OBSTETRICS & GYNECOLOGY

NOTICE: This document contains correspondence generated during peer review and subsequent revisions but before transmittal to production for composition and copyediting:

- Comments from the reviewers and editors (email to author requesting revisions)
- Response from the author (cover letter submitted with revised manuscript)*

**The corresponding author has opted to make this information publicly available.*

Personal or nonessential information may be redacted at the editor's discretion.

Questions about these materials may be directed to the *Obstetrics & Gynecology* editorial office: obgyn@greenjournal.org.

Date: Apr 05, 2022
To: "Sara Naseri" [redacted]
From: "The Green Journal" em@greenjournal.org
Subject: Your Submission ONG-22-429

RE: Manuscript Number ONG-22-429

Screening for high-risk human papilloma virus using passive self-collected menstrual blood

Dear Dr. Naseri:

Thank you for sending us your work for consideration for publication in Obstetrics & Gynecology. Your manuscript has been reviewed by the Editorial Board and by special expert referees. The Editors would like to invite you to submit a revised version for further consideration as a Research Letter.

If you wish to revise your manuscript, please read the following comments submitted by the reviewers and Editors. Each point raised requires a response, by either revising your manuscript or making a clear argument as to why no revision is needed in the cover letter.

To facilitate our review, we prefer that the cover letter you submit with your revised manuscript include each reviewer and Editor comment below, followed by your response. That is, a point-by-point response is required to each of the EDITOR COMMENTS (if applicable), REVIEWER COMMENTS, STATISTICAL EDITOR COMMENTS (if applicable), and EDITORIAL OFFICE COMMENTS below. Your manuscript will be returned to you if a point-by-point response to each of these sections is not included.

The revised manuscript should indicate the position of all changes made. Please use the "track changes" feature in your document (do not use strikethrough or underline formatting).

Your submission will be maintained in active status for 21 days from the date of this letter. If we have not heard from you by Apr 26, 2022, we will assume you wish to withdraw the manuscript from further consideration.

EDITORS COMMENTS:

We would welcome your resubmission after responding to the reviewer comments. However, you rightly describe this in the manuscript text as a 'pilot study' which in the Info for Authors would necessitate that you edit this to a Research Letter category. Thank you!

REVIEWER COMMENTS:

Reviewer #1:

The authors present a prospective observational study comparing q-pad menstrual blood analysis to office testing for HRHPV. The results demonstrate high concordance between the two tests, which is very promising, as the qpad seems to be non-invasive and cost-effective (it would be useful to discuss cost of this tool in the paper as the goal seems to be to deploy it in resource rich settings.) Overall, the design is appropriate and the results could be an important contribution to existing literature to make HPV testing available to the global majority.

Abstract:
line 9: do you mean "HR-HPV screening is a potential..."
line 23-24: "HR-HPV positive samples where the CCS to QPS interval (Q-Pad use) was < 2 month..." this is confusing,

Introduction:
line 46-47: "HR-HPV screening is conventionally performed in the clinic and requires women to undergo a gynecological exam, which is invasive, uncomfortable, and resource intense", this needs citation and a bit more elaboration, particularly about economic costs of this test and limitation of use in resource-poor settings

line 55-59: the description of the QPad can be placed in the methods section, and instead you could just state that utility of a non-invasive, simple and cheap test could improve HR-HPV screening.
line 73: "We found favorable participation in community based screening" was this in a previous study or focus group? please specify
line 74-76: seems out of place here, maybe can be placed in paragraph above describing the q-pad briefly, or in discussion/conclusion

Methods:
line 93: what do you mean by "regularly menstruate", is it those with regular menses; did you then exclude anyone with menstrual disorders, what about those with dysmenorrhea and PMDD who may have a different concentration of inflammatory markers compared to those without menstrual disorders ? please clarify

line 98: I am not able to discern the rationale for including the self-collected swab, the authors make valid points about the limitations of the test, and in my opinion it is not useful to compare the self-swab to the qpad since the standard test is clinician-collected, and comparing that to the qpad is sufficient to demonstrate utility; please provide clarification on why the self-swab was included as part of this study
please include information in methods about statistical analysis

line 161: please provide the total number of participants who could not provide samples within 2 months

line 167-168: for the presence of HR-HPV on the QPad, was there confirmatory testing done (either repeat pap/hr-hpv or colposcopy)? if yes, please provide results, if not please explain why not, as the assumption that the clinician test was a false negative and not that the qpad result was a false positive seems biased

Discussion:
line 199-202: discussion of the number of people who had a delay in submitting qpad results is better suited to the Result section; please provide more detail about spontaneous viral clearance of HR-HPV; did the patients who had a +HRHPV result undergo additional testing to confirm resolution? without confirmation, it is not reasonable to assume that the qpad result indicates clearance rather than a false negative

Reviewer #2:

Thank you for allowing me the opportunity to review "Screening for high risk HPV using passive self-collected menstrual blood" presented for possible publication. In this manuscript, the authors present a pilot study of a new technology, The QPad. This new pad includes a paper-based, dried blood spot (DBS) strip which allows for self-collection of menstrual blood that can be used to screen for high risk types of HPV. The HPV test is the Roche Cobas 4800, which is one of two FDA approved testing platforms that allows for primary HPV testing.

The article is timely. There are currently no US FDA approved devices for HR HPV self-collection despite a growing body of research on this technology. There is a lot of interest in this topic as cervical cancer screening, in this country and worldwide, has become a disease of access. Approximately 50% of patients diagnosed with cervical cancer were not screened in the preceding 5 years. Increasing availability of testing to unreached populations is going to be instrumental (along with increased HPV vaccine uptake) to reducing cervical cancer, a preventable disease.

The authors report that collection through menstrual blood was acceptable to patients (over self collection) and with a high concordance to clinician sampling results. They also report a sensitivity (of HR HPV with clinician collected as reference) of 94% and sensitivity of 82%. Generally sensitivity and specificity are reported in relation to HSIL but the authors only had 7 patients with CIN 2+ in this pilot study.

Some specific comments below:

Introduction:
- (Line 36): Cervical cancers are preventable with a combination of screening and primary prevention. Pap testing is important but is only one step in the prevention of cervical cancer.
- (Line 45): HR HPV is not poised to become a primary screening tool, it already is. From the most recent ACS guidelines, primary HPV screening is the preferred method of testing. ACOG and ASCCP support both primary HR HPV screening and co-testing.
- (Line 71-73): Citing unpublished and anecdotal evidence is not best practice. The authors are not proposing a point of care (POC) test so I'm not sure this statement is even necessary.
- (Line 76) - "Women's physical" is a misleading term. A physical is much more than just a pap test. This term

underrepresents what is done at a "physical exam."

