NR, not reported; Ctrl, control group; T2DM, hAVO, High avocado allotment; lAVO, Low avocado allotment MUFA, monounsaturated fatty acids; CHO, carbohydrate.

Table 3. Overview of outcome measures reported by studies included for review and their inclusion for meta- analysis

		Blood lipids							Body composition					Other cardiometabolic markers						
Author (Date)	Primary outcome	TC	LDL-C	HDL-C	TC: HDL-C	TG	non-HDL-C	ApoB	BMI	Body weight	Body fat %	VAT	WC	BGL	Insulin	HbA1c	HOMA-IR	BP	Ox-LDL	CVD risk
Colquhoun (1992)	Lipids	✓	✓	✓	✓	✓	-	✓	-	-	-	-	-	-	-	-	-	-	-	-
Henning (2019)	Weight loss	✓	✓	✓	-	✓	-	-	✓	✓	✓	✓	-	✓	✓	-	-	-	-	-
Kahn (2021[b])	Abdominal adiposity, insulin resistance, oral glucose tolerance	✓	-	✓	-	✓	-	-	-	✓	✓	✓	-	✓	-	-	✓	-	-	-
Lerman-Garber (1994)	Glycaemic control, lipids	✓	✓	✓	-	✓	-	-	-	✓	-	-	-	✓	-	-	-	-	-	-
Lerman-Garber (1995)	Glycaemic control, lipids	✓	✓	✓	-	✓	-	-	-	✓	-	-	-	✓	-	✓	-	-	-	-
Pacheco (2021)	Nutrient intake	✓	✓	✓	-	✓	-	-	✓	-	-	-	✓	✓	✓	✓	✓	✓	-	-
Pieterse (2005)	Weight loss	✓	✓	✓	-	✓	-	-	✓	✓	✓	-	-	-	-	-	-	✓	-	-
Scott (2017)	Cognition	✓	✓	✓	-	✓	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Wang (2015)	Lipids	✓	✓	✓	✓	✓	✓	✓	-	-	-	-	-	✓	✓	-	✓	✓	✓[a]	✓
Total studies (Total included for meta-analysis)		9 (8)	8 (8)	9 (8)	2 (0)	9 (8)	1 (0)	2 (0)	3 (3)	5 (2)	3 (2)	2 (2)	1 (0)	6 (2)	3 (2)	2 (0)	3 (2)	3 (2)	1 (0)	1 (0)

Underline indicates that results were able to be included for meta-analysis, [a]Results reported in Wang 2020 [46]., [b]Results for lipid data are reported in a subsequent publication Hannon 2020 [45] however format was not able to be used for meta-analysis.

Abbreviations: TC, total cholesterol; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglyceride; apoB, apolipoprotein B; BMI, body mass index; VAT, visceral adipose tissue; WC, waist circumference; BGL, blood glucose levels; HbA1c, glycated haemoglobin; HOMA-IR, homeostatic model of insulin resistance; BP, blood pressure; Ox-LDL, oxidised low-density lipoprotein; CVD, cardiovascular disease.

2.2 Outcome Results

2.2.1 Low-density lipoprotein-cholesterol

Nine studies reported LDL-C as an outcome (Figure 5) and eight were included for meta-analysis (191 participants in the avocado group, 235 participants in the control group). Two of these studies were in hypercholesterolaemic populations [32, 36]. When all studies were pooled, there was no significant difference in LDL-C observed between the avocado and control groups (-3.02 mg/dL (-8.83, 2.78 mg/dL), p = 0.31) (Figure 5). Heterogeneity was moderate (p = 0.08, I^2 = 44%). When subgroup analysis was conducted, comparing studies with hypercholesterolaemic (mean baseline LDL-C >115 mg/dL) vs. normocholesterolaemic (those with LDL-C <115 mg/dL or not reported) participants, a significant difference was seen between subgroups (Chi^2 = 10.62, p = 0.0001). Studies in hypercholesterolaemic participants showed a significant reduction in LDL-C (-9.4 mg/dl (-10.84, -7.95 mg/dL) p = <0.00001) in the avocado vs. control groups whereas studies in normocholesterolaemic participants showed no difference. Comparing studies according to intervention length, studies with an intervention length <8 weeks vs. studies with an intervention length ≥8 weeks, the results were replicated as the subgroups were exactly the same as for the hyper- and normocholesterolaemic subgroups (Figure 6).

Subgroup analysis by sex also showed a subgroup difference (Chi^2 = 8.57, p = 0.003) with studies in females only [32-34] showing a significant reduction in LDL-C (-9.52 mg/dL (-10.99, -8.06 mg/dL), p = <0.00001). Similarly, as above, the female only studies all fell within the hypercholesterolaemic subgroup.

Sensitivity analysis showed a significant reduction in LDL-C in the avocado vs. control group when the Pacheco et al. study was removed (-6.73 mg/dL (-10.92, -2.53 mg/dL), p = 0.002) and the Henning et al. study, which was at “high risk” of bias, -5.35 mg/dl (-10.22, -0.49 mg/dL), p = 0.002. Conversely, when the study by Colquhoun et al. was removed, the results became non-significant in all the above subgroup analyses.

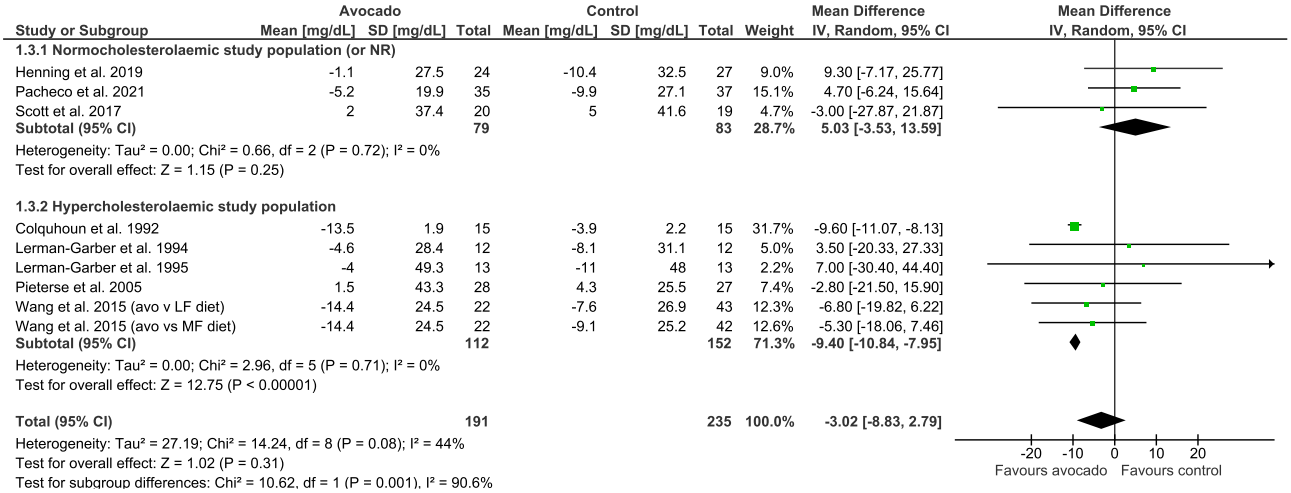

Figure 5. Forest plot of mean (95% CI) difference in LDL-C (mg/dL) between avocado and control groups, stratified for subgroups of hypercholesterolaemic and normocholesterolaemic or not reported study populations.

Avo, avocado diet, LF, low-fat, MF moderate fat. Hypercholesterolaemia was defined as baseline LDL-C >115 mg/dL and normocholesterolaemia as baseline LDL-C <115 mg/dL (or not reported) as per European Society of Cardiology (ESC) and the European Atherosclerosis Society (EAS) guidelines for the management of dislipidemia [6].

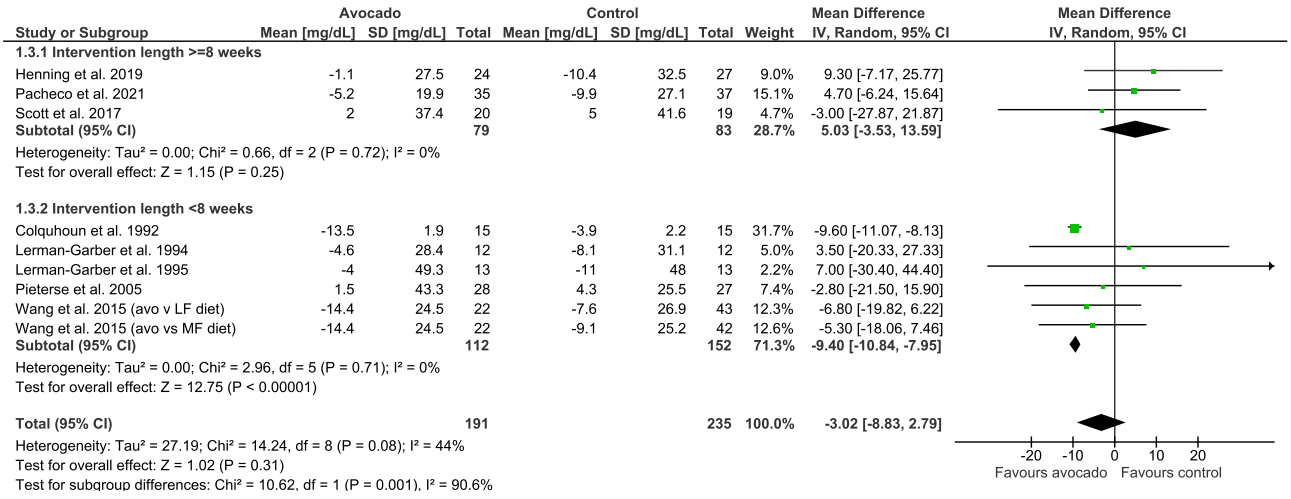

Figure 6. Forest plot of mean (95% CI) difference in LDL-C (mg/dL) between avocado and control groups, stratified for subgroups with intervention length <8 weeks vs. intervention length ≥8 weeks.

Avo, avocado diet, LF, low-fat, MF moderate fat

Meta-regression was performed to examine the study-level relationship between dose of avocado intake and LDL-C (Figure 7, Figure 8). Dose ranged from 99 - 330 g avocado per day. When all studies (n=8 studies, 9 comparisons) were included a significant inverse relationship between avocado dose and LDL-C was observed (i.e. an increased avocado amount resulted in a greater reduction in LDL-C) (regression coefficient -0.049, 95% CI -0.079, -0.019, p = 0.0013) (Figure 7). Given the large weighting (32%) that the Colquhoun et al. study carried in the analysis, combined with its high risk of bias and the high relative dose provided (330 g per day), the meta-regression was also performed with this study excluded (Figure 8). Dose ranged from 99 - 200 g with Colquhoun excluded and the relationship between avocado dose and LDL-C became non-significant (-0.054, 95% CI -0.259, 0.150, p = 0.60).

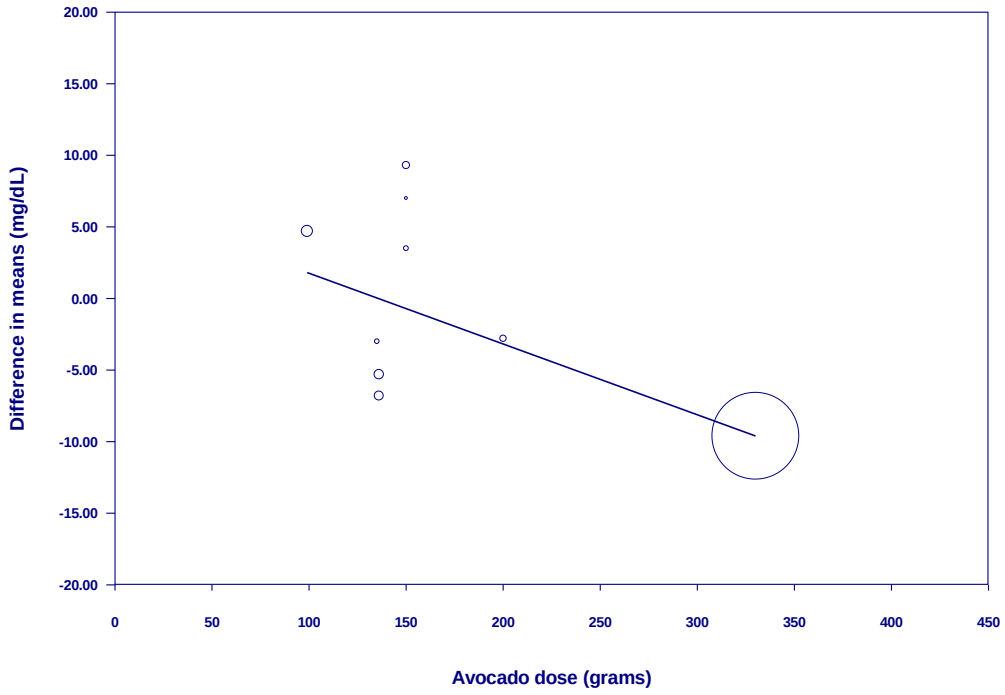

Figure 7. Mean difference in LDL-C (md/dL) between avocado and control groups by avocado dose (n = 8 studies, 9 comparisons).

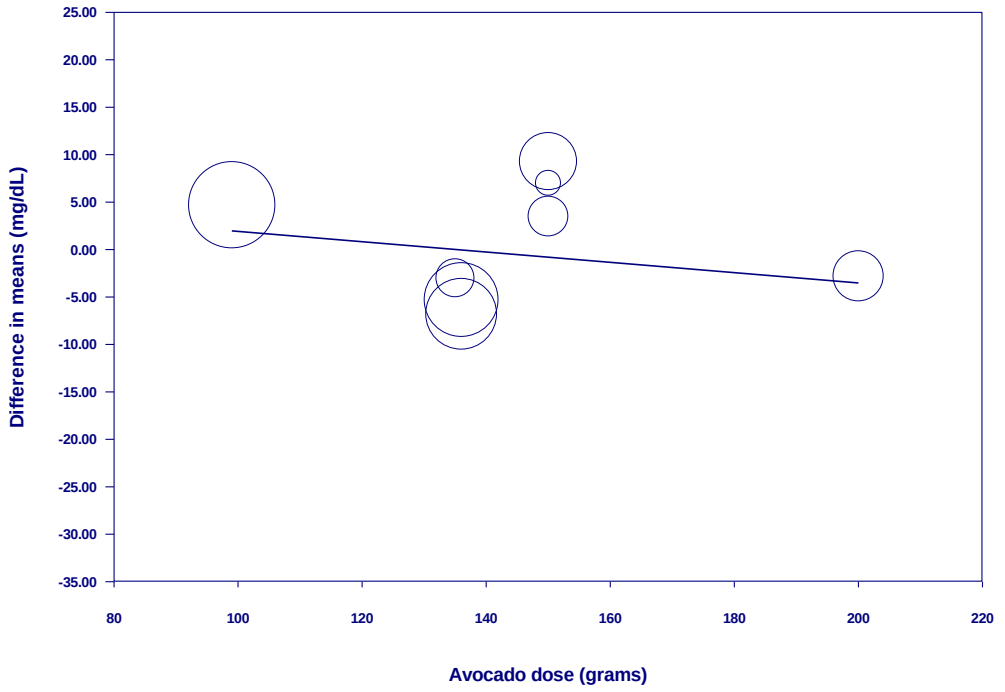

Figure 8. Mean difference in LDL-C (md/dL) between avocado and control groups by avocado dose (n = 7 studies, 8 comparisons) with Colquhoun et al. excluded.

2.2.2 Total cholesterol

Nine studies reported on TC as an outcome (Figure 9) and eight were included for meta-analysis (191 participants in avocado group, 235 in control group). Two of these studies specifically recruited hypercholesterolaemic populations [32, 36]. When all studies were pooled, there was a significant reduction in TC observed between the avocado and control groups (-5.08 mg/dL (-9.29, -0.87 mg/dL), p = 0.02) and heterogeneity was low (p = 0.33, I^2 = 12%) (Figure 9)). Subgroup analysis comparing studies in hypercholesterolaemic populations (mean elevated baseline LDL-C >115 mg/dL) vs. those with normocholesterolaemic populations (LDL-C <115 mg/dL or not reported), or studies with an intervention length ≥8 weeks with those with an intervention length <8 weeks, showed that it was the studies in hypercholesterolaemic populations (or intervention length <8 weeks) which were responsible for the overall effect, with a mean reduction in TC of -7.54 mg/dL (-9.40, -5.68 mg/dL), p < 0.00001) in this subgroup. No subgroup effect was seen for sex.

Sensitivity analysis showed that when the study by Colquhoun et al. which carried a weighting of 62% overall was removed the results became non-significant overall (-0.75 mg/dL, (-7.26, 5.77 mg/dL), p = 0.82) and in the hypercholesterolaemic subgroup (-3.95 mg/dL (-12.93, 5.03 mg/dL) p = 0.39).

Kahn et al., whose results could not be included in the meta-analysis, reported no significant effects on TC by intervention group (or time) [23, 45].

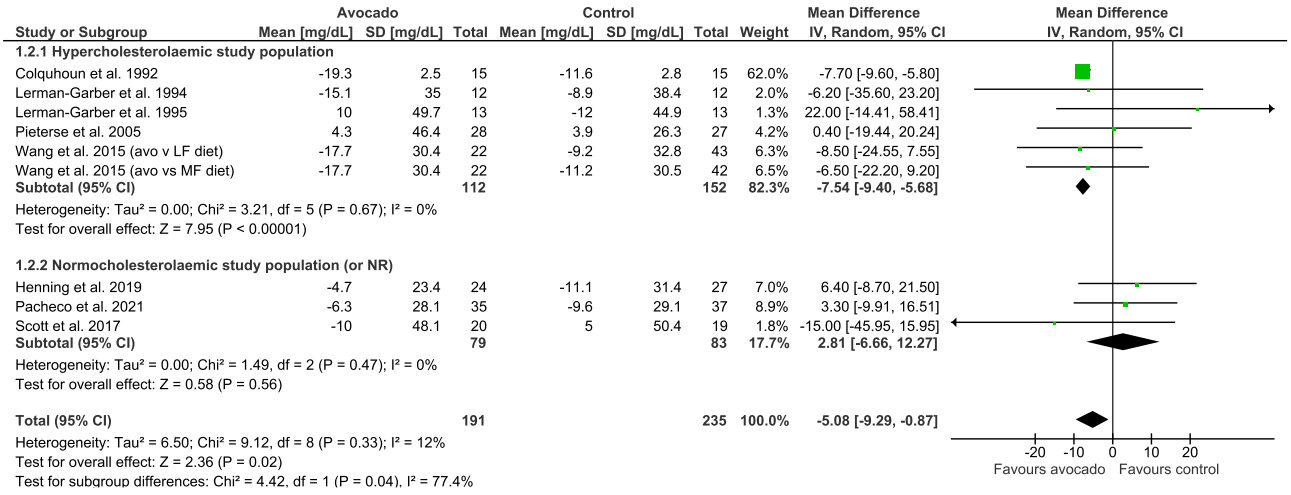

Figure 9. Forest plot of mean (95% CI) difference in total cholesterol (mg/dL) between avocado and control groups, stratified for subgroups of hypercholesterolaemic and normocholesterolaemic study populations.

Avo, avocado diet, LF, low-fat, MF moderate fat. Hypercholesterolaemia was defined as baseline LDL-C >115 mg/dL and normocholesterolaemia as baseline LDL-C <115 mg/dL (or not reported) as per European Society of Cardiology (ESC) and the European Atherosclerosis Society (EAS) guidelines for the management of dislipidemia [6].

Meta-regression was performed to examine the study-level relationship between avocado intake and TC (Figure 10, Figure 11). Dose ranged from 99 - 330 g avocado per day. When all studies (n=8 studies, 9 comparisons) were included a significant inverse relationship between avocado dose and TC was observed (i.e. an increased avocado amount resulted in a greater reduction in TC) (regression coefficient -0.035, 95% CI -0.071, -0.0003, p = 0.048) (Figure 10). Given the large weighting (62%) that the Colquhoun et al. study carried in the analysis, combined with its high risk of bias and the high relative dose provided (330g per day), the meta-regression was also performed with this study excluded (Figure 11). Dose ranged from 99 –

200 g with Colquhoun excluded and the relationship between avocado dose and TC became non-significant (regression coefficient -0.004, 95% CI -0.233, 0.224, p =0.97).

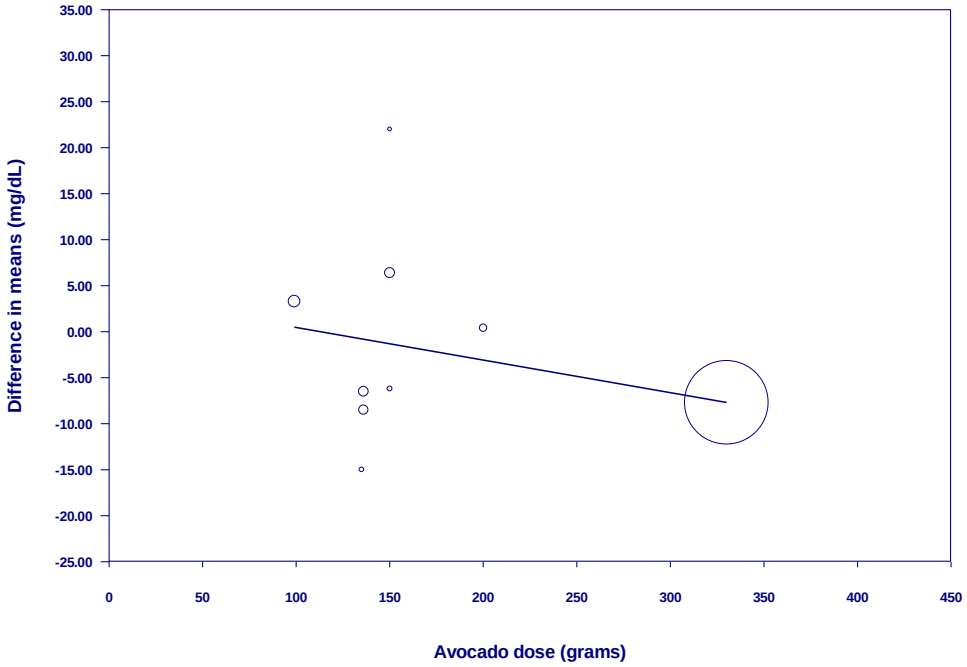

Figure 10. Mean difference in TC (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 8 studies, 9 comparisons).

