Reviewer's report

Title: Iron-fortified lentils to improve iron (Fe) status among adolescent girls in Bangladesh—study protocol for a double-blind community-based randomized control trial

Version: 0 Date: 11 Nov 2018

Reviewer: Andrew Gray

Reviewer's report:

This manuscript describes the protocol for a cluster randomised controlled trial looking at providing iron-fortified lentils to adolescent (aged 10-17) girls (n=1260) in Bangladesh. Outcomes include iron status (all participants, primary outcome), anthropometry (all participants), and cognitive performance (360 participants, primary outcome).

The study is well motivated and mostly well described, although there are some important omissions around the randomisation procedure, details of blinding, what would count as evidence in favour of the fortified lentils, some details in the sample size information, and the statistical analyses.

Page 2, Line 4: Technically the study protocol describes a study with these aims.

Page 2, Line 4: Are you actually concerned with efficacy here (how well can it work under ideal conditions) or effectiveness (how well does it work under real-world conditions)? If the former, a per-protocol focused analysis would be standard with exclusion of non-compliers, if the latter, an intention to treat focused analysis would be standard with compliance not incorporated into that. Page 13, Line 22 makes it clear there that it is effectiveness you are interested in, and not efficacy.

Page 2, Line 6: "improving" compared to what, ordinary lentils or no lentils?

Page 2, Line 10: The phrase "double blind" can be interpreted in many ways (see http://dx.doi.org/10.1136/ebm.7.1.4) and I think it would be useful to add the groups who will be blinded in parentheses here. Note that SPIRIT item 17a refer to exactly who will be blinded.

Page 2, Line 16: It's not entirely clear what the primary question of interest here is. The description of the no-lentil group as the control would suggest that the key question is around the provision of lentils, but the iron-fortified group suggests to me that this is the key intervention here. Somewhere in the methods in the abstract, the reader should be able to find out what the
primary research question is (are lentils useful for iron and cognition versus does fortifying lentils improve iron and cognition?) Line 40 below suggests that it might be the second of the questions that you are most interested in and this matches the online registration.

Page 2, Lines 19-22: The description of "a locally-acceptable recipe" suggests that monotony might be an issue. Could you add some information to the body of the manuscript covering this aspect?

Page 2, Line 29: While it seems likely that the follow-up will be at 4 mo, this should be explicit.

Page 2, Line 31: While it seems likely that the midpoint interim measurement will be at 2 mo, this should be explicit.

Page 2, Lines 30-35: Is the cognitive testing at the interim or only baseline and follow-up times? (This is later clarified on Page 9, Line 12, but should also be clear here.)

Page 2, Line 34: "carried-out"

Page 3, Lines 2-4: I suggest adding "of" after each of these percentages.

Page 3, Line 8: Suggest "ages" (add "s") here.

Page 4, Lines 10-12: Compared to ordinary lentils or compared to no lentils?

Page 4, Line 14: Same comment as above regarding efficacy/effectiveness.

Page 4, Line 16: Again, improving compared to what?

Page 4, Line 32: Same comment as above regarding efficacy/effectiveness.

Page 4, Line 38: Same comment as above regarding efficacy/effectiveness.
Page 4, Line 41: Suggest "the drop-out" (adding "the"). Also, while the drop-out rate was 0%, were there missed meals during the study? This should be mentioned here.

Page 4, Line 50: How often were the uneaten amounts below the 37.5g dry weight proposed here? Perhaps it would be best to give the amounts consumed for each group as means or medians. Also, just checking, this means that those in the 50g group consumed 12.5g more than those in the 37.5g group (if the uneaten amounts were the same)?

Page 5, Line 2: Same comment as above regarding efficacy/effectiveness.

Page 5, Line 7: Same comment as above regarding blinding.

Page 5, Line 8: Same comment as above regarding efficacy/effectiveness.

