

BMJ Open

BMJ Open is committed to open peer review. As part of this commitment we make the peer review history of every article we publish publicly available.

When an article is published we post the peer reviewers' comments and the authors' responses online. We also post the versions of the paper that were used during peer review. These are the versions that the peer review comments apply to.

The versions of the paper that follow are the versions that were submitted during the peer review process. They are not the versions of record or the final published versions. They should not be cited or distributed as the published version of this manuscript.

BMJ Open is an open access journal and the full, final, typeset and author-corrected version of record of the manuscript is available on our site with no access controls, subscription charges or pay-per-view fees (<http://bmjopen.bmj.com>).

If you have any questions on BMJ Open's open peer review process please email info.bmjopen@bmj.com

BMJ Open

Study protocol for a randomised trial for atosiban versus placebo in threatened preterm birth: the APOSTEL 8 study

Journal:	<i>BMJ Open</i>
Manuscript ID	bmjopen-2019-029101
Article Type:	Protocol
Date Submitted by the Author:	14-Jan-2019
Complete List of Authors:	<p>Klumper, Job; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p> <p>Breebaart, Wouter; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p> <p>Roos, Carolien; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p> <p>Naaktgeboren, CA ; Julius Centrum voor Gezondheidswetenschappen en Eerstelijns Geneeskunde, University Medical Centre Utrecht</p> <p>van der Post, Joris; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p> <p>Bosmans, J; Vrije Universiteit Amsterdam, Department of Health Sciences, Amsterdam Public Health research institute</p> <p>van Kaam, Anton; Amsterdam UMC, location AMC and VUmc, Department of neonatology</p> <p>Schuit, Ewoud; Julius Center for Health Sciences and Primary Care, University Medical Centre Utrecht; Stanford University, Stanford Prevention Research Center</p> <p>Mol, Ben; School of Medicine, Monash University, Melbourne, Australia, Department of obstetrics & gynaecology</p> <p>Baalman, Jelle; National Maternity Hospital, Department of obstetrics & gynaecology</p> <p>McAuliffe, Fionnuala; University College Dublin, UCD Perinatal Research Centre; National Maternity Hospital, Department of obstetrics & gynaecology</p> <p>Thornton, Jim; University of Nottingham, Department of obstetrics & gynaecology</p> <p>Kok, Marjolein; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p> <p>Oudijk, Martijn; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p>
Keywords:	preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

SCHOLARONE™
Manuscripts

1 Study protocol for a randomised trial for atosiban versus 2 placebo in threatened preterm birth: the APOSTEL 8 study

3 J. Klumper,¹ W. Breebaart,¹ C. Roos,¹ C.A. Naaktgeboren,² J.A.M van der Post,¹ J.E.
4 Bosmans,³ A.H. van Kaam,⁴ E. Schuit,² B.W. Mol,⁵ J.H. Baalman,⁶ F.M. McAuliffe,⁶ J.G.
5 Thornton,⁸ M. Kok¹ and M.A. Oudijk.¹

6 Affiliations

7 ¹Department of Obstetrics and Gynaecology, Amsterdam UMC, University of Amsterdam, The Netherlands

8 ²Julius Centre for Health Sciences and Primary Care, University Medical Centre Utrecht, Utrecht University, the
9 Netherlands

10 ³Department of Health Sciences, Faculty of Science, Vrije Universiteit Amsterdam, Amsterdam Public Health
11 research institute, The Netherlands

12 ⁴Department of Neonatology, Amsterdam UMC, Vrije Universiteit van Amsterdam, University of Amsterdam, the
13 Netherlands.

14 ⁵The Robinson Institute, School of Paediatrics and Reproductive Health, University of Adelaide, Australia.

15 ⁶UCD Perinatal Research Centre, School of Medicine, University College Dublin, National Maternity Hospital,
16 Dublin, Ireland

17 ⁷Department of Child Health Obstetrics and Gynaecology, Nottingham University Hospitals NHS Trust, University
18 of Nottingham, United Kingdom

19 Corresponding author

20 M.A. Oudijk, MD, PhD

21 Email: m.a.oudijk@amc.nl

22 Amsterdam University Medical Centres, location AMC

23 PO box 22660 | 1100 DD Amsterdam

24 Telephone: +31 20 5667204

25
26 W. Breebaart: breebaart@gmail.com

27 C. Roos: c.dijkstra@amc.uva.nl

28 C.A. Naaktgeboren: c.naaktgeboren@umcutrecht.nl

29 J.A.M van der Post: j.a.vanderpost@amc.uva.nl

30 J.E. Bosmans: j.e.bosmans@vu.nl

31 A.H. van Kaam: a.h.vankaam@amc.uva.nl

32 E. Schuit: e.schuit@umcutrecht.nl

33 B.W. Mol: ben.mol@monash.edu

34 J.H. Baalman: jelle.baalman@gmail.com

35 F.M. McAuliffe: fionnuala.mcauliffe@ucd.ie

36 J.G. Thornton: jim.thornton@nottingham.ac.uk

37 M.A. Oudijk: m.a.oudijk@amc.uva.nl

38 M. Kok: m.kok@amc.uva.nl

39 Abstract

40 **Introduction:** Preterm birth complicates more than 15 million pregnancies annually
41 worldwide. In many countries, women who present with signs of preterm labour are treated
42 with tocolytics for 48 hours. Although this delays birth, it has never been shown to improve
43 neonatal outcome. In 2015 the WHO stated that the use of tocolytics should be reconsidered
44 and that large placebo controlled studies to evaluate the effectiveness of tocolytics are
45 urgently needed.

46 **Methods and analysis:** An international multicentre, randomised, double blinded, placebo-
47 controlled clinical trial.

48 **Participants:** Women with threatened preterm birth (gestational age 30 – 34 weeks) defined
49 as uterine contractions with

- 50 1) a cervical length of ≤ 15 mm or
- 51 2) a cervical length of 15-30 mm and a positive fibronectin test or
- 52 3) in centres where cervical length measurement is not part of the local protocol: a positive
53 fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or
- 54 4) ruptured membranes.

55 **Intervention:** Atosiban infusion for 48 hours

56 **Control:** placebo infusion for 48 hours

57 **Primary outcome:** A composite of perinatal mortality and severe neonatal morbidity.

58 **Analysis:** Analysis will be by intention to treat. A sample size of 1514 participants (757 per
59 group) will detect a reduction in adverse neonatal outcome from 10% to 6% (alpha 0.05, beta
60 0.2). A cost-effectiveness analysis will be performed from a societal perspective.

61 **Ethics and dissemination:** The Ethics Committee of the Amsterdam University Medical
62 Centres, location AMC, has approved this study. The results will be presented at
63 conferences and published in a peer-reviewed journal. Participants will be informed about the
64 results.

65 **Discussion:** This trial will show whether tocolysis with atosiban reduces adverse neonatal
66 outcome in women with threatened preterm birth at 30-34 weeks gestation.

67 **Trial registration number:** NTR6646 (date registration 24-aug-2017)

68 **Keywords:** preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

69 Article summary

70 Strengths and limitations of this study:

- 71 • The primary outcome is perinatal mortality and neonatal morbidity, not prolongation of
72 pregnancy.
- 73 • This is the largest randomised trial comparing atosiban to placebo for women with
74 threatened preterm birth.
- 75 • Over 40 hospitals in Europe will participate.
- 76 • Tocolysis is incorporated in daily routine as it has been the recommendation in many
77 guidelines. This will prove to be a challenge in counselling patients to participate in a
78 placebo controlled trial, especially in an acute setting.

