1 **Original article**

2 **A novel, palatable paediatric oral formulation of midazolam: pharmacokinetics, tolerability, efficacy**

3 **and safety**

4 S. Salman,[1] E. K. Y. Tang,[2] L. C. Cheung,[3,4,5] M.N. Nguyen,[2] D. Sommerfield,[6] L. Slevin,[7] L-Y. Lim,[8] and B. S.

5 von Ungern Sternberg[6,9]

6 [1] Clinical Senior Lecturer, Medical School, Faculty of Health and Medical Sciences, The University of

7 Western Australia, Perth, Australia

8 [2] Research Officer, Division of Pharmacy, School of Allied Health, Faculty of Health and Medical Sciences,

9 University of Western Australia, Perth, Australia

10 [3] Senior Research Officer, Telethon Kids Institute, University of Western Australia, Perth, Australia

11 [4] Lecturer, School of Pharmacy and Biomedical Sciences, Curtin University, Perth, Australia

12 [5] Pharmacist, Department of Pharmacy, Princess Margaret Hospital for Children, Perth, Australia

13 [6] Consultant, Department of Anaesthesia and Pain Management, Princess Margaret Hospital for

14 Children, Perth, Australia

15 [7] Clinical Research Manager, Telethon Kids Institute, University of Western Australia, Perth, Australia

16 [8] Professor, Division of Pharmacy, School of Allied Health, Faculty of Health and Medical Sciences,

17 University of Western Australia, Perth, Australia

18 [9] Chair of Paediatric Anaesthesia, Medical School, Faculty of Health and Medical Sciences, The University

19 of Western Australia, Perth, Australia

20 Correspondence to: B.S. von Ungern Sternberg

21 Email: britta.regli-vonungern@uwa.edu.au

22 Short title: Novel paediatric formulation of midazolam

25   **Summary**

26   Midazolam is one of many bitter drugs where provision of a suitable paediatric formulation, particularly

27   in the pre-anaesthetic setting, remains a challenge. To overcome this problem a novel chocolate-based

28   tablet formulation has been developed with positive pre-clinical results. To further investigate the

29   potential of this formulation, 150 children aged 3-16 years who were prescribed midazolam as a pre-

30   medication were randomised to receive 0.5 mg.kg^{-1} either as the novel formulation or an intravenous

31   solution given orally, which is the current standard at our institution. Tolerability was assessed by each

32   child, parent and nurse using a five-point facial hedonic scale and efficacy was determined as the time to

33   onset of sedation. Blood samples for midazolam and 1-hydroxymidazolam values were analysed using

34   high-performance liquid chromatography. Population pharmacokinetics were evaluated using non-linear

35   mixed effects modelling. The novel formulation had significantly improved tolerability scores from

36   children, parents and nurses (all $p<0.001$). Time to effect was not different between the groups ($p=0.140$).

37   The pharmacokinetics of midazolam and 1-hydroxymidazolam were able to be suitably modelled

38   simultaneously. The novel formulation was subject to a higher estimated first pass metabolism compared

39   with the intravenous solution (8.6 vs 5.0 %) and a significantly lower relative bioavailability of 82.1%

40   ($p=0.013$), with no other significant differences. Exposure relative to dose was in the range previously

41   reported for midazolam syrup. We conclude that the novel chocolate-based formulation of midazolam

42   provides improved tolerability while remaining efficacious with suitable pharmacokinetics when used as

43   a premedicant for children.

44

Introduction

The importance of palatability for paediatric medicines, a previously neglected aspect of drug development, has recently been recognised by regulatory authorities and the pharmaceutical industry [1-3]. Midazolam, a short-acting benzodiazepine with sedative, amnesic and anxiolytic effects, is one example where the importance of this has been highlighted. Oral midazolam is commonly prescribed for children as premedication prior to the induction of anaesthesia. Alone it has a bitter taste, limiting its clinical utility in the paediatric population. Only one commercially available syrup is available in some regions, so it is common practice to use the intravenous (iv) formulation orally. To improve tolerability, the iv formulation has been mixed with various palatable liquids, however masking its taste has only been moderately successful [4,5].

As an alternative solution to this problem a novel chewable Chocolate-based tablet Delivery System (CDS) was developed [6]. Pre-clinical data from both in-vitro and animal studies have suggested an improved acceptability of the novel formulation. Rodent data showed the CDS matrix to be effective at masking the bitter taste of midazolam. The midazolam CDS tablet did not require refrigerated storage and was stable for at least 18 months when wrapped in foil and maintained at room temperature. In vitro drug dissolution experiments showed complete release of the midazolam load from the tablet into simulated gastric fluid in 15 min for pre-crushed tablets (to simulate mastication) and 35 min for intact tablets. Based on these data the CDS formulation was then trialled in a paediatric population who were prescribed midazolam prior to general anaesthesia at our centre. We decided to assess tolerability, efficacy and safety, as well as comparative pharmacokinetics of the novel CDS formulation compared with the iv formulation administered orally.