The size of the circle is proportional to the study weighting.

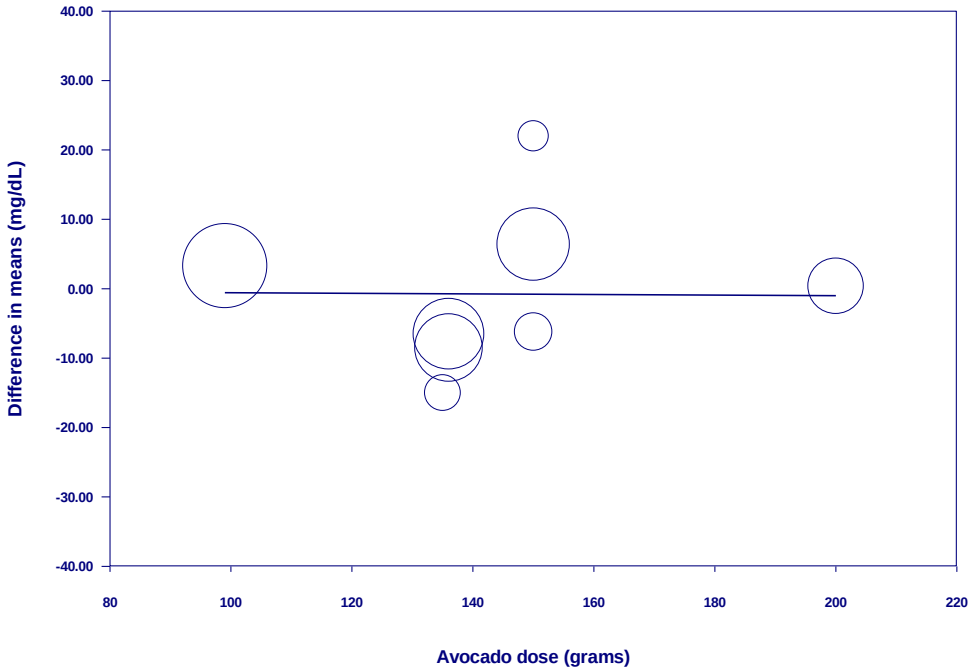

Figure 11. Mean difference in TC (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 7 studies, 8 comparisons) with Colquhoun et al. excluded.

2.2.3 High-density lipoprotein-cholesterol

Nine studies reported HDL-C as an outcome (Figure 12) and 8 were included for meta-analysis (191 participants in the avocado group, 235 participants in the control group). Two of these studies [33, 34] reported low mean baseline HDL-C levels (defined as <46 mg/dL for women and <43 mg/dl for males [6]). When all studies were pooled, there was no difference between the avocado and control groups (1.46 mg/dL (-1.98, 4.90 mg/dL), p = 0.41) (Figure 12). Heterogeneity was considerable (p = <0.00001, I^2 = 88%). None of the subgroup analyses (including baseline HDL-C status, intervention duration, sex and risk of bias) nor sensitivity analysis changed the overall non-significant result.

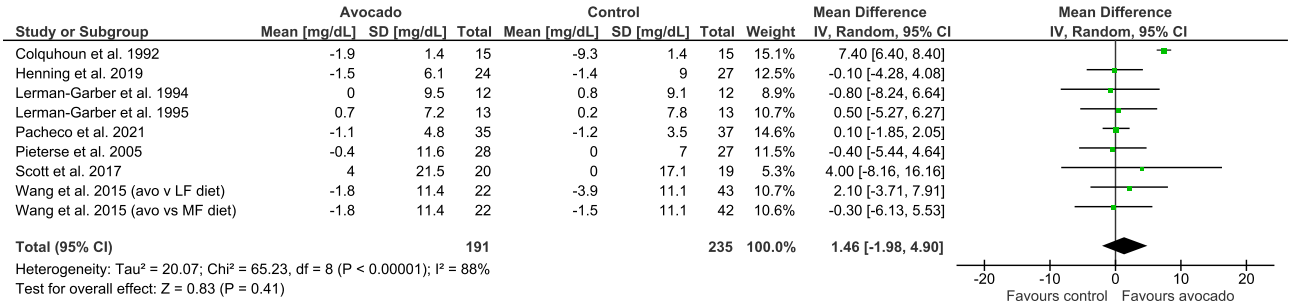

Figure 12. Forest plot of mean (95% CI) difference in HDL-C (mg/dL) between avocado and control groups.
Avo, avocado diet, LF, low-fat, MF moderate fat

Kahn et al., whose results could not be included in the meta-analysis, reported no significant effects on HDL-C by intervention group (or time) [23, 45].

Meta-regression was performed to examine the study-level relationship between avocado intake and HDL-C (Figure 13, Figure 14). Dose ranged between 99 – 330 g. When all studies (n=8 studies, 9 comparisons) were included a significant relationship between avocado dose and HDL-C was observed (regression coefficient 0.034, 95% CI 0.025, -0.042, p = <0.00001) (Figure 13). Given the high relative dose (330 g per day) used in the Colquhoun et al. study combined with high risk of bias, meta-regression was also performed with this study excluded (Figure 14) which resulted in a loss of the significant relationship (regression coefficient -0.002, 95% CI -0.048, 0.044, p = 0.93).

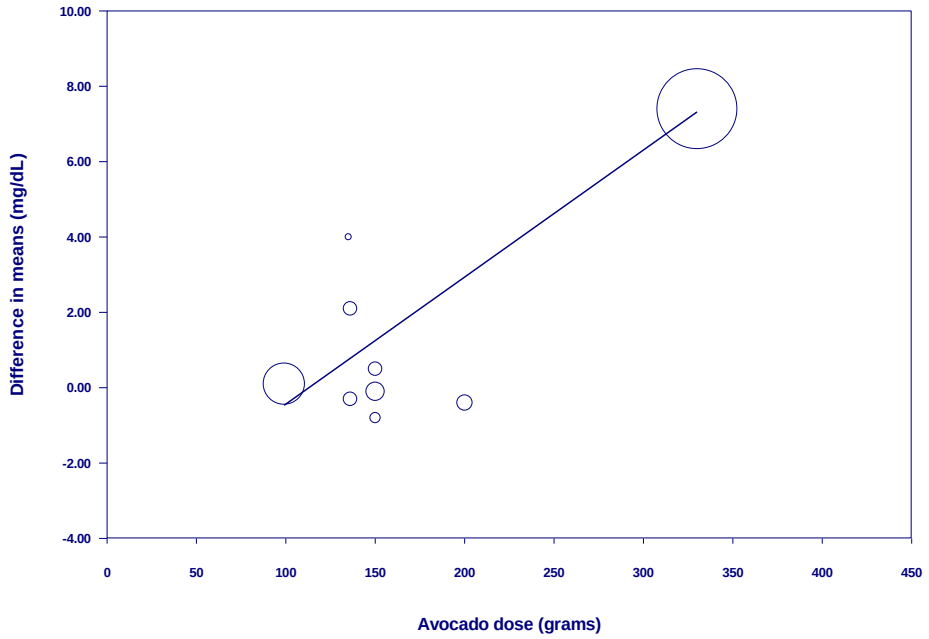

Figure 13. Mean difference in HDL-C (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 8 studies, 9 comparisons).

The size of the circle is proportional to the study weighting.

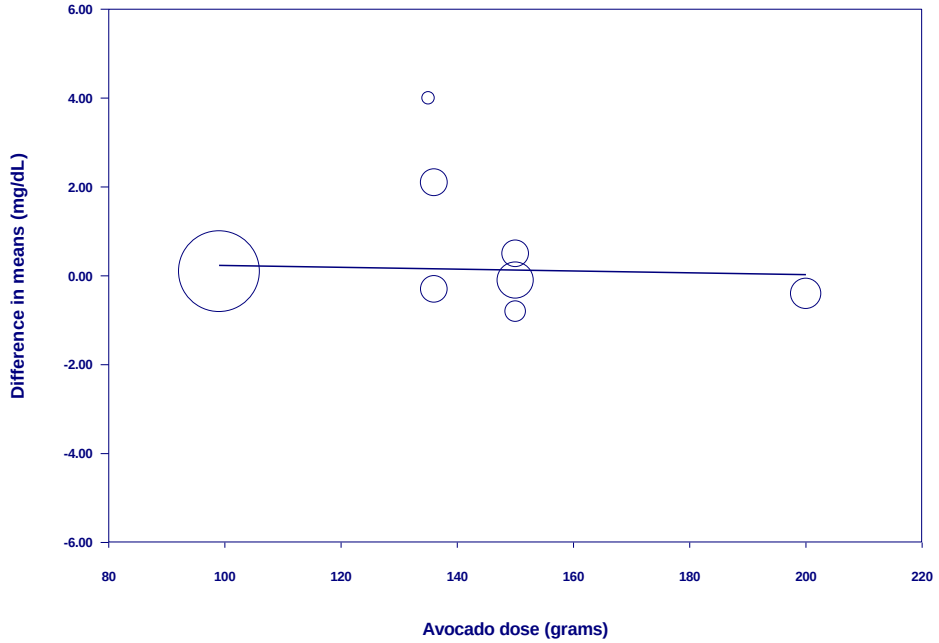

Figure 14. Mean difference in HDL-C (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 7 studies, 8 comparisons) with Colquhoun et al. excluded.

2.2.4 Total cholesterol to High-density lipoprotein-cholesterol ratio

Two studies reported TC to HDL-C ratio [32, 36], both of which were included for meta-analysis (59 participants in the avocado group and 101 participants in the control group). Both were conducted in a hypercholesterolaemic populations. There was a small, significant decrease in total cholesterol to HDL-C ratio (-0.48 (-0.76, -0.20), p = 0.0008) in avocado vs. control groups (Figure 15). Heterogeneity was moderate (p = 0.13, I^2 = 51%).

Due to the minimum number of studies able to be included for this outcome no subgroup or sensitivity analysis were conducted.

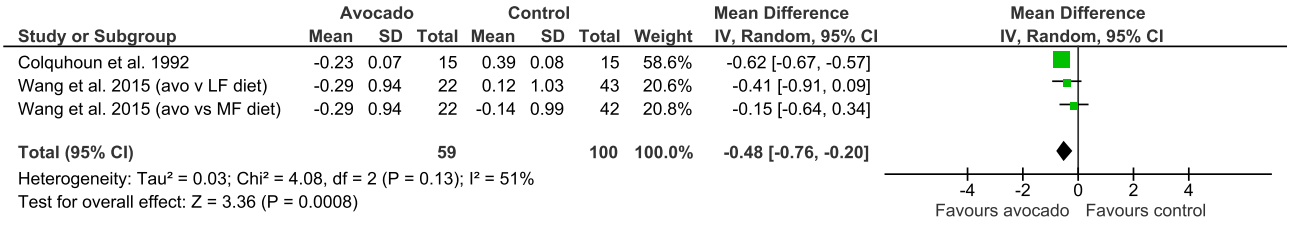

Figure 15. Forest plot of mean (95% CI) difference in total cholesterol to HDL-C ratio between avocado and control groups.

Avo, avocado diet, LF, low-fat, MF moderate fat

2.2.5 Triglycerides

Nine studies reported triglycerides as an outcome and 8 were included for meta-analysis (191 participants in the avocado group and 235 in the control group). Two of these studies specifically recruited a population with hypertriglyceridemia and T2DM [33, 34], while one study reported elevated mean triglyceride concentrations (>150 mg/dL [6]) in the intervention group but not the control group [43]. When all studies were pooled, no significant difference was observed between the avocado and control groups (-1.90 mg/dL (-5.00, 1.21 mg/dL), p = 0.23) (Figure 16). Heterogeneity was low (p = 0.52, I^2 = 0%). When, subgroup analysis was conducted, comparing subgroups with mean baseline triglycerides >150 mg/dL vs. <150 mg/dL [33, 34, 43], female only vs. mixed sex studies, no significant changes in result were seen.

Sensitivity analysis showed a significant reduction in triglycerides in the avocado vs. control group when the Colquhoun et al. study, which was significantly weighted (92.2%) in the meta-analysis, was removed (-13.65 mg/dL (-24.75, -2.56 mg/dL) p = 0.02), (Chi^2 = 2.45, p = 0.93, I^2 = 0%.)

The additional study not included for meta-analysis, Kahn et al., reported no significant effects on triglycerides by group (or time) [23, 45].

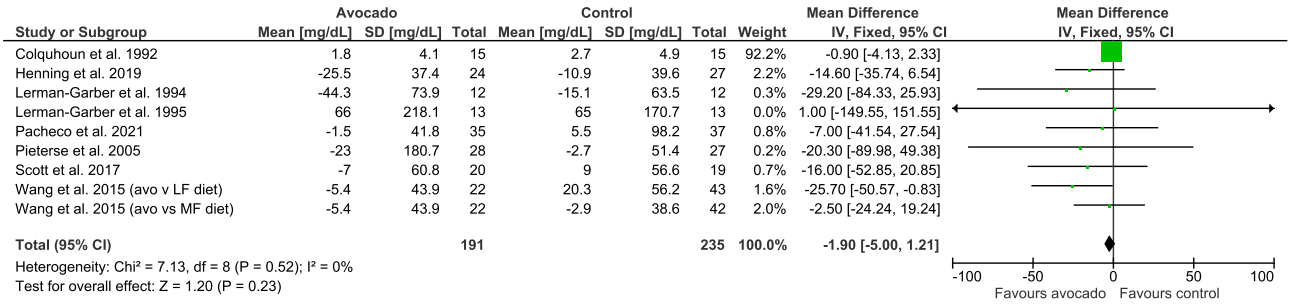

Figure 16. Forest plot of mean (95% CI) difference in triglycerides (mg/dL) between avocado and control groups.

Avo, avocado diet, LF, low-fat, MF moderate fat

Meta-regression was performed to examine the study-level relationship between avocado intake and triglycerides (Figure 17). Dose ranged between 99 g to 330 g. When all studies (n=8 studies, 9 comparisons) were included there was a significant relationship between avocado dose and triglycerides was observed (i.e. triglycerides increased as avocado dose increased) (regression coefficient 0.062, 95% CI 0.001, 0.122, p = 0.046) (Figure 17). Given the high relative dose (330 g per day) used in the Colquhoun et al. study combined with high risk of bias, meta-regression was also performed with this study excluded (Figure 18) which resulted in a loss of the significant relationship (regression coefficient -0.161, 95% CI -0.808, 0.486, p = 0.63).

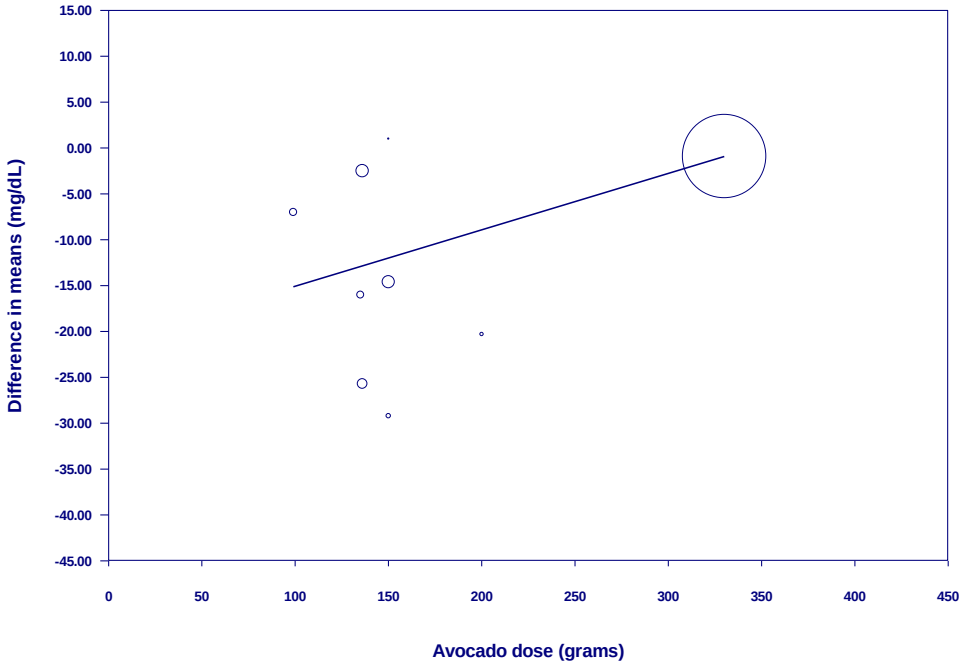

Figure 17. Mean difference in triglycerides (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 8 studies, 9 comparisons).

The size of the circle is proportional to the study weighting.

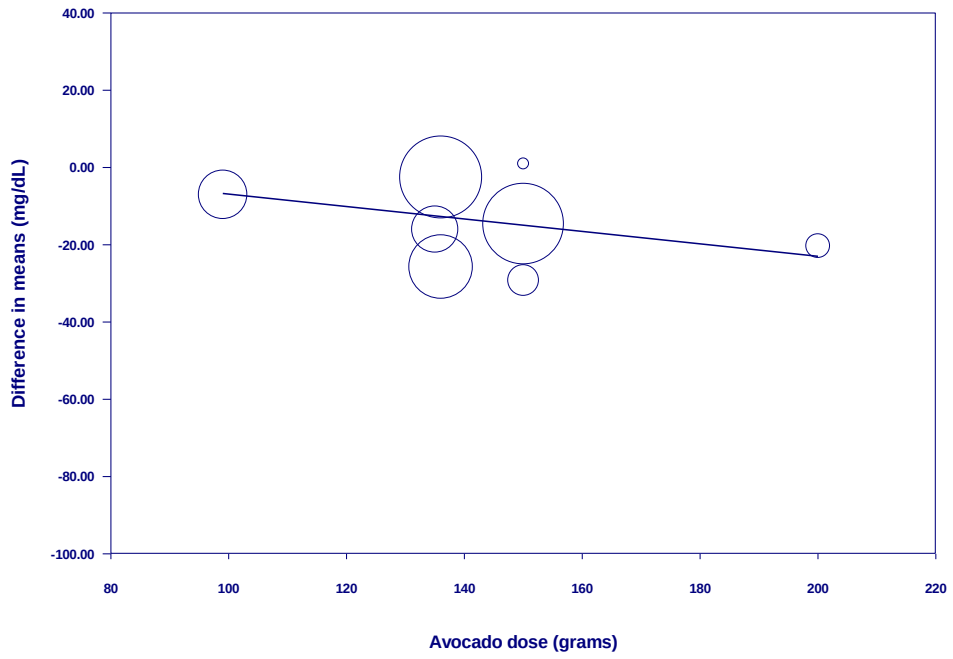

Figure 18. Mean difference in triglycerides (md/dL) between avocado and control groups by avocado dose (grams per day) (n = 7 studies, 8 comparisons) with Colquhoun et al. excluded.

2.2.6 Other blood lipid markers (non-HDL-C, apolipoprotein B)

One study reported non-HDL-C [36] and two studies reported apolipoprotein B [32, 36]. Wang et al. 2015 showed a reduction in non-HDL-C following the avocado intervention, which was significantly greater than was elicited by both the low-fat and moderate-fat (from high oleic oils) control interventions. Wang et al. also reported a reduction in apolipoprotein B that was significantly different from the low-fat diet, but not the moderate-fat diet. Colquhoun showed a reduction in apolipoprotein B in the avocado group during the intervention, however no comparison was made to the control group.

2.2.7 Body mass index

Four studies reported on BMI as an outcome and three were included for meta-analysis (total of 87 participants in the intervention group and 91 participants in the control group). Two studies assessed avocados in the context of weight loss diets [21, 43], and one used an *ad libitum* diet [24]. There was no difference in BMI observed (0.08 kg/m^2 (-0.17, 0.32 kg/m^2) p = 0.52) (Figure 19). Heterogeneity was low (p = 0.74, I^2 = 0%) and no difference was observed when the non-weight loss study was removed.

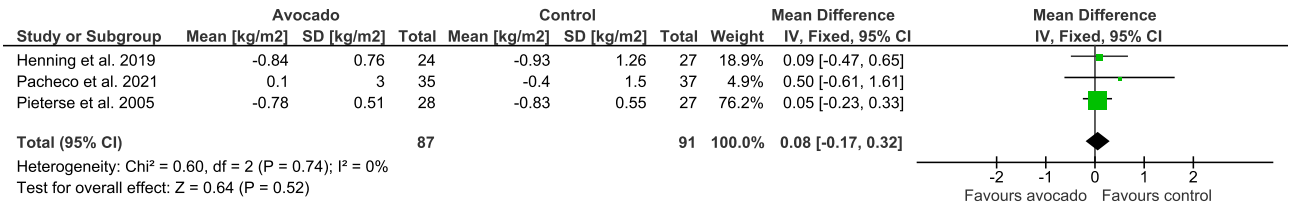

Figure 19. Forest plot of mean (95% CI) difference in BMI (kg/m^2) between avocado and control groups.