Methods
- (Line 92): Why were women under age 21 recruited? The demographic table at the end suggests that no one under 21 yo participated. Further, the demographic table includes women up to age 49, but in the methods section, only women up to age 45 were recruited. This seems to be a discrepancy.
- Was any other demographic data collected? This is a significant limitation and should at least be addressed in the discussion session if no other information was collected. Things such as history of HSIL and/or immunocompromise would increase risk of persistent HR HPV infection and may alter results (especially in those for whom more than 2 months passed from clinician sampling before menstrual blood sampling).
- Presumably all of these patients have a cervix? It should just be clear in the inclusion criteria.
- Is there any data on stability of the testing available from the manufacturer of the strip? An average of 10 days passed, but there is no range provided. Were any samples held for prolonged periods of time? Any relation to longer held samples and false negative or false positive results?
- Was any genotyping performed or is it possible to do this? The Cobas test does offer genotype screening which becomes important when addressing the clinical application of this technology.

Results
- Almost 50 patients (approximately 30% of patients) did not complete the study. This should be addressed as another limitation in the discussion.

Discussion
- (Line 217): It is certainly possible that the HPV was higher in the cervical canal which is why it was missed on clinician sampling. Further, consider the fact that HPV generally is a field effect and may infect the vulva, vagina and anus. Is it possible that the menstrual blood pad may be falsely positive for HPV if the HPV is present in the anal canal?

Reviewer #3:

1. This manuscript describes a novel collection method to collect blood to screen for high-risk HPV, and explains why HPV screening is important and gives the sense that, though this is not yet the current practice, HPV testing alone could become the screening method in use, replacing cytology. The argument for a self-collected, non-invasive screening method is provided, as screening with a clinician involved time, a trained clinician, and an invasive exam, but also may not be available in all settings. The article then proceeds to compare two types of self-screening to samples collected by clinicians in the conventional fashion. It was generally well organized, with few exceptions.

2. The idea is novel, in that there are few published articles about using menstrual blood as a means of screening for HR-HPV. The authors state their conclusions and limitations, about which I have made a few specific comments, but do not overstate most of the conclusions.

3. If self-collected menstrual blood were found to be comparable to testing with conventional clinician collected samples, this could be of huge benefit in places without a clinician or without a nearby lab, as the samples can be shipped and stored without special packaging or handling. A potential issue might be the acceptability to diverse populations. Though menses is universal, cultural beliefs about handling and shipping part of a menstrual pad may vary.

4. There are a few points in the manuscript which are unclear or repetitive, and these are mentioned in the detailed comments.

5. Overall, the manuscript would benefit from attention to style. For example, sometimes it is HR-HPV and sometimes, this is written as HRHPV. Also, the style describing (abbreviations, spelled out) which of the three sample types is being discussed varies throughout the manuscripts, but is generally easier to understand when not abbreviated.

Line 61: This sentence does state that menstrual blood has been used for TSH and A1C, but the mention of the Q-Pad in the middle of the sentence was initially confusing. Perhaps it would be made clearer to write that menses is comparable to serum.
Line 74: The mention of other collection methods is redundant, as this has also been stated in Line 50.
Line 105: This line mentions "a laboratory with standard laboratory equipment" and feels out of place. However, the laboratory methods are then described in great detail, and some of these details may be excessive to readers of the journal.
Line 129: If this section is not revised, this line has a missing word.
Line 147: This line mentions those with biopsies, and one can extrapolate that these participants had biopsies after being found to have HPV, but it is not entirely clear.

Line 167: The positive Q-Pads are described as true positives, with the clinician collected samples then noted to be false negatives. More about this in the discussion.
Line 170: This sentence states that 94% of subjects preferred the Q-Pad but fails to mention the high attrition, which may suggest that the subjects did not like the Q-Pad.
Line 214 and on: This paragraph offers several reasons that the clinician collected sample could be negative and the Q-Pad positive. This is after offering the clinician collected sample as the gold standard and calculating the sensitivity and specificity of the patient collected samples as compared to this gold standard. It is then interesting that the sensitivity and specificity of the clinician samples is not discussed, nor is the possibility of false positive results from the Q-Pad or DBS samples explored, as this would have consequences in use and if present, would warrant further study. Despite not discussing the possibility of false positive results, the following paragraph discusses the need to optimize the Q-Pad analysis.
Line 220: There is a sentence fragment.
Line 228: It is unclear what it would mean to "submit" the entire cervix for culture.
Line 245: As mentioned previously, it may be an overstatement that women found it preferable due to the large number who did not return the Q-Pads.

Table 2 has inconsistent formatting, with a different n for each row, and some missing values. There might be a simpler way to organize the different groups to make it easier to follow.

STATISTICS EDITOR COMMENTS:

Table 1: Should compare those evaluated vs those not evaluated with stats to assure the reader that the evaluable group is representative.

Table 2 and lines 23-28: Need to include 95% CIs for the concordance and the 100% agreement. Also, need to clearly format the CIs as representing 95% CIs.

Table 2: Should expand the comparisons to include metrics beyond agreement, ie, sensitivity, specificity with respective CIs.

EDITORIAL OFFICE COMMENTS:

1. If your article is accepted, the journal will publish a copy of this revision letter and your point-by-point responses as supplemental digital content to the published article online. You may opt out by writing separately to the Editorial Office at em@greenjournal.org, and only the revision letter will be posted.

2. When you submit your revised manuscript, please make the following edits to ensure your submission contains the required information that was previously omitted for the initial double-blind peer review:
* Funding information (ie, grant numbers or industry support statements) should be disclosed on the title page and at the end of the abstract. For industry-sponsored studies, describe on the title page how the funder was or was not involved in the study.
* Include clinical trial registration numbers, PROSPERO registration numbers, or URLs at the end of the abstract (if applicable).
* Name the IRB or Ethics Committee institution in the Methods section (if applicable).
* Add any information about the specific location of the study (ie, city, state, or country), if necessary for context.

3. Obstetrics & Gynecology's Copyright Transfer Agreement (CTA) must be completed by all authors. When you uploaded your manuscript, each coauthor received an email with the subject, "Please verify your authorship for a submission to Obstetrics & Gynecology." Please ask your coauthor(s) to complete this form, and confirm the disclosures listed in their CTA are included on the manuscript's title page. If they did not receive the email, they should check their spam/junk folder.

Requests to resend the CTA may be sent to em@greenjournal.org.

