Reviewer's report

Title: Lower Vitamin D Levels in Saudi Pregnant Women are Associated with Higher Risk of Developing GDM

Version: 2 Date: 18 Jul 2017
Reviewer: Kathryn Hart

Reviewer's report:

General:

Minor grammatical issues throughout - suggest thorough grammar review prior to resubmission, for example 'of' used in place of 'in' in several places.

Abstract:

"among vitamin D deficient and normal women" - replace 'normal' with the relevant specific term - assume you mean Vitamin D 'sufficient'? Since there is controversy about what cut offs are used it is really important to be explicit about the status of your participants.

Could you also state how many were below 25 or 30nmol/L as these are also accepted cut offs for deficiency which might be more informative than a 50nmol cut off which is likely to characterise most people as deficient.

Background:

Pg 5, line 41 - 'prevalence of 26-98% in pregnancy' - can you be explicit whether this range of prevalence is based on the same cut off being applied or represents different cut offs in different studies. If the former are these values all based on a deficiency cut off of 50nmol/L?

Methods:

Pg 6, line 9 'have been as described before' - remove word 'as'.

Appreciate that your methods are described elsewhere but some indication of number approached to achieve 515 and the basis for this sample size would be helpful here.
What social or demographic information was collected - will be important when characterising your population, e.g. how was 'Saudi' defined - is this assumed to be an ethnically homogenous group?

Again, given the controversy surrounding different methods for Vitamin D analysis I think this should be clearly stated in the paper rather than referring the reader to a previous paper - a clear statement of analytic method is required to allow for swift assessment of the study.

Results:

Did the approx. 100 women who did not return for OGTT differ in any way from those who did?

Pg 7, line 17 'rest 90 (17.5%) were normal' - rephrase as 'rest 90 (17.5%) were Vitamin D sufficient'

'Based on the recently recognized clinical categories of vitamin D status [20-23], we classified pregnant women having <50 nmol/l as deficient and those having >50 nmol/l as non-deficient/normal' - the references you refer to are original trials and a meta-analysis NOT clinical guidelines. Given the wealth of recommendations (e.g. IOM, EFSA, SACN, Nordic Nutrition) for appropriate cut offs would it be more appropriate to refer to one of these consensus statements as the source of your proposed cut off?

Pg 7, line 22 'The pregnant women visiting the clinics for the second time during their 24-28th week of pregnancy, and whose anthropometric and biochemical characteristics were already available, were subjected to OGTT and results analyzed according to IADPSG criteria as given in methods' - this is not needed in results. Just state return rate/ take up rate for OGTT and how these women differed (if at all) from non-returners (see previous point) and then state the prevalence of GDM.

Line 25 '116 (27.7%) out of 419 women were diagnosed as positive for GDM, while others 303 (72.3%) were normal.' - avoid use of word 'normal' as previously recommended. Remove word 'others' - not needed.

Line 31 'Various anthropometric and biochemical parameters were determined for the pregnant women using blood samples collected during their first visit, as described in methods. From the OGTT results, the pregnant women were categorized as GDM and non-GDM and two were compared for various anthropometric and clinical parameters (Table 1).’ - again this is not required. Repeats method and results section above.
Line 43 'Also, all the obesity indices determined in this study, pre-pregnancy BMI, BMI, waist size, hip size and WHR were significantly higher among the GDM women compared to non-GDM women.' - give summary p values for this group of results, even if you can only say that they were all <0.05

Your adjustment of odds ratios for potential confounders appears appropriate but were you also able to perform multiple regression analysis to assess the relative contributions of the different variables to GDM risk? Were any of the other variables significantly and uniquely associated with GDM as I assume there is considerable overlap between these as you suggested? What would the best predictive model look like?

Table 1 - p values cannot be 0 - change to <0.001

As well as looking for statistically significant differences and/or associations based on GDM status could you also look in more depth at the clinical significance of some of your findings as based on mean values alone these are difficult to ascertain? For example mean TAG levels (whilst significantly different) appear relatively close in absolute terms. Did you investigate the proportion of women who would be classed as having abnormal/normal TAG and whether this is sig associated with optimal/sub-optimal Vit D.? Similarly whilst it is statistically significant the correlation between serum Vit D and fasting glucose (Fig 1) is weak and the clinical significance of these findings warrants greater discussion.

Discussion:

'Furthermore, all the obesity indices, pre-pregnancy BMI, BMI, waist size, hip size and WHR were significantly higher in the GDM women compared to non-GDM women suggesting a strong confounder effect from obesity.' - I agree so can this confounding effect be quantified (see previous suggestion re: multiple regression)? This is really important to extend our understanding in this field. The general associations you present have been previously shown (as suggested in your intro) so we now need to know which are the primary drivers (is it obesity, for which Vit D deficiency is co-morbid, or is the Vit D deficiency more important?) in order to inform prevention and intervention.

Is there any overlap between the studies covered in the three reviews in refs 23, 24 and 25? If so then this section would benefit from some rewriting to reflect this.

Line 34 - 'However, a systematic review and meta-analysis to study the link between vitamin D and gestational diabetes (2012) indicated a significant inverse relation between serum 25(OH)D and the incidence of GDM.' - this does not need to start 'however' as the pattern of findings (low Vit D, high risk) is the same as described previously.
Line 39 'Also, in a prospective cohort study (Spain, 2015) involving 2382 pregnant women, overall, 31.8% and 19.7% vitamin D insufficiency [25(OH)D3 20-29.99 ng/ml] and deficiency [25(OH)D3 <20 ng/ml], respectively, showed no association between maternal 25(OH)D3 concentration and risk of GDM [26].’ - this should start with 'however or conversely' as this DOES report a different direction of effect.

Line 54 'This was, despite the increase in circulating 25(OH)D levels that was associated with vitamin D supplementation' - again it would be good to put these results in clinical context - how many of the studies were based exclusively on women who were deficient at baseline? Just increasing vitamin D is unlikely to be enough to exert an effect whereas the effect of moving people from deficiency to sufficiency is more clinically important.

Conclusion:

In line with various previous comments I think that, to advance the field, the conclusion needs to be extended beyond the basic association between Vitamin D and GDM to include some recognition of the potential confounders of this relationship or, ideally, the independence of this association from weight or other potential confounders (if these have been clearly proven and reported in the results).

Figure 1. Legend can be simplified - first sentence fragment (starting "Correlational analysis of..) is unnecessary as same information is conveyed in fragment starting 'Scatter plot of…

Figure legend should include sample size (n= )

Table 1 - title is slightly misleading. Perhaps rephrase to be clear that characteristics are at baseline but GDM status was not determined until visit 2. Cut off used to determine deficiency (with associated reference) should be stated in table footnote
Are the methods appropriate and well described?
If not, please specify what is required in your comments to the authors.

No

Does the work include the necessary controls?
If not, please specify which controls are required in your comments to the authors.

Yes

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