The prospective cohort study by Heskey et al. found that among individuals with a normal BMI at baseline, the weight and BMI of avocado consumers increased at a lower rate than non-consumers. For high avocado consumers (intake ≥32 g of avocado/day) this translated to a 0.26% weight increase over 5 years compared with 0.79% for non-consumers. There was no significant effect of avocado on weight or BMI for adults within the cohort who were overweight or obese at baseline. Both low and high consumption of avocados resulted in a reduced risk of becoming overweight and obese, in those people of normal weight at baseline compared to no avocado consumption. The adjusted odds ratio was 0.89 (0.82, 0.96) for adults with low consumption (>0 g and <32 grams per day of avocado) and 0.61 (0.44, 0.85) for those with high consumption, representing an 11% and 39% reduced risk respectively. This result was attenuated when adjusting for baseline BMI. When the results were stratified by age, high avocado consumers aged ≥60 years had a lower reduction in weight change than non-consumers (% change in weight -1.59 for high avocado intake vs. -1.97 for non-consumers).

2.2.8 Body weight

Five studies [21, 23, 33, 34, 43] (52 participants in the avocado group and 54 participants in the control group) reported body weight, two of which were weight loss studies [21, 43] and both of which were included for meta-analysis [21, 43]. There was no significant change in body weight observed (0.46 kg (-0.37, 1.29 kg) p = 0.28) (Figure 20). Heterogeneity was low (p = 0.82, I^2 = 0%).

Due to the minimum number of studies able to be included for this outcome no subgroup or sensitivity analysis were conducted.

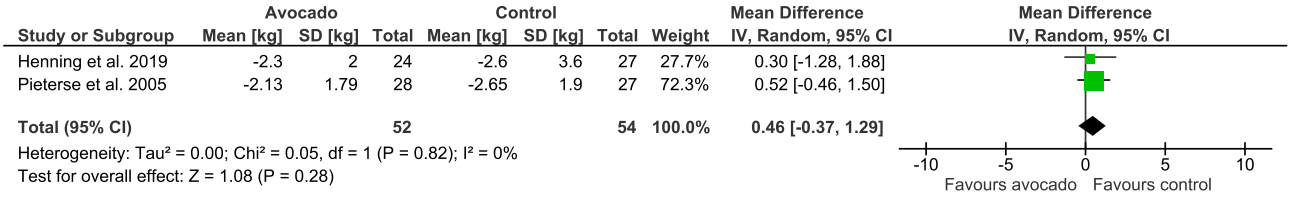

Figure 20. Forest plot of mean (95% CI) difference in body weight (kg) between avocado and control groups.

The remaining studies, which were not included in meta-analysis, did not have weight loss as an intended outcome (i.e. they used a eucaloric energy prescription) [23, 33, 34], however these studies did not report change in weight for the intervention and control groups.

2.2.9 Body fat

Three studies [21, 23, 43] reported on body fat, two of which used Dual-energy X-ray absorptiometry (DXA) [21, 23] and one used estimated body fat equations [43]. Two of these studies were included for meta-analysis [21, 43] (52 participants in the avocado group and 54 participants in the control group) and both were weight loss studies. There was a very small reduction in body fat favouring the control intervention relative to the avocado intervention (0.28%, (0.00, 0.57%), p = 0.05) (Figure 21). Heterogeneity was low (p = 0.88, I^2 = 0%).

Due to the minimum number of studies able to be included for this outcome no subgroup or sensitivity analysis were conducted.

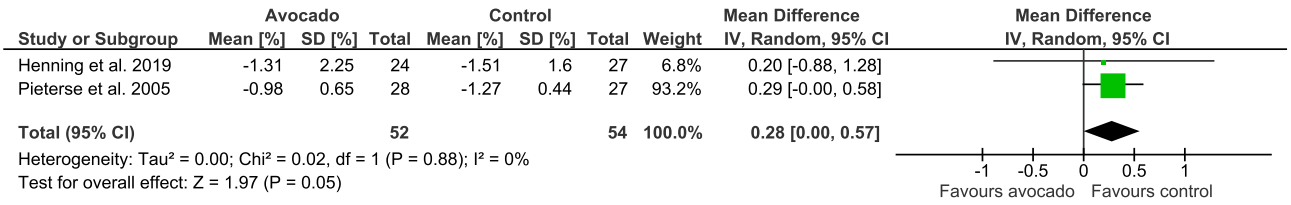

Figure 21. Forest plot of mean (95% CI) difference in body fat (%) between avocado and control groups.

The additional study which measured body fat [23] did not report change in weight for the intervention and control groups.

2.2.10 Visceral adipose tissue

Two studies [21, 23] (76 participants in the avocado group and 80 in the control group) reported on visceral adipose tissue and both were included for meta-analysis. When the studies were pooled, there was no significant change in visceral adipose tissue (-12.32 g (-49.02, 24.38 g), p = 0.51) (Figure 22). Heterogeneity was low (p = 0.32, I^2 = 13%).

Due to the minimum number of studies able to be included for this outcome no subgroup or sensitivity analysis were conducted.

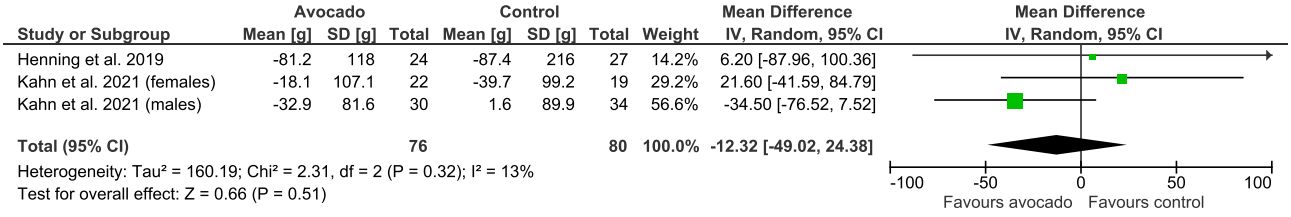

Figure 22. Forest plot of mean (95% CI) difference in visceral adipose tissue (g) between avocado and control groups.

2.2.11 Waist circumference

Only one study reported waist circumference as an outcome measure [24]. No significant difference was reported between the high avocado group vs. the low avocado group (p = 0.85).

2.2.12 Blood glucose levels

Five studies reported fasting blood glucose levels [21, 23, 24, 33, 36], one of which was in participants with T2DM [33]. Two studies were included for meta-analysis [21, 24] (59 participants in the avocado group and 64 participants in the control group). When the studies were pooled, there was no significant difference in blood glucose levels (3.45 mg/dL (-5.69, 12.59 mg/dL) p = 0.46) between the avocado group and control group (Figure 23). Heterogeneity was moderate (p = 0.10, I^2 = 62%).

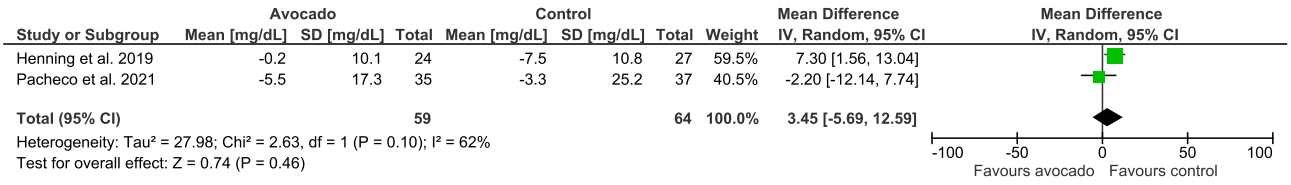

Figure 23. Forest plot of mean (95% CI) difference in blood glucose levels (mg/dL) between avocado and control groups.

The three additional studies which measured fasting blood glucose levels [23, 33, 36] did not report change in glucose for the intervention or control groups. Wang et al. reported no significant difference in glucose between the avocado group and either of the control groups, the low fat and moderate fat diets.

2.2.13 Insulin

Three studies reported plasma fasting insulin [23, 24, 36] however only one of these reported change by treatment group [24], with a small reduction (-0.6 mg/dL (11.8 mg/dL) in the avocado group and a small increase (1 mg/dL (5.85 mg/dL) in the control group. Wang et al. reported no significant difference in the avocado group compared to the control groups, the low and moderate fat diets.

2.2.14 HbA1C

Two studies reported HbA1c [24, 33], however only one had the required data (between group change) for meta-analysis [24]. Pacheco et al. showed no significant difference in HbA1c between the high and low avocado interventions (0.09% (0.98%) p = 0.4).

2.2.15 HOMA-IR

Three studies reported HOMA-IR [23, 24, 36], an indicator of insulin resistance. Two were included for meta-analysis [24, 36] (87 participants in the avocado group and 90 in the control group). When the studies were pooled, there was no significant difference in HOMA-IR (0.29 (-0.05, 0.63) p = 0.10) (Figure 24). Heterogeneity was low (p = 0.50, I^2 = 0%).

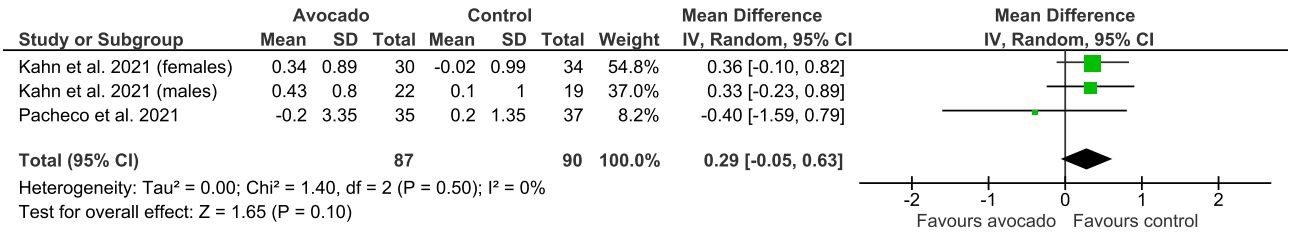

Figure 24. Forest plot of mean (95% CI) difference in HOMA-IR between avocado and control groups.

The additional study not included for meta-analysis [36], reported no significant difference between the avocado group and the control groups, the low and moderate fat diets.

2.2.16 Blood pressure

Three studies reported blood pressure [24, 36, 43], two of which were included for meta-analysis [24, 43] (63 participants in the intervention group and 64 in the control group). When the studies were pooled, there was no significant difference in systolic blood pressure (3.50 mmHg (-1.82, 8.81 mmHg) p = 0.26) (Figure 25) or diastolic blood pressure (2.48 mmHg (-1.32, 6.27 mmHg) p = 0.20) (Figure 26). Heterogeneity was low for both systolic (p = 0.26, I^2 = 21%) and diastolic (p = 0.22, I^2 = 32%) blood pressure.

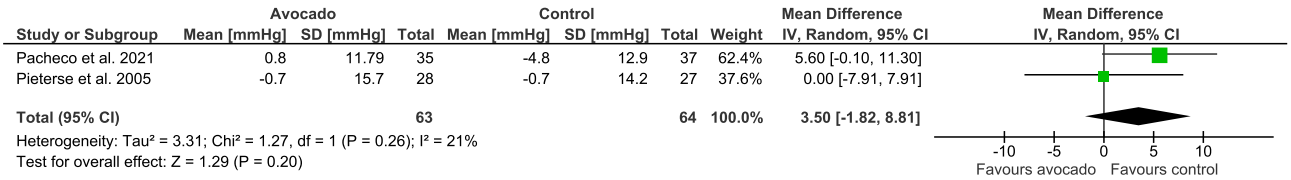

Figure 25. Forest plot of mean (95% CI) difference in systolic blood pressure between avocado and control groups.

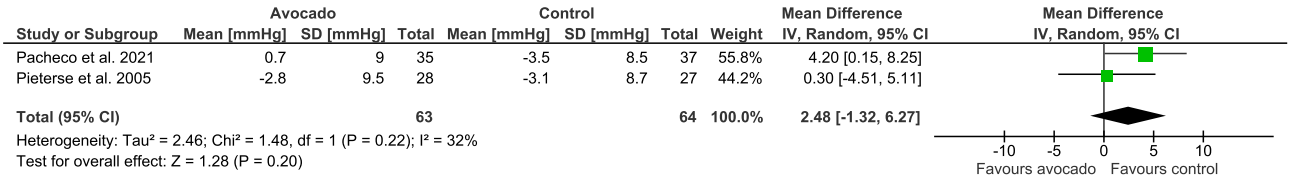

Figure 26. Forest plot of mean (95% CI) difference in diastolic blood pressure between avocado and control groups.

The additional study, not included for meta-analysis [36] reported no significant difference between the avocado group and the control groups, the low, or moderate fat diets. None of the studies measuring blood pressure specifically recruited hypertensive participants.

2.2.17 Oxidative stress markers

One study [46] reported on oxidised LDL, a marker of oxidative stress. The avocado diet significantly decreased circulating oxidised LDL compared to the two control diets, the low-fat diet ($p = 0.03$) and the moderate-fat diet (from high oleic oils) ($p = 0.05$).

2.2.18 Cardiovascular disease risk

One study [36] reported predicted change in cardiovascular disease risk using the 2013 Prevention Guidelines Atherosclerotic Cardiovascular Disease Risk Estimator which considers participant's age, sex, race, smoking status, diabetes status, hypertension treatment, blood pressure, total cholesterol, and HDL-C. A significant improvement for overall 10 year and lifetime risk was seen in the avocado group compared to the Baseline Average American Diet but no comparisons with the moderate fat or low fat diet (the comparisons of interest for the present study) were made.

2.2.19 Adverse effects

In those studies which reported on adverse effects [21, 24, 33-35], no adverse effects were reported.

2.2.20 Publication bias- funnel plots

Funnel plots were examined for risk of publication bias (Figure 27). As the number of studies were below the recommended ≥10 (n=8) for conducting a funnel plot, these results should be interpreted with caution. The small number of studies and the fact that all studies were small in size probably explain the lack of appearance of an inverse funnel (where larger studies that generally have smaller SEs are gathered at the top of the funnel and smaller studies that generally have larger SEs spread at the bottom). Studies were generally spread on either side of the mean effect, and this was true for those studies reporting smaller and larger SEs, therefore publication bias was not suspected, although it cannot be ruled out.

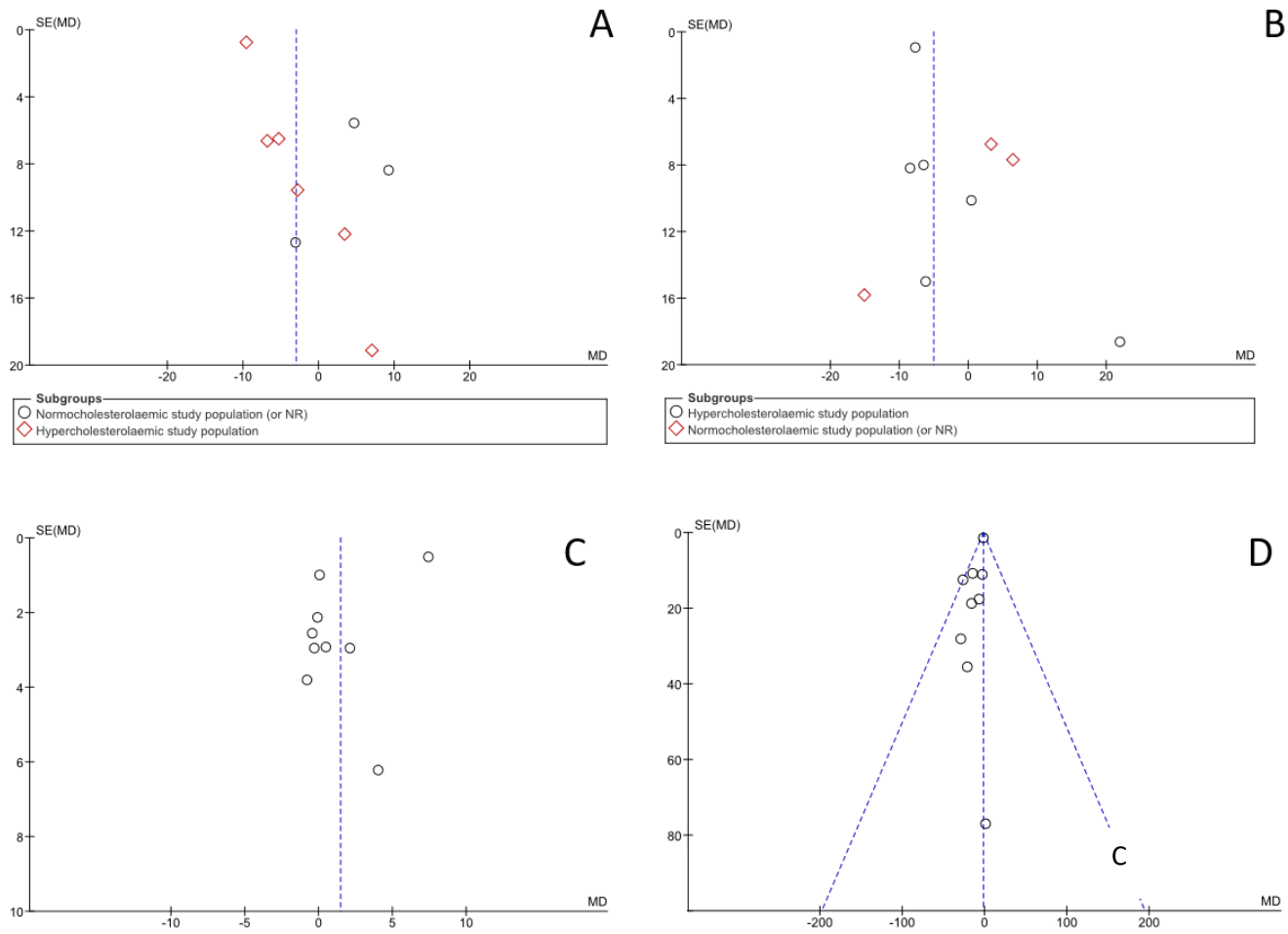

Figure 27. Funnel plot for (A) LDL-C (mg/dL); (B) TC (mg/dL); (C) HDL-C (mg/dL); (D) triglycerides (mg/dL).

SE, standard error; MD, mean difference between avocado and control groups.

2.2.21 Quality of the total body of evidence

Table 4 summarises the process of rating the quality of the body of evidence according to the GRADE guidelines and provides a rating that reflects the degree of certainty in the relationship between avocado intake and blood lipids. The degree of certainty was rated for each blood lipid outcome in mixed populations (all studies pooled) and for LDL-C and TC, stratified according to studies conducted in hyper- and normocholesterolaemic populations. The degree of certainty that daily intake of avocado compared to no or low avocado intake reduced TC and has no effect on LDL-C, HDL-C or triglycerides in a mixed population of healthy, overweight, obese, T2DM, normo- and dyslipidaemic (including hypercholesterolaemic and triglyceridaemic) adults (all studies pooled) were rated as very low meaning that we have very little confidence in the effect estimate: the true effect is likely to be substantially different from the estimate of effect. The certainty was downgraded because of risk of bias, inconsistency of study results and imprecision of the results (wide confidence intervals which crossed zero in most studies).

In the subgroup of studies in adults with hypercholesterolaemia, the degree of certainty that daily intake of avocado compared to no or low avocado intake reduces LDL-C or TC were rated as very low, meaning we have very little confidence in the effect estimate: the true effect is likely to be substantially different from the estimate of effect. Certainty was downgraded due to risk of bias, imprecision and indirectness.

The degree of certainty that in the subgroup of generally healthy normocholesterolaemic adults, daily intake of avocado compared to no or low avocado intake has no effect on LDL-C and TC, was rated as moderate, meaning that we are moderately confident in the effect estimate: the true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different. The certainty was downgraded due to risk of bias. It was only possible to conduct GRADE assessments of the evidence for lipids, due to the low number of studies reporting on outcomes other than lipids. For body weight related outcomes (body weight and BMI) there was emerging evidence, meaning a small number of studies with consistent findings. For all other outcomes relating to body composition and other cardiometabolic health markers, the evidence was limited due to the small number of studies and inconsistent findings. Further well-designed studies would be required to strengthen the evidence for these outcomes.

