# Referee: 1

Thank you for inviting me to review this manuscript and I want to firstly and importantly acknowledge the huge volume of work that has been undertaken. In general, this is well written and easy to follow. I do have some concerns regarding statements throughout that implicate MR is always a better source of evidence compared to other studies designs. Surely, for example, meta-analyses of observational data is also of value? My specific comments are below.

# Major comments

## Key Messages: I suggest the authors edit the point “Evidence for the causal effects of adiposity and health outcomes has not been comprehensively evaluated.” A multitude of evidence (MR studies, observational studies, systematic reviews of observational studies etc) suggests that adiposity is casually associated with adverse health outcomes. What the current manuscript adds is further evidence to support this statement looking exclusively at MR studies – this certainly adds value but is not the only way to assess causality; hence to ‘comprehensively evaluate causality’ the authors would in fact need to expand the review to beyond just MR studies. This point applies throughout the text as well e.g., last sentence of 1st paragraph of the ‘Introduction’ and 1st sentence of “Strengths and limitations”.

**These lines have been removed.**

## I note the authors have considered as exposures several ‘adjusted’ factors for example, WHR, WC, HC etc adjusted for BMI. I am sure the authors are aware of the potential issues of examining these exposures in an MR context (see <https://doi.org/10.1093/ije/dyaa266>). Given the inherent bias in these associations why focus on them and meta-analyse them together? I would argue that these exposures should be dropped (with an explanation as to why they were excluded). A meta-analysis of biased associations will be biased.

**This is an important point and we thank the reviewer for pointing this out. We have changed the first line of the Meta-analysis and quality assessment section to include reference to this as below and have removed adjusted measures from the meta-analyses:**

**In total, 66 studies from 34 articles were included in 29 meta-analyses – studies investigating the effect of adjusted variables (i.e., WHRadjBMI) in two-sample settings were excluded given recent evidence of biased estimates when using adjusted traits in MR studies.**

## I suggest the authors reconsider the statement “These inconsistencies may reflect the robustness of MR estimates to unmeasured confounding between the exposure and outcome in comparison to observational studies or ….” This is not entirely true, is it? Have the authors assessed pleiotropy in all included studies and only included in the meta-analyses studies which suggest that there is no pleiotropy? In any case, in both MR and observational studies one can never be completely confident in the assumption of ‘no unmeasured confounding’ and therefore MR is not (in my view) more robust to this assumption.

**This is a good point and upon reflection we think it best to remove this sentence entirely as it is implied we have assessed pleiotropy and we have not.**

## Some comments on (i) the variability of instruments used from study to study is warranted and (ii) the fact that non-linear associations were not considered in the literature is important to flag – because it is not just overweight that is a concern, but underweight is also a serious health problem.

**We have added the following paragraph in regards to instrument variability and non-linear effects. We have refrained from reference to underweight as this is a distinct health condition and not merely the opposite of adiposity.**

***Most studies employed similar instrumentation approaches, using a p-value threshold of 5 x 10-8 and a linkage disequilibrium R2 threshold of 0.0001 (the default for the TwoSampleMR R package) to identify independent instruments. This has the advantage that many studies will likely have used the same SNPs for the same exposure. Similarly, most studies used the same methodologies, however, there was little investigation of non-linear effects.***

# Minor comments:

## For clarity, can the authors confirm in lines 390-393, would results be biased towards the confounded observational estimate, or the observational estimate more generally?

**The text is correct and says: The majority of the 31 meta-analyses included just two MR analyses, this was primarily a result of overlapping outcome samples across studies which would ultimately bias results towards the confounded observational estimate.**

## Completely acknowledging the tediousness of referencing a manuscript, please double-check the reference library because at various places “(Error! Reference source not found.)” appears.