4. For studies that report on the topic of race or include it as a variable, authors must provide an explanation in the manuscript of who classified individuals' race, ethnicity, or both, the classifications used, and whether the options were defined by the investigator or the participant. In addition, describe the reasons that race and ethnicity were assessed in the Methods section and/or in table footnotes. Race and ethnicity must have been collected in a formal or validated way. If it was not, it should be omitted. Authors must enumerate all missing data regarding race and ethnicity as in some cases missing data may comprise a high enough proportion that it compromises statistical precision and bias of analyses by race.

Use "Black" and "White" (capitalized) when used to refer to racial categories.

List racial and ethnic categories in tables in alphabetic order. Do not use "Other" as a category; use "None of the above" instead.

Please refer to "Reporting Race and Ethnicity in Obstetrics & Gynecology" at https://edmgr.ovid.com/ong/accounts/Race_and_Ethnicity.pdf.

5. Figure 1: okay. Figure 2: Please provide letter of permission to use in print and online formats : Has this been previously published in another source? If yes, both print and electronic (online) rights must be obtained from the holder of the copyright (often the publisher, not the author), and credit to the original source must be included in your manuscript. Many publishers have online systems for submitting permissions requests; please consult the publisher directly for more information.

6. Responsible reporting of research studies, which includes a complete, transparent, accurate and timely account of what was done and what was found during a research study, is an integral part of good research and publication practice and not an optional extra. Obstetrics & Gynecology supports initiatives aimed at improving the reporting of health research, and we ask authors to follow specific guidelines:

CHEERS: economic evaluations of health interventions
CHERRIES: studies reporting results of Internet e-surveys
CONSERVE: reporting trial protocols and completed trials modified due to the COVID-19 pandemic and other extenuating circumstances
CONSORT: randomized controlled trials
MOOSE: meta-analyses and systematic reviews of observational studies
PRISMA: meta-analyses and systematic reviews of randomized controlled trials
PRISMA for harms: PRISMA for harms
RECORD: observational studies using ICD-10 data
STARD: studies of diagnostic accuracy
STROBE: observational studies
SQUIRE 2.0: quality improvement in health care studies

Include the appropriate checklist for your manuscript type upon submission, if applicable, and indicate in your cover letter which guideline you have followed. Please write or insert the page numbers where each item appears in the margin of the checklist. Further information and links to the checklists are available at www.equator-network.org/.

7. Standard obstetric and gynecology data definitions have been developed through the reVITALize initiative, which was convened by the American College of Obstetricians and Gynecologists and the members of the Women's Health Registry Alliance. Obstetrics & Gynecology has adopted the use of the reVITALize definitions. Please access the obstetric data

definitions at https://www.acog.org/practice-management/health-it-and-clinical-informatics/revitalize-obstetrics-data-definitions and the gynecology data definitions at https://www.acog.org/practice-management/health-it-and-clinical-informatics/revitalize-gynecology-data-definitions. If use of the reVITALize definitions is problematic, please discuss this in your point-by-point response to this letter.

8. Make sure your manuscript meets the following word limit. The word limit includes the précis, abstract, text, tables, boxes, and figure legends, but excludes the title page, reference list, and supplemental digital content. Figures are not included in the word count.

Research Letters: 600 words (do not include more than two figures and/or tables [2 items total])

9. Specific rules govern the use of acknowledgments in the journal. Please review the following guidelines and edit your title page as needed:

* All financial support of the study must be acknowledged.
* Any and all manuscript preparation assistance, including but not limited to topic development, data collection, analysis, writing, or editorial assistance, must be disclosed in the acknowledgments. Such acknowledgments must identify the entities that provided and paid for this assistance, whether directly or indirectly.
* All persons who contributed to the work reported in the manuscript, but not sufficiently to be authors, must be acknowledged. Written permission must be obtained from all individuals named in the acknowledgments, as readers may infer their endorsement of the data and conclusions. Please note that your response in the journal's electronic author form verifies that permission has been obtained from all named persons.
* If all or part of the paper was presented at the Annual Clinical and Scientific Meeting of the American College of Obstetricians and Gynecologists or at any other organizational meeting, that presentation should be noted (include the exact dates and location of the meeting or indicate whether the meeting was held virtually).
* If your manuscript was uploaded to a preprint server prior to submitting your manuscript to Obstetrics & Gynecology, add the following statement to your title page: "Before submission to Obstetrics & Gynecology, this article was posted to a preprint server at: [URL]."
* Do not use only authors' initials in the acknowledgement or Financial Disclosure; spell out their names the way they appear in the byline.

10. Provide a short title of no more than 45 characters, including spaces, for use as a running foot. Do not start the running title with an abbreviation.

11. Be sure that each statement and any data in the abstract are also stated in the body of your manuscript, tables, or figures. Statements and data that appear in the abstract must also appear in the body text for consistency. Make sure there are no inconsistencies between the abstract and the manuscript, and that the abstract has a clear conclusion statement based on the results found in the manuscript.

In addition, the abstract length should follow journal guidelines. Please provide a word count.

Research Letter: 125 words

12. Only standard abbreviations and acronyms are allowed. A selected list is available online at http://edmgr.ovid.com/ong/accounts/abbreviations.pdf. Abbreviations and acronyms cannot be used in the title or précis. Abbreviations and acronyms must be spelled out the first time they are used in the abstract and again in the body of the manuscript.

13. The journal does not use the virgule symbol (/) in sentences with words, except with ratios. Please rephrase your text to avoid using "and/or," or similar constructions throughout the text. You may retain this symbol if you are using it to express data or a measurement.

14. In your abstract, manuscript Results sections, and tables, the preferred citation should be in terms of an effect size, such as odds ratio or relative risk or the mean difference of a variable between two groups, expressed with appropriate confidence intervals. When such syntax is used, the P value has only secondary importance and often can be omitted or noted as footnotes in a Table format. Putting the results in the form of an effect size makes the result of the statistical test more clinically relevant and gives better context than citing P values alone.

Please standardize the presentation of your data throughout the manuscript submission. For P values, do not exceed three decimal places (for example, "P = .001").

Express all percentages to one decimal place (for example, 11.1%"). Do not use whole numbers for percentages.

15. Please review the journal's Table Checklist to make sure that your tables conform to journal style. The Table Checklist is available at http://edmgr.ovid.com/ong/accounts/table_checklist.pdf.

16. Please review examples of our current reference style at https://edmgr.ovid.com/ong/accounts/ifa_suppl_refstyle.pdf. Include the digital object identifier (DOI) with any journal article references and an accessed date with website references.

Unpublished data, in-press items, personal communications, letters to the editor, theses, package inserts, submissions, meeting presentations, and abstracts may be included in the text but not in the formal reference list. Please cite them on the line in parentheses.