69 **Methods**

70 We performed a prospective, open-label, single centre, randomised, single treatment trial at Princess

71 Margaret Hospital, the only paediatric tertiary referral centre in Western Australia. Institutional research

72 ethics approval was obtained from the Princess Margaret Hospital and the University of Western Australia

73 (2014102EP and RA/4/1/7610, respectively). Written informed parental or guardian consent and assent

74 from the child (where appropriate) was obtained prior to enrolment in the study.

75 Computer-generated block randomisation, stratified according to age (< 7 years old or >7 years old) and

76 gender, was produced by the clinical trials pharmacy in order to assign patients, and was performed

77 independently from the study team. Children received either the midazolam CDS tablet or iv midazolam

78 solution (Pfizer, Australia) orally using 1:1 randomisation. Both formulations were dosed according to local

79 institutional guidelines, in a target dose of 0.5 mg.kg^{-1}, as prescribed by the treating anaesthetist.

80 Midazolam CDS tablets used in the clinical trial were manufactured within the Department of Pharmacy,

81 Princess Margaret Hospital. We excluded children allergic to midazolam or chocolate (CDS base is nut free)

82 or when informed consent could not be obtained.

83 During administration of the study drug, a member of the research team recorded whether the whole

84 dose was swallowed, partially expelled or totally refused by the patient. For tolerability, the child was

85 asked immediately after administration to record how much he or she liked the sample by putting a mark

86 on a five-point facial hedonic scale (ranging from 1; dislike very much, to 5; liked very much) [7] and

87 whether they would be happy to take the drug again if required. The parent and accompanying nurse

88 were also asked independently to give a score, using separate five-point hedonic scales, based on their

89 perception of how the child reacted to the taste of the assigned formulation. If the child expelled the dose

90 immediately, the treating anaesthetist was free to decide whether a repeat dose was required, in line

91 with current routine management. If a second dose was recommended the treating anaesthetist, together

4

92   with the parent and/or child, decided whether to repeat the dose or administer the alternative form. The

93   time to sedation onset (clinical effect of midazolam) was recorded for all patients.

94   Monitoring as recommended by the AAGBI and Australian and New Zealand College of Anaesthetists

95   (ANZCA) (electrocardiography, non-invasive blood pressure measurements, capnography and pulse

96   oximetry) was commenced and general anaesthesia was induced either by a consultant anaesthetist or

97   under their direct supervision. The choice of anaesthetic agents and analgesia was left to the discretion

98   of the anaesthetist. Oxygen saturation was continuously monitored throughout surgery and in the post

99   anaesthesia care unit until patient discharge. One to one nursing was guaranteed at all times in the post

100   anaesthesia care unit with at least one additional circulating nurse present at all time.

101   Given the paediatric study population undergoing otherwise usual care, a flexible, sparse venous blood

102   sampling approach was utilised for PK sampling. The first sample was collected as soon as possible after

103   the child lost consciousness (approximately 30 minutes after the midazolam premedication had been

104   administered). The second and third samples were collected between 45-60 minutes and 90-120 minutes,

105   respectively. The final sample was collected as late as possible before the procedure was completed whilst

106   the patient was still anaesthetised. At each time point 3ml venous blood was withdrawn from a cannula

107   and emptied into an EDTA tube. The number of samples varied according to the length of the procedure,

108   with a maximum of four samples, and total blood volume not exceeding 12ml. After collection blood

109   samples were centrifuged at 2,000 rpm at 4 °C for 10 minutes before the plasma was separated into

110   Eppendorf tubes for storage at -80 °C until analysis was performed.

111   The method for quantifying midazolam and 1-hydroxymidazolam in plasma was adapted from Juřica et al.

112   [8] with minor modifications. Differences in the pre-analytical step were that the stock solutions were

113   prepared at a lower concentration (10 µg.ml^{-1} for diazepam, 25 µg.ml^{-1} for midazolam and 1-

114   hydroxymidazolam), dilutions were made with methanol alone, the dynamic range for both analytes was

115   17 – 333 ng.ml^{-1}, 20 µl of 0.5 M NaOH was used for alkalisation, samples were spiked with 30 µl of internal

116   standard, a second reconstitution/evaporation step was performed and final dissolution utilised 100 µl

117   methanol, of which 30 µl was injected for analysis.

118   The HPLC system comprised an Agilent 1260 Infinity HPLC (Agilent Technologies Australia, NSW, Australia)

119   with a Hypersil BDS column (250 × 4.6 mm, 5 µm, Thermo Fisher Scientific Australia, WA, Australia). The

120   mobile phase consisted of 20 mM potassium phosphate buffer, pH 5.0, and acetonitrile. A double gradient

121   elution method was applied as follows: 35% acetonitrile ramping to 45% in 12 min, held isocratic for 9

122   min, followed by another ramp from 45% acetonitrile to 100% in 7 min, held isocratic for 5 min before

123   bringing the acetonitrile concentration down to 35% in 2 min, and holding for at least 7 min prior to next

124   sample injection. The flow rate was 1 ml.min^{-1}, at ambient temperature, and eluent was monitored at 245

125   nm.