79 Introduction

80 Preterm birth, defined as birth before 37 weeks gestation, is a major contributor to perinatal
81 mortality and morbidity, complicating over 15 million pregnancies worldwide.^{1,2} Of all infant
82 deaths before the age of 5 years, more than one third can be attributed to preterm birth.³ In
83 addition, spontaneous preterm birth is the leading cause of neonatal morbidity, mostly due to
84 respiratory immaturity, intracranial haemorrhage and infections.^{4,5} These conditions can have
85 long-term neurodevelopmental sequelae such as cognitive impairment, cerebral palsy and
86 visual and hearing deficiencies. Preterm birth is one of the largest single contributors to the
87 Global Burden of Disease because of the high mortality early in life and the morbidity of
88 lifelong impairment.⁶

89 Maternal administration of corticosteroids to accelerate fetal lung maturation is an effective
90 treatment for women with threatened preterm birth.⁷ Since steroids have their maximum
91 effect if birth is delayed by 48 hours, many obstetricians administer a tocolytic drug alongside
92 the steroids to allow maximal steroid effect and facilitate transport of the mother to a centre
93 with neonatal intensive care unit facilities if needed. Several tocolytics are used, including β
94 adrenoceptor agonists, cyclooxygenase inhibitors (COX), magnesium sulphate, calcium-
95 channel blockers and oxytocin receptor antagonists. Though more or less effective in
96 delaying delivery, no tocolytics used in obstetrical practice are proven effective in reducing
97 neonatal morbidity and mortality.^{8,9} The two most commonly used tocolytic drugs, atosiban
98 and nifedipine, showed comparable perinatal outcome in the APOSTEL III study.¹⁰ However,
99 neonatal mortality was higher in the nifedipine group, although not significant (5.4% vs. 2.4%
100 RR 2.20; 95% CI 0.91-5.33).

101 The oxytocin receptor antagonist atosiban has fewer maternal side effects in head to head
102 comparison with alternative drugs.¹¹ In placebo controlled trials, atosiban has not shown a
103 reduction in perinatal mortality (RR 2.25, 95% CI 0.79 to 6.38; two studies with 729 infants)
104 or major neonatal morbidity.¹²

1
2
3 105 One explanation might be that since spontaneous preterm birth is associated in 40-70% of
4 106 cases with chorioamnionitis,^{13,14} tocolysis may prolong fetal exposure to an infectious
5 107 environment, which may worsen neonatal outcome.

6
7
8 108 Perinatal outcome has also markedly improved over the last few decades, in part due to
9 109 postnatal interventions such as exogenous surfactant treatment which reduces mortality and
10 110 respiratory morbidity in preterm infants.¹⁵ This might also limit the potential benefit of
11 111 tocolytics.

12
13
14
15 112 Worldwide, practice varies widely. Several large institutions in countries like Canada,
16 113 Scotland and Ireland, rarely use tocolytics, while in the USA, cyclooxygenase inhibitors
17 114 (indomethacin) and calcium channel blockers (nifedipine) are popular. In Europe, nifedipine
18 115 and the oxytocin antagonist, atosiban, are both widely used.

19
20
21
22
23 116 In conclusion, current widespread use of tocolytic drugs for this indication is not supported by
24 117 the available evidence. The primary goal of tocolysis should not be prolongation of
25 118 pregnancy, but improvement of neonatal outcome. This view is supported by the WHO, as
26 119 they state in their 2015 guidelines on preterm birth that the effectiveness of tocolytics is not
27 120 proven, and that placebo controlled studies are urgently needed.¹⁶ Based on the results of
28 121 the APOSTEL III study,¹⁰ the associated editorial,¹⁷ and its safety profile we chose to
29 122 evaluate atosiban in the APOSTEL 8 study.

30 31 32 33 34 35 123 **Objective**

36
37 124 To test the hypothesis that tocolysis with atosiban in late preterm birth (30 to 34 weeks)
38 125 reduces neonatal mortality and morbidity and is cost-effective compared with placebo.

39
40
41 126

42 43 44 127 **Methods and analysis**

45 46 47 128 **Design and setting**

48
49 129 We will conduct an international, multicentre, double-blind, randomised, placebo-controlled
50 130 clinical trial, performed in The Netherlands, Belgium, United Kingdom and Ireland.

51 52 53 131 **Participants/eligibility criteria**

54
55
56 132 Women, aged ≥ 18 years, with threatened preterm birth and a gestational age between 30⁺⁰
57 133 and 33⁺⁶ weeks are eligible. Threatened preterm birth is defined as uterine contractions with
58 134 1) a cervical length of ≤ 15 mm or

- 1
2
3 135 2) a cervical length of 15-30 mm and a positive fibronectin test or
4 136 3) Or in centres where cervical length measurement is not part of the local protocol: a
5 137 positive fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or
6 138 4) ruptured membranes.

9
10 139 Both women with singleton and multiple pregnancies are eligible.
11 140 Women with a contra-indication for tocolysis, signs of fetal distress, clinical signs of intra-
12 141 uterine infection, previous treatment for threatened preterm birth with corticosteroids in the
13 142 current pregnancy and known fetal chromosomal or severe structural abnormalities are not
14 143 eligible.

19 144 **Procedures, recruitment, randomization and collection of data**

20
21 145 Potential participants will be identified by the local research co-ordinators and/or the staff of
22 146 participating hospitals. Women eligible for the trial will be counselled by doctors, midwives or
23 147 research nurses trained in 'good clinical practice', and will be given a patient information form
24 148 to read. Those who wish to participate, will be asked to give for written informed consent, and
25 149 are registered within the central trial database. Randomisation will be performed by using
26 150 sequentially numbered medication packs available in each centre. Only the independent data
27 151 manager has access to the computer-generated randomization list in which the medication
28 152 numbers are linked with atosiban or placebo. Treatment allocation is blinded to investigators,
29 153 participants, clinicians and research coordinators. Randomisation will be balanced with
30 154 varying block sizes of 2 and 4, and stratified by centre.

31
32 155 At study entry, baseline demographic, past obstetric and medical history will be recorded into
33 156 the web-based Case Report Form (CRF) accessible through a secure central website
34 157 (Castor Electronic Data Capture, Ciwit B.V.)¹⁸ Details of delivery, maternal and neonatal
35 158 assessments during pregnancy and post-partum period will be recorded on the same
36 159 system. All data will be coded, processed and stored with adequate precautions to ensure
37 160 patient confidentiality.

48 161 **Interventions**

49
50
51 162 Participants are allocated to atosiban or matching placebo (0.9% saline) for 48 hours. The
52 163 medication will be administered by a bolus injection of 6.75 mg/0.9 ml in one minute followed
53 164 by a continuous infusion of 18 mg/hour for 3 hours followed by a continuous infusion of 6
54 165 mg/hour for the remaining 45 hours. Participating women will otherwise be treated according
55 166 to local protocol based on national guidelines, including corticosteroids and antibiotics if
56 167 needed.