**This is an issue of IJE’s linking system for in text cross-referencing for figures/tables, not an issue of referencing.**

## Figure 3 does not appear in the pdf (but does in the html version of the manuscript).

**This is an issue of IJE’s upload system. I have changed the format of the figure and this shouldn’t happen again.**

# Referee: 2

In this study the authors performed a systematic review and meta-analysis of a series of outcomes previously associated with a causal effect of adiposity.  Although a large number of relevant papers exist, the authors followed a well though procedure to select the most reliable evidence which they meta-analyse. The results confirm several suggested associations. An attempt to provide a narrative of studies not used in the meta-analysis was also attempted. This is an interesting idea that will help our identification of risk factors in the future but some limitations are obvious.

## 1) My main concern is the period reviewed.  The authors state that they covered publications up to February 2019.  Although I appreciate the effort and time that a systematic review requires, if this manuscript is to be published in IJE it will appear almost 4 years after the review time cut-off.  This would have been OK for clinical trials that are few and take years to complete, but for a rapidly progressing topic as the causal impact of adiposity, I am afraid that this delay negatively impacts the work done.  This is more of an issue if we consider that adiposity is a well-known factor of COVID-19 severity with different MRs recently published looking at the causal associations of related traits and outcomes.

**During the recent pandemic there was an explosion of work focused on body mass index related traits and outcomes/intermediates or infection impact. This is extremely important, but is complicated by both the parameterisation of infection as a target and the nature of exhaustive genetic instruments for adiposity. This work has been brought together to recount the body of work undertaken immediately before this event and hence presents a pre-pandemic overview of the literature. We agree, further work is now – of course – needed to distil the post-pandemic literature, however that is not within the remit of this review. We believe that this work provides a robust assessment of the causal landscape of adiposity based on the extensive literature published prior to the COVID-19 pandemic.**

## 2) The authors have included pre-prints in their sources, which is commendable.  Though the authors restrict their search to bioRxiv and do not mention medRxiv, which also publishes MR studies.

**medRxiv was launched in June 2019. The literature search was performed on February 18th 2019.**

## 3) Line 255: “The remaining 26 tests had positive effect estimates, 15 of which had CIs that did not span the null”. This description can be misunderstood easily. If the CIs span the null, do we have evidence to say that the effect is positive or negative, or should we just describe it as indistinguishable from 0? In my view, under a frequentist setting suggesting that these associations have positive or negative effect can be problematic.

**This is a good point. Given the large number of studies included in this work we use effect estimates to describe directions of effect in the meta-analyses so that these descriptions are consistent with the narrative synthesis where it is not possible to examine all 2,000+ CIs. We have changed the wording of this sentence to make it clearer:**

***14 of the remaining tests had positive effect estimates with CIs that did not span the null. The remaining 10 tests had positive effect estimates with CIs that spanned the null.***

## 4) Line 29. Why three outcomes? The results provide analyses for many more.

**In response to the other reviewer I have excluded WHRadjBMI from the meta-analyses and have thus changed this paragraph of the abstract as follows:**

***Body mass index (BMI) was the predominant exposure used and was primarily associated with an increase in investigated outcomes; the largest effect in the meta-analyses was observed for the association between BMI and polycystic ovary syndrome (estimates reflect odds ratios (OR) per standard deviation change in each adiposity measure): OR = 2.55; 95% confidence interval (CI) = 1.22–5.33. Only colorectal cancer was investigated with two exposures in the meta-analysis: BMI (OR = 1.18; 95% CI = 1.01–1.37) and waist-hip ratio (WHR; OR = 1.48; 95% CI = 1.08–2.03). Broadly, results were consistent across the meta-analyses and narrative synthesis.***

## 5) Line 112: “If a study focused on adiposity alongside other exposures, the effect of each adiposity measure was reported separately if available. If it was not available, the joint effect was reported”. I am not sure what the authors mean here. Have they done this in the manuscript? At which point?