Table 4. Quality of the body of evidence according to the GRADE guidelines

Evidence statement	Outcomes	Effect estimate Mean Difference (95% CI) (mg/dL)	№ of participants (studies)	Certainty of the evidence (GRADE)
Daily intake of avocado compared to no or low avocado intake does not affect LDL cholesterol in a mixed population of healthy, overweight, obese, diabetic, normo- and dyslipidaemic adults	LDL-C	**3.02 lower** (8.83 lower to 2.79 higher)	426 (9 RCTs)	⨁◯◯◯ Very low[b,c,d,g,h]
Daily intake of avocado compared to no or low avocado intake reduces total cholesterol in a mixed population of healthy, overweight, obese, diabetic, normo- and dyslipidaemic adults	Total cholesterol	**5.08 lower** (9.29 lower to 0.87 lower)	426 (9 RCTs)	⨁◯◯◯ Very low[a,b,c,d]
Daily intake of avocado compared to no or low avocado intake does not affect HDL cholesterol in a mixed population of healthy, overweight, obese, diabetic, normolipidemic and dyslipidaemic adults	HDL-C	**1.46 higher** (1.98 lower to 4.9 higher)	426 (9 RCTs)	⨁◯◯◯ Very low[b,c,g,i]
Daily intake of avocado compared to no or low avocado intake does not affect triglycerides in a mixed population of healthy, overweight, obese, diabetic, normolipidemic and dyslipidaemic adults	Triglycerides	**1.9 lower** (5.0 lower to 1.21 higher)	426 (9 RCTs)	⨁⨁◯◯ Low[a,c,d]
Daily intake of avocado compared to no or low avocado intake reduces LDL cholesterol in a population of adults with hypercholesterolaemia[1] +/- T2DM	LDL-C	**9.4 lower** (10.84 lower to 7.95 lower)	264 (6 RCTs)	⨁◯◯◯ Very low[a,d,e]
Daily intake of avocado compared to no or low avocado intake reduces total cholesterol in a population of adults with hypercholesterolaemia[1] +/- T2DM	Total cholesterol	**7.52 lower** (9.38 lower to 5.66 lower)	273 (6 RCTs)	⨁◯◯◯ Very low[a,d,e]
Daily intake of avocado compared to no or low avocado intake does not affect total cholesterol in a population of generally healthy normocholesterolaemic[1] adults	Total cholesterol	**2.81 higher** (6.6 lower to 12.27 higher)	162 (3 RCTs)	⨁⨁⨁◯ Moderate[f]
Daily intake of avocado compared to no or low avocado intake does not affect LDL cholesterol in a population of generally healthy normocholesterolaemic[1] adults	LDL-C	**5.03 higher** (3.53 lower to 13.59 higher)	162 (3 RCTs)	⨁⨁⨁◯ Moderate[f]
	GRADE Working Group grades of evidence **High certainty:** we are very confident that the true effect lies close to that of the estimate of the effect. **Moderate certainty:** we are moderately confident in the effect estimate: the true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different. **Low certainty:** our confidence in the effect estimate is limited: the true effect may be substantially different from the estimate of the effect.			

	Very low certainty: we have very little confidence in the effect estimate: the true effect is likely to be substantially different from the estimate of effect.

Explanations

a. The result is driven by one study (Colquhoun et al) which has a high risk of bias (risk of bias)
b. Point estimates differed in whether they favoured avocado or control (inconsistency)
c. Comparison diets varied in their composition, often including other elements of dietary change (i.e. improvement) (indirectness)
d. Confidence intervals crossed zero (imprecision)
e. Three of the five studies included in this subgroup were in female only populations and had durations less than 8 weeks (suggested by European Food Safety Authority as an appropriate timeframe to test sustainability of blood lipids) (indirectness)
f. One of the three studies had a high risk of bias and the remaining two studies had some concerns (risk of bias)
g. Two of the eight studies were rated as high risk of bias with the remaining six studies having some concerns (risk of bias)
h. Heterogeneity was moderate as indicated by I^2 value (inconsistency)
i. Heterogeneity was high as indicated by the I^2 value and P <0.001 (inconsistency)

GRADE, Grading of Recommendations Assessment, Development and Evaluation, CI, confidence interval; MD, mean difference, [1]Hypercholesterolaemia was defined as baseline LDL-C >115 mg/dL and normocholesterolaemia as baseline LDL-C <115 mg/dL (or not reported) as per European Society of Cardiology (ESC) and the European Atherosclerosis Society (EAS) guidelines for the management of dyslipidaemia [6].

3 Discussion

Blood lipids

The main findings of this study were that when combining all studies conducted in mixed populations (including healthy, overweight, obese, T2DM, normo- and dyslipidaemic adults) daily intake of avocado compared to no or low avocado intake did not affect LDL-C, HDL-C or triglycerides, however a small reduction in TC was observed (-5.08 mg/dL (-9.29, -0.87 mg/dL) p = 0.02). The reduction in TC was driven by the studies conducted in participants with hypercholesterolaemia, as shown by subgroup analyses. A similar result was observed for adults with hypercholesterolaemia for LDL-C, with avocado intake resulting in a significant reduction in LDL-C (-9.4 mg/dL [95%CI -10.84, -7.95 mg/dL]) compared to control diets. However, due to various limitations in the current evidence, the degree of certainty in the above findings were rated as very low. This means our confidence in the effect estimate is currently limited and the true effect may substantially differ when more studies of better quality become available in future.

The lack of effect on LDL-C and triglycerides when all studies were combined, was in agreement with the most recent meta-analysis by Mahmassani et al. However our results differed for HDL-C, with Mahmassani et al. reporting a significant improvement (i.e. increase) in HDL-C while the present review showed no effect. These results were also in contrast with the meta-analyses performed by Peou et al., who showed a significant decrease in TC, LDL-C and TG [19]. However as previously noted, the conclusions from the Peou et al. meta-analysis are biased as they only considered the change from baseline to end within the avocado group while not accounting for the control group. A control group is critical to the randomised controlled trial design, helping to ensure that any changes observed are in fact the result of the intervention and not a result of chance or other factors which may have changed during the study period.

Mahmassani et al. conducted subgroup analysis based on cardiometabolic risk at baseline and reported no differences in results between the main and subgroup analyses. Whereas the present meta-analysis showed significant differences in LDL-C and TC between studies conducted on hyper- vs. normocholesterolaemic populations as defined by using a clinically relevant cut off for LDL-C [6] at baseline. Studies in hypercholesterolaemic populations showed significantly reduced LDL-C and TC with avocado intake vs. control. While studies in normocholesterolaemic populations showed no effect of avocado consumption vs. control diet on LDL-C and TC. This is consistent with the notion that it is generally more difficult to show a significant improvement in a biomarker that already falls within normal reference ranges. The difference in LDL-C of -9.4 mg/dL [95%CI -10.84, -7.95 mg/dL] between avocado and control interventions in the hypercholesterolaemic subgroup is of modest clinical importance. A reduction of LDL-C by 38.6 mg/dL (1 mmol/L) was shown to reduce risk of death due to coronary heart disease by 20% [47]. Hence, a reduction of 9.4 mg/dL is estimated to be associated with a reduced risk of death due to coronary heart disease of ~5%. Although this is small, it may still make an important contribution to reducing cardiovascular disease risk when in the context of an overall healthy diet combined with other foods with LDL-C lowering properties.

Subgroup analysis based on sex and study duration were also significantly different with studies in females and ≤8 weeks duration favouring LDL-C and TC in response to avocado intake vs. control. However, as these studies all fell into the hypercholesterolaemic subgroup, it is not possible to differentiate which criteria primarily affected the results. Furthermore, the favourable results on LDL-C and TC from subgroup analysis were largely driven by one study that was clearly an outlier, Colquhoun et al. Sensitivity analysis showed that the favourable effects in subgroup analysis on LDL-C and TC with avocado intake disappeared when

Colquhoun et al. was excluded from the meta-analysis. This study was evaluated to have a high risk of bias. In addition, SDs of lipid outcomes reported by Colquhoun were questioned as they were considerably lower, by a magnitude of ~10x, compared to all other studies (i.e. for LDL-C, 2 mg/dL compared to other studies ranging from 25 to 49 mg/dL). Lower SDs (implying lower variation in the point estimate) in part drives the weighting that a study carries in the meta-analysis and therefore the contribution it makes to the overall meta-analysis results. If these SDs were indeed wrongly reported, it may contribute to biased conclusions.

In the present meta-analysis, HDL-C was not affected by avocado intake compared to control diets. This finding is inconsistent with the meta-analysis by Mahmassani et al. who showed an increase in HDL-C with avocado intake compared to no avocado intake. The conflicting findings likely reflect the slightly different inclusion of studies across the two meta-analyses. Mahmassani et al. included seven studies in their meta-analyses on serum HDL-C, four of which were also included in our meta-analysis [32, 35, 36, 43]. The present review included an additional four studies not included in the Mahmassami review, all showing no effect of avocado intake on HDL-C [21, 24, 33, 34]. While the Mahmassani et al. review included three studies not included in the present meta-analysis [48-50], two of which showed significant increases in HDL-C favouring avocado [48, 49]. Two of these studies did not meet our English-language inclusion criteria [49, 51] while Carranza-Madrigal (2008), did not provide baseline data to allow for within group change to be calculated [48]. It is possible, but not guaranteed, that the inclusion of these studies may change the overall HDL-C result in the current study.

The finding for triglycerides, which showed no significant difference between avocado and control groups when all studies were combined, was in line with the results of the Mahmassani review, however sensitivity analysis showed that removing the study by Colquhoun et al. resulted in a significant reduction in triglycerides that favoured the avocado group vs. control. The lack of effect on triglycerides in the Colquhoun study may have been due to low baseline triglyceride levels (<150 mg/dL). Due to the questionable (see above explanation) high weight the Colquhoun studied carried in the meta-analysis it resulted in an overall non-significant effect. Austin et al. showed that 1 mmol/L (equal to 88.5 mg/dL) increase in triglycerides were associated with a 32% and 76% increase in cardiovascular disease risk in men and women, respectively [52]. This suggest that a decrease of 13.65 mg/dL (-24.75, -2.56 mg/dL) seen in the current meta-analysis (excluding Colquhoun et al) may be clinically meaningful, particularly in women. Factors that may favourable affect triglyceride responses include increased baseline triglyceride levels as well as a low-carbohydrate diet [53]. Sub-group analysis comparing studies on hyper- vs. normotriglyceridaemic populations may not have achieved significance due to a loss of statistical power within sub-groups. Because of large intraindividual variations, triglycerides studies generally require larger study populations [54]. Several of the studies in the current meta-analysis used high-carbohydrate, low-fat diets as control diets [33-36] which may have increased triglycerides relative to the higher-fat, lower-carbohydrate diet caused by incorporating avocado into the diet. This potential triglyceride lowering effect of avocados is worth further investigation within larger studies.

A significant reduction in TC to HDL-C ratio was seen which favoured the avocado group compared to the control group (-0.48 [-0.76, -0.20]). However, this outcome was only reported by 2 studies (n=3 comparisons) [32, 36] which included the high risk of bias Colquhoun study. The effect of avocado consumption was investigated on non-HDL-C in one study [32, 36] and apolipoprotein B in two studies [32, 36], all showing significant reductions in these two variables following avocado consumption. Evidence is accumulating that these emerging biomarkers, TC to HDL-C ratio, non-HDL-C and apolipoprotein B, are stronger predictors of cardiovascular disease than LDL-C [55, 56]. However few studies have included these biomarkers.

There are a number of mechanisms by which avocado intake may reduce LDL-C and TC. This includes the substitution of MUFA/PUFA for SFA in the diet. Only three studies [23, 24, 32] reported the change in nutrient intakes with interventions, all of which confirmed an increase in MUFA and decrease in SFA in the avocado relative to the control group. In addition, avocados are a source of several bioactives that may, in combination, contribute to LDL-C and TC lowering effects, including dietary fibre (containing 4.3 g per 100g [11], containing ~30% soluble fibre [12, 57] and phytosterols [58].

There was some evidence of a favourable dose response relationship between avocado intake and TC, LDL-C and HDL-C, and some evidence of an unfavourable relationship between avocado intake and triglycerides. However, these relationships were only present when the study by Colquhoun et al. was included which provided the highest daily avocado dose, estimated at 330 grams per day. Given the absence of these relationships when this study was excluded and the paucity of evidence at higher doses, these findings need to be interpreted with caution. The dose range used in the intervention was estimated to range between 99 g – 330 grams per day, which does not provide any direct evidence of health impacts that may come from increasing the average serving size from 50 g to 75 g. However there appears to be an absence of any negative effects on blood lipids from daily amounts greatly exceeding 75 g and in those studies which reported information on adverse events, none were reported. Taken together, it seems unlikely that there would be any adverse health outcomes to increasing the recommended serving size from 50 g to 75 g.

The European Food Safety Authority (EFSA), responsible for scientific advice and support related to food regulation in the European Union, recommend studies of 4 weeks in duration to reach stabilisation in lipid outcomes, but to provide evidence on sustainability of the effect, studies should be conducted over longer periods of time (e.g. 8 weeks) [39]. EFSA further recommends that studies in hypercholesterolaemic participants treated with lifestyle measures (e.g. diet, not drugs) can be used for scientific substantiation of dietary interventions on lipid outcomes.

To improve the degree of certainty in the outcomes of the present meta-analysis, larger, longer duration, randomised controlled studies should be conducted. As individuals with hypercholesterolaemia are most likely to benefit from a dietary intervention focused on reducing LDL-C, consideration should be given to test the effects in hypercholesterolaemic populations.

The addition of the results of the recently completed (October 2020), but yet to be published, Habitual Diet and Avocado Trial, a large (n=1008), multicentre randomised controlled trial [59] may change the results of the present meta-analysis and increase the certainty of the overall results.

Body composition

No significant differences were seen between the avocado intervention and control intervention in the majority of body composition outcomes (weight, BMI, VAT or WC). A very small decrease (0.28% (0.00, 0.57%)) in body fat favouring the control group was seen when the two weight loss studies were combined, but this is unlikely to be of any clinical significance. These two studies both had a primary aim to assess weight loss and used avocado in the context of an energy restricted diet [21, 43] and their findings concurred, with similar weight loss achieved in the avocado group compared to the control.

The prospective observational study showed that adult avocado consumers had a slower rate of weight gain compared to non-consumers over a follow up period of 4 to 11 years, although this was only seen in those people with a BMI in the normal weight at baseline, therefore there may be other aspects of diet that differed by weight status which were not adjusted for. This finding is in line with observational studies from Australia and the USA which have both found a lower BMI and body weight in avocado consumers vs. non-consumers [15, 16].

The absence of a significant difference in body composition outcomes is a meaningful finding, indicating that the consumption of avocado, even relatively high amounts, well above the current usual daily consumption, consumed on a daily basis, did not result in unfavourable changes to body composition. Avocados are a nutritionally dense food [60] although may be perceived to contribute to weight gain [61], perhaps related to the historic and now outdated notion that foods rich in fat should be avoided. However, the present review demonstrates that across both energy-restricted (i.e. weight loss) diets and uncontrolled *ad libitum* diets, there was no weight gain effect seen. Only three [21, 23, 24] of the six studies exploring weight related outcomes (body weight or BMI) were of durations of 12 weeks or more, commonly considered to be a minimum intervention length for these outcomes [62]. The evidence should be considered emerging, with further research required to strengthen the evidence base, however these findings do support the absence of any negative health effects from avocado doses ranging between 99 – 200 g, which is in excess of the proposed revised serving size of 75 g per day.

Interestingly, the prospective cohort study showed age related differences in weight change with avocado consumption, with a protective effect against weight loss observed in older adults. Unintentional age-related weight decline has many negative health outcomes including loss of muscle mass and associated function [63]. It is possible that the nutrient and energy density of avocado could be a contributing factor to reducing age-related weight loss, however this would require further evidence from studies in an at-risk population. It is also possible that avocado intake is a marker of overall diet quality.

Other markers of cardiometabolic health

Very few (<4) studies assessed any of the remaining outcomes of interest: blood glucose levels, insulin, HbA1c, insulin resistance, blood pressure, oxidised-LDL and CVD risk. Where these outcomes were measured, effects observed were small and inconsistent in direction. The limited body of evidence combined with the inconsistency of findings does not allow for any conclusions to be made on the effect of avocados on these outcomes.

Strengths and limitations

There were a number of strengths to the present study. This includes the rigorous methodological approach to the review which used a comprehensive search strategy, a duplicate screening process, data checking by a second reviewer, quality assessment of studies conducted in duplicate and overall grading of the quality of evidence, where possible.

Reported compliance with the interventions was generally high and the dose of avocado used across the intervention studies was relatively high, between 99 - 330 g of avocado per day (~0.75 - 2.2 average whole avocados), meaning any null results are not likely to be a result an insufficient dose to produce an effect. However, given the substantial gap between current population intakes (~16 g per day in consumers who comprised only 16% of Australian adults) and the dose of avocados used in the intervention studies, a significant increase in consumption would likely be required to bring about health benefits such as a TC or LDL-C reduction. Furthermore, the high dose of avocado used in the interventions limited the ability to assess the impact of lower doses of avocado that align with the current industry recommended serving size (50 g) or proposed revised serving size (75g). These amounts may be more realistic targets for regular consumption.

Other limitations included the size of the studies. All the studies were small (<40 participants per arm). We were unable to include some studies published in languages other than English. We cannot be certain about the impact on the findings if these studies were included in the meta-analyses, however the grading of the evidence as low to very low certainty for most of the findings related to lipids outcomes indicates

that the addition of more studies may change the overall affect, and this is observed in the different findings for HDL-C between the recent review by Mahmassani et al. and the present review.

There was substantial heterogeneity across studies because of the clinical diversity of study populations, e.g. sex of participants, baseline lipid status, weight status, presence or absence of co-conditions such as T2DM. In some respects, this is a strength, making the results more generalisable to the population at large, however it introduces a large amount of variability in responses to the intervention (particularly in the context of small sample sizes), leading to inconsistency and imprecision of results across studies.

Diversity in study design was also high, including variation in the composition of control diets, study design and study duration. Not all studies provided the avocado dose used in the intervention as a specific weight. For the meta-regression to be conducted, weights were estimated for these studies [21, 24, 32-34] based on several assumptions previously described which may not be accurate.

The present review included a range of comparator diets including those lower in MUFA than the avocado intervention diet (e.g. a high carbohydrate diet), those which were comparable in MUFA to the avocado intervention but from an alternate source (e.g. high oleic oils), and those where avocado was added to a habitual diet with no macronutrient targets prescribed. The difference in comparator diet is likely to impact the between group change for some outcomes, such as in the case of triglycerides. Due to the limited number of studies for each comparator, and infrequent reporting of MUFA content achieved in the diets, it was not possible to do subgroup analysis by comparator diet.

Quality assessment revealed that all studies were considered to have at least one flaw which increased their risk of bias, with three studies being considered to have a high risk of bias. This included the study by Colquhoun et al. which was considered particularly problematic as this study was often responsible for singularly influencing the overall result of the meta-analyses and meta-regression as shown by sensitivity analysis. The relatively small number of studies reporting on outcomes other than blood lipids, has limited the ability to draw any conclusions about the effects of avocados on these risk factors for cardiometabolic health.

To better understand the effects of avocados on cardiometabolic health there is a need for more well-designed studies. These studies should give careful consideration to the health attributes of the study population given that the effect may be different for those at a higher level of risk such as those with elevated blood lipids at baseline.

Part II Scoping review of avocados and other health outcomes

4 Introduction

To date, avocados have most commonly been studied for their role in cardiovascular health; the focus of Part I. Given the broad range of nutrients and bioactive substances found in avocados, it is logical to consider that health benefits may extend beyond cardiometabolic health outcomes and recent research has expanded to explore other possible health benefits of avocado consumption.

The purpose of this scoping review was to provide an overview of avocado trials and observational studies focusing on outcomes other than risk factors for cardiometabolic health.

5 Methods

5.1 Aim

A scoping review was conducted to examine the effects of avocado intake on health outcomes other than cardiometabolic health. The effects of consuming diets containing avocado compared to control diets containing no avocado, lower avocado, or an alternative dietary fat source (MUFA or other source), were investigated in adults who were healthy or at increased risk or diagnosed with CVD or type 2 diabetes. Where applicable, the scoping review was conducted and reported in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) criteria [26].

5.2 Data sources and study eligibility

5.2.1 Literature search

A comprehensive literature search was conducted on 25 November 2021 across four scientific journal databases (PubMed, Web of Science (core collection), Scopus and ProQuest) combining terms that included the exposure (avocado) and the outcomes of interest (terms related to health effects other than cardiometabolic health, see Table 5. The search included original literature published from January 1990 through October 2021. In addition, a search of Google Scholar retrieving the first 400 results was conducted to ensure that relevant studies were captured. Appendix C provides the search strategy.

5.2.2 Study inclusion and exclusion criteria

Studies published from 1990 onwards and available in English were included if they met the inclusion and exclusion criteria summarised in Table 5.

5.2.3 Study selection

All duplicate publications were removed. Relevant studies were identified by one reviewer (GJM). Covidence software was used [28] to screen titles and abstracts followed by review of the full-text publications. Any uncertainties were resolved by consensus with a second reviewer (PB).

Table 5. Inclusion and exclusion criteria for scoping review of avocado and general health effects

Population	Adults (aged ≥18 years) who were healthy (including those who were overweight and obese) or had metabolic syndrome, an increased risk of, or a diagnosis of CVD or type 2 diabetes
Intervention/Exposure	Avocado-enriched diet which uses the avocado fruit (i.e. flesh)
Comparison	No avocado intake (i.e. usual diet), low avocado intake (defined as an amount of ≤50% of the intervention dose) or contained an alternative dietary fat source (MUFA or otherwise)
Outcomes	Changes in the gut microbiome, faecal metabolites e.g. SCFA Measures of cognitive functions e.g. memory, attention, executive function Health effects (to broadly capture anything else e.g. eye health, inflammation, bone health, reproductive health)
Study design	Intervention studies (i.e. parallel and cross-over RCTs, non-randomised controlled trials) ≥3 weeks Prospective cohort studies Cross-sectional studies
Other inclusion criteria	English language Jan 1990 - Jan 2022
Exclusion criteria	Animal studies *In vitro* studies Acute studies Studies in children Studies in adults with disease states other than those specified in the inclusion criteria Studies using components of avocado other than flesh (e.g. avocado oil, avocado extract, avocado seed) Other observational study designs (e.g. case-control)

Abbreviations: CVD, cardiovascular disease; MUFA, monounsaturated fatty acid; SCFA, short-chain fatty acids; RCT, randomised controlled trial

5.3 Data extraction

Data from each study were extracted by one investigator (PB). The following data were extracted: first author's family name; year of publication; geographic location; primary objective; study design (duration, randomisation procedures, participant information (sample size, sex distribution, mean age, mean body mass index (BMI)); potential confounding factors that were not controlled for were identified; details regarding intervention/exposure and control treatments; mean dietary intakes (energy, macronutrients); methods used to assess outcomes; outcome results; and the conclusions reported by authors.