168 Outcome measures

169 Outcome parameters are in line with the core outcome set for studies on prevention of
170 preterm birth defined by members of GONet and the Core Outcomes in Women's health
171 (CROWN) initiative (www.crown-initiative.org).¹⁹

172 The primary outcome measure is a composite of adverse perinatal outcome composed of
173 perinatal in-hospital mortality and six severe perinatal morbidities: bronchopulmonary (BPD)
174 dysplasia at 36 weeks postmenstrual age, periventricular leucomalacia (PVL) > grade 1,
175 intraventricular haemorrhage > grade 2, necrotizing enterocolitis (NEC) \geq stage 2,
176 retinopathy of prematurity > grade 2 or needing laser therapy, and culture proven sepsis.
177 The diagnosis of BPD will be made according to the international consensus guideline as
178 described by Jobe and Bancalari.²⁰ PVL > grade 1 and intraventricular haemorrhage > grade
179 2 will be diagnosed by repeated cranial ultrasound according to the guidelines on neuro
180 imaging described by de Vries et al.²¹ and Ment et al.²² NEC \geq stage 2 will be diagnosed
181 according to Bell.²³ Culture proven sepsis is diagnosed on the combination of clinical signs
182 and positive blood cultures. The components of the composite adverse perinatal outcome will
183 also be assessed separately.

184 Secondary infant outcomes will be birth within 48 hours, time to birth, gestational age at birth,
185 birth weight, number of days on invasive mechanical ventilation, length of NICU stay,
186 convulsions, asphyxia, meningitis, pneumothorax until hospital discharge.

187 Maternal outcomes will be mortality, infection of inflammation, prelabor rupture of
188 membranes and harm to mother from interventions (side effects). Side effects are defined as
189 admission to intensive care, anaphylactic shock, dyspnoea, hypotension (leading to CTG
190 abnormalities), liver test abnormalities (elevated ASAT or ALAT), general side effects
191 (nausea, vomiting, headache), post-partum haemorrhage defined as > 500 ml blood loss and
192 maternal mortality.

193 We will ask informed consent to approach the parents for long-term follow-up of the children.
194 We intend, subject to funding, to use standardized questionnaires at 2 years and 5 years of
195 age.

196 Maternal quality of life will be assessed at randomisation and at three months baby corrected
197 age using the EuroQol (EQ-5D-5L) questionnaire. This consists of five dimensions (mobility,
198 self-care, usual activities, pain/discomfort, anxiety/depression) that are rated using five levels
199 (no problems, slight problems, moderate problems, severe problems, extreme problems).

200 Societal costs will be assessed using adapted versions of the iMTA Medical and Productivity
201 Cost Questionnaires at 40 weeks postmenstrual age and 3 months corrected baby age. Cost
202 data include costs of the intervention, other health care utilization, patient and family costs
203 and costs of productivity losses.

204 **Withdrawal of subjects**

205 Participants can cease study treatment at any time for any reason if they wish to do so.
206 Unless they refuse to allow further data collection, such participants will continue to be
207 followed-up and will be analysed in the group to which they were originally allocated.
208 Participants who decline follow-up will have no further trial data collected. Any results
209 collected up to the point at which they decline follow up will be analysed. Study medication
210 will be discontinued in patients with signs of intra-uterine infections or signs of fetal distress
211 (abnormal CTG, meconium stained amniotic fluid). Data of such participants will continue to
212 be analysed. Further management will be left to the expertise of the responsible clinician.
213 The responsible clinician can contact a perinatologist from the project group in case of
214 suspected side effects or other medical problems. If necessary, treatment will be
215 discontinued.

216 **Monitoring and Safety**

217 An independent data safety monitoring board (DSMB) will focus on both effectiveness and
218 safety. Serious Adverse Events (SAEs) will be collected from the first study-related
219 procedure until 3 months after delivery. All SAEs will be reported to the DSMB within 7 days.
220 Whenever there is proof of effectiveness (at interim analysis) or safety issues (increased
221 (serious) adverse events in one of the two treatment arms) the DSMB will advise whether the
222 trial should be stopped or continued. The data safety monitoring board will be blinded when
223 first analysing the data, but unblinded before reaching a decision.

224 The advice(s) of the DSMB will only be sent to the sponsor of the study. Should the sponsor
225 decide not to fully implement the advice of the DSMB, the sponsor will send the advice to the
226 reviewing Medical Ethics Committee, including a note to substantiate why (part of) the advice
227 of the DSMB will not be followed.

228 A formal interim analysis is planned after data collection of 500 and of 1000 women. At these
229 interim analyses, the Haybittle-Peto alpha spending function will be used, which means that
230 an effect at interim with a p-value <0.001 is considered statistically significant.

231 **Sample size**

1
2
3 232 Based on the APOSTEL III data, the proportion of adverse perinatal outcome in women
4 233 randomized between 30 and 34 weeks gestation and treated with atosiban was 6%.¹⁰ To
5 234 show a 40% reduction (10% in the placebo group to 6% in the atosiban group) we need to
6 235 randomise 1438 women (beta-error 0.2; alpha error 0.05). Assuming a 5% drop-out or loss-to
7 236 follow up rate, we will randomize 1514 women (757 in each arm).

11 237 **Statistical analysis**

14 238 **Data analysis**

16 239 Data analysis will be performed according to the intention-to-treat principle. In the baseline
17 240 table, categorical variables will be expressed as a number with the percentage of the total
18 241 allocation arm. Continuous variables will be presented as mean with standard deviation, as
19 242 geometric mean with 95% confidence interval (CI) or as median with interquartile range,
20 243 whichever appropriate.

24 244 The main outcome 'adverse neonatal outcome' will be assessed on the infant level, using a
25 245 log-binomial generalized estimating equations model (GEEs), resulting in a relative risk (RR)
26 246 with accompanying 95% confidence interval (CI). To account for stratified randomization by
27 247 centre, we will also take centre into the model if the model converges.²⁴ We will account for
28 248 interdependence between outcomes in multiple pregnancies by considering the mother as a
29 249 cluster variable.²⁵

35 250 The other secondary outcome measures on the child level will be analyzed similarly to the
36 251 primary outcome measure. Outcomes on the maternal level will be assessed by using a
37 252 binomial regression model with log-link function. When a statistically significant difference in
38 253 primary outcome is found between both groups, we will calculate the number needed to treat
39 254 (NNT). Time to delivery will be evaluated by Kaplan-Meier estimates and Cox proportional
40 255 hazard analysis, taking into account the different durations of gestation at study entry, and
41 256 will be tested with the log rank test. A p-value of less than 0.05 will be considered to indicate
42 257 statistical significance.

48 258 **Subgroup analyses**

50 259 The following subgroup analyses are planned:

- 52 260 1) singleton versus multiple pregnancy,
- 55 261 2) cervical length < 15 mm, versus cervical length 15 - 30 mm and a positive fibronectin test
56 262 (or no cervical length measurement and a positive Fibronectin test or Partus test),
- 59 263 3) ruptured or unruptured membranes at entry

1
2
3 264 5) previous preterm birth.
4

5 265 **Sensitivity analysis**

7 266 A sensitivity analysis will be performed excluding multiple pregnancies and pregnancies
8 267 complicated by preterm premature rupture of membranes.