126   The retention times for midazolam, 1-hydroxymidazolam and diazepam (internal standard) were 13.9,

127   10.2 and 17.0 min, respectively. Calibration curves for midazolam and 1-hydroxymidazolam were linear

128   from 17 to 333 ng.ml^{-1} ($R^2 \geq 0.99$). Inter- and intra-day accuracy and precision were suitable within this

129   range with bias <9% and coefficient of variability <15%. The lower limit of quantification was 13 ng.ml^{-1}

130   for both analytes.

131   Log$_e$ plasma concentration-time datasets for midazolam and 1-hydroxymidazolam were analysed by

132   nonlinear mixed effects modelling using NONMEM (v 7.2.0, ICON Development Solutions, Ellicott City,

133   MD, USA) with an Intel Visual FORTRAN 10.0 compiler. The first order conditional estimate with

134   interaction (FOCE with INTER) method was used. The minimum value of the objective function (OFV) and

135   visual predictive checks were used to choose suitable models during the model-building process. A

136   significance level of p<0.05 was set for comparison of nested models. Allometric scaling for body weight

137 (WT) was employed a priori, with volume terms multiplied by $(WT/70)^{1.0}$ and clearance terms by

138 $(WT/70)^{0.75}$ [9]. Residual variability (RV) was estimated as additive error for the log-transformed data. Base

139 models were parameterised using V_C (central volume of distribution), CL (clearance), V_P and Q (peripheral

140 volumes of distribution and their respective inter-compartmental clearances). A time to event (onset of

141 effect) was planned, however this could not be established due to a lack of concentration data prior to

142 onset of effect.

143 Given the primary purpose of the analysis was to compare the two formulations, different bioavailability

144 and absorption parameters were estimated in the model. The bioavailability of the CDS formulation

145 relative to the iv formulation was included as a parameter while different absorption parameters for each

146 of the formulations were included, where supported by available data.

147 Initial modelling was performed on the midazolam data set alone and one-, two- and three- compartment

148 models (ADVAN 2, 4 and 12, respectively) were assessed. Given that the absorption profile between

149 subjects varied, several absorption models were tested, including single- and double-phase absorption

150 with zero- and first-order rates, with and without an initial lag time, as well as a transit compartment

151 model. In this model, the dose passes through a series of transit compartments before entering the

152 absorption compartment to model the delay often associated with drug absorption. A single rate constant

153 (k_{tr}) describes the entry and exit for all transit compartments. Using a previously described

154 implementation of the transit compartment model in NONMEM [10], the number of transit

155 compartments (NN) and the mean transit time (MTT = (1+NN) / k_{tr}) were estimated as continuous

156 variables. Once a suitable structural model for midazolam was established, 1-hydroxymidazolam plasma

157 concentration-time data were added and custom general linear disposition models were constructed

158 using ADVAN5. Modelling of midazolam and 1-hydroxymidazolam was then performed simultaneously.

159 To allow PK identifiability in the parent drug-metabolite model, complete conversion of midazolam to 1-

160 hydroxymidazolam was assumed. Although this assumption is not biologically correct, it only represents

161 a scaling factor for the 1-hydroxymidazolam modelling. Therefore, all midazolam parameters were

162 relative to bioavailability (F) while all 1-hydroxymidaolam parameters were relative to F x metabolic

163 conversion (F*). One and two additional compartments were tested for 1-hydroxymidazolam. Models

164 with first-pass metabolism, estimated separately for each of the formulations, were also assessed. Once

165 the base structure of the models was established, inter-individual variability (IIV) as well as correlations

166 between IIV terms, were evaluated for each suitable parameter and included where supported by the

167 data. Inter-individual variability was exponentially modelled for all parameters.

168 Relationships between model parameters and age, BMI and sex were assessed through inspection of

169 scatterplots and boxplots of individual parameters vs covariate, and subsequently evaluated within

170 NONMEM. A stepwise forward inclusion and backward elimination method was used with a significance

171 level of $p<0.05$ required for inclusion of a covariate relationship and $p<0.01$ to retain a covariate

172 relationship.

173 Once modelling for the primary PK population (those patients reported to have ingested the entire dose)

174 was completed, a secondary analysis was performed to estimate the related absorption of the dose in

175 those reported to have partially ingested the dose but who did not receive a further dose. For this analysis

176 all other parameters in the model were fixed to the values in the primary final model. Details of model

177 evaluation are provided in the supplementary material available online.