For each outcome reported in experimental studies, the mean and standard deviation (SD) at baseline, end of the intervention, and change were extracted for the intervention and control arms. When multiple time points were reported only the end of intervention point was used. For the cross-sectional study, the mean and standard deviation (SD) of each variable were extracted for avocado-consumer and non-consumer groups.

6 Results

Figure 28 outlines the number of studies that were assessed at each stage of the screening process. A total of six publications, reporting on five unique studies (or cohorts), were included in the review. Five studies were experimental trials and one was a cross-sectional study.

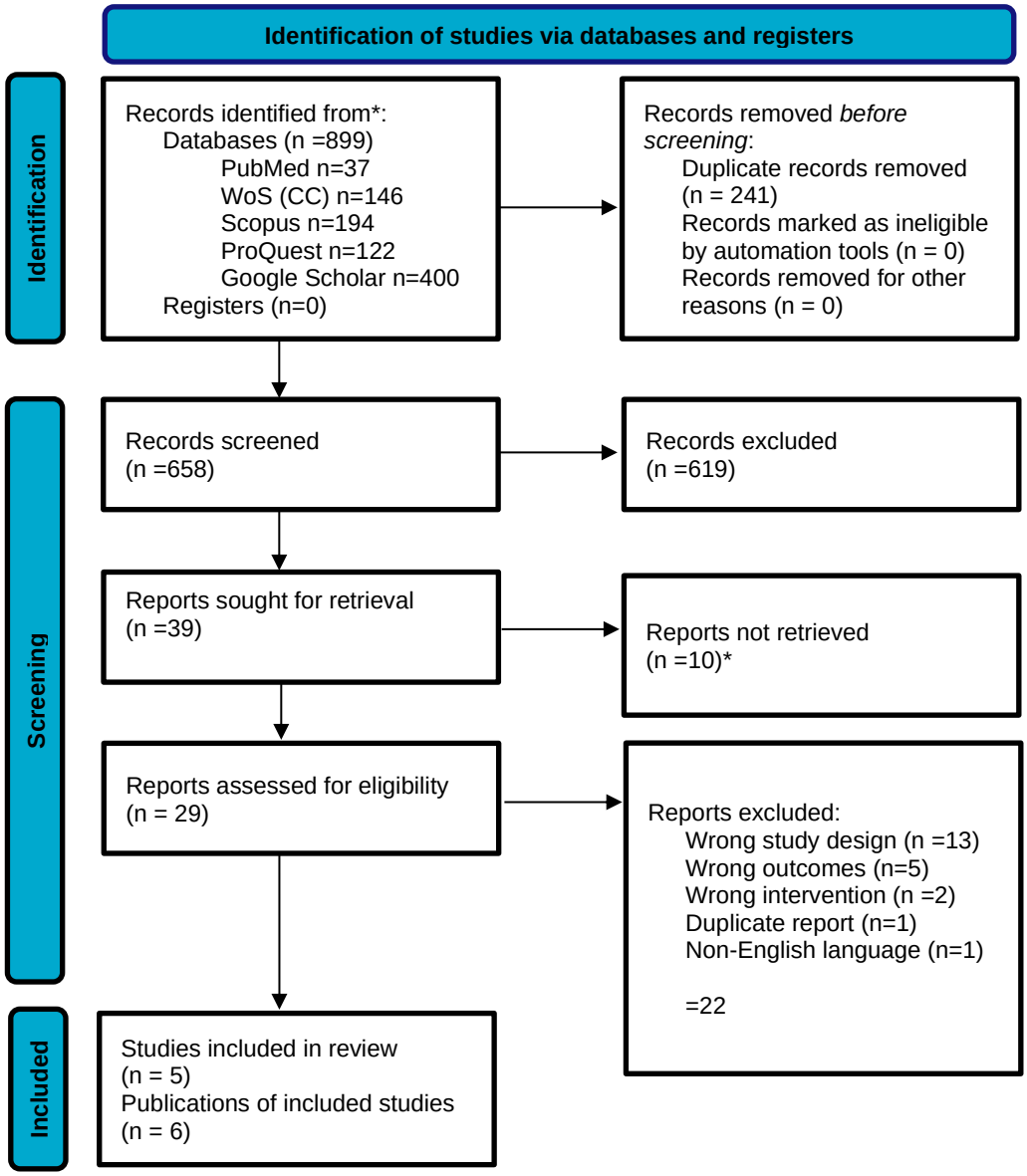

Figure 28. Study flow diagram showing a summary of the literature search and publication selection using the PRISMA format [26]

*Reports not retrieved were citation only and abstract only records

6.1 Description of trials included for review

Five experimental trials and one cross-sectional study were included in the review. One study had a sample of females only [64], while all other studies included both males and female adults in their sample population. All studies were performed in the United States of America. The study details and participants' characteristics of the included studies are presented in Table 6.

The experimental trials all used a randomised, controlled, parallel design, ranging in length from eight [64] to 24 weeks [35]. Two of the experimental trials [65, 66] presented secondary outcomes from another study (Persea Americana for Total Health (PATH); presented in Part I) [23], and three [21, 35, 64] were unique studies. Four trials studied adults with overweight and/or obesity [21, 65, 66].

The dietary comparisons and prescriptions assessed in studies varied. One trial tested the health impacts of avocado in the context of an energy-restricted diet (i.e. weight loss), while the other studies were eucaloric in their energy prescription. All studies compared iso-caloric (i.e., energy matched) interventions, meaning that the intervention arms contained a similar number of kilojoules. The majority of studies controlled the energy intake of participants through the provision of structured dietary advice or eating plans, with the exception of one trial, in which participants consumed an *ad libitum* diet [35].

The cross-sectional study used data from a nationally representative survey (National Health and Nutrition Examination Survey (NHANES; US)) [67], focusing on a cohort of older adults (≥60 y) between 2011-2014. The study compared adults who consumed avocados and/or guacamole ('avocado consumers') to adults who did not consume avocados ('non-consumers').

The dose of avocado provided in the intervention groups of the experimental trials ranged from 135 to 175 grams per day. In the cross-sectional study (i.e., a free-living population), avocado consumers were defined as participants who reported consuming any avocado/guacamole in either of the two 24-h dietary recalls. Among the avocado consumers, the mean intake of avocado was 73.6 grams per day and the mean intake of guacamole was 53.4 g per day.

Table 6. Baseline study and participant characteristics of trials included in the review

First author, year (ref) country	Study design (intervention duration)	n Enrolled (n Analysed)	Male, %	Ethnicity	BMI, kg/m² (SD)	Weight status	Age range Mean years (SD) [range]	Co-conditions	Diet comparison	Avocado g/day	Energy prescription	Isocaloric treatments	Ad libitum diet
Experimental studies													
Scott et al., 2017 [35] USA	Randomised, controlled, parallel (24 wk)	48 (40)	63	NR	Exp: 24.1 (3.1) Con: 24.2 (2.4)	NR	Avo: 63.3 (11.1) [NR] Con: 62.5 (9.2) [NR]	Nil reported	Avocado vs. control (chickpea and/or potato)	135	Eucaloric	Yes	Yes
Henning et al., 2019 [21] USA	Randomised, controlled, parallel (12 wk)	63 (51) Exp: n=24 Con: n=27	Exp: 17 Con: 26	NR	Exp: 30.1 (3.2) Con: 30.0 (3.7)	Overweight and obese	Avo: 42.5 (12.7) [NR] Con: 36.4 (10.8) [NR]	Nil reported	Hypocaloric diet with avocado vs. hypocaloric diet without avocado	150[1]	Negative (i.e., energy restricted)	Yes	No
*Edwards et al., 2020 [65] USA	Randomised, controlled, parallel (12 wk)	163 (84) Exp: n=47 Con: n=37	37 Exp: 32 Con: 43	NR	Exp: 32.5 (5.8) Con: 31.3 (5.5)	Overweight and obese	25-45	Nil reported	Avocado containing meal vs. avocado free meal	Exp: 140 (F); 175 (M) Con: 0	Eucaloric	Yes	No
*Thompson et al., 2021 [66] USA	Randomised, controlled, parallel (12 wk)	163 (109) Exp: n=55 Con: n=54	36	77% Caucasian	32.8 (0.5) F: 33.7 (0.6) M:31.3 (0.9)	Overweight and obese	25-45	Nil reported	Avocado containing meal vs. avocado free meal	Exp: 140 (F); 175 (M) Con: 0	Eucaloric	Yes	No

First author, year (ref) country	Study design (intervention duration)	n Enrolled (n Analysed)	Male, %	Ethnicity	BMI, kg/m² (SD)	Weight status	Age range Mean years (SD) [range]	Co-conditions	Diet comparison	Avocado g/day	Energy prescription	Isocaloric treatments	Ad libitum diet
Henning et al., 2022 [64] USA	Randomised, controlled, parallel (8 wk)	41 (39) Exp: n=20 Con: n=19	0	Exp: White 29% Ctrl: White 40%	Exp: 30.9 (6.9) Con: 31.6 (6.4)	Overweight	27-73 Exp: 43.1 (13.1) [NR] Con: 48.4 (11.3) [NR]	Nil reported	Daily avocado consumption vs. habitual (control) diet	150[1]	NR	NR	Yes
Observational studies													
Cheng et al., 2021 [67] USA	Cross-sectional, nationally representative survey (NHANES, 2011-2014)	n=2,886 adults ≥60 Avo consumer: n=193 Non-consumer: n=2,693	46 Avo consumers: 38 Non-consumers: 47	79.5% Non-Hispanic white	29.1 (0.2) Avo consumers: 27.5 (0.7) Non-consumers: 29.3 (0.2)	NR	≥ 60	Nil reported	Avocado consumers (including guacamole) or non-consumers	Avo consumers: 73.6 (avo, mean intake); 53.4 (guacamole, mean intake) Non-consumers: 0	N/A	N/A	N/A

Grey shading indicates studies were also included in Part I of the review focused of cardiovascular health outcomes; * indicates publications were from a single study; [1] Calculated value based on average weight of an avocado 150g [14].
Abbreviations: BMI, body mass index; M, male; F, female; Exp, experimental group; Con, control group; Avo, avocado; NHANES, National Health and Nutrition Examination Survey; Avo, avocado; NR, not reported; USA, United States of America; wk, weeks; SD, standard deviation.

The outcome measures examined in the studies were grouped into five categories: (i) gut microbiome; (ii) cognitive function; (iii) eye health; (iv) inflammation; and (v) skin health. Table 7 presents the number of studies which reported data for each of the outcomes. Two studies reported measures relating to gut microbiome, both of which included analyses on faecal microbiota and faecal metabolites. Three studies reported measures associated with cognitive function, two studies reported measures related to eye health (macular pigment optical density, MPOD), and two reported on inflammation. Only one study investigated the effect of avocado consumption on skin health.

The study aims, outcome measures and results of trials included in the review are presented in Table 8.

Table 7. Overview of outcome measures reported by studies included for review

		Gut microbiome						Blood lipids							Body composition					Other metabolic markers						
	Primary outcome	**Faecal microbiota**	**Faecal metabolites**	**Cognitive function**	**MPOD**	**Inflammation**	**Skin health**	**TC**	**LDL-C**	**HDL-C**	**non-HDL-C**	**TC: HDL-C**	**TG**	**apoB**	**BW**	**BMI**	**WC**	**Body fat %**	**VAT**	**CVD risk**	**BGL**	**HbA1c**	**Insulin**	**HOMA**	**BP**	**Ox-LDL**
Scott (2017)	Cognition	-	-	✓	✓	✓	-	✓	✓	✓	-	-	✓	-	-	-	-	-	-	-	-	-	-	-	-	✓
Henning (2019)	Weight loss	✓	✓	-	-	✓	-	✓	✓	✓	-	-	✓	-	✓	✓	-	✓	✓	-	✓	-	✓	-	-	-
Edwards et al., (2020)	Cognitive function	-	-	✓	✓	-	-	-	-	-	-	-	-	-	-	✓	-	✓	-	-	-	-	-	-	-	-
Thompson et al., (2021)	Intestinal microbiota	✓	✓	-	-	-	-	-	-	-	-	-	-	-	✓	✓	-	-	✓	-	-	-	-	-	-	-
Henning et al., (2022)	Skin health	-	-	-	-	-	✓	-	-	-	-	-	-	-	✓	✓	-	-	-	-	-	-	-	-	-	-
Cheng et al., (2021)	Cognitive function	-	-	✓	-	-	-	-	-	-	-	-	-	-	-	✓	-	-	-	-	-	-	-	-	-	-
Total - Part II		**2**	**2**	**3**	**2**	**2**	**1**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**2**	**4**	**0**	**1**	**1**	**0**	**0**	**0**	**0**	**0**	**0**	**0**

Underlined text indicates that results were included in Part I of the review focused of cardiovascular health outcomes.

Abbreviations: MPOD, macular pigment optical density; TC, total cholesterol; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglyceride; apoB, apolipoprotein B; BMI, body mass index; VAT, visceral adipose tissue; WC, waist circumference; BGL, blood glucose levels; HbA1c, glycated haemoglobin; HOMA-IR, homeostatic model of insulin resistance; BP, blood pressure; Ox-LDL, oxidised low-density lipoprotein; CVD, cardiovascular disease.

Table 8. Study aims, outcome measures and results of trials included in the review

First author, year	Study aim/s	Outcome measures	Results	Author's conclusions & Notes
Experimental studies				
Scott et al., 2017	The primary objective was to test the effects of the intake of avocado versus potatoes or chickpeas on cognition in older adults. A secondary objective was to investigate the effect of avocado consumption on anti-oxidation, anti-inflammation, and the lipoprotein profile.	Primary outcome: • Cognitive health (serum lutein, zeaxanthin, cognitive tests (attention; visual memory; and executive function, working memory and planning tests), MPOD)	• Serum lutein concentrations significantly increased in both groups (avocado group, 25% increase, p = 0.001; control group 15% increase, p = 0.03). • Avocado consumption did not change serum zeaxanthin. However, serum zeaxanthin concentrations significantly increased in the control group (by >20%, p = 0.005). • There were no significant changes in Attention Tests (choice reaction time or Rapid Visual Information Processing) in either group. • There were improvements in one test of Visual Memory (Paired Associates Learning) in both groups (avocado, p = 0.018; control, p = 0.001), but not in Delayed Matching to Sample. • There were improvements in some tests of Executive Function, Working Memory, and Planning (avocado, Stockings of Cambridge, p = 0.002; control, Spatial Span Reverse, p = 0.028). There were no other significant changes in Executive Function, Working Memory, and Planning tests (Spatial Span or Spatial Working Memory) in either group. • MPOD increased in the avocado group (25%, p = 0.001). There were changes in MPOD in the control group at 3-months, which were not sustained at six months. • Improvements in MPOD were associated with changes (improvements) in cognitive outcomes (Spatial Working Memory and the efficiency in approaching a problem) in the avocado group.	A dietary intervention with avocados was particularly effective in increasing MPOD levels, a biomarker of brain lutein. Increases in MPOD were related to better cognitive performance. Dietary recommendations including avocados may be an effective strategy for cognitive health in the aging population.

First author, year	Study aim/s	Outcome measures	Results	Author's conclusions & Notes
		Secondary outcome/s: • Anti-oxidation (LDL Oxidative lagtime) • Anti-inflammation (CRP, β-Amyloid) • Lipoprotein profile	• There were no changes in inflammatory biomarkers (CRP, β-Amyloid) in either group. • Anti-oxidation and lipoprotein profile results are reported in Part I. Briefly, there was no treatment by time interaction or main effect of treatment or time for HDL, LDL, or total cholesterol. There were no changes in oxidative stress (LDL Oxidative lagtime)	
Henning et al., 2019	To investigate if including 1 Hass avocado per day in a 500- calorie-deficient diet supports weight loss by changing the composition of the intestinal microbiota. Other analyses were conducted to determine if consuming 1 Hass avocado daily improves anti-inflammatory and metabolic markers compared with consuming a hypocaloric control diet.	Primary outcome: • Weight loss • Body composition (BMI, VAT)	• Results reported in Part I. Briefly, both groups reported significant reductions in weight, BMI, total body fat, and VAT.	Daily Hass avocado consumption as part of a hypocaloric diet supported weight loss, a decrease in serum HGF, and an increase in the abundance of bacteria involved in plant polysaccharide fermentation.
		Secondary outcome/s: • Intestinal microbiome (faecal microbiota) • Serum metabolic (glucose, lipids; Reported in Part I) and inflammatory markers	• Compared with the control group, daily avocado intake resulted in an increase in *Firmicutes* (p = 0.016), a decrease in *Bacteroides* (p = 0.023) and trend to increase in *Prevotellaceae* (p = 0.08). This represents a shift in the microbiota to one more characteristic for a dietary pattern of plant-based fibre and fat compared with the control group that experiencing an increase in *Bacteroides* characteristic for animal protein and fat intake. • There were no significant changes in any markers of inflammation in either group.	

First author, year	Study aim/s	Outcome measures	Results	Author's conclusions & Notes
*Edwards et al., 2020	The primary aim was to determine the impact of 12-week daily consumption of fresh Hass avocado on behavioural and neuroelectric indices of cognitive control among persons with overweight and obesity. Secondary analyses were conducted to explore whether the cognitive benefits derived from daily avocado consumption were dependent on changes in circulating (i.e., serum) and retinal (i.e., MPOD) lutein concentrations.	Primary outcome/s: • Cognitive function (serum lutein, attention (Flanker test) and inhibition (Oddball task), MPOD)	• Avocado consumption significantly increased (i.e., improved) serum lutein status ($p \leq 0.001$). There was no difference in the control group ($p = 0.3$). • Avocado consumption was associated with improvements in cognitive performance. Specifically, significant improvements in accuracy during an attentional inhibition task (assessed via the Flanker test; overall accuracy %, $p = 0.01$). There was no change in the control group. • There were no significant changes in response inhibition, and no difference between groups. • There were no significant changes in MPOD, and no difference between groups. • There were no relationships between the change in serum lutein, nor change in MPOD, and changes in cognition in either the behavioural or the neuroelectric outcomes ($p > 0.2$, all).	This study used data from PATH study (primary results reported in Kahn et al., 2021 (and Part I)) *The primary objective of the PATH study was to assess glycaemic control and abdominal adiposity.* Daily avocado intake over 12 weeks, after controlling for covariates, improved attentional inhibition and increased serum lutein concentrations among adults with overweight and obesity. However, the cognitive benefits were independent of changes in lutein concentrations. Additional work is necessary to determine noncarotenoid, or carotenoid interactive, mechanisms by which avocados may influence cognitive function.
		Secondary outcome/s: • Anthropometry and adiposity	• There were no changes in BMI in either group (avocado, $p = 0.8$; control, $p = 0.3$).	
*Thompson et al., 2021	The primary aim was to determine the impact of 12-week daily consumption of fresh Hass avocado on the faecal microbiota in adults with overweight and obesity. Secondary outcomes of the study include microbial metabolites, and relationship between metabolic	Primary outcome: • Intestinal microbiota (faecal microbiota)	Per-protocol results presented: • Faith Phylogenetic Diversity, a measure of microbiota α- diversity (within sample diversity), was greater among the avocado group ($p = 0.02$) compared to control. • There were changes in relative abundance of a number of bacteria within the Firmicutes phylum. At the genus level, *Faecalibacterium*, *Lachnospira*, and *Alistipes* increased in abundance in the avocado group, while the *Roseburia* and *Ruminococcus* were diminished.	This study used data from PATH study (primary results reported in Kahn et al., 2021 (Part I)) *The primary objective of the PATH study was to assess glycaemic control and abdominal adiposity.* Daily avocado consumption resulted in lower faecal bile acid concentrations, greater faecal fatty acid and SCFAs, and greater relative abundances of bacteria capable of fibre

First author, year	Study aim/s	Outcome measures	Results	Author's conclusions & Notes
	health markers, microbial taxa, and metabolites.	Secondary outcome/s: • Microbially derived metabolites (faecal fatty acid and bile acid concentrations)	• Faecal acetate (a SCFA) concentrations were 18% greater in the avocado group compared to the control group (p = 0.01). • Despite reporting higher total dietary fat intake, the avocado group had lower faecal bile acid concentrations compared with control group, including 91% and 57% lower cholic acid and chenodeoxycholic acid concentrations.	fermentation, providing evidence that this nutrient dense food affects digestive physiology, as well as the composition and metabolic functions of the intestinal microbiota.
Henning et al., 2022	To investigate the effects of consumption of one avocado daily for 8 weeks compared with no avocado control on skin health including elasticity, firmness, hydration, pigmentation, and UVB resistance in women with increased abdominal circumference and elevated BMI.	• Skin health (facial skin elasticity (R1, R2, R5, R6, R7), firmness (R0, R8), tiring (R3, R4, R9), pigmentation, sebum, and hydration at two sites: forehead and under-eye) assessed via cutometer	• There was a statistically significant difference between groups for skin firmness at the forehead (R0 only) from baseline to post-intervention, favouring the avocado group (p = 0.04). • Skin firmness significantly increased at the forehead (R0 and R8) and under-eye (R0 only) in the avocado group. • Skin elasticity significantly increased at the forehead (avocado group only) and under-eye (both groups) (R1), and R7 at the forehead for the control group only. There were no significant differences for R2, R5 or R6. • Tiring of skin (R3 and R4 only) significantly reduced at the forehead and under-eye in the avocado group, and at the under-eye (R4 only) in the control group. There were no significant differences for R9. • Melanin index significantly decreased at under-eye (both groups), and at the forehead (control group only) • Erythema significantly increased at under-eye (both groups), and at the forehead (control group only). • There were no significant within- or between-group differences in sebum and hydration.	Daily oral avocado consumption may lead to enhanced elasticity and firmness of the facial skin in healthy women

First author, year	Study aim/s	Outcome measures	Results	Author's conclusions & Notes
			• There were no significant changes in body weight or BMI from baseline to post-intervention in either group.	
Observational studies				
Cheng et al., 2021	This study aims to examine how avocado relates to cognitive function among individuals aged 60 or older from the NHANES 2011–2014.	• Avocado and guacamole intake (24-h dietary recalls) • Cognitive function (CERAD: IWR and DWR; AFT; DSST)	• Average consumption of avocados by consumers was 73.6 g/day. • Average consumption of guacamole by consumers was 53.4 g/day. • In the unadjusted model, avocado consumers had significantly better cognitive performance across all cognitive tests and the global cognition score ($p < 0.05$). • Avocado consumers had significantly better (higher z-) scores for the CERAD IWR and DWR tests and the overall global cognition score and remained significant when controlling for relevant confounders (in all adjusted models), recalling an average of 1.8 more words across the three learning trials for the IWR and 0.9 more words on the DWR than non-consumers. • The mean differences in AFT and DSST scores were attenuated in the adjusted models and were no longer significant (Model 2 AFT, $p = 0.11$, DSST, $p = 0.19$; Model 3, AFT, $p = 0.25$, DSST, $p = 0.21$).	The researchers adjusted data for (1) age, sex, ratio of family income to poverty, race, and marital status (Model 2); and (2) further included adjusted for smoking status, alcohol consumption, work activity, recreational activities, BMI, Mediterranean Diet score, self-reported physician diagnosis of prediabetes or diabetes, self-reported physician diagnosis of coronary heart disease, self-reported physician diagnosis of high blood pressure, and self-reported physician diagnosis of stroke (Model 3). Avocado consumption was associated with significantly better IWR, DWR, and the overall global cognition score, which remained significant when controlling for all relevant confounders.

