**Supplemental Table 6: Risk of bias assessment signaling questions.**

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| **Bias domain** | **Signaling question1 and skip logic** | | **Response options** |
| Confounding2 | 1.1) Did the investigators use an appropriate analysis method that adjusted for all the critically important baseline confounding areas? Common adjustment methods include standardization, Mantel-Haenszel pooling, stratification, and multivariable regression. | | Y / PY / PN / N / NI |
| If Y / PY to 1.1 | 1.2) Were baseline confounding areas that were adjusted for measured validly and reliably by the variables available in this study? | Y / PY / PN / N / NI |
| 1.3) Was exposure assessed at multiple time points? | | Y / N |
| If Y to 1.3 | 1.4) Was the analysis based on splitting follow-up time according to exposure status? In other words, did the analysis account for potentially time-varying exposure status? | Y / PY / PN / N / NI |
| If Y to 1.3 | 1.5) Were exposure changes likely to be related to factors that are prognostic for/predictive of/associated with the outcome? | Y / PY / PN / N / NI |
| If Y / PY to 1.5 | 1.6) Did the investigators use an appropriate analysis method that adjusted for all the critically important confounding areas and for time-varying confounding? Common methods for adjusting for time-varying confounding include the G formula/G estimation, marginal structural models/inverse probability of treatment weighting/inverse probability of exposure weighting. | Y / PY / PN / N / NI |
| If Y / PY to 1.6 | 1.7) Were time-varying confounding areas that were adjusted for measured validly and reliably by the variables available in this study? | Y / PY / PN / N / NI |
| If Y / PY to 1.1  OR  If Y / PY to 1.6 | 1.8) Did the investigators avoid adjusting for post-exposure variables (other than any time-varying confounders)? | Y / PY / PN / N / NI |
| 1.9) Risk of bias due to confounding. Use your answers to the above signaling questions about potential confounding and adjustment for confounding when assessing the risk of bias due to confounding. | | L / M / S / C / NI |
| Selection bias | 2.1) Was the selection of participants into the study (or into the analysis) based on variables measured after exposure assessment? | | Y / PY / PN / N / NI |
| If Y / PY to 2.1 | 2.2) Were the post-exposure variables that influenced selection associated with exposure? | Y / PY / PN / N / NI |
| If Y / PY to 2.2 | 2.3) Were the exposure variables that influenced selection influenced by the outcome or a cause of the outcome? | Y / PY / PN / N / NI |
| If cohort | 2.4) Was there loss to follow-up? If the study was not longitudinal, please select Not Applicable. | Y / PY / PN / N / NI / NA |
| If Y / PY to 2.4 | 2.5) Was the loss to follow-up similarly distributed between participants who developed the outcome and those who did not (i.e. non-differential loss to follow-up)? | Y / PY / PN / N / NI / NA |
| If case-control | 2.6) Were cases selected for the study a representative sample of all cases in the source population? | Y / PY / PN / N / NI |
| If case-control | 2.7) Were controls selected for the study from the same source population as the cases? In other words, if a control had instead had the outcome, would they likely have been identified as a case for the study? If the study is a nested case-control study, the answer will likely be yes. | Y / PY / PN / N / NI |
| If case-control | 2.8) Were controls selected for the study a representative sample of people who did not develop the outcome in the source population? | Y / PY / PN / N / NI |
| If Y / PY to 2.2 AND Y / PY to 2.3  OR  If N / PN to 2.4  OR  If N / PN to 2.6  OR  If N / PN to 2.7  OR  If N / PN to 2.8 | 2.9) Did the investigators use adjustment techniques that are likely to correct for the presence of selection bias? Common methods to adjust for selection bias is inverse probability weighting, controlling for covariates associated with selection, and bias analysis. | Y / PY / PN / N / NI |
| 2.10) Risk of bias due to selection bias. Use your answers to the above signaling questions about potential selection bias and adjustment for selection bias when assessing the risk of selection bias. | | L / M / S / C / NI |
| Exposure assessment | 3.1) Is exposure status well defined? | | Y / PY / PN / N / NI |
| 3.2) Was information used to define exposure status recorded prior to outcome assessment? | | Y / PY / PN / N / NI |
| 3.3) Could classification of exposure status have been affected by knowledge of the outcome or risk of the outcome? | | Y / PY / PN / N / NI |