178 Statistical analysis was performed using R version 2.14.2 (R Foundation for Statistical Computing, Vienna,

179 Austria) software. Two-sample comparisons for non-normally distributed variables were made using

180 Mann Whitney U-test. Unless otherwise stated, all p values are two-tailed and unadjusted for multiple

181 comparisons. Power analysis was performed using the Monte-Carlo Mapped Power (mcmp) method

182 automated through Perl speaks NONMEM (PsN) using PK parameters from a previous study [11] to

183   determine if the proposed sampling schedule is sufficient to determine a 20% difference between the two

184   formulations (a clinically relevant difference). Fifty patients in each group would achieve 80% power with

185   an α value of 0.05, while 75 in each group corresponded to a power of 90%. Similar results were obtained

186   when the ka of the chocolate formulation was set 50% lower (54 and 80 in each group for a power of 80%

187   and 90%, respectively). In order to account for drop outs and missing PK data points a target study

188   population of 150 was set.

189

190

191

192

193

194

195

196

197

198

199

200

201

202

203

204

205

206

207 **Results**

208 One hundred and fifty children were included in the study and baseline characteristics are shown in Table

209 1. Twenty participants did not fully ingest the first dose, with 6 in the chocolate group and 12 in the iv

210 formulation group partially ingesting the dose, and an additional 2 in the iv formulation group completely

211 refusing the dose. Therefore approximately 8% in the chocolate group did not entirely ingest the dose

212 compared with 18% in the iv formulation group, p=0.065. For three patients in the iv group who did not

213 entirely ingest the first dose, a decision was made by the treating anaesthetist to give a second dose, and

214 all were given the CDS tablets. Two of the three entirely ingested the CDS tablets while the other patient

215 partially ingested this dose. Participants who did not entirely ingest the dose had a lower mean age than

216 those who did (6.1 vs 7.7 years, p=0.046), and there was no difference with regards to the formulation

217 they had been assigned.

218 Although all children were included in the taste and safety analysis, only the 130 children who completely

219 ingested the dose were included in the primary efficacy analysis (Table 1). This consisted of 67 in the

220 chocolate group and 62 in the iv formulation group. One participant in the chocolate group did not have

221 any PK samples taken and was excluded from the primary PK analysis. An additional 17 children (6 in the

222 chocolate group and 11 in the iv formulation group) who partially ingested the dose, but did not receive

223 further doses, were included in the secondary PK analysis.

224 The CDS tablet had acceptable tolerability with significantly improved scoring compared with the iv

225 formulation on the 5-point scale for children, parents and clinical staff, p<0.001 (Figure 1). Five children

226 (all younger than 4.5 years, four of whom were in the iv formulation group) were unable to provide a

227 score, and were scored low by the parent (≤2). Mean (SD) scores given by the children, parents and nurses

228 for the CDS tablet were 3.16 (1.45), 3.52 (1.25) and 3.36 (1.29), compared with corresponding mean (SD)

229 scores of 1.71 (1.13), 1.71 (1.00) and 1.97 (1.00) for the iv formulation. These significant differences were

230   noted across age and gender strata with p values <0.050. Significantly more children in the chocolate

231   group than in the iv formulation group (62% vs 39%), indicated they would take the same formulation

232   again, p = 0.007.

233   Despite a higher mean (SD) administered dose for patients in the iv formulation group, there was no

234   significant difference in sedation onset time between the groups. Median (interquartile range [range])

235   time to onset of sedation was 13 (10 – 17 [5-31]) min in the CDS group compared with 12 (9 – 16 [4-30])

236   min in the iv formulation group, p=0.140. We observed no serious adverse events during the study.

237   From the 129 children included in the primary PK analysis there were 294 individual plasma midazolam

238   concentrations (160 and 134 from the chocolate and iv formulation groups, respectively) and 317 1-

239   hydroxymidazolam concentrations (172 and 145 from the chocolate and iv formulation groups,

240   respectively) available for analysis. Of these, 3% and 9%, respectively, were measurable but below the

241   lower limit of quantification. Given these were <10% of the total dataset they were kept at their measured

242   values for the purposes of analysis [12]. There was an additional 45 midazolam and 48 1-

243   hydroxymidazolam concentrations in the secondary analysis estimating the degree of absorption with

244   partial dose ingestion. A two-compartment model for midazolam was most appropriate, with no benefit

245   from additional compartments, p>0.05. The absorption was best represented with a transit compartment

246   model with a different MTT parameter for the two formulations, MTT_{CDS} for the chocolate formulation

247   and MTT for the iv formulation. A single additional compartment was adequate to describe the disposition

248   of 1-hydroxymidazolam. The inclusion of first-pass metabolism, with a separate parameter for each

249   formulation, resulted in significantly lower objective function value and improved appearance of

250   diagnostic plots.

251   Inter-individual variability was estimable for CL/F_{MDZ}, V_C/F_{MDZ}, V_P/F_{MDZ}, $CL/F*_{OHMDZ}$, MTT and MTT_{CDS}.

252   Correlation between V_C/F_{MDZ} and V_P/F_{MDZ} was estimated to be close to one, therefore this was fixed to

253   unity. Otherwise a full covariance matrix was estimable between CL/F_{MDZ}, V_C/F_{MDZ} and CL/F^*_{OHMDZ}. None

254   of the tested covariate relationships improved the fit of the model.