**This is referring to studies in which an adiposity measure, for example BMI, is used in an MR analysis alongside another trait such as in a multivariable MR analysis. For example, if a study reports the multivariable MR effect estimate of BMI adjusted for metabolite1 on colorectal cancer, we would extract the BMI estimate alone if available otherwise we would extract the adjusted BMI estimate. This situation did not occur for studies included in the meta-analyses. For the handful of situations where this did occur, it was possible to extract data on the adiposity effect (not the joint effect).**

## 6) Line 216: Some of the references in the copy I downloaded appear with an error.

**This is an issue of IJE’s linking system for in text cross-referencing for figures/tables, not an issue of referencing.**

## 7) Figure 3: The in-text Figure 3 does not render correctly. It is OK at the end of the manuscript.

**This is an issue of IJE’s upload system. I have changed the format of the figure and this shouldn’t happen again.**

## 8) In Figures 4 and 5 it will be good to add that the p-values are for the heterogeneity test.

**This is a good point of clarity and I have added this information to the legends.**

## 9) Line 281: “BMI was the predominant exposure and was found to be associated with an increase in the risk of all cancers tested (colorectal, endometrial, lung, ovarian, and prostate”.  This is not true for breast cancer. As CIs cross 0 it is not correct to describe an association with prostate cancer either. The correct statement is similar to: no evidence of association were observed with prostate cancer.

**BMI was not investigated with breast cancer in the meta-analyses; birthweight was investigated with breast cancer and did show a negative direction of effect. The reviewer is correct that the CIs associated with the effect of BMI and prostate cancer do cross the null. However, there is also a positive effect estimate and we describe this as an increase to reflect the direction of the effect estimate. This description keeps the meta-analyses consistent with the narrative synthesis where it was not possible to provide details on all 2,000+ studies and their CIs, instead we summarised directions of effect.**

## 10) I am not convinced that the Narrative analysis adds any additional information to this work. I suggest that the authors remove this section.

**We appreciate the reviewer’s point of view here but think that for the sake of transparency, it is important to provide an overview of all the results. These details may also be useful for researchers who are interested in outcomes that were not represented in the meta-analysis.**

## 11) Line 325: General statements on the association of adiposity to cancer are not taking into account the vast heterogeneity between cancer types and should be avoided.

**This is an important point and we follow this line with wording on line 326 which mentions the heterogeneity of cancer type and subtype**

***Broadly, results suggest adiposity increases overall cancer risk and risk of mortality. However, this risk is modulated by cancer type and subtype.***

## 12) Line 358: HDL and LDL are described as metabolites (from previous sentence). This is not correct.  All studies used for HDL actually use HDL-C instead of the particle.  For LDL, one study appears to use LDL as particle while the other LDL-C in which case the meta-analysis is not looking at the same outcome and should be revised.

**This is correct and an omission on our part. I have corrected the use of HDL-C and LDL-C in the text and figures. In regards the latter point, I believe the reviewer is referring to the study by Wurtz et al. In the methods/results they state ‘LDL’ which may have caused this confusion. In the supplementary data, where we obtained the data, it is stated as ‘LDL cholesterol’ – as can be seen in row 2 page 2** [**https://storage.googleapis.com/plos-corpus-prod/10.1371/journal.pmed.1001765/1/pmed.1001765.s007.pdf?X-Goog-Algorithm=GOOG4-RSA-SHA256&X-Goog-Credential=wombat-sa%40plos-prod.iam.gserviceaccount.com%2F20221120%2Fauto%2Fstorage%2Fgoog4\_request&X-Goog-Date=20221120T082803Z&X-Goog-Expires=86400&X-Goog-SignedHeaders=host&X-Goog-Signature=**](https://storage.googleapis.com/plos-corpus-prod/10.1371/journal.pmed.1001765/1/pmed.1001765.s007.pdf?X-Goog-Algorithm=GOOG4-RSA-SHA256&X-Goog-Credential=wombat-sa%40plos-prod.iam.gserviceaccount.com%2F20221120%2Fauto%2Fstorage%2Fgoog4_request&X-Goog-Date=20221120T082803Z&X-Goog-Expires=86400&X-Goog-SignedHeaders=host&X-Goog-Signature=)