| 3.4) Were exposure assessment methods robust (including sampling, storage, laboratory assays, data processing, and exposure classification)? 16s rRNA gene sequencing and cpn60 sequencing methods are generally considered robust methods for assessing microbiota composition. | | Y / PY / PN / N / NI |
| 3.5) Were exposure assessment methods comparable for participants with and without the outcome? | | Y / PY / PN / N / NI |
| 3.6) Risk of bias due to exposure assessment. Use your answers to the above signaling questions about exposure assessment and potential misclassification when assessing the risk of information bias due to exposure assessment. | | L / M / S / C / NI |
| Outcome assessment | 4.1) Could the outcome measure have been influenced by knowledge of exposure status? | | Y / PY / PN / N / NI |
| 4.2) Was outcome measurement sensitive? Sensitivity = TP / (TP + FN). | | Y / PY / PN / N / NI |
| 4.3) Were outcome assessors unaware of exposure status? | | Y / PY / PN / N / NI |
| 4.4) Were methods of outcome assessment comparable across exposure statuses? | | Y / PY / PN / N / NI |
| 4.5) Were any systematic errors in outcome assessment related to exposure status? | | Y / PY / PN / N / NI |
| If case-control | 4.6) Were cases selected for the study incident cases? | Y / PY / PN / N / NI |
| 4.7) Risk of bias due to outcome assessment. Use your answers to the above signaling questions about outcome assessment and potential misclassification when assessing the risk of information bias due to outcome assessment. | | L / M / S / C / NI |
| Missing data | 5.1) Were participants excluded due to missing outcome data? | | Y / PY / PN / N / NI |
| 5.2) Were participants excluded due to missing exposure data? | | Y / PY / PN / N / NI |
| 5.3) Were participants excluded due to missing data on other variables used in the analysis? | | Y / PY / PN / N / NI |
| If Y / PY to 5.1  OR  If Y / PY to 5.2  OR  If Y / PY to 5.3 | 5.4) Are the proportion of participants with missing data and reasons for missing data similar across exposure statuses? | Y / PY / PN / N / NI |
| If Y / PY to 5.1  OR  If Y / PY to 5.2  OR  If Y / PY to 5.3 | 5.5) Are the proportion of participants with missing data and reasons for missing data similar across outcome statuses? | Y / PY / PN / N / NI |
| If Y / PY to 5.1  OR  If Y / PY to 5.2  OR  If Y / PY to 5.3 | 5.6) Were appropriate statistical methods used to account for missing data? Methods to account for missing data include single imputation (mean substitution - missing values replaces with the mean for the variable, worst-case analysis - impute worst-case value for missing values, last observation carried forward, hot deck imputation - impute missing value from similar observations, single regression imputation) and multiple imputation. Multiple imputation is considered much more robust than single imputation. | Y / PY / PN / N / NI |
| 5.7) Risk of bias due to missing data. Use your answers to the above signaling questions about missing data and methods to account for missingness when assessing the risk of bias due to missing data. | | L / M / S / C / NI |
| Selection of reported results | 6.1) Is the reported effect estimate likely to be selected on the basis of results from multiple outcome measurements within the outcome domain? If the outcome was only measured with one method, please select No. | | Y / PY / PN / N / NI |
| 6.2) Is the reported effect estimate likely to be selected on the basis of results from multiple analyses of the exposure-outcome relationship? | | Y / PY / PN / N / NI |
| 6.3) Is the reported effect estimate likely to be selected on the basis of results from different subgroups? | | Y / PY / PN / N / NI |
| 6.4) Risk of bias due to selection of reported results. Use your answers to the above signaling questions when assessing the risk of bias due to selection of reported results. | | L / M / S / C / NI |
| Overall | 6.5) Overall risk of bias. Use your judgements on the risk of bias in individual bias domains (confounding, selection bias, exposure misclassification, missing data, outcome misclassification, selection of reported results) to evaluate the overall risk of bias for the study. A conservative approach is to classify the study at the level of risk of bias that matches the highest risk of bias you assigned for an individual bias domain. | | L / M / S / C / NI |

1Adapted from Morgan et al. Environ Int. 2018;120:382-387.

2Confounders of interest were determined a priori and included vaginal sex (e.g. frequency, number of partners, condom use), hormonal contraception, vaginal washing, race/ethnicity, and the variables race/ethnicity may be a proxy for (e.g. SES, site/region).

Y, yes; PY, probably yes; PN, probably no; N, no; NI, not enough information; L, low; M, moderate; S, serious; C, critical; NA, not applicable; TP; true positive; FN, false negative.