255   The final model parameter estimates and the bootstrap results are summarised in Table 2. The relative

256   bioavailability for the CDS tablets was lower than for the iv formulation, specifically, 82.1% with an

257   empirical 95% confidence interval of 69.3 – 95.1% (p=0.013 for the difference between formulations), co-

258   incident with higher first-pass metabolism (8.6 % vs 5.0 %, respectively). A clinically insignificant trend for

259   slower absorption of the CDS tablets (mean transit time estimated to be 2 minutes longer) with slightly

260   larger interindividual variability (78 vs 68%) was also observed. When the children with partially ingested

261   doses were added to the analysis, the estimated dose absorbed was 70% and 58% for the CDS tablet and

262   iv formulation, respectively. The dose normalised AUCs for both formulations were within the range which

263   has previously been reported for midazolam syrup (Figure 2) [13,14].

264

Discussion

265

266 This study demonstrates that the novel paediatric formulation of midazolam as a chocolate-based tablet

267 has improved tolerability, whilst maintaining similar efficacy, compared with the iv solution given orally,

268 the current standard at our institution. A population PK model, using sparse sampling accommodating for

269 usual care in this paediatric population, was successfully created. This model was used to estimate the

270 relative bioavailability between the formulations as well as to investigate differences in the absorption

271 profiles of the two formulations.

272 Consistent with in vitro and pre-clinical data [6], we found a significant improvement in tolerability of

273 midazolam when incorporated into the CDS tablet. Taste scores using a 5-point facial hedonic scale were

274 significantly better for the CDS tablet when scored by children, their parents and nursing staff caring for

275 the child. This scale has been used for taste evaluation in children from 3 to 12 years old and for parents

276 of children aged 4 to 16 years [7]. Although other methods can be used, such as a visual analogue scale,

277 this method is more commonly used, particularly in younger children. The hospital environment can be

278 stressful for the very young and it is not surprising that they may not comply with such scoring, particularly

279 if they dislike the taste of the premedication. Significantly more children in the chocolate group were

280 willing to have the same formulation again, further demonstrating an improved tolerability of this

281 formulation.

282 There was no significant difference in the primary measure of efficacy between the two formulations.

283 Although median time to onset of sedation was 1.5 minutes longer in the chocolate group there was

284 reasonable variability within each group. This difference was not statistically significant and could not be

285 considered clinically significant either. This was so despite a 13% lower average mg.kg^{-1} dose for patients

286 in the chocolate group. The population PK model mirrored these findings. The estimated mean transit

287 time for the CDS tablets was slightly longer than the iv formulation (13 vs 11 min) with significant inter-

288   individual variability (>60% for both formulations). This difference was not significant in the PK model,

289   with overlapping 95% confidence intervals.

290   Other differences between the two formulations identified in the PK model were with respect to

291   absorption parameters. Relative bioavailability was estimated to be 82%. Considering the low reported

292   absolute bioavailability of midazolam syrup in children (15-37%) [13,14], a reduced absorption for a solid

293   formulation (CDS tablets) when compared with a high concentration liquid (iv formulation) is not

294   surprising. Consistent with these differences there was lower estimated first pass metabolism for the iv

295   formulation group (5.0% vs 8.6% in the chocolate group) possibly due to greater buccal absorption which

296   avoids first pass effect. No other differences were noted between the two formulations. More

297   importantly, there was no formulation effect on the clearance of midazolam or 1-hydroxymidazolam. The

298   differences between the two tested formulations are of unclear clinical significance given there was no

299   difference in the primary efficacy measure. In patients who partially ingested the dose there was a 42%

300   reduced exposure in the iv solution group compared with 30% in the chocolate group. Although this

301   difference was not statistically significant it does suggest that children are more likely to expel the iv

302   formulation compared with the CDS tablets.

303   There have been two previously published reports on the PK of a midazolam syrup in paediatric patients.

304   Payne et al [14] compared several different formulations of midazolam in anaesthetised children aged 3

305   to 10 years that included 3 different doses for an oral syrup compounded from the iv formulation. They

306   reported an absolute bioavailability of 15-27% for the syrup administered via nasogastric tube, with lower

307   bioavailability for 0.45 mg.kg^{-1} and 1.0 mg.kg^{-1} compared with 0.15 mg.kg^{-1} dose. Reed et al [13] reported

308   the PK of Versed® Syrup administered orally in children aged 6 months to 16 years old. The absolute

309   bioavailability was estimated to be 37% with an interindividual variability of 28%. The range of dose-

310   normalised AUC for both formulations in the present study was within the range reported in these

311   previous studies and had a similar spread to that of Reed et al. Therefore, with equivalent dosing it would

312   be expected that the novel CDS formulation would result in similar overall exposure to midazolam.