If you cite ACOG documents in your manuscript, be sure the references you are citing are still current and available. Check the Clinical Guidance page at https://www.acog.org/clinical (click on "Clinical Guidance" at the top). If the reference is still available on the site and isn't listed as "Withdrawn," it's still a current document. In most cases, if an ACOG document has been withdrawn, it should not be referenced in your manuscript.

Please make sure your references are numbered in order of appearance in the text.

17. Figure 1: okay
Figure 2: Please provide letter of permission to use in print and online formats.

18. Authors whose manuscripts have been accepted for publication have the option to pay an article processing charge and publish open access. With this choice, articles are made freely available online immediately upon publication. An information sheet is available at http://links.lww.com/LWW-ES/A48. The cost for publishing an article as open access can be found at https://wkauthorservices.editage.com/open-access/hybrid.html.

If your article is accepted, you will receive an email from the Editorial Office asking you to choose a publication route (traditional or open access). Please keep an eye out for that future email and be sure to respond to it promptly.

***

If you choose to revise your manuscript, please submit your revision through Editorial Manager at http://ong.editorialmanager.com. Your manuscript should be uploaded as a Microsoft Word document. Your revision's cover letter should include a point-by-point response to each of the received comments in this letter. Do not omit your responses to the EDITOR COMMENTS (if applicable), the REVIEWER COMMENTS, the STATISTICAL EDITOR COMMENTS (if applicable),

or the EDITORIAL OFFICE COMMENTS.

If you submit a revision, we will assume that it has been developed in consultation with your coauthors and that each author has given approval to the final form of the revision.

Again, your manuscript will be maintained in active status for 21 days from the date of this letter. If we have not heard from you by Apr 26, 2022, we will assume you wish to withdraw the manuscript from further consideration.

Sincerely,

John O. Schorge, MD
Deputy Editor, Gynecology

2020 IMPACT FACTOR: 7.661
2020 IMPACT FACTOR RANKING: 3rd out of 83 ob/gyn journals

_____

Dear Dr. Schorge,

We are pleased to provide a revised manuscript of our submission ONG-22-429. We believe we have addressed all the reviewers' and editors' comments, as well as those of the statistician and the editorial staff. Our responses are provided at the end of every reviewer's comment. We have also submitted a track-changed version of the manuscript as well as revised Tables and Figures. Importantly, per our correspondence with the journal, this revised manuscript is being submitted as an original research paper and not as a research letter.

Thank you for the opportunity to re-submit this manuscript.

Sincerely

P. D. Blumenthal, MD, MPH
Professor of Obstetrics and Gynecology, Emeritus
Stanford University

Responses to reviewers, editors and statistician

RE: Manuscript Number ONG-22-429

Screening for high-risk human papilloma virus using passive self-collected menstrual blood

The revised manuscript should indicate the position of all changes made. Please use the "track changes" feature in your document (do not use strikethrough or underline formatting). Done.

Your submission will be maintained in active status for 21 days from the date of this letter. If we have not heard from you by **Apr 26, 2022**, we will assume you wish to withdraw the manuscript from further consideration.

EDITORS COMMENTS:

We would welcome your resubmission after responding to the reviewer comments. However, you rightly describe this in the manuscript text as a 'pilot study' which in the Info for Authors would necessitate that you **edit this to a Research Letter category**. Thank you!
Per our correspondence with the journal this revised manuscript is being submitted as an original research paper and not as a research letter.

REVIEWER COMMENTS:

Reviewer #1:

The authors present a prospective observational study comparing q-pad menstrual blood analysis to office testing for HRHPV. The results demonstrate high concordance between the two tests, which is very promising, as the qpad seems to be non-invasive and cost-effective (it would be useful to discuss cost of this tool in the paper as the goal seems to be to deploy it in resource rich settings.) Overall, the design is appropriate and the results could be an important contribution to existing literature to make HPV testing available to the global majority.

Abstract:

line 9: do you mean "HR-HPV screening is a potential..." Yes thank you for pointing this out. Has been added.
line 23-24: "HR-HPV positive samples where the CCS to QPS interval (Q-Pad use) was < 2 month..." this is confusing. We have edited this to be more clear.

Introduction:
line 46-47: "HR-HPV screening is conventionally performed in the clinic and requires women to undergo a gynecological exam, which is invasive, uncomfortable, and resource intense", this needs citation and a bit more elaboration, **particularly about economic costs of this test and limitation of use in resource-poor settings** Thank you for your comment. We have added references regarding the relative invasiveness, discomfort and anxiety related to pelvic exams. A discussion about cost and potentially cost-effectiveness is beyond the scope of this paper which is primarily focused on establishing the potential utility of menstrual blood in this setting. Addressing resource-poor settings is also something we plan to investigate in future studies but, eliminating the need for a clinic room, a clinician, and all the accessories associated with clinical specimen collection would seem to logically reduce resource requirements.

line 55-59: the description of the QPad can be placed in the methods section, and instead you could just state that utility of a non-invasive, simple and cheap test could improve HR-HPV screening. Thank you for this comment. We have revised the manuscript to make it more streamlined and less redundant.
line 73: "We found favorable participation in community based screening" was this in a previous study or focus group? please specify This was in a previous study but since this was unpublished we have removed this section.
line 74-76: seems out of place here, maybe can be placed in paragraph above describing the q-pad briefly, or in discussion/conclusion Thank you for this comment. We have moved this section to a point in the manuscript where we describe the Q-Pad.

Methods:
line 93: what do you mean by "regularly menstruate", is it those with regular menses; did you then exclude anyone with menstrual disorders, what about those with dysmenorrhea and PMDD who may have a different concentration of inflammatory markers compared to those without menstrual disorders ? please clarify. We have edited this to say regular menses. Our aim was to include women who were likely to menstruate within a few weeks to a month of having their pap-smear in order to get a Q-Pad specimen back in a timely manner. Women with menstrual disorders such as dysmenorrhea or PMDD were not excluded but we were also not looking for inflammatory markers in this study, only for HR-HPV.

line 98: I am not able to discern the rationale for including the self-collected swab, the authors make valid points about the limitations of the test, and in my opinion it is not useful to compare the self-swab to the qpad since the standard test is clinician-collected, and comparing that to the qpad is sufficient to demonstrate utility; please provide clarification on why the self-swab was included as part of this study please include information in methods about statistical analysis. We thank the reviewer for this comment. We included self-swab specimens because there is a lot of interest in self-swab specimens as a potential future screening approach and we wanted to compare results of selv-swab specimens to menstrual specimens both in terms of accuracy as well as acceptability. Given the above mentioned interest in self-swabbing it is important to provide this comparison in addition to comparison with the current conventional clinician collect specimen.

line 161: please provide the total number of participants who could not provide samples within 2 months We have edited this to be more clear and added a specific number.