10
11 268 To assess whether a subgroup effect is present we will add an interaction term between the
12 269 subgrouping variables and the treatment allocation to the regression model. When an
13 270 interaction term is statistically significant ($p < 0.05$), we will estimate the treatment effect within
14 271 strata of the subgrouping variable.

17
18 272 Details of the statistical analysis will be describes in separate statistical analysis plan that will
19 273 be completed before data lock.

21
22 274 **Cost-effectiveness analysis**

23 275 The cost-effectiveness analysis will be done according to the intention-to treat principle.
24 276 Missing cost and effect data will be imputed using multiple imputation according to the MICE
25 277 algorithm developed by van Buuren.²⁶ Rubin's rules will be used to pool the results from the
26 278 different multiply imputed datasets. Bivariate regression analyses will be used to estimate
27 279 cost and effect differences between atosiban and placebo while adjusting for confounders if
28 280 necessary. Incremental cost-effectiveness ratios (ICERs) will be calculated by dividing the
29 281 difference in the mean total costs between the treatment groups by the difference in mean
30 282 effect between the treatment groups. Bias-corrected and accelerated bootstrapping with
31 283 5000 replications will be used to estimate statistical uncertainty. Uncertainty surrounding
32 284 ICERs will be graphically presented on cost-effectiveness planes. Cost-effectiveness
33 285 acceptability curves will be estimated showing the probability that atosiban is cost-effective in
34 286 comparison with placebo for a range of different ceiling ratios thereby showing decision
35 287 uncertainty. Sensitivity analyses will be performed to assess the robustness of the results
36 288 using different assumptions regarding costs and effects.

37
38
39
40
41
42
43
44
45
46 289 **Patient and Public Involvement**

48 290 The preterm birth research line of the Dutch Consortium is in close collaboration with two
49 291 Dutch patient associations, the VOC (Vereniging van Ouders van Couveusekinderen, freely
50 292 translated to society of parents of children admitted to NICU) and the NVOM (Nederlandse
51 293 Vereniging van Ouders van Meerlingen, freely translated to Dutch society of parents of
52 294 multiples). They are involved in the design of new studies, updated on progress of running
53 295 trials, and informed of study results. Project members are invited speakers at yearly
54 296 conferences of these societies to present on the progress of our preterm birth research line.
55 297 At these conferences, surveys are being performed on patient preferences on study ideas.
56
57
58
59
60

298 Tocolysis was deemed an important research issue. Both associations have written support
299 letters to the funding agency ZonMw (The Netherlands organization for health research and
300 development) for the APOSTEL 8 study.

301 A project panel of parents that experienced a spontaneous preterm birth consisting of 6
302 couples was involved in the design of our study. A survey was performed during the design
303 of the study amongst members of the closed Facebook group of the VOC, to address
304 questions on whether they would be interested in participation in the APOSTEL 8 study.
305 The Dutch consortium has a website where it publishes all results of completed studies, and
306 publishes the protocols of currently recruiting studies.
307 Presentations will be held at yearly conferences at patient organizations and updates on
308 research are being published in the journal of the VOC.

309 **Ethics and dissemination**

310 The Medical Ethics Committee at the Amsterdam University Medical Centres, location AMC,
311 approved this study. Additional regional approval was obtained for the remaining participating
312 hospitals in The Netherlands. For Ireland and United Kingdom, both national and local
313 authorities approved this trial according to national regulations.

314 This trial is registered with the Netherlands Trial Register, NTR6646.

315 A manuscript with the results of the primary study will be published in a peer-reviewed
316 journal. A separate manuscript will be written on the cost effectiveness analysis.

317 The results of this clinical trial will be presented at conferences and disseminated through
318 publication in a peer-reviewed journal.

319 **Acknowledgements**

320 We would like to thank the collaborators of the study group; the gynecologists of the
321 participating centres for their help as local investigators for the APOSTEL 8 study.

322 We also thank the patient associations VOC and NVOW for their input in this study.

323 **Author contributions**

324 CAN, JAMP, JEB, AHLcVcK, BWM, JHB, FMcA, JGT, MAO, MK were involved in conception
325 and design of the study.

326 JK and WB drafted the manuscript.

327 CR, CAN, JAMP, JEB, AHLcVcK, BWM, JHB, FMcA, JGT, MAO, MK reviewed and edited the
328 manuscript. All authors mentioned in the manuscript are member of the APOSTEL study

1
2
3 329 group or collaborators. They participated in the design of the study during several meetings
4 330 and are local investigators in the participating centres. All authors edited the manuscript and
5 331 read and approved the final manuscript.
6
7

8 9 332 **Funding**

10
11 333 This study is funded by ZonMw (The Netherlands organization for health research and
12 334 development), grant number 848041004 and the United Kingdom National Institute for Health
13 335 Research, Clinical Research Network.
14
15
16

17 336 **Competing interests statement**

18
19
20 337 JGT received a number of lectures and medical advisory fees from Ferring pharmaceuticals
21 338 between 2000 and 2016.
22
23

24 339 **References**

- 25
26
27 340 1. Howson CP, Kinney MV, Lawn JE (editors). Born Too Soon: The Global Action
28 341 Report on Preterm Birth. Geneva: World Health Organization, 2012
29
30 342 2. Liu L, Johnson HL, Cousens S, Perin J, Scott S, Lawn JE, et al. Global, regional, and
31 343 national causes of child mortality: an updated systematic analysis for 2010 with time
32 344 trends since 2000. *Lancet* 2012;379(9832):2151–61
33
34 345 3. Matthews TJ, Macdorman MF, Thoma ME. Infant Mortality Statistics From the 2013
35 346 Period Linked Birth/Infant Death Data Set. *Natl Vital Stat Rep*. 2015;64(9):1-30
36
37 347 4. Hack M, Fanaroff AA. Outcomes of children of extremely low birthweight and
38 348 gestational age in the 1990's. *Early Hum Dev*. 1999;53(3):193-218.
39
40 349 5. Moore T, Hennessy EM, Myles J, et al. Neurological and developmental outcome in
41 350 extremely preterm children born in England in 1995 and 2006: the EPICure studies.
42 351 *BMJ*. 2012;345:e7961.
43
44 352 6. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235
45 353 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the
46 354 Global Burden of Disease Study 2010. *Lancet*. 2012;380(9859):2095-128.
47
48 355 7. Roberts D, Brown J, Medley N, Dalziel SR. Antenatal corticosteroids for accelerating
49 356 fetal lung maturation for women at risk of preterm birth. *Cochrane Database Syst*
50 357 *Rev*. 2017;3:CD004454.
51
52 358 8. Haas DM, Imperiale TF, Kirkpatrick PR, Klein RW, Zollinger TW, Golichowski AM.
53 359 Tocolytic therapy: a meta-analysis and decision analysis. *Obstet Gynecol* 2009; 113:
54 360 585–94.
55
56
57
58
59
60