313   Our study has some limitations. The use of a sparse sampling approach and avoiding discomfort to the

314   child by delaying sampling until after they were anaesthetised resulted in poorer precision PK parameters

315   relating to absorption. In particular, first pass metabolism and the number of transit compartments had a

316   relative standard error (RSE) of >50%. Despite this, key PK parameters in this study, including relative

317   bioavailability and clearance parameters, were well estimated with RSE <30%. A further potential

318   limitation was the use of iv formulation as the comparator, given paediatric formulations of midazolam

319   syrups exist. Use of the iv formulation is the standard of care within our institution, as the commercial

320   midazolam syrup (Versed® Syrup) is not available in Australia. Despite this we would expect the

321   bioavailability of a high concentration, pH unadjusted, small volume liquid to be no worse and likely

322   greater, given reduced first-pass metabolism, compared with the oral syrup formulation. Consistent with

323   this, when a prepared mixture of the iv formulation was compared with the syrup formulation it resulted

324   in a 45% higher serum concentration up to 90 minutes after administration [5]. Therefore, we would

325   expect that the CDS tablets would have higher relative bioavailability when compared with the

326   commercial midazolam syrup.

327   Developing suitable medications for children raises specific challenges, particularly with regard to

328   ensuring tolerability and performing ethically appropriate studies in order to allow for their evaluation.

329   The present study was designed to have minimal impact on our usual care of children while still providing

330   key information to evaluate a new paediatric formulation. The novel CDS tablets were found to have

331   favourable tolerability for midazolam, a drug with a bitter taste, while remaining equally efficacious, safe

332   and with adequate relative bioavailability. Not only does this provide a better alternative for pre-

333   medication but establishes a methodology that could be applied for other bitter medications in children.

334

335 **Acknowledgements**

341

342 **Competing interests**

348

349

350   **References**

351   1.   Zajicek A, Fossler MJ, Barrett JS, et al. A report from the pediatric formulations task
352   force: perspectives on the state of child-friendly oral dosage forms. *AAPS J* 2013; **15:**
353   1072-81.
354   2.   European Parliament and the Council of the European Union Regulation (EC) n.
355   1902 / 2006 of the European Parliament and of the Council of 20 December 2006
356   amending Regulation 1901 / 2006 on medicinal products for paediatric use. *Official*
357   *Journal of the European Union* 2006; **L378:** 20-1.
358   3.   Committee for Medicinal Products for Human Use Reflection paper: formulations of
359   choice for the paediatric population. *European Medicines Agency* 2006.
360   4.   Isik B, Baygin O, Bodur H Effect of drinks that are added as flavoring in oral
361   midazolam premedication on sedation success. *Pediatric Anesthesia* 2008; **18:** 494-
362   500.
363   5.   Brosius KK, Bannister CF Midazolam premedication in children: a comparison of
364   two oral dosage formulations on sedation score and plasma midazolam levels.
365   *Anesthesia and Analgesia* 2003; **96:** 392-5, table of contents.
366   6.   Cheung LC, Nguyen M, Tang E, et al. Taste evaluation of a novel midazolam tablet for
367   pediatric patients: In vitro drug dissolution, in vivo animal taste aversion and
368   clinical taste perception profiles. *International Journal of Pharmaceutics* 2017; **535:**
369   194-200.
370   7.   Davies EH, Tuleu C Medicines for children: a matter of taste. *Journal of Pediatrics*
371   2008; **153:** 599-604, e1-2.
372   8.   Jurica J, Dostalek M, Konecny J, Glatz Z, Hadasova E, Tomandl J HPLC determination
373   of midazolam and its three hydroxy metabolites in perfusion medium and plasma
374   from rats. *Journal of Chromatography. B, Analytical Technologies in the Biomedical*
375   *and Life Sciences* 2007; **852:** 571-7.
376   9.   Anderson BJ, Holford NH Mechanistic basis of using body size and maturation to
377   predict clearance in humans. *Drug Metabolism and Pharmacokinetics* 2009; **24:** 25-
378   36.
379   10.   Savic RM, Jonker DM, Kerbusch T, Karlsson MO Implementation of a transit
380   compartment model for describing drug absorption in pharmacokinetic studies.
381   *Journal of Pharmacokinetics and Pharmacodynamics* 2007; **34:** 711-26.
382   11.   Reed MD, Rodarte A, Blumer JL, et al. The single-dose pharmacokinetics of
383   midazolam and its primary metabolite in pediatric patients after oral and
384   intravenous administration. *J Clin Pharmacol* 2001; **41:** 1359-69.
385   12.   Bergstrand M, Karlsson MO Handling data below the limit of quantification in mixed
386   effect models. *AAPS J* 2009; **11:** 371-80.
387   13.   Reed MD, Rodarte A, Blumer JL, et al. The single-dose pharmacokinetics of
388   midazolam and its primary metabolite in pediatric patients after oral and
389   intravenous administration. *The Journal of Clinical Pharmacology* 2001; **41:** 1359-
390   69.
391   14.   Payne K, Mattheyse FJ, Liebenberg D, Dawes T The pharmacokinetics of midazolam
392   in paediatric patients. *European Journal of Clinical Pharmacology* 1989; **37:** 267-72.

393

394 **Table 1.** Baseline characteristics for children administered midazolam orally as either CDS tablets or iv
395 formulation. Data are mean (SD) or number (proportion).