Note: Grey shading indicates studies were included in Part I; * indicates publications were from a single study.
Abbreviations: NHANES, National Health and Nutrition Examination Survey; Avo, avocado; Con, control; MPOD, macular pigment optical density; VAT, visceral adipose tissue; BMI, body mass index; CRP, C-reactive protein; HGF, hepatocyte growth factor; SCFA, short-chain fatty acids; PATH, Persea Americana for Total Health; UVB, ultraviolet B; CERAD, Consortium to Establish a Registry for Alzheimer's disease; IWR, immediate recall; DWR, delayed recall; AFT, animal fluency test; DST, digit symbol substitution test.

6.2 Gut microbiome

Two randomised controlled parallel studies reported measures relating to the gut microbiome [21, 66]. Both studies were conducted over 12-weeks and included participants with overweight/obesity. Faecal microbiota was assessed with 16S ribosomal RNA gene sequencing. Faecal fatty acid extraction was performed using an established method, and metabolite profiles were generated via gas chromatography. Overall, there is some evidence to suggest that avocado consumption may change the microbial diversity and abundances in individuals on both regular (i.e. not energy restricted), and energy-restricted, weight loss diets.

One study [66] compared faecal microbiota and metabolites at baseline and post-intervention between groups of participants (mean age 35 y) consuming a eucaloric diet including a daily avocado (140 - 175 grams per day, intervention) to a diet without any avocado (control) for 12 weeks in a sample of 151 participants with overweight/obesity. Faith Phylogenetic Diversity, a measure of microbiota α- diversity, was significantly greater among the avocado group ($p = 0.02$), relative to the control. There was a trend ($p = 0.009$) for the avocado group to have a greater measure of β-diversity at post-intervention, relative to control. At the genus level, the relative abundances of *Faecalibacterium, Lachnospira*, and *Alistipes* were enriched in the avocado group (26% and 65%), and the relative abundances of *Roseburia* and *Ruminococcus* were diminished, compared to the control group and post-intervention. The avocado group had higher faecal acetate (18%, $p = 0.01$) with a trend for higher faecal total short-chain fatty acids and greater faecal stearic acid (70%) and palmitic acid (98%) concentrations compared to the control group.

Another randomised controlled parallel study [21] compared hypocaloric (500 kcal deficit) diets with the consumption of an avocado each day (~150 grams per day, intervention) to a hypocaloric diet without avocado (control) for 12 weeks in a sample of 51 participants with overweight/obesity. Both groups lost weight during the intervention, and there was no difference in weight loss between the avocado and control groups. Compared to the control group, daily avocado intake resulted in an increase in *Firmicutes* ($p = 0.016$), a decrease in *Bacteroides* ($p = 0.023$) and trend to increase in *Prevotellaceae* ($p = 0.08$). At the genus level, there were significant differences between groups in *Bacteroides* ($p = 0.012$), *Clostridium* ($p = 0.05$), *Methanosphaera* ($p = 0.012$), and *Candidatus Soleaferrea* ($p = 0.04$) from baseline to post-intervention. In the avocado group, linolenic acid was decreased significantly at post-intervention from baseline ($p = 0.042$), and this change was different to the change in the control group ($p = 0.001$). There was a trend for a decrease in palmitic acid and an increase in oleic acid in the avocado group and the reverse trend in the control group (of an increase in palmitic acid and decrease in oleic acid) at post-intervention compared to baseline.

6.3 Inflammation

Two randomised controlled parallel studies reported the effects of avocado consumption on measures associated with inflammation [21, 35]. Daily avocado consumption did not change any markers of inflammation in either study.

One randomised controlled parallel study [35] compared eucaloric diets with either avocado (135 grams per day, intervention) or potato/chickpeas (one cup/day, control) added to diets over 24-weeks in a sample of 48 healthy adults. β-Amyloid and C-reactive protein were measured in serum as markers of inflammation using a direct solid-phase enzyme immunoassay at baseline, 3- and 6-months. There were no changes in either inflammatory biomarkers (C-reactive protein, β-Amyloid) in the avocado or control groups.

Another randomised controlled parallel study [21] compared hypocaloric (500 kcal deficit) diets with the consumption of an avocado each day (~150 grams per day, intervention) to a diet without avocado

(control) for 12 weeks in a sample of 51 participants with overweight/obesity. Circulating inflammatory markers including IL-6, TNF-α, IL-1β, monocyte chemoattractant protein-1, and C-reactive protein were analysed using the human adipocyte panel on a MagPix analyzer (Luminex). There were no significant changes in any inflammatory markers in either group. IL-1β and C-reactive protein showed a trend to decrease from baseline to the end of the intervention in the avocado group ($p = 0.08$ and $p = 0.085$, respectively). There was also a trend toward a decrease in systemic inflammatory markers IL-1β ($p = 0.070$) and C-reactive protein ($p = 0.074$) in the avocado group, relative to the control group.

6.4 Cognitive function

Three studies reported measures associated with cognitive function; two were randomised, controlled, parallel trials [35, 65] and one was a cross-sectional study [67]. The experimental trials included a variety of measures to assess cognitive function. Both experimental studies measured circulating (serum) and retinal (MPOD) lutein concentrations, which have been related to cognitive function in both young and older adults [68-70]. Scott et al. [35] also measured serum zeaxanthin concentrations and cognitive tests to assess attention, visual memory, and executive function, working memory and planning. Edwards et al., [65] measured attention and inhibition. Although 'attention' was assessed in both studies, their method of measurement was different. The cross-sectional study [67] included subjective, but established, assessments of cognitive function to evaluate participants' memory (via the CERAD – IWR and DWR), executive function (via the AFT), and processing speed and attention (via the DSST). Overall, there is some evidence to suggest that avocado consumption may improve specific domains in cognitive function.

One randomised controlled parallel study [35] compared cognitive test scores at baseline, mid- (three months) and post-intervention (six months) between groups of participants (mean age 63 y) consuming a eucaloric diet with either avocado (135 grams per day, intervention) or potato/chickpeas (one cup/day, control). Of the eight tests, Paired Associates Learning (total errors) decreased (indicating an improvement) in both groups from baseline to mid- (avocado, $p = 0.002$; control, $p = 0.001$) and post-intervention (avocado, $p = 0.018$; control, $p = 0.001$). Relative to baseline, there was an improvement in Stockings of Cambridge (number completed in minimum moves) scores in the avocado group ($p = 0.002$), and Span Reverse (highest span) scores in the control group ($p = 0.028$). Despite the significant within-group changes, there were no significant differences between the avocado and control groups for any of the eight tests.

Another randomised controlled parallel study [65] compared eucaloric diets including a daily meal with avocado (140 - 175 grams per day, intervention) to a diet without avocado (control) for 12 weeks in a sample of 84 participants with overweight/obesity. Participants in the avocado group significantly improved incongruent accuracy (only) in the Flanker task (a measure of attentional inhibition) by 3% ($p = 0.01$). There were no significant changes in any other cognitive test scores, and there were no significant differences between the avocado and control groups for any task. Regarding markers of lutein concentrations, there were no significant changes in MPOD, and no difference between groups. There was, however, a significant increase in serum lutein concentrations in the avocado group over the intervention period ($p \leq 0.001$).

In their nationally representative sample of 2,886 older adults (≥60 y) from the National Health and Nutrition Examination Survey 2011-2014, Cheng et al. [67] compared the cognitive performance of avocado/guacamole consumers (n=193) and non-consumers (n=2,693). In the unadjusted model, avocado consumers had significantly higher (better) scores across all cognitive tests and the global cognition score ($p < 0.05$). When the test scores were adjusted for potential confounders, such as age, sex, and diet quality, scores assessing memory remained significant; avocado consumers recalled an average of 1.8 more words across the three learning trials for the IWR and 0.9 more words on the DWR than non-consumers ($p < 0.05$). However, assessments of executive function and processing speed and attention were no longer different

between avocado consumers and non-consumers when confounders were controlled for in the adjusted model.

6.4.1 Associations between lutein concentrations and cognitive measures

Macular pigment optical density is a non-invasive biomarker for brain concentrations of lutein and has been associated with cognitive function in both young and older adults. Scott et al. [35] reported changes in MPOD were associated with changes in Spatial Working Memory ($r = 0.46$, $p = 0.041$) and the efficiency in approaching a problem ($r = 0.47$, $p = 0.036$) in the avocado group. There were no relationships between the change in MPOD and the change in cognition in the control group. Edwards et al. [65] did not find any relationships between the change in circulatory (serum), nor retinal (MPOD) lutein, and changes in any measures of cognition ($p > 0.2$, all).

6.5 Eye health

Two randomised, controlled, parallel trials reported measures related to eye health (macular pigment density) [35, 65], and findings were mixed.

In the study by Scott et al. [35], MPOD increased from baseline by more than 25% at both mid- (three months) and post-intervention (six months) in the group consuming a eucaloric diet with daily avocado (intervention group, $p = 0.001$). MPOD significantly increased from baseline by approximately 17% ($p = 0.005$) at mid-intervention, but this was not sustained at the end of the intervention in the group consuming a eucaloric diet with daily potato/chickpeas (control group).

In both their unadjusted and adjusted models, Edwards et al. [65] did not report any change to MPOD in either the intervention (daily meal with avocado) or the control groups over the 12-week intervention.

6.6 Skin health

Henning et al., 2022 [64] investigated the effects of daily avocado consumption over eight weeks on skin health in a sample of 39 female adults (27-73 years) with an increased waist-circumference (≥35 inches). Measures of skin health, including facial skin elasticity, firmness, pigmentation (melanin index and erythema), sebum, and hydration were assessed using a cutometer on the forehead and under-eye at baseline, 4- and 8-weeks. Throughout the study, participants were instructed to maintain their habitual diet, usual activity level, avoid sun exposure, and not consume any avocados outside the study.

Compared with a control group consuming a 'habitual diet with no avocado', participants in the avocado (intervention) group showed a stronger (positive) change in forehead skin firmness marker R0 from baseline to week 8 ($p = 0.04$).

There were positive changes in most markers of skin health (firmness, elasticity and tiring, melanin index and erythema) in both groups over time. However, none of these changes were significantly different between avocado and control groups. There were also no significant changes in hydration and sebum formation in either group during the study.

7 Discussion

The purpose of this scoping review was to provide an overview of avocado trials and observational studies focusing on health outcomes other than risk factors for cardiometabolic health. The scoping review identified five areas (reported across six publications) where the effects of avocado consumption have been investigated: (i) gut microbiome; (ii) cognitive function; (iii) eye health; (iv) inflammation; and (v) skin health. Overall, there is a small amount of preliminary evidence to suggest that avocado consumption may change the microbial diversity and abundances, and may improve specific domains in cognitive function. Regarding avocado consumption and eye health, findings were mixed; one study reported that avocado consumption may increase retinal lutein (MPOD), but the second of two studies in this area reported no change. The review found no effect of avocado consumption on markers of inflammation. One study reported a marker of skin firmness on the forehead showed positive changes in those who consumed avocado, relative to the no-avocado control group. There were positive changes in other markers of skin health over time, but these changes were similar between the avocado and control groups, suggesting there may be other variables contributing to this change.

Gut microbiome research, which focuses on the behaviour, interactions, and function of microbial communities within a specified environment, has been a growing area of research over the past two decades [71]. However, the research is still considered to be in its infancy, with most analyses exploratory. Consequently, a large number of studies published recently have reported differences in the microbiome under different conditions [72]. The effect of avocado consumption on the gut microbiome (faecal microbiota and metabolites) was investigated by Henning et al. [21] as a secondary outcome of their weight-loss study, and by Thompson et al. [66] as the focus of a secondary analysis of the PATH study. Avocado intake resulted in changes in relative abundances and diversity of bacteria within the *Firmicutes* and *Bacteroides* phyla; the two most abundant bacterial phyla in humans (40-60% and 20-40%, respectively). Bacteria from the phyla Firmicutes are thought to be more efficient in energy extraction and the Firmicutes/Bacteroidetes ratio is frequently cited in the scientific literature as a hallmark of obesity, although this has been questioned recently [73]. The shift in the microbiota among the avocado group is characteristic of a dietary pattern of plant-based fibre and fat resulting in an increase in *Firmicutes*, whereas the increase in *Bacteroides* among the control group is characteristic of animal protein and fat intake. Avocado consumption also increased faecal acetate concentrations; a short-chain fatty acid produced as a by-product of microbial fermentation of dietary fibre. These SCFAs are absorbed by the colon with a range of health impacts, locally and systematically (Fukuda et al., 2011). However, additional research is required to better understand the metabolic role of SCFAs within the context of overweight and obesity. Henning et al. (2019) found that both intervention groups lost similar amounts of weight in their weight loss trial, therefore changes to microbiome observed in this study were under negative energy conditions. As energy restricted diets have been shown to alter the gut microbiota, it is difficult to separate the influence of avocado over and above energy restriction [74]. The viscous and prebiotic fibre found in diets with avocados can act as a prebiotic to help restore the balance of the colonic microbiota towards a higher anti-inflammatory profile by increasing the ratio of Bacteroidetes to Firmicutes, increasing microflora diversity, and lowering levels of primary and secondary bile acids, all of which are important for maintaining overall health [25]. While current research is limited, studies have described changes observed in the microbiota (in response to a stimulus), however, firm conclusions regarding the effect of the stimulus on the microbiome and the association with health benefits or specific diseases are insufficient [71]. Future research should include more diverse samples of individuals to investigate the effect of avocado consumption in healthy weight adults, and include dietary assessment methods to support the interpretation of study findings.

The effect of avocado consumption on cognition was investigated as a primary outcome in three studies; two experimental studies [35, 65] and one cross-sectional study [67]. While the study by Edwards et al. (2020) focussed on cognitive function, it was also a secondary analysis of the PATH study [23]. There were a broad range of measures used to assess cognitive function, with some evidence to suggest that avocado consumption may improve specific domains in cognitive function, such as immediate and delayed recall. However, there were also improvements in these domains for the control groups. The proposed relationship between avocado consumption and improvements in cognitive function is suggested to be due to the high unsaturated to saturated fatty acid ratio, highly bioavailable carotenoids, and source of prebiotic type fibre [25]. MPOD, a non-invasive biomarker for brain concentrations of lutein, has been associated with cognitive function in both young and older adults [75]. The proposed mechanisms by which lutein benefits cognitive function in the elderly may involve its role as an antioxidant or anti-inflammatory agent [76]. Lutein supplementation has been shown to significantly improve verbal fluency scores in healthy older adults [77]. However, the effect of consuming lutein-containing foods, such as avocados, on cognitive function is not yet fully understood [12]. Both experimental trials included in this review measured retinal lutein (MPOD); one reported no change in either group [65], and the other reported improvements in MPOD in both the avocado and control groups [35]. These improvements in MPOD were also associated with changes (improvements) in cognitive outcomes (Spatial Working Memory and the efficiency in approaching a problem) in the avocado group. The difference in findings between the two studies may be due to different demographic characteristics; Scott and colleagues (2017) examined the relationship between avocado consumption and MPOD in a sample of healthy older adults (mean age 63 years), whereas Edwards et al., (2019) included young-to-middle aged adults (mean age 34 years) with overweight/obesity. The baseline MPOD values between the two studies also varied, and as suggested by Edwards et al., (2019) there may be a ceiling effect with MPOD levels, meaning participants with higher baselines levels have little room for improvement. It is also possible that a higher avocado dose may be necessary to change MPOD status among persons with overweight/obesity.

Daily avocado consumption did not change any of the inflammatory biomarkers measured in the two randomised controlled parallel studies included in the scoping review [21, 35]. Both studies measured inflammation as a secondary outcome and were within a normal range at the start of the study, therefore improvements in anti-inflammatory status may have been difficult to detect. Fatty acids play an important role in inflammatory processes [78], so it is feasible that the consumption of foods high in unsaturated fatty acids, such as avocados, may be beneficial. However, to date this has not been supported by research trials. Others have hypothesised that the consumption of unsaturated fatty acids in combination with the carotenoids, lutein and zeaxanthin in avocados may increase carotenoid absorbability and improve skin health. To date, only one study has investigated the effect of daily avocado consumption on skin health and parameters of skin ageing. There we positive changes in markers of skin health, such as firmness, elasticity and tiring, melanin index and erythema in both the avocado and control groups, suggesting no benefits specifically relating to the avocado intake, but possibly a result of the other advice all participants were given around avoiding sun exposure and maintaining usual skincare. While mechanism have been proposed, more research is needed to understand the role of avocado consumption on outcomes such as skin health and inflammation. Generally, across all the outcomes, other than outcomes of cardiovascular disease assessed in the meta-analysis, the number of studies available is limited, and the heterogeneity in design and how the outcomes are measured means there is not an adequate and consistent body of evidence at this stage to support a beneficial effect of avocados on these health-related outcomes. More rigorously controlled studies, purposefully designed to detect differences between groups using consistent and gold standard methods to assess outcomes are needed to better understand the unique role of avocado on outcomes such as gut health, cognition, eye and skin health.

The strengths of the review include a broad search strategy, the systematic approach to the search and selection of studies for inclusion in the review, and the inclusion of both experimental and observation

evidence. Despite its strengths, several limitations of the review must be acknowledged. The studies were not scored for quality or assessed by statistical analysis of mean effects or heterogeneity for each health effect. There were only six publications eligible for inclusion in the review, all of which were performed in the USA, had small sample sizes and were considered relatively short in duration. Finally, studies included in the review were limited to those published in English. As a result of the limited available evidence, it is difficult to make clear recommendations.

The scoping review identified five areas where the effects of avocado consumption have been investigated: (i) gut microbiome; (ii) cognitive function; (iii) eye health; (iv) inflammation; and (v) skin health. There is some preliminary evidence to suggest avocado consumption may change the microbial diversity and abundances, and may improve some specific domains in cognitive function, however the number of studies investigating this relationship are limited. For a better understanding of the effect of avocado consumption on the health outcomes examined here, more well-designed studies with larger sample sizes, longer in duration, and with different doses of avocado are required.

8 Conclusion and recommendations

This review found that daily avocado consumption may reduce TC and LDL-C in higher risk adults with hypercholesterolaemia compared to a control diet. In more diverse populations (including healthy, overweight, obese, T2DM, normo- and dyslipidaemic adults), avocado consumption resulted in a minor reduction in TC, but had no effect on LDL-C, HDL-C or triglycerides compared to a control diet. However, due to a number of limitations with the current evidence, the degree of certainty in the above findings were rated as very low. This means our confidence in the effect estimate is currently limited and the true effect may be substantially different when more studies of better quality become available. The findings in relation to HDL-C differ from the recent systematic literature review by Mahamassami et al., most likely because of differences in the inclusion criteria for the two reviews. This further supports the need for more larger, well-designed studies which will help to increase the level of certainty in the synthesis of results. The results of a large RCT [59] are expected to be published in the coming months and the addition of these results may have an impact on the conclusions and certainty in the results for cholesterol.