- 1
2
3 361 9. Haas DM, Caldwell DM, Kirkpatrick P, McIntosh JJ, Welton NJ. Tocolytic therapy for
4 362 preterm delivery: systematic review and network meta-analysis. *BMJ* 2012; 345:
5 363 e6226.
6
7 364 10. Van Vliet EO, Nijman TA, Schuit E, et al. Nifedipine versus atosiban for threatened
8 365 preterm birth (APOSTEL III): a multicentre, randomised controlled trial. *Lancet*.
9 366 2016;387(10033):2117-24.
10 367 11. De Heus R, Mol BW, Erwich JJ, et al. Adverse drug reactions to tocolytic treatment
11 368 for preterm labour: prospective cohort study. *BMJ*. 2009;338:b744.
12 369 12. Flenady V, Reinebrant HE, Liley HG, Tambimuttu EG, Papatsonis DNM. Oxytocin
13 370 receptor antagonists for inhibiting preterm labour. *Cochrane Database of Systematic*
14 371 *Reviews* 2014, Issue 6. Art. No.: CD004452.
15 372 13. Tita AT, Andrews WW. Diagnosis and management of clinical chorioamnionitis. *Clin*
16 373 *Perinatol*. 2010;37(2):339-54.
17 374 14. Romero R, Dey SK, Fisher SJ. Preterm labor: one syndrome, many causes. *Science*.
18 375 2014;345(6198):760-5.
19 376 15. Soll R, Ozek E. Prophylactic protein free synthetic surfactant for preventing morbidity
20 377 and mortality in preterm infants. *Cochrane Database Syst Rev*. 2010;(1):CD001079.
21 378 16. WHO (2015, November 17). WHO recommendation on the use of tocolytic treatment
22 379 for inhibiting preterm labour. Retrieved from:
23 380 [https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
24 381 [care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
25 382 [treatment-inhibiting-preterm-labour](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
26 383 17. Walker KF, Thornton JG. Tocolysis and preterm labour. *Lancet*.
27 384 2016;387(10033):2068-2070.
28 385 18. Castor: Amsterdam, Ciwit BV the N. Castor electronic data capture. 2017.
29 386 19. van 't Hooft J, Duffy JM, Daly M, Williamson P, Meher S, Thom E, et al. A core
30 387 outcome Set for evaluation of interventions to prevent preterm birth. *Obstet Gynecol*.
31 388 2016;127(1):49–58.
32 389 20. Jobe AH, Bancalari E: Bronchopulmonary dysplasia. *Am J Respir Crit Care. Med*
33 390 2001, 163:1723–1729.
34 391 21. de Vries LS, Eken P, Dubowitz LM: The spectrum of leukomalacia using cranial
35 392 ultrasound. *Behav Brain Res* 1992, 49:1–6.
36 393 22. Ment LR, Bada HS, Barnes P, Grant PE, Hirtz D, Papile LA, Pinto-Martin J, Rivkin M,
37 394 Slovis TL: Practice parameter: neuroimaging of the neonate: report of the Quality
38 395 Standards Subcommittee of the American Academy of Neurology and the Practice
39 396 Committee of the Child Neurology Society. *Neurology* 2002, 58(12):1726–1738.
40 397 23. Bell MJ: Neonatal necrotizing enterocolitis. *Ann Surg* 1978, 187:1–7.

- 1
2
3 398 24. Kahan BC. Accounting for centre-effects in multicentre trials with a binary outcome -
4 399 when, why, and how?. *BMC Med Res Methodol*. 2014;14:20.
5
6 400 25. Gates S, Brocklehurst P. How should randomised trials including multiple
7 pregnancies be analysed?. *BJOG*. 2004;111(3):213-9.
8 401
9 402 26. Van Buuren, S., and Groothuis-Oudshoorn, K. (2011). MICE: Multivariate Imputation
10 by Chained Equations in R. *Journal of Statistical Software* 45(3), 1-68
11 403
12

13 404 **Word count**

14
15
16 405 2611 words
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60



SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description
Administrative information		
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry
	2b	All items from the World Health Organization Trial Registration Data Set
Protocol version	3	Date and version identifier
Funding	4	Sources and types of financial, material, and other support
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors
	5b	Name and contact information for the trial sponsor
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)
Introduction		
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention
	6b	Explanation for choice of comparators
Objectives	7	Specific objectives or hypotheses
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)

Methods: Participants, interventions, and outcomes

Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size

Methods: Assignment of interventions (for controlled trials)

Allocation:

Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions
---------------------	-----	--

1			
2	Allocation	16b	Mechanism of implementing the allocation sequence (eg, central
3	concealment		telephone; sequentially numbered, opaque, sealed envelopes),
4	mechanism		describing any steps to conceal the sequence until interventions are
5			assigned
6			
7	Implementation	16c	Who will generate the allocation sequence, who will enrol participants,
8			and who will assign participants to interventions
9			
10	Blinding	17a	Who will be blinded after assignment to interventions (eg, trial
11	(masking)		participants, care providers, outcome assessors, data analysts), and
12			how
13		17b	If blinded, circumstances under which unblinding is permissible, and
14			procedure for revealing a participant's allocated intervention during
15			the trial
16			
17			
18			
19			

Methods: Data collection, management, and analysis

20			
21	Data collection	18a	Plans for assessment and collection of outcome, baseline, and other
22	methods		trial data, including any related processes to promote data quality (eg,
23			duplicate measurements, training of assessors) and a description of
24			study instruments (eg, questionnaires, laboratory tests) along with
25			their reliability and validity, if known. Reference to where data
26			collection forms can be found, if not in the protocol
27		18b	Plans to promote participant retention and complete follow-up,
28			including list of any outcome data to be collected for participants who
29			discontinue or deviate from intervention protocols
30			
31	Data	19	Plans for data entry, coding, security, and storage, including any
32	management		related processes to promote data quality (eg, double data entry;
33			range checks for data values). Reference to where details of data
34			management procedures can be found, if not in the protocol
35			
36	Statistical	20a	Statistical methods for analysing primary and secondary outcomes.
37	methods		Reference to where other details of the statistical analysis plan can be
38			found, if not in the protocol
39		20b	Methods for any additional analyses (eg, subgroup and adjusted
40			analyses)
41		20c	Definition of analysis population relating to protocol non-adherence
42			(eg, as randomised analysis), and any statistical methods to handle
43			missing data (eg, multiple imputation)
44			
45			
46			
47			
48			
49			
50			
51			

Methods: Monitoring

52			
53	Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role
54			and reporting structure; statement of whether it is independent from
55			the sponsor and competing interests; and reference to where further
56			details about its charter can be found, if not in the protocol.
57			Alternatively, an explanation of why a DMC is not needed
58			
59			
60			

1			
2		21b	Description of any interim analyses and stopping guidelines, including
3			who will have access to these interim results and make the final
4			decision to terminate the trial
5			
6	Harms	22	Plans for collecting, assessing, reporting, and managing solicited and
7			spontaneously reported adverse events and other unintended effects
8			of trial interventions or trial conduct
9			
10	Auditing	23	Frequency and procedures for auditing trial conduct, if any, and
11			whether the process will be independent from investigators and the
12			sponsor
13			
14			

