PEER REVIEW HISTORY

BMJ Paediatrics Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

ARTICLE DETAILS

TITLE (PROVISIONAL)	Pharmacokinetics of prednisolone in children: An open-label, randomised two-treatment cross-over trial investigating the bioequivalence of different prednisolone formulations in children with airway disease – protocol of the POP child
AUTHORS	Haslund-Krog, Sissel; Schmidt, Maria; Mathot, Ron; Kryger Jensen, Andreas; Jørgensen, Inger Merete; Holst, Helle

VERSION 1 – REVIEW

REVIEWER	Reviewer name: Hussain Mulla Institution and Country: University Hospitals of Leicester England Competing interests: No
REVIEW RETURNED	31-May-2019

GENERAL COMMENTS	I am a little bit confused by the study design and the role of the control group. Classically, BE studies of formulations are cross-over studies, typically 2-way or 3-way. BE studies can be parallel design in some circumstances e.g. long half-life. But the sample size required is much greater. The study design presented here appears to be a combination of both. But the subject numbers are unlikely to be sufficient for a BE assessment from parallel design. Following on from this, the statistical methods section lacks detail - it is not clear how the AUC and Cmax parameters will be calculated (single subject compartmental modelling? population PK modelling?) and how BE assessment will be conducted. Moreover, it is stated that the objective is to investigate the BE of different formulations in three different age groups (6-23 months, 2-5 yrs, 6-11 yrs). However, the control group is aged 6-11 yrs. So it does not seem possible to investigate an age effect (assuming BE assessment involves a comparison with the control group). I am concerned that with the lack of in-study 'controls' around the time of dosing and post-dosing will make precise determination of AUC and Cmax quite difficult. For example, if there is no control on food intake, absorption profiles could be very erratic because of increased variability in gastric emptying (though I appreciate it is probably not possible to make young children fast for a prolonged period, but it may be possible to limit food intake around the time of dosing). Similarly, if children are not supine but ambulatory, this could significantly increase the variability in the absorption profile(again I appreciate difficult in children). There could be substantial variability in the PK data and as a consequence the study could lack power.

REVIEWER	Reviewer name: Hassan Almoazen PhD Institution and Country: Assistant Professor and PhD Director University of Tennessee USA

	Competing interests: No competing interests
REVIEW RETURNED	27-Jun-2019

GENERAL COMMENTS	There is no BE data to evaluate the significance of this project. Although it is novel to evaluate saliva instead of plasma samples, but the lack of data makes it hard to evaluate the novelty of the manuscript.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1

Comments to the Author

I am a little bit confused by the study design and the role of the control group. Classically, BE studies of formulations are cross-over studies, typically 2-way or 3-way. BE studies can be parallel design in some circumstances e.g. long half-life. But the sample size required is much greater. The study design presented here appears to be a combination of both. But the subject numbers are unlikely to be sufficient for a BE assessment from parallel design.

Response: We fully acknowledge that according to The BE guideline it is recommended to compare the formulations as, a randomised, two/three-period, two/three-sequence single dose crossover design and the treatment periods should be separated by a wash out period sufficient to ensure that drug concentrations are below the lower limit of bioanalytical quantification in all subjects at the beginning of the second period. At the same time the variability of response to pain, distress and fear between children should be taken into consideration and investigations/interventions should be limited to the minimum required for obtaining valid data and performed using size-/age-appropriate material and devices, including limiting in advance the number of attempts for sampling. This is elaborated in the limitations, please see p. 8. In order to meet the latter, we decided in conjunction with the PI to limit the study to a two-way cross-over with sufficient wash out period ($t_{1/2}$ = 2.5±0.5 h) to determine the PK and the derived parameter. According to the statistical analysis plan it has been prespecified that the comparison will be done in a stepwise manor, initially comparing the various subpopulation, e.g. the 6-23 moth old. If these data are comparable, we will pool the data and compare to the older age groups. The oldest age group will only then serve as control-group across age population. We have added this to the design, please see p. 7. If they do not show BE, the bioavailability data will be reported separately for each age groups

Following on from this, the statistical methods section lacks detail - it is not clear how the AUC and Cmax parameters will be calculated (single subject compartmental modelling? population PK modelling?) and how BE assessment will be conducted. Moreover, it is stated that the objective is to investigate the BE of different formulations in three different age groups (6-23 months, 2-5 yrs, 6-11 yrs). However, the control group is aged 6-11 yrs. So it does not seem possible to investigate an age effect (assuming BE assessment involves a comparison with the control group).

Response: the AUC(0-t) and Cmax parameters will be calculated using single subject compartmental modelling. Actual time of sampling will be used in the estimation of the pharmacokinetic parameters. The assessment of bioequivalence will be based upon 90% confidence intervals for the ratio of the population geometric means (test/reference), with the null hypothesis of bioequivalence at the 5% significance level (acceptance interval of 80.00- 125.00%). Due to the limited wash-out period, a test for carry-over will be addressed by examination of the pre-treatment plasma concentrations in period 2.

The control group will only be used across age as explained above. The above have been added to the manuscript, please see p. 7.

I am concerned that with the lack of in-study 'controls' around the time of dosing and post-dosing will make precise determination of AUC and Cmax quite difficult. For example, if there is no control on food intake, absorption profiles could be very erratic because of increased variability in gastric emptying (though I appreciate it is probably not possible to make young children fast for a prolonged period, but it may be possible to limit food intake around the time of dosing).

Response: In the Danish SMPC of the prednisolone tablets there are no impact of food intake. Most, but not all children, will take the prednisolone in the morning. In the eCRF time for last meal for every dose is stated, and if at all possible, the dosing will be before feeding, but in the very young, we foresee it will not always be possible.

Similarly, if children are not supine but ambulatory, this could significantly increase the variability in the absorption profile (again I appreciate difficult in children). There could be substantial variability in the PK data and as a consequence the study could lack power.

Response: All children are supine while administrated the drug, but the study will reflect real life setting in the sense, they will not be restricted to lie down for a longer period of time.

Response to Reviewer 2, Assistant Professor and PhD Director Hassan Almoazen, University of Tennessee USA

Thank you for your review of our paper. We have answered each of your points below.

Reviewer: 2

Comments to the Author

There is no BE data to evaluate the significance of this project. Although it is novel to evaluate saliva instead of plasma samples, but the lack of data makes it hard to evaluate the novelty of the manuscript.

Response: We find that the two published similar adult studies from 1982 and 1984 highlights the fact that new data on this subject is needed in a target population. Although, prednisolone is an old drug, prednisolone treatment in the paediatric population can still be improved.