However, before this theory can be fully supported, issues of ambiguity that were identified in this systematic review should be addressed. Brusselaers et al aimed to assess the association between vaginal dysbiosis and cervical cancer. Concerns arise with respect to the definition of vaginal dysbiosis that was used and the study selection criteria.

First, vaginal dysbiosis was defined as a deviation from a *Lactobacillus*-dominant microbiota. However, a definitive cut off point for *Lactobacillus* dominance was never identified, and this may vary from study to study. Further confusion occurs because the definition that was used by Brusselaers et al uses microscopy-based assessment of *Lactobacillus* dominance, but the authors included studies that used Amsel's criteria. Amsel's criteria does not directly assess *Lactobacillus* dominance, rather it assesses the presence or absence of clue cells via microscopy.[2] Additionally, the presence of clue cells is not required for the diagnosis of vaginal dysbiosis. Three of the 4 Amsel's criteria are required to be diagnosed with vaginal dysbiosis, of which presence of clue cells is only 1.[2] This may have resulted in an overestimation of results because of misclassification. It should also be noted that it is possible to have a *Lactobacillus*-dominant vaginal microbiota and be classified as unhealthy or exhibit characteristics that are similar to that of a vaginal microbiota that has deviated from *Lactobacillus* dominance.[3]

Second, we are concerned that Brusselaers et al[1] acknowledge that vaginal dysbiosis commonly is also referred to as bacterial vaginosis; however, this term was not included in the search strategy. Furthermore, although the authors used MESH and Emtree terms, CINAHL headings were not included in the search strategy for the CINAHL database. Further clarity is also needed with respect to the inclusion of grey literature. Conference abstracts were cross checked for relevant full text papers; the time frame and conferences that were searched were not included. We acknowledge the efforts of Brusselaers et al to assess bias; however, a customized tool was used for assessment of risk of bias, and the validity of the tool could not be verified. It was not expressed clearly whether this tool was validated or piloted before its use. Additionally, the authors failed to assess publication bias. Funnel plots could have been used to assess publication bias graphically. Brusselaers et al amply highlight the need for further investigation into the association between vaginal dysbiosis and the risk of human papillomavirus and cervical cancer. ■

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REPLY

We thank Drs Coudray and Kiplagat for their interest in our work and agree that our study has limitations, as described earlier.[1]

There is an important debate ongoing considering the vaginal microbiome, in particular how to categorize this, for example to use community state types or not.[2] Yet, because of the almost discrete distribution of community state types, there is a clear distinction between *lactobacilli*-dominated vaginal microbiota compositions, and those not dominated by *lactobacilli*, which makes cut-offs redundant. Beside the difficulties in categorising, it remains to be established which microbiome composition is to be considered healthy, which does not necessarily equal asymptomatic. To date, it remains to be established firmly which vaginal microbiota composition (ie, which specific *lactobacilli spp*) should be preferred and how a dysbiotic state can be restored from an "unhealthy" microbiome composition.

There is increasing evidence that the vaginal microbiome plays an important role in many outcomes, including HPV acquisition and persistence, yet many subquestions remain to be answered. What came first? The vaginal dysbiosis or the human papillomavirus? Therefore, longitudinal data are required to assess changes over time in the same individuals. Until the present, most published microbiome studies that have assessed the vaginal microbiota composition are relatively small and cross-sectional; to date, longitudinal data based on molecular studies remain too scarce to be implemented in a metaanalysis.

We also would like to point out that, although the Amsel criteria do not measure the presence of lactobacilli directly, individuals with a positive result will not present with a large abundance of *lactobacilli*. The presence of clue cells and positive Amsel tests have shown to be highly specific for bacterial vaginosis (98% and 99%, respectively), although sensitivity is suboptimal.[3]

Considering the literature search: The conference abstracts were those retrieved through the Web of Science, which also indexes abstracts of major conferences. No additional hand searching was conducted, because we aimed to include

full-length papers only. Because this is a relatively recent research field, we do believe that we have conducted a thorough and broad search, including backward and forward citation tracking. Combined with the extensive expertise of the team of authors in vaginal dysbiosis, human papillomavirus, and conducting metaanalyses, we fully do support our reported search strategy and have not identified any papers so far that may have been missed.

Quality assessment is indeed an important part of the process of conducting systematic reviews. In our experience, we did not find the customized quality assessment tools sufficiently sensitive or specific for our research question. The purpose of quality assessment should be to explore reasons for heterogeneity and bias in the original studies, not purely as a summary score claiming that "x%" of our studies were "very high quality" based on what the authors reported, giving ourselves good marks for our own study. It is well-known that most quality assessment tools are focusing on quality of reporting, not necessarily methodologic quality.[4] By targeting items that may be of high relevance for our specific research question, this customized approach enables a more thorough assessment and understanding of the reasons of heterogeneity, which enabled us to differentiate between the different concerns that were related to the methodologic quality of the studies and to assess the potential impact in more depth. In addition, we did not use the quality assessment as a tool to exclude studies that were scored as "lower quality" from our analysis, which is an approach that should be discouraged for metaanalyses that are based on observational studies because it does increase the risk of selection bias.

Funnel plots were not used because of the low number of studies, which is also supported by many researchers in the field.[5] ■

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