Ethics and dissemination

15			
16			
17	Research ethics	24	Plans for seeking research ethics committee/institutional review board
18	approval		(REC/IRB) approval
19			
20	Protocol	25	Plans for communicating important protocol modifications (eg,
21	amendments		changes to eligibility criteria, outcomes, analyses) to relevant parties
22			(eg, investigators, REC/IRBs, trial participants, trial registries, journals,
23			regulators)
24			
25			
26	Consent or assent	26a	Who will obtain informed consent or assent from potential trial
27			participants or authorised surrogates, and how (see Item 32)
28			
29		26b	Additional consent provisions for collection and use of participant data
30			and biological specimens in ancillary studies, if applicable
31			
32	Confidentiality	27	How personal information about potential and enrolled participants will
33			be collected, shared, and maintained in order to protect confidentiality
34			before, during, and after the trial
35			
36			
37	Declaration of	28	Financial and other competing interests for principal investigators for
38	interests		the overall trial and each study site
39			
40	Access to data	29	Statement of who will have access to the final trial dataset, and
41			disclosure of contractual agreements that limit such access for
42			investigators
43			
44			
45	Ancillary and	30	Provisions, if any, for ancillary and post-trial care, and for
46	post-trial care		compensation to those who suffer harm from trial participation
47			
48	Dissemination	31a	Plans for investigators and sponsor to communicate trial results to
49	policy		participants, healthcare professionals, the public, and other relevant
50			groups (eg, via publication, reporting in results databases, or other
51			data sharing arrangements), including any publication restrictions
52			
53		31b	Authorship eligibility guidelines and any intended use of professional
54			writers
55			
56		31c	Plans, if any, for granting public access to the full protocol, participant-
57			level dataset, and statistical code
58			
59			
60			

Appendices

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "[Attribution-NonCommercial-NoDerivs 3.0 Unported](#)" license.

For peer review only

BMJ Open

Study protocol for a randomised trial for atosiban versus placebo in threatened preterm birth: the APOSTEL 8 study

Journal:	<i>BMJ Open</i>
Manuscript ID	bmjopen-2019-029101.R1
Article Type:	Protocol
Date Submitted by the Author:	11-Jun-2019
Complete List of Authors:	<p>Klumper, Job; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Breebaart, Wouter; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Roos, Carolien; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Naaktgeboren, CA ; Julius Centrum voor Gezondheidswetenschappen en Eerstelijns Geneeskunde, University Medical Centre Utrecht van der Post, Joris; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Bosmans, J; Vrije Universiteit Amsterdam, Department of Health Sciences, Amsterdam Public Health research institute van Kaam, Anton; Amsterdam UMC, location AMC and VUmc, Department of neonatology Schuit, Ewoud; Julius Center for Health Sciences and Primary Care, University Medical Centre Utrecht; Stanford University, Stanford Prevention Research Center Mol, Ben; School of Medicine, Monash University, Melbourne, Australia, Department of obstetrics & gynaecology Baalman, Jelle; National Maternity Hospital, Department of obstetrics & gynaecology McAuliffe, Fionnuala; University College Dublin, UCD Perinatal Research Centre; National Maternity Hospital, Department of obstetrics & gynaecology Thornton, Jim; University of Nottingham, Department of obstetrics & gynaecology Kok, Marjolein; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Oudijk, Martijn; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p>
Primary Subject Heading:	Obstetrics and gynaecology
Secondary Subject Heading:	Obstetrics and gynaecology
Keywords:	preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60



1 Study protocol for a randomised trial for atosiban versus 2 placebo in threatened preterm birth: the APOSTEL 8 study

3 J. Klumper,¹ W. Breebaart,¹ C. Roos,¹ C.A. Naaktgeboren,² J.A.M van der Post,¹ J.E.
4 Bosmans,³ A.H. van Kaam,⁴ E. Schuit,² B.W. Mol,⁵ J.H. Baalman,⁶ F.M. McAuliffe,⁶ J.G.
5 Thornton,⁸ M. Kok¹ and M.A. Oudijk.¹

6 Affiliations

7 ¹Department of Obstetrics and Gynaecology, Amsterdam UMC, University of Amsterdam, The Netherlands

8 ²Julius Centre for Health Sciences and Primary Care, University Medical Centre Utrecht, Utrecht University, the
9 Netherlands

10 ³Department of Health Sciences, Faculty of Science, Vrije Universiteit Amsterdam, Amsterdam Public Health
11 research institute, The Netherlands

12 ⁴Department of Neonatology, Amsterdam UMC, Vrije Universiteit van Amsterdam, University of Amsterdam, the
13 Netherlands.

14 ⁵The Robinson Institute, School of Paediatrics and Reproductive Health, University of Adelaide, Australia.

15 ⁶UCD Perinatal Research Centre, School of Medicine, University College Dublin, National Maternity Hospital,
16 Dublin, Ireland

17 ⁷Department of Child Health Obstetrics and Gynaecology, Nottingham University Hospitals NHS Trust, University
18 of Nottingham, United Kingdom

19 Corresponding author

20 M.A. Oudijk, MD, PhD

21 Email: m.a.oudijk@amsterdamumc.nl

22 Amsterdam University Medical Centres, location AMC

23 PO box 22660 | 1100 DD Amsterdam

24 Telephone: +31 20 5667204

25
26 J. Klumper: j.klumper@amc.uva.nl

27 W. Breebaart: breebaart@gmail.com

28 C. Roos: c.dijkstra@amc.nl

29 C.A. Naaktgeboren: c.naaktgeboren@umcutrecht.nl

30 J.A.M van der Post: j.a.vanderpost@amc.uva.nl

31 J.E. Bosmans: j.e.bosmans@vu.nl

32 A.H. van Kaam: a.h.vankaam@amc.uva.nl

33 E. Schuit: e.schuit@umcutrecht.nl

34 B.W. Mol: ben.mol@monash.edu

35 J.H. Baalman: jelle.baalman@gmail.com

36 F.M. McAuliffe: fionnuala.mcauliffe@ucd.ie

37 J.G. Thornton: jim.thornton@nottingham.ac.uk

38 M. Kok: m.kok@amc.uva.nl

39 Abstract

40 **Introduction:** Preterm birth complicates more than 15 million pregnancies annually
41 worldwide. In many countries, women who present with signs of preterm labour are treated
42 with tocolytics for 48 hours. Although this delays birth, it has never been shown to improve
43 neonatal outcome. In 2015 the WHO stated that the use of tocolytics should be reconsidered
44 and that large placebo controlled studies to evaluate the effectiveness of tocolytics are
45 urgently needed.

46 **Methods and analysis:** An international multicentre, randomised, double blinded, placebo-
47 controlled clinical trial.

48 **Participants:** Women with threatened preterm birth (gestational age 30 – 34 weeks) defined
49 as uterine contractions with

50 1) a cervical length of ≤ 15 mm or

51 2) a cervical length of 15-30 mm and a positive fibronectin test or

52 3) in centres where cervical length measurement is not part of the local protocol: a positive
53 fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or

54 4) ruptured membranes.

55 **Intervention:** Atosiban infusion for 48 hours

56 **Control:** placebo infusion for 48 hours

57 **Primary outcome:** A composite of perinatal mortality and severe neonatal morbidity.

58 **Analysis:** Analysis will be by intention to treat. A sample size of 1514 participants (757 per
59 group) will detect a reduction in adverse neonatal outcome from 10% to 6% (alpha 0.05, beta
60 0.2). A cost-effectiveness analysis will be performed from a societal perspective.

61 **Ethics and dissemination:** The Research Ethics Committee (REC) of the Amsterdam
62 University Medical Centres, location AMC, has approved this study. The study is currently
63 under review by the local REC in Dublin, and the REC in the United Kingdom. The results will
64 be presented at conferences and published in a peer-reviewed journal. Participants will be
65 informed about the results.