The quality of the current scientific evidence for the effects of avocado on lipid outcomes are generally poor. These studies have small sample sizes and are heterogenous, varying in population characteristics and study design. All the studies had at least one element of their design that increased their risk of bias. There were more serious concerns relating to the data from Colquhoun et al. [32] which appeared to be responsible for influencing the result of the meta-analyses and meta-regression as demonstrated through sensitivity analysis. Hence, the results when this study is included need to be interpreted with caution.

There was emerging evidence to suggest that daily avocado consumption compared to a control diet does not result in weight gain or an increase in BMI. The findings from RCTs were supported by one prospective observational study, which found that avocado consumers with a normal BMI at baseline had a lower rate of change in BMI overtime, and this finding is consistent with cross-sectional observational studies which have shown inverse associations between avocado intake and BMI. While only present in one observation study, the finding of a reduction in age-related weight decline in avocado consumers was novel and may be an area of interest for further research. For other body composition outcomes (including body fat and waist circumference) and other markers of metabolic health (such as blood pressure and blood glucose) there was insufficient evidence combined with inconsistent findings, so no conclusions could be made about the effect of avocados on these health outcomes.

The scoping review identified five areas where the effects of avocado consumption have been investigated: (i) gut microbiome; (ii) cognitive function; (iii) eye health; (iv) inflammation; and (v) skin health. Due to the limited number of studies all these areas of evidence were considered emerging. Despite the small number of studies, consumption of avocados was reported to change the relative abundance and diversity of gut bacteria in both an energy restricted (i.e. weight loss) and an *ad libitum* (i.e. unrestricted) diet. Until there are further advances in the scientific understanding of the gut microbiome, it is difficult to ascertain the implications of the impact of the observed changes in the gut microbiome as a result of avocado consumption.

There are potentially beneficially effects of consuming avocados on cognitive function, with some evidence of improvement in specific domains relating to immediate and delayed recall. Avocados have a high unsaturated to saturated fatty acid ratio, contain highly bioavailable carotenoids, and are a source of prebiotic type fibre which in combination may potentially contribute to this effect on cognitive outcomes. The research findings on the effect of avocado consumption on eye health was mixed and did not support the role of avocados in reducing inflammation, however markers of inflammation were not elevated at

baseline, limiting the likelihood of observing an improvement. Only one study to date has included skin health as an outcome and it reported some positive changes compared to a control diet. Generally, for these other health outcomes examined in the scoping review, further research is needed to understand the effects of consuming avocados.

While it was not possible to elucidate an optimal dose of avocado for any of the health outcomes assessed, there were no negative effects observed in blood lipids or body composition, and no adverse effects reported in the RCTs. Given these studies used avocado doses well above the potentially revised serving size of 75 g per day, it would appear unlikely that there would be any adverse effects of an increase in serving size from the industry serving size of 50 grams to 75 grams.

The strengths of the review include the broad strategy and the systematic approach to the searches. The systematic review and meta-analyses were conducted using a best practice approach, with study quality assessed and sensitivity examined where possible. The daily doses of avocado provided in the studies were relatively high (6-20 times the current consumption levels among Australian consumers), therefore the high levels of compliance reported across most studies was important. However, there were a number of limitations that should be acknowledged. The overall number of studies for most outcomes was limited, with small sample sizes and diverse health characteristics among the study populations. The study designs and dietary comparisons used in the interventions varied so direct comparisons between studies was difficult. Overall, this has limited the ability to draw conclusions on many outcomes and where it was possible, the results need to be interpreted with caution.

To increase the strength of the evidence and certainty in the findings, larger, well-designed studies are needed. For some outcomes, including lipids and inflammation there is a greater likelihood of showing a favourable effect in parameters if the study population has elevated levels of these markers at baseline. Therefore, study populations should be selected with careful consideration. However, this needs to be weighed up against the generalisability of results as a more selective sample makes inferences to the broader population more difficult.

In designing future studies, consideration should also be given to those outcomes shown to have the most meaningful clinical impact. For cholesterol, this includes TC to HDL-C ratio, non-HDL-C and apolipoprotein B, which to date have been reported by few studies. There would be benefits to conducting more large scale, controlled trials to examine the clinical outcomes associated with increased avocado consumption, and more prospective observational studies are also an alternative way to examine the health benefits in a more real world setting with “realistic” level of avocado consumption. There will always be the challenge of demonstrating the effects of avocados independent of other dietary components, so it is critical that all future studies are well thought through and designed in a way that gives confidence in the interpretation of results.

A.1 Appendix A: Search strategy (Part I)

Supplementary Table 1. Search strategy for Part 1, review of avocado and cardiometabolic health effects

Database searched [date of search]	Search Terms	Filters / Limiters applied
PubMed [10.11.2021]	(Avocado*[tiab] OR persea*[tiab] OR "alligator pear"[tiab] OR Persea[MeSH Terms]) AND ("Cardiovascular disease"[tiab] OR Cardiovascular Diseases[MeSH Terms] OR "heart disease"[tiab] OR "heart attack"[tiab] OR "myocardial infarction"[tiab] OR "myocardial ischemia"[tiab] OR arteriosclerosis[tiab] OR atherosclerosis[tiab] OR "coronary artery disease"[tiab] OR stroke[tiab] OR Stroke[MeSH Terms] OR "serum lipids"[tiab] OR LDL-C[tiab] OR HDL-C[tiab] OR "total cholesterol"[tiab] OR triglyceride*[tiab] OR Triglycerides[MeSH Terms] OR TC:HDL-C[tiab] OR non-HDL-C[tiab] OR apoB[tiab] OR "apolipoprotein B"[tiab] OR LDL[tiab] OR Cholesterol, LDL[MeSH Terms] OR Cholesterol, HDL[tiab] OR "blood glucose"[tiab] OR blood glucose[MeSH Terms] OR HbA1c[tiab] OR Glycated Hemoglobin A[MeSH Terms] OR hs-CRP[tiab] OR insulin[tiab] OR insulin[MeSH Terms] OR QUICKI[tiab] OR "homeostasis model assessment-insulin resistance"[tiab] OR HOMA[tiab] OR diabetes[tiab] OR Diabetes Mellitus[MeSH Terms] OR blood pressure[tiab] OR blood pressure[MeSH Terms] OR hypertension[tiab] OR metabolic syndrome[tiab] OR metabolic syndrome[MeSH Terms] OR weight[tiab] OR bodyweight[tiab] OR body weight[tiab] OR "Body Weight"[MeSH Terms] OR BMI[tiab] OR "body mass index"[tiab] OR Body Mass Index[MeSH Terms] OR "waist circumference"[tiab] OR Waist Circumference[MeSH Terms] OR "hip to waist"[tiab] OR hip-to-waist[tiab] OR Waist-Hip Ratio[MeSH Terms] OR "fat mass"[tiab] OR "fat free mass"[tiab] OR fat-free mass[tiab] OR body fat[tiab] OR Adipose Tissue[MeSH Terms]) AND (randomized controlled trial[pt] OR controlled clinical trial[pt] OR clinical trial[pt] OR observational study[pt] OR randomi*[tiab] OR placebo*[tiab] OR clinical trials as topic[MeSH Terms:noexp] OR randomly[tiab] OR trial[tiab] OR trials[tiab] OR prospective[tiab] OR cohort[tiab] OR cross-over[tiab]) NOT (animals[MeSH Terms] NOT humans[MeSH Terms])	post 1990
Web of Science (Core Collection) [10.11.2021]	TS=(avocado* OR persea* OR "alligator pear") AND TS=(“Cardiovascular disease*” OR “heart disease*” OR “heart attack” OR “myocardial infarction” OR “myocardial ischemia” OR arteriosclerosis OR atherosclerosis OR “coronary artery disease” OR stroke OR “serum lipids” OR LDL-C OR HDL-C OR “total cholesterol” OR triglyceride* OR TC:HDL-C OR non-HDL-C OR apoB OR “apolipoprotein B” OR LDL OR “blood glucose” OR HbA1c OR "Glycated Hemoglobin A" OR hs-CRP OR insulin OR QUICKI OR “homeostasis model assessment-insulin resistance" OR HOMA OR diabetes OR "blood pressure" OR hypertension OR "metabolic syndrome" OR weight OR bodyweight OR BMI OR “body mass index” OR “waist circumference” OR “hip to waist” OR "Waist-Hip Ratio" OR “fat mass” OR “fat free mass” OR "body fat" OR "Adipose Tissue") AND TS=("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over)	post 1990
Scopus [10.11.2021]	(TITLE-ABS-KEY (avocado* OR persea* OR "alligator pear")) AND (TITLE-ABS-KEY ("Cardiovascular disease*" OR "heart disease*" OR "heart attack" OR "myocardial infarction" OR "myocardial ischemia" OR arteriosclerosis OR atherosclerosis OR "coronary artery disease" OR stroke OR "serum lipids" OR ldl-c OR hdl-c OR "total cholesterol" OR triglyceride* OR tc:hdl-c OR non-hdl-c OR apob OR	post 1990

	"apolipoprotein B" OR ldl OR "blood glucose" OR hba1c OR "Glycated Hemoglobin A" OR hs-crp OR insulin OR quicki OR "homeostasis model assessment-insulin resistance" OR homa OR diabetes OR "blood pressure" OR hypertension OR "metabolic syndrome" OR weight OR bodyweight OR bmi OR "body mass index" OR "waist circumference" OR "hip to waist" OR "Waist-Hip Ratio" OR "fat mass" OR "fat free mass" OR "body fat" OR "Adipose Tissue")) AND (TITLE-ABS-KEY ("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over))	
ProQuest (NTIS, Agriculture Science Database, Biological Science Database, Continental Europe Database, East & South Asia Database, East Europe, Central Europe Database, Health & Medical Collection, Healthcare Administration Database, India Database, Middle East & Africa Database, Nursing & Allied Health Database, Public Health Database, Science Database, Turkey Database, UK & Ireland Database) [10.11.2021]	(ab(Avocado* OR persea* OR "alligator pear") AND ab("Cardiovascular disease*" OR "heart disease*" OR "heart attack" OR "myocardial infarction" OR "myocardial ischemia" OR arteriosclerosis OR atherosclerosis OR "coronary artery disease" OR stroke OR "serum lipids" OR LDL-C OR HDL-C OR "total cholesterol" OR triglyceride* OR TC:HDL-C OR non-HDL-C OR apoB OR "apolipoprotein B" OR LDL OR "blood glucose" OR HbA1c OR "Glycated Hemoglobin A" OR hs-CRP OR insulin OR QUICKI OR "homeostasis model assessment-insulin resistance" OR HOMA OR diabetes OR "blood pressure" OR hypertension OR "metabolic syndrome" OR weight OR bodyweight OR BMI OR "body mass index" OR "waist circumference" OR "hip to waist" OR "Waist-Hip Ratio" OR "fat mass" OR "fat free mass" OR "body fat" OR "Adipose Tissue") AND ab("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over)) OR (ti(Avocado* OR persea* OR "alligator pear") AND ab(("cardiovascular disease" OR "cardiovascular diseases") OR ("heart disease" OR "heart diseases") OR "heart attack" OR "myocardial infarction" OR "myocardial ischemia" OR arteriosclerosis OR atherosclerosis OR "coronary artery disease" OR stroke OR "serum lipids" OR LDL-C OR HDL-C OR "total cholesterol" OR triglyceride* OR TC:HDL-C OR non-HDL-C OR apoB OR "apolipoprotein B" OR LDL OR "blood glucose" OR HbA1c OR "Glycated Hemoglobin A" OR hs-CRP OR insulin OR QUICKI OR "homeostasis model assessment-insulin resistance" OR HOMA OR diabetes OR "blood pressure" OR hypertension OR "metabolic syndrome" OR weight OR bodyweight OR BMI OR "body mass index" OR "waist circumference" OR "hip to waist" OR "Waist-Hip Ratio" OR "fat mass" OR "fat free mass" OR "body fat" OR "Adipose Tissue") AND ab("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over))Limits applied (Scholarly Journals, Reports, Theses after 1990)	Scholarly Journals, Theses, Reports, post 1990
Google Scholar (via Publish or Perish) [10.11.2021]	(Avocado* OR persea* OR “alligator pear”) AND (“Cardiovascular disease*” OR “heart disease*” OR “heart attack” OR “myocardial infarction” OR “myocardial ischemia” OR arteriosclerosis OR atherosclerosis OR “coronary artery disease” OR stroke OR “serum lipids” OR LDL-C OR HDL-C OR “total cholesterol” OR triglyceride* OR TC:HDL-C OR non-HDL-C OR apoB OR “apolipoprotein B” OR LDL OR “blood glucose” OR HbA1c OR "Glycated Hemoglobin A" OR hs-CRP OR insulin OR QUICKI OR “homeostasis model assessment-insulin resistance" OR HOMA OR diabetes OR "blood pressure" OR hypertension OR "metabolic syndrome" OR weight OR bodyweight OR BMI OR “body mass index” OR “waist circumference” OR “hip to waist” OR "Waist-Hip Ratio" OR “fat mass” OR “fat free mass” OR "body fat" OR "Adipose Tissue") AND ("clinical trial*" OR "comparative stud*" OR "evaluation stud*" OR "controlled trial*" OR "follow-up stud*" OR "prospective stud*" OR random* OR placebo* OR "single blind* OR "double blind*" OR "observational study" OR cohort OR cross-over OR crossover)	1990 onwards

A.2 Appendix B: Avocado serving sizes

Supplementary Table 2. Average serving sizes of avocados reported from food composition databases

Food description	Size description	Weight (g)	Data source	Notes
Avocado, raw	Small	101	AUSNUT Food measures file [79]	Supermarket search undertaken by FSANZ in 2011. Assumes 72% edible portion.
Avocado, raw	Medium	159	AUSNUT Food measures file [79]	Supermarket search undertaken by FSANZ in 2011. Assumes 72% edible portion.
Avocado, raw	Large	216	AUSNUT Food measures file [79]	Supermarket search undertaken by FSANZ in 2011. Assumes 72% edible portion.
Avocados, raw, California[1]	1 fruit without skin and seeds	136	USDA, SR legacy [14]	
Avocados, raw, Florida	1 fruit without skin and seeds	304	USDA, SR legacy [14]	
Avocado, raw	1 fruit	150	USDA, Survey (FNDDS) [14]	

[1] California avocados are the Hass variety.

In Australia, the current industry standard for a serving size is 50g (~1/4 of a large whole avocado or ⅓ of a small) [80] while the current serving size for a vegetable according to the Australian Dietary Guidelines is 75g [9].

A.3 Appendix C: Search strategy (Part II)

Supplementary Table 3. Search strategy for part 2, review of avocado and general health effects

Database searched	Search Terms	Filters / Limiters applied	# of Records retrieved
PubMed	(Avocado*[Title/Abstract] OR persea*[Title/Abstract] OR "alligator pear"[Title/Abstract] OR "Persea"[MeSH Terms]) AND ("Human health"[Title/Abstract] OR "disease risk"[Title/Abstract] OR "health effect"[Title/Abstract] OR "health effects"[Title/Abstract] OR "health promoting"[Title/Abstract] OR health-promoting[Title/Abstract] OR "health impact"[Title/Abstract] OR "health impacts"[Title/Abstract] OR "health benefit"[Title/Abstract] OR "health benefits"[Title/Abstract] OR "nutritive value"[Title/Abstract] OR "Inflammation"[MeSH Terms] OR inflammation[Title/Abstract] OR inflammatory[Title/Abstract] OR allergenicity[Title/Abstract] OR "Sarcopenia"[MeSH Terms] OR Sarcopenia[Title/Abstract] OR "Frailty"[MeSH Terms] OR frailty[Title/Abstract] OR "renal function"[Title/Abstract] OR "Bone and Bones"[MeSH Terms] OR "bone health"[Title/Abstract] OR "bone density"[Title/Abstract] OR "bone mineralisation"[Title/Abstract] OR "musculoskeletal health"[Title/Abstract] OR "genetic toxicology"[Title/Abstract] OR "DNA Damage"[MeSH Terms] OR "DNA Damage"[Title/Abstract] OR "DNA health"[Title/Abstract] OR "Telomere"[MeSH Terms] OR "Telomere length"[Title/Abstract] OR "Gastrointestinal Microbiome"[MeSH Terms] OR "gastrointestinal microbiome"[Title/Abstract] OR "gut microbiome"[Title/Abstract] OR "gastrointestinal microbiota"[Title/Abstract] OR "gut health"[Title/Abstract] OR "fecal metabolite*"[Title/Abstract] OR "faecal metabolite*"[Title/Abstract] "short-chain fatty acid*"[Title/Abstract] OR SCFA*[Title/Abstract] OR "Cognition"[MeSH Terms] OR cognition[Title/Abstract] OR "cognitive ability"[Title/Abstract] OR "Affect"[MeSH Terms] OR affect[Title/Abstract] OR mood[Title/Abstract] OR "brain health"[Title/Abstract] OR "Memory"[MeSH Terms] OR memory[Title/Abstract] OR "Attention"[MeSH Terms] OR attention[Title/Abstract] OR "Executive Function"[MeSH Terms] OR "executive function"[Title/Abstract] OR "Eye"[MeSH Terms] OR "eye health"[Title/Abstract] OR eyesight[Title/Abstract] OR "Eye Diseases"[MeSH Terms] OR "macular degeneration"[Title/Abstract] OR "Healthy		37

	Aging"[MeSH Terms] OR "healthy aging"[Title/Abstract] OR "Gynecology"[MeSH Terms] OR "Reproductive Physiological Phenomena"[MeSH Terms] OR "Musculoskeletal System"[MeSH Terms] OR "Endocrine System"[MeSH Terms] OR "Kidney"[MeSH Terms] OR "Mortality"[MeSH Terms] OR "Nervous System Diseases"[MeSH Terms] OR estrogen[Title/Abstract] OR testosterone[Title/Abstract] OR gynecology[Title/Abstract] OR gynaecology[Title/Abstract] OR gynecological[Title/Abstract] OR gynaecological[Title/Abstract] OR endometriosis[Title/Abstract] OR fertility[Title/Abstract] OR infertility[Title/Abstract] OR pregnancy[Title/Abstract] OR breastfeeding[Title/Abstract] OR "breast milk"[Title/Abstract] OR "sperm count"[Title/Abstract] OR "sperm motility"[Title/Abstract] OR "sperm morphology"[Title/Abstract] OR "reproductive health"[Title/Abstract] OR "menstrual cycle"[Title/Abstract] OR menopause[Title/Abstract] OR menopausal[Title/Abstract] OR premenopause[Title/Abstract] OR premenopausal[Title/Abstract] OR perimenopause[Title/Abstract] OR perimenopausal[Title/Abstract] OR postmenopause[Title/Abstract] OR postmenopausal[Title/Abstract] OR osteoporosis[Title/Abstract] OR "thyroid function"[Title/Abstract] OR "bone health"[Title/Abstract] OR endocrine[Title/Abstract] OR renal[Title/Abstract] OR kidney[Title/Abstract] OR mortality[Title/Abstract] OR morbidity[Title/Abstract] OR neurology[Title/Abstract] OR neurological[Title/Abstract] OR "nervous system"[Title/Abstract]) AND (randomized controlled trial[pt] OR controlled clinical trial[pt] OR clinical trial[pt] OR observational study[pt] OR randomi*[Title/Abstract] OR placebo*[Title/Abstract] OR "clinical trials as topic"[MeSH Terms:noexp] OR randomly[Title/Abstract] OR trial[Title/Abstract] OR trials[Title/Abstract] OR prospective[Title/Abstract] OR cohort[Title/Abstract] OR cross-over[Title/Abstract] OR crossover[Title/Abstract]) NOT (animals [MeSh Terms] NOT humans [MeSH Terms])		
Web of Science (Core Collection)	TS=(Avocado* OR persea* OR “alligator pear”) AND TS=(“Human health” OR “disease risk” OR “health effect” OR “health effects” OR “health promoting” OR health-promoting OR “health impact” OR “health impacts” OR “health benefit” OR “health benefits” OR “nutritive value” OR inflammation OR inflammatory OR allergenicity OR Sarcopenia OR frailty OR “renal function” OR “bone health” OR “bone density” OR “bone mineralisation” OR “musculoskeletal health” OR “genetic toxicology” OR “DNA Damage” OR “DNA health” OR “Telomere length” OR “gastrointestinal microbiome” OR “gut	post 1990	146

	microbiome" OR "gastrointestinal microbiota" OR "gut health" OR "fecal metabolite*" OR "faecal metabolite*" "short-chain fatty acid*" OR SCFA* OR cognition OR "cognitive ability" OR affect OR mood OR "brain health" OR memory OR attention OR "executive function" OR "eye health" OR "eye disease*" OR eyesight OR "macular degeneration" OR "healthy aging" OR estrogen OR testosterone OR gynecology OR gynaecology OR gynecological OR gynaecological OR endometriosis OR fertility OR infertility OR pregnancy OR breastfeeding OR "breast milk" OR "sperm count" OR "sperm motility" OR "sperm morphology" OR "reproductive health" OR "menstrual cycle" OR menopause OR menopausal OR premenopause OR premenopausal OR perimenopause OR perimenopausal OR postmenopause OR postmenopausal OR osteoporosis OR "thyroid function" OR "bone health" OR endocrine OR renal OR kidney* OR mortality OR morbidity OR neurology OR neurological OR "nervous system" OR musculoskeletal) AND TS=("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over OR crossover)		
Scopus	TS=(Avocado* OR persea* OR "alligator pear") AND TS=("Human health" OR "disease risk" OR "health effect" OR "health effects" OR "health promoting" OR health-promoting OR "health impact" OR "health impacts" OR "health benefit" OR "health benefits" OR "nutritive value" OR inflammation OR inflammatory OR allergenicity OR Sarcopenia OR frailty OR "renal function" OR "bone health" OR "bone density" OR "bone mineralisation" OR "musculoskeletal health" OR "genetic toxicology" OR "DNA Damage" OR "DNA health" OR "Telomere length" OR "gastrointestinal microbiome" OR "gut microbiome" OR "gastrointestinal microbiota" OR "gut health" OR "fecal metabolite*" OR "faecal metabolite*" "short-chain fatty acid*" OR SCFA* OR cognition OR "cognitive ability" OR affect OR mood OR "brain health" OR memory OR attention OR "executive function" OR "eye health" OR "eye disease*" OR eyesight OR "macular degeneration" OR "healthy aging" OR estrogen OR testosterone OR gynecology OR gynaecology OR gynecological OR gynaecological OR endometriosis OR fertility OR infertility OR pregnancy OR breastfeeding OR "breast milk" OR "sperm count" OR "sperm motility" OR "sperm morphology" OR "reproductive health" OR "menstrual cycle" OR menopause OR menopausal OR premenopause OR premenopausal OR perimenopause OR perimenopausal OR postmenopause OR postmenopausal OR osteoporosis OR "thyroid function" OR "bone health" OR endocrine OR renal OR kidney* OR mortality OR morbidity OR neurology OR neurological OR "nervous system" OR musculoskeletal) AND TS=("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over OR crossover)	post 1990	194