line 167-168: for the presence of HR-HPV on the QPad, was there confirmatory testing done (either repeat pap/hr-hpv or colposcopy)? if yes, please provide results, if not please explain why not, as the assumption that the clinician test was a false negative and not that the qpad result was a false positive seems biased. Thank you for this comment. We did not perform confirmatory tests after obtaining the COBAS test results from both the clinician collected specimen, the self-vaginal-swab and the Q-Pad results. To be sure we had considered clinician collected specimens to be a reference standard and were surprised by the Q-Pad and the self-vaginal-swabs results that were positive when the clinician collected swabs were negative. In the case of the self-vaginal-swab it's certainly possible that the swab was picking up HR-HPV in the vagina which we also comment on in the discussion. But there were still an additional 8 Q-Pad test that were positive when both the self-vaginal-swab and clinician collected specimen was negative. This led us to at least hypothesize that the Q-Pad could be picking up HR-HPV from a point higher in the cervix (but through which menstrual blood flows) that was not sampled by the clinician, especially given the relatively high proportion of nulliparous patients. We have attempted to add clarifying points to this in the discussion.

Discussion:
line 199-202: discussion of the number of people who had a delay in submitting qpad results is better suited to the Result section; please provide more detail about spontaneous viral clearance of HR-HPV; did the patients who had a +HRHPV result undergo additional testing to confirm resolution? without confirmation, it is not reasonable to assume that the qpad result indicates clearance rather than a false

negative Thank you for this comment. We hope we have made it clear that we are not assuming that viral clearance occurred but only that it was a possible explanation for our results. With respect to the effect of the COVID Pandemic on our study we do mention in the result section however we thought its impact on the study was potentially significant enough that we felt it important to mention it again in the discussion. Unfortunately we did not have the opportunity to do repeat testing to specifically confirm the hypothesis about viral clearance. We acknowledge this as a limitation in the discussion.

Reviewer #2:

Thank you for allowing me the opportunity to review "Screening for high risk HPV using passive self-collected menstrual blood" presented for possible publication. In this manuscript, the authors present a pilot study of a new technology, The QPad. This new pad includes a paper-based, dried blood spot (DBS) strip which allows for self-collection of menstrual blood that can be used to screen for high risk types of HPV. The HPV test is the Roche Cobas 4800, which is one of two FDA approved testing platforms that allows for primary HPV testing.

The article is timely. There are currently no US FDA approved devices for HR HPV self-collection despite a growing body of research on this technology. There is a lot of interest in this topic as cervical cancer screening, in this country and worldwide, has become a disease of access. **Approximately 50% of patients diagnosed with cervical cancer were not screened in the preceding 5 years. Increasing availability of testing to unreached populations is going to be instrumental (along with increased HPV vaccine uptake) to reducing cervical cancer, a preventable disease.**

The authors report that collection through menstrual blood was acceptable to patients (over self collection) and with a high concordance to clinician sampling results. They also report a sensitivity (of HR HPV with clinician collected as reference) of 94% and sensitivity of 82%. Generally sensitivity and specificity are reported in relation to HSIL but the authors only had 7 patients with CIN 2+ in this pilot study.

Some specific comments below:

Introduction:

- (Line 36): Cervical cancers are preventable with a combination of screening and primary prevention. Pap testing is important but is only one step in the prevention of cervical cancer. We could not agree more and inserted a mention of primary prevention as well as secondary prevention.
- (Line 45): HR HPV is not poised to become a primary screening tool, it already is. From the most recent ACS guidelines, primary HPV screening is the preferred method of testing. ACOG and ASCCP support both primary HR HPV screening and co-testing. Thank you for this comment. We appreciate that HR-HPV screening is increasingly prevalent as a primary test and is recommended by a number of societies and guidelines. However it is not accurate to say that it is the primary screening clinical settings even in the United States. We have revised the paper to reflect what we feel is the current reality.

- (Line 71-73): Citing unpublished and anecdotal evidence is not best practice. The authors are not proposing a point of care (POC) test so I'm not sure this statement is even necessary. We realize this is not best practice and have removed the sentence.
- (Line 76) - "Women's physical" is a misleading term. A physical is much more than just a pap test. This term underrepresents what is done at a "physical exam." We have revised the sentence to be more precise.

Methods
- (Line 92): Why were women under age 21 recruited? The demographic table at the end suggests that no one under 21 yo participated. Further, the demographic table includes women up to age 49, but in the methods section, only women up to age 45 were recruited. This seems to be a discrepancy. Thank you for this comment. With respect to lower age eligibility we realize that routine screening of women under age 21 is uncommon and not generally recommended. However in our case we were assessing the utility of menstrual blood for screening purposes and thus felt it justified to deviate from standard guidelines. That said it turned that no one under 21 happened to present to our clinic for follow-up of positive HR-HPV or for a routine exam during the recruitment period. With the upper age limit we have corrected this error in the manuscript to be consistent with the inclusion criteria approved by the IRB.

- Was any other demographic data collected? This is a significant limitation and should at least be addressed in the discussion session if no other information was collected. Things such as history of HSIL and/or immunocompromise would increase risk of persistent HR HPV infection and may alter results (especially in those for whom more than 2 months passed from clinician sampling before menstrual blood sampling). We appreciate this comment. Table 1 displays the variety of demographic data we were able to collect which is similar to other studies of this type. We initially sought women

who were known to be HR-HPV positive for this study and eventually elected to also include women of unknown current HR-HPV status and known history of HR-HPV positive results in the past. We reported in the results section that 66% of our population had a history of HR-HPV positive results in the past. That said, the objective of the study was simply to assess whether HR-HPV could be detected in a menstrually derived DBS sample and not whether it was predictive of or related to the presence of dysplasia. With respect to the issue of immunocompromise or other potential confounders that might have prevented HR-HPV clearance over time we did not observe this phenomenon but rather the reverse in this generally health population specifically that the longer the interval between clinician collected pap smear and Q-pad collection the correlation decreased. To our knowledge none of the patients in our study were immunocompromised.

- Presumably all of these patients have a cervix? It should just be clear in the inclusion criteria. We have revised the manuscript to reflect the fact all participants were cervix owners.
- Is there any data on stability of the testing available from the manufacturer of the strip? An average of 10 days passed, but there is no range provided. Were any samples held for prolonged periods of time? Any relation to longer held samples and false negative or false positive results? Unfortunately the laboratory did not record this and thus we are unable to correlate the specimen processing interval once it got the laboratory with the results. However it is well established in the literature that DBS samples are stable well beyond 2-3 months when it comes to DNA detection (i.e. HR-HPV). We have added a reference (Aitken S 2015 [PMID: 26147689]) about this in the manuscript.
- Was any genotyping performed or is it possible to do this? The Cobas test does offer genotype screening which becomes important when addressing the clinical application of this technology. Thanks for this comment. First, genotyping is possible from this type of specimen and we have those results because, as the reviewer may know, the COBAS analyzer reports on this automatically. We did not report the results of genotyping for purposes of brevity and also because our primary objective was just to assess the feasibility of deriving HR-HPV results of any type for this kind of specimen. Should the use of this technology become more common it will be possible to report on genotype as for any other COBAS processed specimen.