66 **Discussion:** This trial will show whether tocolysis with atosiban reduces adverse neonatal
67 outcome in women with threatened preterm birth at 30-34 weeks gestation.

68 **Trial registration number:** NTR6646 (date registration 24-aug-2017)

69 **Protocol version:** 2.0, dated 27-02-2019

70 **Keywords:** preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

71 **Article summary**

72 **Strengths and limitations of this study:**

- 73 • The primary outcome is perinatal mortality and neonatal morbidity, not prolongation of
74 pregnancy.
- 75 • This is the largest randomised trial comparing atosiban to placebo for women with
76 threatened preterm birth.
- 77 • Over 40 hospitals in Europe will participate.
- 78 • Tocolysis is incorporated in daily routine as it has been the recommendation in many
79 guidelines.
- 80 • It will prove to be a challenge in counselling patients to participate in a placebo
81 controlled trial, especially in an acute setting.

82 **Introduction**

83 Preterm birth, defined as birth before 37 weeks gestation, is a major contributor to perinatal
84 mortality and morbidity, complicating over 15 million pregnancies worldwide.^{1,2} Of all infant
85 deaths before the age of 5 years, more than one third can be attributed to preterm birth.³ In
86 addition, spontaneous preterm birth is the leading cause of neonatal morbidity, mostly due to
87 respiratory immaturity, intracranial haemorrhage and infections.^{4,5} These conditions can have
88 long-term neurodevelopmental sequelae such as cognitive impairment, cerebral palsy and
89 visual and hearing deficiencies. Preterm birth is one of the largest single contributors to the
90 Global Burden of Disease because of the high mortality early in life and the morbidity of
91 lifelong impairment.⁶

92 Maternal administration of corticosteroids to accelerate fetal lung maturation is an effective
93 treatment for women with threatened preterm birth.⁷ Since steroids have their maximum
94 effect if birth is delayed by 48 hours, many obstetricians administer a tocolytic drug alongside
95 the steroids to allow maximal steroid effect and facilitate transport of the mother to a centre
96 with neonatal intensive care unit facilities if needed. Several tocolytics are used, including β
97 adrenoceptor agonists, cyclooxygenase inhibitors (COX), magnesium sulphate, calcium-
98 channel blockers and oxytocin receptor antagonists. Though more or less effective in
99 delaying delivery, no tocolytics used in obstetrical practice are proven effective in reducing
100 neonatal morbidity and mortality.^{8,9} None of the studies so far have been powered to show
101 such an effect.

102 The two most commonly used tocolytic drugs, atosiban and nifedipine, showed comparable
103 perinatal outcome in the APOSTEL III study.¹⁰ However, neonatal mortality was higher in the
104 nifedipine group, although not significant (5.4% vs. 2.4% RR 2.20; 95% CI 0.91-5.33).

1
2
3 105 The oxytocin receptor antagonist atosiban has fewer maternal side effects in head to head
4 106 comparison with alternative drugs,¹¹ and showed similar effectiveness in delaying birth
5
6 107 compared to ritodrine.¹² In placebo controlled trials, a Cochrane review showed that atosiban
7
8 108 did not reduce perinatal mortality (RR 2.25, 95% CI 0.79 to 6.38; two studies with 729
9
10 109 infants) or major neonatal morbidity¹³, although the quality of this review has been
11 110 questioned.¹⁴

12
13 111 One explanation might be that since spontaneous preterm birth is associated in 40-70% of
14 112 cases with chorioamnionitis,^{15,16} tocolysis may prolong fetal exposure to an infectious
15
16 113 environment, which may worsen neonatal outcome.

17
18
19 114 Perinatal outcome has also markedly improved over the last few decades, in part due to
20 115 postnatal interventions such as exogenous surfactant treatment which reduces mortality and
21 116 respiratory morbidity in preterm infants.¹⁷ This might also limit the potential benefit of
22 117 tocolytics.

23
24
25
26 118 Worldwide, practice varies widely. Several large institutions in countries like Canada,
27 119 Scotland and Ireland, rarely use tocolytics, while in the USA, cyclooxygenase inhibitors
28 120 (indomethacin) and calcium channel blockers (nifedipine) are popular. In Europe, nifedipine
29 121 and the oxytocin antagonist, atosiban, are both widely used.

30
31
32
33 122 In conclusion, current widespread use of tocolytic drugs for this indication is not supported by
34 123 the available evidence. The primary goal of tocolysis should not be prolongation of
35 124 pregnancy, but improvement of neonatal outcome. This view is supported by the WHO, as
36 125 they state in their 2015 guidelines on preterm birth that the effectiveness of tocolytics is not
37 126 proven, and that placebo controlled studies are urgently needed.¹⁸ Based on the results of
38 127 the APOSTEL III study,¹⁰ the associated editorial,¹⁹ and its safety profile we chose to
39 128 evaluate atosiban in the APOSTEL 8 study.

45 129 **Objective**

46
47
48 130 To test the hypothesis that tocolysis with atosiban in late preterm birth (30 to 34 weeks)
49 131 reduces neonatal mortality and morbidity and is cost-effective compared with placebo.

50
51
52 132

54 133 **Methods and analysis**

57 134 **Design and setting**

1
2
3 135 We will conduct an international, multicentre, double-blind, randomised, placebo-controlled
4 136 clinical trial, performed in The Netherlands, United Kingdom and Ireland.

7 137 **Participants/eligibility criteria**

9 138 Women, aged ≥ 18 years, with threatened preterm birth and a gestational age between 30^{+0}
11 139 and 33^{+6} weeks are eligible. Threatened preterm birth is defined as uterine contractions with
12
13 140 1) a cervical length of ≤ 15 mm or
14 141 2) a cervical length of 15-30 mm and a positive fibronectin test or
15
16 142 3) Or in centres where cervical length measurement is not part of the local protocol: a
17 143 positive fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or
18 144 4) ruptured membranes.

21 145 These inclusion criteria are based on the results of the APOSTEL I study.²⁰ Moreover, our
22 146 previous APOSTEL III study showed that half of the women with these criteria deliver within
23 147 seven days,¹⁰ validating this definition of women at high risk for preterm birth.

27 148 Both women with singleton and multiple pregnancies are eligible.

28 149 Women with a contra-indication for tocolysis, signs of fetal distress, clinical signs of intra-
29 150 uterine infection, previous treatment for threatened preterm birth with corticosteroids in the
30 151 current pregnancy and known fetal chromosomal or severe structural abnormalities are not
31 152 eligible.

36 153 **Procedures, recruitment, randomization and collection of data**

38 154 Potential participants will be identified by the local research co-ordinators and/or the staff of
39 155 participating hospitals. Women eligible for the trial will be counselled by doctors, midwives or
40 156 research nurses trained in 'good clinical practice', and will be given a patient information form
41 157 to read. Those who wish to participate, will be asked to give for written informed consent, and
42 158 are registered within the central trial database. Randomisation will be performed by using
43 159 sequentially numbered medication packs available in each centre. Only the independent data
44 160 manager has access to the computer-generated randomization list in which the medication
45 161 numbers are linked with atosiban or placebo. Treatment allocation is blinded to investigators,
46 162 participants, clinicians and research coordinators. Randomisation will be balanced with
47 163 varying block sizes of 2 and 4, and stratified by centre.