396

	All randomised children (tolerability and safety analysis)			Primary effic
	CDS formulation	iv formulation	p value	CDS formulation
	n=76	n=74		n=67[b]
Weight; kg	29 (14)	30 (17)	0.790	29 (14)
Age; years	7.4 (3.1)	7.5 (3.6)	0.860	7.5 (3.2)
Sex; male	36 (49%)	37 (49%)	0.870	32 (48%)
Height; cm	125 (19)[a]	124 (22)[a]	0.660	125 (19)[c]
Dose; mg of midazolam base		-	-	10.2 (3.2)
Weight adjusted dose; mg.kg^{-1} midazolam base		-	-	0.38 (0.09)

397

398 [a] Height missing for one participant in each group.

399 [b] One additional 6-year-old male with no evaluable PK data included in the efficacy population, inclusion
400 of this participant did not alter the between group comparisons.

401 [c] Height missing for one participant.

402

403

404

405

406

407

408

409

410

411 **Table 2.** Final population pharmacokinetic estimates and bootstrap results for midazolam and 1-

412 hydroxymidazolam in plasma from children prior to surgery

Parameter	Mean	RSE%	Bootstrap median [95% CI]
Objective Function Value	-265.078		-300.214 [-458.616 - -161.877]
Structural model parameters			
F_{CDS} (%)	82.1	8	82.7 [69.3 - 95.1]
MTT (h)	0.186	26	0.187 [0.105 - 0.266]
MTT_{CDS} (h)	0.216	20	0.216 [0.123 - 0.300]
NN	2.45	53	2.45 [0.43 - 8.55]
CL/F_{MDZ} ($l.h^{-1}.70kg^{-1}$)	90.4	30	90.8 [38.3 - 142]
V_C/F_{MDZ} ($l.70kg^{-1}$)	165	9	161 [126 - 213]
Q/F_{MDZ} ($l.h^{-1}.70kg^{-1}$)	90.4	31	95.5 [52.1 - 156.3]
V_P/F_{MDZ} ($l.70kg^{-1}$)	290	63	279 [86.7 – 1,000]
FP (%)	4.99	57	4.99 [0.65 - 12.8]
FP_{CDS} (%)	8.63	50	8.76 [1.00 - 17.7]
CL/F^*_{OHMDZ} ($l.h^{-1}.70kg^{-1}$)	127	28	129 [51.6 - 192]
V/F^*_{OHMDZ} ($l.70kg^{-1}$)	67.4	26	69.8 [24.8 - 127]
Variable model parameters [shrinkage%]			
IIV in CL/F_{MDZ}	65 [19]	28	61 [27 - 99]
IIV in V_C/F_{MDZ}	50 [16]	31	55 [15 - 100]
IIV in CL/F^*_{OHMDZ}	58 [21]	20	56 [27 - 83]
IIV in V_P/F_{MDZ}	36 [16]	30	40 [3 - 137]
IIV in MTT	65 [54]	20	64 [39 - 91]
IIV in MTT_{CDS}	78 [50]	15	77 [50 - 108]
$r(V_C/F_{MDZ}, V_P/F_{MDZ})$	1		FIXED
$r(V_C/F_{MDZ}, CL/F_{MDZ})$	0.762		0.753 [0.112 - 0.947]
$r(V_C/F_{MDZ}, CL/F^*_{OHMDZ})$	0.614		0.594 [-0.199 - 0.922]
$r(CL/F_{MDZ}, CL/F^*_{OHMDZ})$	0.718		0.684 [-0.319 - 0.905]
RV MDZ (%)	29 [26]	10	30 [23 - 36]
RV OHMDZ (%)	30 [26]	13	29 [16 - 37]

413
414 F_{CDS} (bioavailability of CDS formulation relative to iv formulation), MTT (mean transit time), NN (number
415 of transit compartments), CL/F_{MDZ} (clearance of midazolam), V_C/F_{MDZ} (central volume of distribution of
416 midazolam), Q/F_{MDZ} (intercompartmental clearance of midazolam), V_P/F_{MDZ} (peripheral volume of
417 distribution of midazolam), FP (degree of first-pass metabolism), CL/F^*_{OHMDZ} (clearance of 1-
418 hydroxymidazolam), V_C/F^*_{OHMDZ} (volume of distribution of 1-hydroxymidazolam), r (correlation

419   coefficient), IIV (inter-individual variability) and RV (residual variability). IIV and RV is presented as

420    $100\% \times \sqrt{\text{variability estimate}}$.

421

422

423   **Figure Legends:**

424

425   **Figure 1.** Histogram of patient, parent and nurse scoring of tolerability for the CDS tablet IV solution

426   (black bars) and iv solution CDS tablet (grey bars) formulations with the 5-point facial hedonic scale used

427   for assessments.

428

429   **Figure 2.** Mean and standard deviation of dose normalised midazolam area under the curve for CDS

430   tablet (black with star), IV solution given orally (black with triangle) and various dose levels in previously

431   published reports [13] (grey with square) and [14] (grey with circle).