Results

- Almost 50 patients (approximately 30% of patients) did not complete the study. This should be addressed as another limitation in the discussion. We appreciate this comment and have addressed this in the limitations section of the discussion. That said, it is important to know that there were no significant demographic differences between those who were evaluated and those who were enrolled.

Discussion
- (Line 217): It is certainly possible that the HPV was higher in the cervical canal which is why it was missed on clinician sampling. Further, consider the fact that HPV generally is a field effect and may infect the vulva, vagina and anus. Is it possible that the menstrual blood pad may be falsely positive for HPV if the HPV is present in the anal canal? Thank you for this comment. We amended the manuscript to reflect this possibility. However the fluid mechanics of the Q-Pad (the strip is located under a top sheet and for sample to be collected on strip it would require fluid to soak through the top sheet first) and the location of the strip relative to the anus would appear to make this very unlikely.

Reviewer #3:

1. This manuscript describes a novel collection method to collect blood to screen for high-risk HPV, and explains why HPV screening is important and gives the sense that, though this is not yet the current practice, HPV testing alone could become the screening method in use, replacing cytology. The argument for a self-collected, non-invasive screening method is provided, as screening with a clinician involves time, a trained clinician, and an invasive exam, but also may not be available in all settings. The article then proceeds to compare two types of self-screening to samples collected by clinicians in the conventional fashion. It was generally well organized, with few exceptions.

2. The idea is novel, in that there are few published articles about using menstrual blood as a means of screening for HR-HPV. The authors state their conclusions and limitations, about which I have made a few specific comments, but do not overstate most of the conclusions.

3. If self-collected menstrual blood were found to be comparable to testing with conventional clinician collected samples, this could be of huge benefit in places without a clinician or without a nearby lab, as the samples can be shipped and stored without special packaging or handling. A potential issue might be the acceptability to diverse populations. Though menses is universal, cultural beliefs about handling and shipping part of a menstrual pad may vary.

4. There are a few points in the manuscript which are unclear or repetitive, and these are mentioned in the detailed comments.

5. Overall, the manuscript would benefit from attention to style. For example, sometimes it is HR-HPV and sometimes, this is written as HRHPV. Also, the style describing

(abbreviations, spelled out) which of the three sample types is being discussed varies throughout the manuscripts, but is generally easier to understand when not abbreviated. We have attempted to fix stylistic inconsistencies in the manuscript and thank you for these observations. We have removed the abbreviations related to specimen collection types in the main text and abstract.

Line 61: This sentence does state that menstrual blood has been used for TSH and A1C, but the mention of the Q-Pad in the middle of the sentence was initially confusing. Perhaps it would be made clearer to write that menses is comparable to serum. We attempted to clarify and reduce confusion.
Line 74: The mention of other collection methods is redundant, as this has also been stated in Line 50. We apologize but could not find the potentially redundant mention of other collection methods to which the reviewer refers.
Line 105: This line mentions "a laboratory with standard laboratory equipment" and feels out of place. However, the laboratory methods are then described in great detail, and some of these details may be excessive to readers of the journal. Thank you for this comment. The line about standard laboratory equipment has been removed.
Line 129: If this section is not revised, this line has a missing word. We have completed this sentence.
Line 147: This line mentions those with biopsies, and one can extrapolate that these participants had biopsies after being found to have HPV, but it is not entirely clear. Thank you for this comment. We have clarified when the biopsy was taken relative to participation in our study.
Line 167: The positive Q-Pads are described as true positives, with the clinician collected samples then noted to be false negatives. More about this in the discussion. Thank you for this comment. Reviewer 1 asked a similar question. We have added more information about this in the discussion. To be sure, we had considered clinician collected specimens to be a reference standard and were surprised by the Q-Pad and the self-vaginal-swab results that were positive when the clinician collected swabs were negative. In the case of the self-vaginal-swab it's certainly possible that the swab was picking up HR-HPV in the vagina which we also comment on in the discussion. But there were still an additional 8 Q-Pad test that were positive when both the self-vaginal-swab and clinician collected specimen was negative. This led us to at least hypothesize that the Q-Pad could be picking up HR-HPV from a point higher in the cervix (but through which menstrual blood flows) that was not sampled by the clinician, especially given the relatively high proportion of nulliparous patients.

Line 170: This sentence states that 94% of subjects preferred the Q-Pad but fails to mention the high attrition, which may suggest that the subjects did not like the Q-Pad. Thanks for this comment. First we hope it is clear that the 94% only refers to those who

completed the study. With respect to the high attrition rate it is theoretically possible that those who did not return the Q-Pad were “voting with their feet”, indicating possible unacceptability of the collection system. However, given that the COVID-19 pandemic interceded during the course of this study it is more likely that pandemic related reasons were the cause of loss to follow-up and not displeasure or low acceptability of study technology. For example we know that a number of Q-Pad specimens were never delivered to us because the Stanford mail room was not receiving mail for 3 months of the study period. Further, given the demographic similarities between those who enrolled and those evaluated it seems unlikely that the Q-Pad would have been highly unacceptable among those who did not complete the study.

Line 214 and on: This paragraph offers several reasons that the clinician collected sample could be negative and the Q-Pad positive. This is after offering the clinician collected sample as the gold standard and calculating the sensitivity and specificity of the patient collected samples as compared to this gold standard. It is then interesting that the sensitivity and specificity of the clinician samples is not discussed, nor is the possibility of false positive results from the Q-Pad or DBS samples explored, as this would have consequences in use and if present, would warrant further study. Despite not discussing the possibility of false positive results, the following paragraph discusses the need to optimize the Q-Pad analysis. The reviewer brings up a very interesting point. We certainly acknowledge that it is possible that the self-vaginal swab and the Q-Pad are picking up vaginal or even vulvar HR-HPV when the clinician obtained cervical specimen did not. We acknowledge this possibility in the discussion. As we also mention in the discussion, the COBAS analyzer is very unlikely to produce a positive HR-HPV result when there is no HR-HPV present which brings up the possibility that we also discussed sampling error for the clinician collected swab. Historically when a pap smear was negative but the patient was shown to have cervical dysplasia or worse it was not uncommon for pathologists to say that the pap smear was not wrong but that the clinician had incompletely sampled the cervix. With respect to the issue of reference standards we were careful to say “**If** the test performed on the clinical collected swab is considered as a reference standard…” because while we are daily certain that the COBAS analyzer is accurate we are not convinced that the human error potentially associated with clinician collected swabs is equally infallible. That is why we indicated that the real reference standard here would be viral culture of the cervix. We have added more language in the discussion to try to clarify this.