Page 5, Line 8: Suggest "consuming" rather than "consumed"

Page 5, Line 14: Cluster RCTs don't eliminate selection biases. These still exist in the selection of the clusters and can arise in the consenting/assenting of participants within clusters (e.g. Page 5, Line 50). The main arguments in favour of cluster randomisation are convenience and minimising contamination, with the price paid for these being reflected in the design effect and consequently larger sample sizes.

Page 5, Line 14: Reliability is not a property of RCTs versus other designs. Reliability refers to whether the measured result would be the same if the measurement was repeated (at the individual or study level). For this at the study level, the sample size is crucial as is the reliability of the measurements at the individual level.

Page 5, Line 22: Same comment as above regarding efficacy/effectiveness.

Page 5, Lines 22-26: I don't think that "BRAC-previously known as 'Bangladesh Rural Advancement Committee' is the #1 non-government organization (NGO) in the world for the third year running [17]." is relevant to the protocol and suggest its deletion. Adding a reference to the previous sentence would be appropriate though.

Page 5, Line 31: I suggest replacing "between ages" with "aged".
Page 5, Lines 46-50: This isn't a complete sentence, perhaps delete "who"?

Page 5, Line 52: Given the clubs are for those aged 10-19 (Line 33 above), presumably those aged 18 or 19 will also be excluded from analyses?

Page 7, Line 54: I think it is a little confusing to say "three lentil-based…arms" when one does not receive lentils.

Page 7, Line 54: Same comment as above regarding efficacy/effectiveness.

Page 7, Line 58: Note again that the no-lentils group would be a control for the lentil-related questions but not the fortification ones. Note also that the point in the online registration that they can still consume lentils is important and should be incorporated here.

Page 8, Line 5: "lentils" (add "s")

Page 8, Line 28: Presumably this means "including an additional 20% to account for loss to follow-up"?

Page 8, Lines 20-40: In order for the calculations here to be replicable, you also need to provide the SD of ferritin, the detectable cognitive effect size, the SD of this effect, and the number of clusters/number of girls per cluster for the cognitive sub-study. With this information, I should be able to produce the same numbers as you have. The number of girls needing to be invited should also incorporate a non-consenting/assenting rate(s). I am assuming that the study is powered for all three pairwise-comparisons? This would raise the issue of multiplicity. It is important that the "success criteria" for the study are made absolutely clear in the manuscript, either in this section or elsewhere. If the study affects Fe status only, will it be declared a success? What if only cognition shows evidence of response? Given that both these are listed as primary outcomes, my expectation is that unless both show evidence of improvement, the study will not be regarded as showing evidence in favour of iron-fortified lentils? This point needs to be made very clear in the protocol and will have implications for adjustments for multiplicity and so sample size.

Page 8, Lines 42-46: Same comment as above regarding blinding.

Page 8, Line 51: If I'm understanding correctly, the fortified and non-fortified lentils will be in different coloured bags but always of the same colour? If so, anyone reading this text would be
unblinded to the intervention (and so the colours must be deleted from the text). Using the same colours at all sites also means that if someone is unblinded at one site, all sites are potentially unblinded. If the colours vary between sites (yellow=fortified for some and non-fortified for others) or over time, that would strengthen this aspect of the design and should be made clear here.

Page 9, Line 2: I'd suggest not capitalising "URGENT" here.

Page 9, Line 58-Page 10, Line 2: Unless this is going to be used to define a criterion for being included in analyses (must consumed a mean of 30g or more, for example), this is not an efficacy trial but is instead a pragmatic effectiveness trial. I think that effectiveness is the better question, but it does mean that the numerous references to efficacy would need to be changed if this is the case (which it does seem to be based on the statistical methods).

Page 10, Line 19: "detailed" (add "ed")

Page 11, Lines 17-20: Presumably this intends to say that cognitive performance data will be collected from a subsample of the girls who provide venous blood?

Page 11, Line 26: Are not the clubs the clusters? If these are different, this wasn't clear to me earlier.