ProQuest (NTIS, Agriculture Science Database, Biological Science Database, Continental Europe Database, East & South Asia Database, East Europe, Central Europe Database, Health & Medical Collection, Healthcare Administration Database, India Database, Middle East & Africa Database, Nursing & Allied Health Database, Public Health Database, Science Database, Turkey Database, UK & Ireland Database)	TS=(Avocado* OR persea* OR “alligator pear”) AND TS=(“Human health” OR “disease risk” OR “health effect” OR “health effects” OR “health promoting” OR health-promoting OR “health impact” OR “health impacts” OR “health benefit” OR “health benefits” OR “nutritive value” OR inflammation OR inflammatory OR allergenicity OR Sarcopenia OR frailty OR “renal function” OR “bone health” OR “bone density” OR “bone mineralisation” OR “musculoskeletal health” OR “genetic toxicology” OR “DNA Damage” OR “DNA health” OR “Telomere length” OR “gastrointestinal microbiome” OR “gut microbiome” OR “gastrointestinal microbiota” OR “gut health” OR “fecal metabolite*” OR “faecal metabolite*” “short-chain fatty acid*” OR SCFA* OR cognition OR “cognitive ability” OR affect OR mood OR “brain health” OR memory OR attention OR “executive function” OR “eye health” OR “eye disease*” OR eyesight OR “macular degeneration” OR “healthy aging” OR estrogen OR testosterone OR gynecology OR gynaecology OR gynecological OR gynaecological OR endometriosis OR fertility OR infertility OR pregnancy OR breastfeeding OR “breast milk” OR “sperm count” OR “sperm motility” OR “sperm morphology” OR “reproductive health” OR “menstrual cycle” OR menopause OR menopausal OR premenopause OR premenopausal OR perimenopause OR perimenopausal OR postmenopause OR postmenopausal OR osteoporosis OR “thyroid function” OR “bone health” OR endocrine OR renal OR kidney* OR mortality OR morbidity OR neurology OR neurological OR “nervous system” OR musculoskeletal) AND TS=("observational study" OR random* OR placebo* OR trial* OR prospective OR cohort OR cross-over OR crossover)	Scholarly Journals, Theses, Reports, post 1990	122
Google Scholar (via Publish or Perish)	(Avocado* OR persea* OR “alligator pear”) AND (“Human health” OR “disease risk” OR “health effect” OR “health effects” OR “health promoting” OR health-promoting OR “health impact” OR “health impacts” OR “health benefit” OR “health benefits” OR “nutritive value” OR inflammation OR inflammatory OR allergenicity OR Sarcopenia OR frailty OR “renal function” OR “bone health” OR “bone density” OR “bone mineralisation” OR “musculoskeletal health” OR “genetic toxicology” OR “DNA Damage” OR “DNA health” OR “Telomere length” OR “gastrointestinal microbiome” OR “gut microbiome” OR “gastrointestinal microbiota” OR “gut health” OR “fecal metabolite*” OR “faecal metabolite*” OR “short-chain fatty acid*” OR SCFA* OR cognition OR “cognitive ability” OR affect OR mood OR “brain health” OR memory OR attention OR “executive function” OR “eye health” OR “eye disease*” OR eyesight OR “macular degeneration” OR “healthy aging” OR estrogen OR testosterone OR gynecology OR gynaecology OR gynecological OR gynaecological OR endometriosis OR fertility OR infertility OR pregnancy OR	1990 onwards	400

	breastfeeding OR “breast milk” OR “sperm count” OR “sperm motility” OR “sperm morphology” OR “reproductive health” OR “menstrual cycle” OR menopause OR menopausal OR premenopause OR premenopausal OR perimenopause OR perimenopausal OR postmenopause OR postmenopausal OR osteoporosis OR “thyroid function” OR “bone health” OR endocrine OR renal OR kidney* OR mortality OR morbidity OR neurology OR neurological OR “nervous system” OR musculoskeletal) AND ("clinical trial*" OR "comparative stud*" OR "evaluation stud*" OR "controlled trial*" OR "follow-up stud*" OR "prospective stud*" OR random* OR placebo* OR "single blind* OR "double blind*" OR "observational study" OR cohort OR cross-over OR crossover)		

References

1. Roth, G.A., et al., *Global Burden of Cardiovascular Diseases and Risk Factors, 1990–2019: Update From the GBD 2019 Study.* Journal of the American College of Cardiology, 2020. **76**(25): p. 2982-3021.
2. Australian Bureau of Statistics. *Causes of Death, 2018, Cat. no. 3303.0*. 2019; Available from: https://www.abs.gov.au/ausstats/abs@.nsf/0/47E19CA15036B04BCA2577570014668B?Opendocument].
3. AIHW, *Australian Burden of Disease Study: impact and causes of illness and death in Australia 2015—summary report.* , in *Australian Burden of Disease Study series no. 18. Cat. no. BOD 21*. 2019, AIHW: Canberra.
4. *Global burden of 87 risk factors in 204 countries and territories, 1990-2019: a systematic analysis for the Global Burden of Disease Study 2019.* Lancet, 2020. **396**(10258): p. 1223-1249.
5. Heart Foundation, *Dietary Fat & Heart Healthy Eating*. NR.
6. Catapano, A.L., et al., *2016 ESC/EAS guidelines for the management of dyslipidaemias.* European heart journal, 2016. **37**(39): p. 2999-3058.
7. WHO and FAO, *Fats and Fatty Acids in human nutrition. Report of an expert consultation.* . 2010, FAO: Rome.
8. National Health and Medical Research Council, *Nutrient Reference Values for Australia and New Zealand*. 2008, Australian Government: Department of Health and Ageing.: Canberra.
9. National Health and Medical Research Council, *Australian Dietary Guidelines Summary*. 2013, National Health and Medical Research Council: Canberra.
10. Australian Bureau of Statistics, *Australian Health Survey: Nutrition First Results - Foods and Nutrients, 2011-12*, in *cat. no. 4364.0.55.007.* 2017, ABS: Canberra.
11. Horticulture Innovation Ltd, *Horticulture Innovation analysis 2021 [unpublished raw data]*. 2021.
12. Dreher, M.L. and A.J. Davenport, *Hass avocado composition and potential health effects.* Critical reviews in food science and nutrition, 2013. **53**(7): p. 738-750.
13. Food Standards Australia New Zealand, *Australian Food Composition Database – Release 2.* . 2022, FSANZ: Canberra.
14. U.S. Department of Agriculture, A.R.S., *FoodData Central*. 2019.
15. Guan, V.X., E.P. Neale, and Y.C. Probst, *Consumption of avocado and associations with nutrient, food and anthropometric measures in a representative survey of Australians: a secondary analysis of the 2011–2012 National Nutrition and Physical Activity Survey.* British Journal of Nutrition, 2021.
16. Fulgoni, V.L. and C.E. O'Neil, *Avocado consumption by adults is associated with better nutrient intake, diet quality, and some measures of adiposity: National Health and Nutrition Examination Survey (NHANES) 2001–2008.* Internal Medicine Review, 2017.
17. Fulgoni, V.L., M. Dreher, and A.J. Davenport, *Avocado consumption is associated with better diet quality and nutrient intake, and lower metabolic syndrome risk in US adults: results from the National Health and Nutrition Examination Survey (NHANES) 2001–2008.* Nutrition Journal, 2013.
18. Avocados Australia. *Facts at a Glace 2019/20 for the Australian Avocado Industry*. 2020; Available from: https://avocado.org.au/wp-content/uploads/2020/11/Avocados-Australia-Facts-at-a-Glance.pdf.
19. Peou, S., B. Milliard-Hasting, and S.A. Shah, *Impact of avocado-enriched diets on plasma lipoproteins: A meta-analysis.* Journal of Clinical Lipidology, 2016. **10**(1): p. 161-171.
20. Mahmassani, H.A., et al., *Avocado consumption and risk factors for heart disease: a systematic review and meta-analysis.* The American journal of clinical nutrition, 2018. **107**(4): p. 523-536.
21. Henning, S.M., et al., *Hass Avocado Inclusion in a Weight-Loss Diet Supported Weight Loss and Altered Gut Microbiota: A 12-Week Randomized, Parallel-Controlled Trial.* Current Developments in Nutrition, 2019. **3**(8): p. 1.

22. Heskey, C., K. Oda, and J. Sabaté, *Avocado Intake, and Longitudinal Weight and Body Mass Index Changes in an Adult Cohort.* Nutrients, 2019. **11**(3).
23. Khan, N.A., C.G. Edwards, and S.V. Thompson, *Avocado Consumption, Abdominal Adiposity, and Oral Glucose Tolerance Among Persons with Overweight and Obesity.* The Journal of …, 2021.
24. Pacheco, L.S., et al., *Effects of Different Allotments of Avocados on the Nutritional Status of Families: A Cluster Randomized Controlled Trial.* Nutrients, 2021. **13**(11): p. 4021.
25. Dreher, M.L., F.W. Cheng, and N.A. Ford, *A Comprehensive Review of Hass Avocado Clinical Trials, Observational Studies, and Biological Mechanisms.* Nutrients, 2021. **13**(12): p. 4376.
26. Page, M.J., et al., *The PRISMA 2020 statement: an updated guideline for reporting systematic reviews.* Bmj, 2021. **372**.
27. U.S. National Library of Medicine. *Clinicaltrials.gov*. 2021; Available from: https://clinicaltrials.gov/.
28. Veritas Health Innovation, *Covidence systematic review software*. 2021: Melbourne, Australia.
29. The Cochrane Collaboration, *Review Manager (RevMan)*. 2020, The Cochrane Collaboration.
30. Wan, X., et al., *Estimating the sample mean and standard deviation from the sample size, median, range and/or interquartile range.* BMC medical research methodology, 2014. **14**(1): p. 1-13.
31. Higgins, J.T., et al. *Cochrane Handbook for Systematic Reviews of Interventions version 6.2 (updated February 2021)*. 2021; Available from: www.training.cochrane.org/handbook.
32. Colquhoun, D.M., D. Moores, and S.M. Somerset, *Comparison of the effects on lipoproteins and apolipoproteins of a diet high in monounsaturated fatty acids, enriched with avocado, and a high-carbohydrate diet.* The American Journal of Clinical Nutrition, 1992.
33. Lerman-Garber, I., et al., *Response to high carbohydrate and high monounsaturated fat diets in hypertriglyceridemic non-insulin dependent diabetic patients with poor glycemic control.* Diabetes, Nutrition and Metabolism - Clinical and Experimental, 1995. **8**(6): p. 339-345.
34. Lerman-Garber, I., et al., *Effect of a high- monounsaturated fat diet enriched with avocado in NIDDM patients.* Diabetes Care, 1994. **17**(4): p. 311-315.
35. Scott, T.M., et al., *Avocado consumption increases macular pigment density in older adults: a randomized, controlled trial.* Nutrients, 2017.
36. Wang, L., et al., *Effect of a moderate fat diet with and without avocados on lipoprotein particle number, size and subclasses in overweight and obese adults: a randomized, controlled trial.* J Am Heart Assoc, 2015. **4**(1): p. e001355.
37. Deeks, J., J. Higgins, and D. Altman, *Chapter 10: Analysing data and undertaking meta-analyses.*, in *Cochrane Handbook for Systematic Reviews of Interventions version 6.2 (updated February 2021)*, J. Higgins, et al., Editors. 2021, Cochrane.
38. Borenstein, M., et al., *Comprehensive Meta-Analysis Version 3*. 2013.
39. EFSA Panel on Dietetic Products, N., et al., *Guidance for the scientific requirements for health claims related to antioxidants, oxidative damage and cardiovascular health: (Revision 1).* EFSA Journal, 2018. **16**(1): p. e05136.
40. Sterne, J.A., et al., *RoB 2: a revised tool for assessing risk of bias in randomised trials.* BMJ, 2019. **366**.
41. Balshem, H., et al., *GRADE guidelines: 3. Rating the quality of evidence.* Journal of Clinical Epidemiology, 2011. **64**(4): p. 401-406.
42. Bell, L.K., et al., *Supporting strategies for enhancing vegetable liking in the early years of life: an umbrella review of systematic reviews.* The American journal of clinical nutrition, 2021. **113**(5): p. 1282-1300.
43. Pieterse, Z., et al., *High monounsaturated fatty acid avocado for mixed dietary fats during an energy-restricted diet: effects on weight loss, serum lipids, fibrinogen, and vascular function.* Nutrition, 2005.
44. Wells, G.A., et al., *The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses*. 2000, Oxford.
45. Hannon, B.A., et al., *Single Nucleotide Polymorphisms Related to Lipoprotein Metabolism Are Associated with Blood Lipid Changes following Regular Avocado Intake in a Randomized Control Trial among Adults with Overweight and Obesity.* J Nutr, 2020. **150**(6): p. 1379-1387.

46. Wang, L., et al., *A Moderate-Fat Diet with One Avocado per Day Increases Plasma Antioxidants and Decreases the Oxidation of Small, Dense LDL in Adults with Overweight and Obesity: A Randomized Controlled Trial.* The Journal of Nutrition, 2020. **150**(2): p. 276-284.
47. Baigent, C., et al., *Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials.* Lancet (London, England), 2010. **376**(9753): p. 1670-1681.
48. Carranza-Madrigal, J., et al., *Effects of a vegetarian diet vs. a vegetarian diet enriched with avocado in hypercholesterolemic patients.* Archives of medical research, 1997. **28**(4): p. 537-541.
49. Carranza, J., et al., *Effects of avocado on the level of blood lipids in patients with phenotype II and IV dyslipidemias.* Archivos del Instituto de Cardiología de México, 1995. **65**(4): p. 342-348.
50. Carranza Madrigal, J., et al., *Efectos del aguacate como fuente de ácidos grasos monoinsaturados en lipidos séricos, metabolismo de la glucosa y reologia en pacientes con diabetes tipo 2.* Medicina Interna de México, 2008. **24**(4).
51. Madrigal, J.C. and S.M.L. Correa, *El síndrome metabólico en México.* Medicina Interna de México, 2008. **24**(4): p. 251-261.
52. Austin, M. and J.E. Hokanson, KL, *Hypertriglyceridemia as a cardiovascular risk factor.* The American journal of cardiology, 1998. **81**(4): p. 7B-12B.
53. Chawla, S., et al., *The effect of low-fat and low-carbohydrate diets on weight loss and lipid levels: a systematic review and meta-analysis.* Nutrients, 2020. **12**(12): p. 3774.
54. Demacker, P., et al., *Intra-individual variation of serum cholesterol, triglycerides and high density lipoprotein cholesterol in normal humans.* Atherosclerosis, 1982. **45**(3): p. 259-266.
55. Arsenault, B.J., et al., *Beyond low-density lipoprotein cholesterol: respective contributions of non–high-density lipoprotein cholesterol levels, triglycerides, and the total cholesterol/high-density lipoprotein cholesterol ratio to coronary heart disease risk in apparently healthy men and women.* Journal of the American College of Cardiology, 2009. **55**(1): p. 35-41.
56. van Den Bogaard, B., et al., *On-treatment lipoprotein components and risk of cerebrovascular events in the Treating to New Targets study.* European journal of clinical investigation, 2011. **41**(2): p. 134-142.
57. Brown, L., et al., *Cholesterol-lowering effects of dietary fiber: a meta-analysis.* The American Journal of Clinical Nutrition, 1999. **69**(1): p. 30-42.
58. EFSA Panel on Dietetic Products, N. and Allergies, *Scientific opinion on the substantiation of a health claim related to 3 g/day plant sterols/stanols and lowering blood LDL-cholesterol and reduced risk of (coronary) heart disease pursuant to Article 19 of Regulation (EC) No 1924/2006.* EFSA Journal, 2012. **10**(5): p. 2693.
59. Reboussin, D.M., et al., *The design and rationale of a multi-center randomized clinical trial comparing one avocado per day to usual diet: The Habitual Diet and Avocado Trial (HAT).* Contemporary Clinical Trials, 2021. **110**: p. 106565.
60. Drewnowski, A., *Concept of a nutritious food: toward a nutrient density score.* The American Journal of Clinical Nutrition, 2005. **82**(4): p. 721-732.
61. Oakes, M.E., *Filling yet fattening: stereotypical beliefs about the weight gain potential and satiation of foods.* Appetite, 2006. **46**(2): p. 224-233.
62. EFSA Panel on Dietetic Products, *EFSA Panel on Dietetic Products Nutrition and Allergies (NDA). Guidance on the scientific requirements for health claims related to appetite ratings, weight management, and blood glucose concentrations. .* EFSA journal, 2012. **10**: p. 2604.
63. Miller, S.L. and R.R. Wolfe, *The danger of weight loss in the elderly.* The Journal of Nutrition Health and Aging, 2008. **12**(7): p. 487-491.
64. Henning, S.M., et al., *Avocado Consumption Increased Skin Elasticity and Firmness in Women-A Pilot Study.* Journal of cosmetic dermatology, 2022.
65. Edwards, C.G., et al., *Effects of 12-week avocado consumption on cognitive function among adults with overweight and obesity.* Int J Psychophysiol, 2020. **148**: p. 13-24.
66. Thompson, S.V., et al., *Avocado Consumption Alters Gastrointestinal Bacteria Abundance and Microbial Metabolite Concentrations among Adults with Overweight or Obesity: A Randomized Controlled Trial.* J Nutr, 2021. **151**(4): p. 753-762.

67. Cheng, F.W., N.A. Ford, and M.K. Taylor, *US Older Adults That Consume Avocado or Guacamole Have Better Cognition Than Non-consumers: National Health and Nutrition Examination Survey 2011 –2014.* Frontiers in Nutrition, 2021.
68. Feeney, J., et al., *Low macular pigment optical density is associated with lower cognitive performance in a large, population-based sample of older adults.* Neurobiology of aging, 2013. **34**(11): p. 2449-2456.
69. Renzi, L.M., et al., *Relationships between macular pigment optical density and cognitive function in unimpaired and mildly cognitively impaired older adults.* Neurobiology of aging, 2014. **35**(7): p. 1695-1699.
70. Vishwanathan, R., et al., *Macular pigment optical density is related to cognitive function in older people.* Age and ageing, 2014. **43**(2): p. 271-275.
71. Cani, P.D., *Human gut microbiome: hopes, threats and promises.* Gut, 2018. **67**(9): p. 1716-1725.
72. Cullen, C.M., et al., *Emerging Priorities for Microbiome Research.* Frontiers in Microbiology, 2020. **11**.
73. Magne, F., et al., *The Firmicutes/Bacteroidetes Ratio: A Relevant Marker of Gut Dysbiosis in Obese Patients?* Nutrients, 2020. **12**(5): p. 1474.
74. Rinninella, E., et al., *Gut Microbiota during Dietary Restrictions: New Insights in Non-Communicable Diseases.* Microorganisms, 2020. **8**(8): p. 1140.
75. Johnson, E.J., et al., *Relationship between serum and brain carotenoids,-tocopherol, and retinol concentrations and cognitive performance in the oldest old from the Georgia Centenarian Study.* Journal of aging research, 2013. **2013**.
76. Johnson, E.J., *Role of lutein and zeaxanthin in visual and cognitive function throughout the lifespan.* Nutrition reviews, 2014. **72**(9): p. 605-612.
77. Johnson, E.J., et al., *Cognitive findings of an exploratory trial of docosahexaenoic acid and lutein supplementation in older women.* Nutritional neuroscience, 2008. **11**(2): p. 75-83.
78. Liu, A.G., et al., *A healthy approach to dietary fats: understanding the science and taking action to reduce consumer confusion.* Nutrition journal, 2017. **16**(1): p. 53-53.
79. Food Standards Australia New Zealand, *AUSNUT 2011–13 – food measures database file* 2014, FSANZ: Canberra.
80. Australian Avocados. *Australian Avocados: Health and Nutrition*. 2022; Available from: https://australianavocados.com.au/health-nutrition/.

As Australia's national science agency and innovation catalyst, CSIRO is solving the greatest challenges through innovative science and technology.

CSIRO. Unlocking a better future for everyone.

Contact us

1300 363 400
+61 3 9545 2176
csiro.au/contact
csiro.au

For further information

Human Health
Genevieve James-Martin
+61 8 8303 882
genevieve.james-martin@csiro.au
csiro.au/humanhealth