Line 220: There is a sentence fragment. Thank you for this comment. This has been revised in the manuscript.

Line 228: It is unclear what it would mean to "submit" the entire cervix for culture. We have tried to clarify this in the discussion.
Line 245: As mentioned previously, it may be an overstatement that women found it preferable due to the large number who did not return the Q-Pads. We have addressed this point again at the very end of the discussion but also in the limitation section. See also our response to your query from line 170.

Table 2 has inconsistent formatting, with a different n for each row, and some missing values. There might be a simpler way to organize the different groups to make it easier to follow. We have tried to improve the formatting of the table consistent with suggestions from the editors. We do not see a way to avoid having different n for each row because they represent different group sizes.

STATISTICS EDITOR COMMENTS:

Table 1: Should compare those evaluated vs those not evaluated with stats to assure the reader that the evaluable group is representative. Thank you for your comment. We have added p-values to the table.

Table 2 and lines 23-28: Need to include 95% CIs for the concordance and the 100% agreement. Also, need to clearly format the CIs as representing 95% CIs. Thank you for your comment. We have shown 95% CIs for concordance and 100% agreement in table 2 and have indicated in the titel of the table that the data are shown as percent "Percent agreement (95% CI)". We have also added the 95% CI's in the old line 23-28 (now line 30). We were uncertain as to whether 95% CI was desired for the simple proportions relating to acceptability.

Table 2: Should expand the comparisons to include metrics beyond agreement, ie, sensitivity, specificity with respective CIs. We focus on the levels of agreement because, in this case, we were uncertain whether another screening method (e.g. clinician collected swab) could really be considered a reference standard against which sensitivity and specificity could be measured. In the discussion we mention the possibility of interpreting our data as if the clinician collected swab was in fact a reference standard but specifically did not include this table 2 because the paper is really about concordance. In the text were we do calculate an hypothetical sensitivity and specificity we do provide 95% CIs.

EDITORIAL OFFICE COMMENTS:

1. If your article is accepted, the journal will publish a copy of this revision letter and your point-by-point responses as supplemental digital content to the published article online. You may opt out by writing separately to the Editorial Office at em@greenjournal.org, and only the revision letter will be posted. Noted. We have no problem with this policy.

2. When you submit your revised manuscript, please make the following edits to ensure your submission contains the required information that was previously omitted for the initial double-blind peer review:

* Funding information (ie, grant numbers or industry support statements) should be disclosed on the title page and at the end of the abstract. For industry-sponsored studies, describe on the title page how the funder was or was not involved in the study. Noted and included in the title page and abstract.
* Include clinical trial registration numbers, PROSPERO registration numbers, or URLs at the end of the abstract (if applicable). Noted and included at the end of the abstract.
* Name the IRB or Ethics Committee institution in the Methods section (if applicable). Noted and present in manuscript.
* Add any information about the specific location of the study (ie, city, state, or country), if necessary for context. Noted and present in manuscript.

3. Obstetrics & Gynecology's Copyright Transfer Agreement (CTA) must be completed by all authors. When you uploaded your manuscript, each coauthor received an email with the subject, "Please verify your authorship for a submission to Obstetrics & Gynecology." Please ask your coauthor(s) to complete this form, and confirm the disclosures listed in their CTA are included on the manuscript's title page. If they did not receive the email, they should check their spam/junk folder. Requests to resend the CTA may be sent to em@greenjournal.org.
We believe this is completed.

4. For studies that report on the topic of race or include it as a variable, authors must provide an explanation in the manuscript of who classified individuals' race, ethnicity, or both, the classifications used, and whether the options were defined by the investigator or the participant. In addition, describe the reasons that race and ethnicity were assessed in the Methods section and/or in table footnotes. Race and ethnicity must have been collected in a formal or validated way. If it was not, it should be omitted.

Authors must enumerate all missing data regarding race and ethnicity as in some cases missing data may comprise a high enough proportion that it compromises statistical precision and bias of analyses by race. Race is not variable in this study.

Use "Black" and "White" (capitalized) when used to refer to racial categories. Done.

List racial and ethnic categories in tables in alphabetic order. Do not use "Other" as a category; use "None of the above" instead. Done.

Please refer to "Reporting Race and Ethnicity in Obstetrics & Gynecology" at https://edmgr.ovid.com/ong/accounts/Race_and_Ethnicity.pdf.

5. Figure 1: okay. Figure 2: Please provide letter of permission to use in print and online formats : Has this been previously published in another source? If yes, both print and electronic (online) rights must be obtained from the holder of the copyright (often the publisher, not the author), and credit to the original source must be included in your manuscript. Many publishers have online systems for submitting permissions requests; please consult the publisher directly for more information. The figure has been modified from our original submission to ensure originality. This is original art and has not been published elsewhere. A letter of permission to use the figure accompanies the revised manuscript.

6. Responsible reporting of research studies, which includes a complete, transparent, accurate and timely account of what was done and what was found during a research study, is an integral part of good research and publication practice and not an optional extra. Obstetrics & Gynecology supports initiatives aimed at improving the reporting of health research, and we ask authors to follow specific guidelines:

CHEERS: economic evaluations of health interventions
CHERRIES: studies reporting results of Internet e-surveys
CONSERVE: reporting trial protocols and completed trials modified due to the COVID-19 pandemic and other extenuating circumstances
CONSORT: randomized controlled trials
MOOSE: meta-analyses and systematic reviews of observational studies
PRISMA: meta-analyses and systematic reviews of randomized controlled trials
PRISMA for harms: PRISMA for harms
RECORD: observational studies using ICD-10 data
STARD: studies of diagnostic accuracy

STROBE: observational studies
SQUIRE 2.0: quality improvement in health care studies

Include the appropriate checklist for your manuscript type upon submission, if applicable, and indicate in your cover letter which guideline you have followed. Please write or insert the page numbers where each item appears in the margin of the checklist. Further information and links to the checklists are available at www.equator-network.org/.
We have provided a completed STROBE checklist in the revised materials.