Page 12, Figure 2: Why are cognitive assessments repeated at week 29? The figure also does not show the interim assessments.

Page 13, Lines 7-10: More information is needed around anthropometry. What equipment is being used (stadiometers, scales, tapes), how many measurements will be used, how will these be combined, what is the measurement point for waist circumference? Ideally, quality control should also be discussed here (e.g., inter- and intra-rater reliability).

Page 13: The randomisation method has not yet been described and must be added. See the CONSORT guidelines for examples of the necessary information. The SPIRIT items 16a, 16b, and 16c indicates pages 10 and 11 for these details, but I cannot see them there or elsewhere.

Page 13, Line 22: The clarification here that intention to treat will be used appears to confirm that this is an effectiveness and not an efficacy study.
Page 13, Line 28: You cannot use ANCOVAs or other simple statistical models here that assume independence between observations. Because of the clustering within clubs, you must use other options such as mixed models or generalised estimating equations. Given the interim measurements, these will also need to be incorporated into the analyses. This might be what you are referring to on Line 35 ("multilevel linear regression") but this needs to be clearer (what would be the random and what would be the fixed effects? Is there a residual covariance structure you have in mind for the longitudinal data?)

Page 13, Line 31: "as appropriate" is a very unclear phrase. What would make their inclusion appropriate?

Page 13, Line 33: Again, Chi-squared tests and logistic regression models assume independence and some form of modelling that incorporates the clustering within clubs is needed.

Page 13, Line 33: One of the main strengths of randomisation is that it eliminates confounding. While this can be introduced through differential attrition mechanisms, this needs to be addressed by a formal approach to missing data such as multiple imputation with perhaps selection or pattern-mixture models for informative missingness.

Page 13, Line 37: A brief description of model diagnostics that will be used should be added around here.

Page 13, Line 37: Same comment on confounding as above.

Page 13, Lines 36-39: A complete case analysis is not best practice. I'd strongly suggest that you look at MI and pattern-mixture or selection models to deal with missing data.

Page 13, Line 44: It is traditional to also note the level of significance in this section.

Page 13, Line 50: Suggest deleting "the" before "adolescent girls".

Page 14, Line 36: These days, I think "layperson's" would be more appropriate.
**Level of interest**  
Please indicate how interesting you found the manuscript:

An article whose findings are important to those with closely related research interests

**Quality of written English**  
Please indicate the quality of language in the manuscript:

Acceptable

**Quality of figures**  
All images and figures within the manuscript should be genuine i.e. without evidence of manipulation. No specific feature within an image may be enhanced, obscured, moved, removed, or introduced. If you have concerns about the veracity of the figures you should choose the first option below.

**Statistical review**  
Is it essential that this manuscript is seen by an expert statistician? If so, please give your reasons in your report.

**Declaration of competing interests**  
Please complete a declaration of competing interests, considering the following questions:

1. Have you in the past five years received reimbursements, fees, funding, or salary from an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

2. Do you hold any stocks or shares in an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

3. Do you hold or are you currently applying for any patents relating to the content of the manuscript?

4. Have you received reimbursements, fees, funding, or salary from an organization that holds or has applied for patents relating to the content of the manuscript?

5. Do you have any other financial competing interests?

6. Do you have any non-financial competing interests in relation to this paper?
If you can answer no to all of the above, write 'I declare that I have no competing interests' below. If your reply is yes to any, please give details below.

I declare that I have no competing interests

I agree to the open peer review policy of the journal. I understand that my name will be included on my report to the authors and, if the manuscript is accepted for publication, my named report including any attachments I upload will be posted on the website along with the authors' responses. I agree for my report to be made available under an Open Access Creative Commons CC-BY license (http://creativecommons.org/licenses/by/4.0/). I understand that any comments which I do not wish to be included in my named report can be included as confidential comments to the editors, which will not be published.

I agree to the open peer review policy of the journal

Were you mentored through this peer review?

No