432

433

1 # **Supplemental material**

2 **Novel, palatable paediatric oral formulation of midazolam: pharmacokinetics,**

3 **tolerability, efficacy and safety**

4

5 *Model evaluation method*

6 For model evaluation, plots of observed vs individual- and population-predicted values, and time

7 vs WRES, were first assessed. A bootstrap using Perl speaks NONMEM (PSN) with 1,000

8 samples was performed, and the parameters derived from this analysis summarised as median

9 and 2.5^{th} and 97.5^{th} percentiles (95% empirical CI) to facilitate evaluation of final model

10 parameter estimates. In addition, prediction corrected visual predictive checks (pcVPCs)

11 stratified according to formulation, were performed with 1,000 datasets simulated from the final

12 models. The observed 10th, 50th, and 90th percentiles were plotted with their respective simulated

13 90% CIs to assess the predictive performance of the model and to evaluate any major bias.

14 Shrinkage of population variability parameters and residual variability was assessed to help

15 determine whether models were over-parameterised and to determine the reliability of diagnostic

16 plots [1].

17 *Model evaluation results*

18 Bias was less than 9% for all fixed and random model parameters, except for IIV of V_P/F_{MDZ} for

19 which it was less than 15%. Supplemental Figures 1 and 2 show goodness-of-fit plots for

20 midazolam and 1-hydroxymidazolam respectively, with no bias evident. Supplemental Figure 3

21 presents the pcVPC plots. The actual 10th, 50th and 90th percentiles fell within their respective

22 95% CI for both midazolam and 1-hydroxymidazolam for both formulations, therefore there is

23   suitable predictive performance of the model.

24

25

26 **Supplemental figure captions.**

27 **Supplemental Figure 1.** Goodness-of-fit plots for midazolam given to children prior to surgery.

28 The observed plasma concentration has been plotted against population (A) and individual (B)

29 predicted plasma concentrations, and weighted residuals against time (C) and population

30 predicted plasma concentrations (D). Concentrations are on a $\log_{10}$ scale.

31 **Supplemental Figure 2.** Goodness-of-fit plots for 1-hydroxymidazolam after oral midazolam

32 given to children prior to surgery. The observed plasma concentration has been plotted against

33 population (A) and individual (B) predicted plasma concentrations, and weighted residuals

34 against time (C) and population predicted plasma concentrations (D). Concentrations are on a

35 $\log_{10}$ scale.

36 **Supplemental Figure 3.** Prediction corrected visual predictive check for midazolam (**A**, **B**) and

37 1-hydroxymidazolam (**C**, **D**) (ng.ml^{-1} $\log_{10}$ scale) for children prior to surgery receiving oral

38 midazolam either as IV solution formulation (**A**, **C**) or novel CDS formulation (**B**, **D**). Plots

39 demonstrate observed 50th (solid line), 10th and 90th (dotted lines) percentiles within their

40 simulated 95% CI (grey shaded areas) with overlying data points (○).
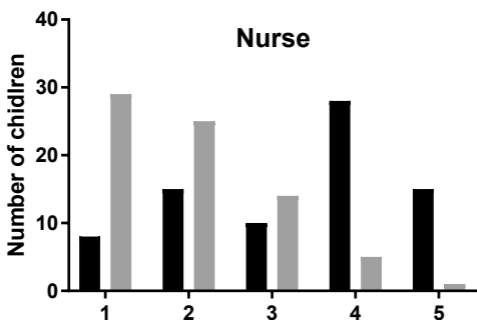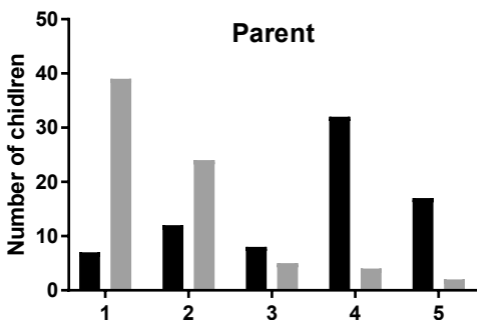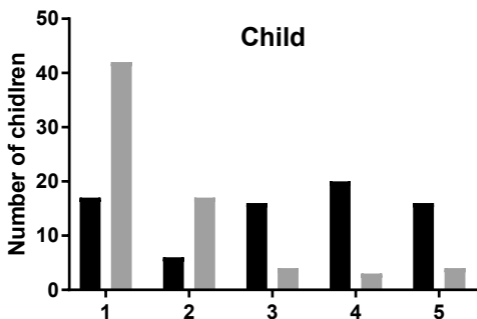
41

42

43

44   ***References***

45   1.   Savic RM, Karlsson MO Importance of shrinkage in empirical bayes estimates for
46   diagnostics: problems and solutions. *AAPS J* 2009; **11:** 558-69.

47

Child

Parent

Nurse

Number of chidlren

Facial scale score

1	2	3	4	5
Dislike very much	Dislike a little	Not sure	Like a little	Like very much