7. Standard obstetric and gynecology data definitions have been developed through the reVITALize initiative, which was convened by the American College of Obstetricians and Gynecologists and the members of the Women's Health Registry Alliance. Obstetrics & Gynecology has adopted the use of the reVITALize definitions. Please access the obstetric data definitions at https://www.acog.org/practice-management/health-it-and-clinical-informatics/revitalize-obstetrics-data-definitions and the gynecology data definitions at https://www.acog.org/practice-management/health-it-and-clinical-informatics/revitalize-gynecology-data-definitions. If use of the reVITALize definitions is problematic, please discuss this in your point-by-point response to this letter.
We have reviewed these definitions and do not see any deviation from the reVITALize definitions.

8. Make sure your manuscript meets the following word limit. The word limit includes the précis, abstract, text, tables, boxes, and figure legends, but excludes the title page, reference list, and supplemental digital content. Figures are not included in the word count.

Research Letters: 600 words (do not include more than two figures and/or tables [2 items total])
Per email from editorial staff (04/21/22) we have resubmitted as an original research paper and meet those word limit requirements.

9. Specific rules govern the use of acknowledgments in the journal. Please review the following guidelines and edit your title page as needed:

* All financial support of the study must be acknowledged.

* Any and all manuscript preparation assistance, including but not limited to topic development, data collection, analysis, writing, or editorial assistance, must be disclosed in the acknowledgments. Such acknowledgments must identify the entities that provided and paid for this assistance, whether directly or indirectly.
* All persons who contributed to the work reported in the manuscript, but not sufficiently to be authors, must be acknowledged. Written permission must be obtained from all individuals named in the acknowledgments, as readers may infer their endorsement of the data and conclusions. Please note that your response in the journal's electronic author form verifies that permission has been obtained from all named persons.
* If all or part of the paper was presented at the Annual Clinical and Scientific Meeting of the American College of Obstetricians and Gynecologists or at any other organizational meeting, that presentation should be noted (include the exact dates and location of the meeting or indicate whether the meeting was held virtually).
* If your manuscript was uploaded to a preprint server prior to submitting your manuscript to Obstetrics & Gynecology, add the following statement to your title page: "Before submission to Obstetrics & Gynecology, this article was posted to a preprint server at: [URL]."
* Do not use only authors' initials in the acknowledgement or Financial Disclosure; spell out their names the way they appear in the byline.
We have added an acknowledgement section in the revised manuscript.

10. Provide a short title of no more than 45 characters, including spaces, for use as a running foot. Do not start the running title with an abbreviation.
Menstrual blood for cervical HR-HPV detection.

11. Be sure that each statement and any data in the abstract are also stated in the body of your manuscript, tables, or figures. Statements and data that appear in the abstract must also appear in the body text for consistency. Make sure there are no inconsistencies between the abstract and the manuscript, and that the abstract has a clear conclusion statement based on the results found in the manuscript.

In addition, the abstract length should follow journal guidelines. Please provide a word count.

Research Letter: 125 words
Per email from editorial staff (04/21/22) we have resubmitted as an original research paper and meet those word limit requirements.

12. Only standard abbreviations and acronyms are allowed. A selected list is available online at http://edmgr.ovid.com/ong/accounts/abbreviations.pdf. Abbreviations and acronyms cannot be used in the title or précis. Abbreviations and acronyms must be spelled out the first time they are used in the abstract and again in the body of the manuscript.
This has been done.

13. The journal does not use the virgule symbol (/) in sentences with words, except with ratios. Please rephrase your text to avoid using "and/or," or similar constructions throughout the text. You may retain this symbol if you are using it to express data or a measurement.
We believe we are in compliance.

14. In your abstract, manuscript Results sections, and tables, the preferred citation should be in terms of an effect size, such as odds ratio or relative risk or the mean difference of a variable between two groups, expressed with appropriate confidence intervals. When such syntax is used, the P value has only secondary importance and often can be omitted or noted as footnotes in a Table format. Putting the results in the form of an effect size makes the result of the statistical test more clinically relevant and gives better context than citing P values alone.

Please standardize the presentation of your data throughout the manuscript submission. For P values, do not exceed three decimal places (for example, "P = .001").

Express all percentages to one decimal place (for example, 11.1%"). Do not use whole numbers for percentages.

We believe we are in compliance.

15. Please review the journal's Table Checklist to make sure that your tables conform to journal style. The Table Checklist is available at http://edmgr.ovid.com/ong/accounts/table_checklist.pdf.

We believe we are in compliance.
16. Please review examples of our current reference style at https://edmgr.ovid.com/ong/accounts/ifa_suppl_refstyle.pdf. Include the digital object identifier (DOI) with any journal article references and an accessed date with website references.

Unpublished data, in-press items, personal communications, letters to the editor, theses, package inserts, submissions, meeting presentations, and abstracts may be included in the text but not in the formal reference list. Please cite them on the line in parentheses.

If you cite ACOG documents in your manuscript, be sure the references you are citing are still current and available. Check the Clinical Guidance page at https://www.acog.org/clinical (click on "Clinical Guidance" at the top). If the reference is still available on the site and isn't listed as "Withdrawn," it's still a current document. In most cases, if an ACOG document has been withdrawn, it should not be referenced in your manuscript.

Please make sure your references are numbered in order of appearance in the text.
We believe we are in compliance.

17. Figure 1: okay
Figure 2: Please provide letter of permission to use in print and online formats.
We have provided this letter.

18. Authors whose manuscripts have been accepted for publication have the option to pay an article processing charge and publish open access. With this choice, articles are made freely available online immediately upon publication. An information sheet is available at http://links.lww.com/LWW-ES/A48. The cost for publishing an article as open access can be found at https://wkauthorservices.editage.com/open-access/hybrid.html.

If your article is accepted, you will receive an email from the Editorial Office asking you to choose a publication route (traditional or open access). Please keep an eye out for that future email and be sure to respond to it promptly.

Acknowledged.

***

If you choose to revise your manuscript, please submit your revision through Editorial Manager at http://ong.editorialmanager.com. Your manuscript should be uploaded as a Microsoft Word document. Your revision's cover letter should include a point-by-point response to each of the received comments in this letter. Do not omit your responses to the EDITOR COMMENTS (if applicable), the REVIEWER COMMENTS, the

STATISTICAL EDITOR COMMENTS (if applicable), or the EDITORIAL OFFICE COMMENTS.

If you submit a revision, we will assume that it has been developed in consultation with your coauthors and that each author has given approval to the final form of the revision.

Again, your manuscript will be maintained in active status for 21 days from the date of this letter. If we have not heard from you by Apr 26, 2022, we will assume you wish to withdraw the manuscript from further consideration.

Sincerely,

John O. Schorge, MD
Deputy Editor, Gynecology

2020 IMPACT FACTOR: 7.661
2020 IMPACT FACTOR RANKING: 3rd out of 83 ob/gyn journals