55 164 At study entry, baseline demographic, past obstetric and medical history will be recorded into
56 165 the web-based Case Report Form (CRF) accessible through a secure central website
57 166 (Castor Electronic Data Capture, Ciwit B.V.)²¹ Details of delivery, maternal and neonatal
58 167 assessments during pregnancy and post-partum period will be recorded on the same

168 system. All data will be coded, processed and stored with adequate precautions to ensure
169 patient confidentiality. This is described in a separate data management plan.

170 **Interventions**

171 Participants are allocated to atosiban or matching placebo (0.9% saline) for 48 hours. The
172 medication will be administered by a bolus injection of 6.75 mg/0.9 ml in one minute followed
173 by a continuous infusion of 18 mg/hour for 3 hours followed by a continuous infusion of 6
174 mg/hour for the remaining 45 hours. Participating women will otherwise be treated according
175 to local protocol based on national guidelines, including corticosteroids and antibiotics if
176 needed.

177 **Outcome measures**

178 Outcome parameters are in line with the core outcome set for studies on prevention of
179 preterm birth defined by members of GONet and the Core Outcomes in Women's health
180 (CROWN) initiative (www.crown-initiative.org).²²

181 The primary outcome measure is a composite of adverse perinatal outcome composed of
182 perinatal in-hospital mortality and six severe perinatal morbidities: bronchopulmonary (BPD),
183 periventricular leucomalacia (PVL) > grade 1, intraventricular haemorrhage > grade 2,
184 necrotizing enterocolitis (NEC) \geq stage 2, retinopathy of prematurity > grade 2 or needing
185 laser therapy, and culture proven sepsis.

186 The diagnosis of BPD will be made according to the international consensus guideline as
187 described by Jobe and Bancalari.²³ PVL > grade 1 and intraventricular haemorrhage > grade
188 2 will be diagnosed by repeated cranial ultrasound according to the guidelines on neuro
189 imaging described by de Vries et al.²⁴ and Ment et al.²⁵ NEC \geq stage 2 will be diagnosed
190 according to Bell.²⁶ Culture proven sepsis is diagnosed on the combination of clinical signs
191 and positive blood cultures. The components of the composite adverse perinatal outcome will
192 also be assessed separately.

193 Secondary infant outcomes will be birth within 48 hours, time to birth, gestational age at birth,
194 birth weight, number of days on invasive mechanical ventilation, length of NICU stay,
195 convulsions, asphyxia, meningitis, pneumothorax until hospital discharge.

196 Maternal outcomes will be mortality, infection of inflammation and harm to mother from
197 interventions (side effects). Side effects are defined as admission to intensive care,
198 anaphylactic shock, dyspnoea, hypotension (leading to CTG abnormalities), liver test
199 abnormalities (elevated ASAT or ALAT), general side effects (nausea, vomiting, headache),
200 post-partum haemorrhage defined as > 500 ml blood loss and maternal mortality.

1
2
3 201 We will ask informed consent to approach the parents for long-term follow-up of the children.
4 202 We intend, subject to funding, to use standardized questionnaires at 2 years and 5 years of
5 203 age.

6
7
8 204 Maternal quality of life will be assessed at randomisation and at three months baby corrected
9 205 age using the EuroQol (EQ-5D-5L) questionnaire. This consists of five dimensions (mobility,
10 206 self-care, usual activities, pain/discomfort, anxiety/depression) that are rated using five levels
11 207 (no problems, slight problems, moderate problems, severe problems, extreme problems).

12
13 208 Societal costs will be assessed using adapted versions of the iMTA Medical and Productivity
14 209 Cost Questionnaires at three months corrected baby age. Cost data include costs of the
15 210 intervention, other health care utilization, patient and family costs and costs of productivity
16 211 losses.

22 212 **Withdrawal of subjects**

23
24
25 213 Participants can cease study treatment at any time for any reason if they wish to do so.
26 214 Unless they refuse to allow further data collection, such participants will continue to be
27 215 followed-up and will be analysed in the group to which they were originally allocated.
28 216 Participants who decline follow-up will have no further trial data collected. Any results
29 217 collected up to the point at which they decline follow up will be analysed. Study medication
30 218 will be discontinued in patients with signs of intra-uterine infections or signs of fetal distress
31 219 (abnormal CTG, meconium stained amniotic fluid). Data of such participants will continue to
32 220 be analysed. Further management will be left to the expertise of the responsible clinician.
33 221 The responsible clinician can contact a perinatologist from the project group in case of
34 222 suspected side effects or other medical problems. If necessary, treatment will be
35 223 discontinued.

36 224 **Monitoring and Safety**

37
38 225 An independent data safety monitoring board (DSMB) will focus on both effectiveness and
39 226 safety. Serious Adverse Events (SAEs) will be collected from the first study-related
40 227 procedure until 3 months after delivery. All SAEs will be reported to the DSMB within 7 days.
41 228 Whenever there is proof of effectiveness (at interim analysis) or safety issues (increased
42 229 (serious) adverse events in one of the two treatment arms) the DSMB will advise whether the
43 230 trial should be stopped or continued. The data safety monitoring board will be blinded when
44 231 first analysing the data, but unblinded before reaching a decision.
45 232 The advice(s) of the DSMB will only be sent to the sponsor of the study. Should the sponsor
46 233 decide not to fully implement the advice of the DSMB, the sponsor will send the advice to the
47
48
49
50
51
52
53
54
55
56
57
58
59
60

1
2
3 234 reviewing Medical Ethics Committee, including a note to substantiate why (part of) the advice
4 235 of the DSMB will not be followed.

6
7 236 A formal interim analysis is planned after data collection of 500 and of 1000 women. At these
8 237 interim analyses, the Haybittle-Peto alpha spending function will be used, which means that
9 238 an effect at interim with a p-value <0.001 is considered statistically significant.

12 239 **Sample size**

15 240 Based on the APOSTEL III data, the proportion of adverse perinatal outcome in women
16 241 randomized between 30 and 34 weeks gestation and treated with atosiban was 6%.¹⁰ To
17 242 show a 40% reduction (10% in the placebo group to 6% in the atosiban group) we need to
18 243 randomise 1438 women (beta-error 0.2; alpha error 0.05). Assuming a 5% drop-out or loss-to
19 244 follow up rate, we will randomize 1514 women (757 in each arm).

23 245 **Statistical analysis**

26 246 **Data analysis**

28 247 Data analysis will be performed according to the intention-to-treat principle. In the baseline
29 248 table, categorical variables will be expressed as a number with the percentage of the total
30 249 allocation arm. Continuous variables will be presented as mean with standard deviation, as
31 250 geometric mean with 95% confidence interval (CI) or as median with interquartile range,
32 251 whichever appropriate.

36 252 The main outcome 'adverse neonatal outcome' will be assessed on the infant level, using a
37 253 log-binomial generalized estimating equations model (GEEs) to take into account the
38 254 correlation of outcomes in multiples, resulting in a relative risk (RR) with accompanying 95%
39 255 confidence interval (CI). To account for stratified randomization by centre, we will also take
40 256 centre into the model if the model converges.²⁷ We will account for interdependence between
41 257 outcomes in multiple pregnancies by considering the mother as a cluster variable.²⁸

46 258 The other secondary outcome measures on the child level will be analyzed similarly to the
47 259 primary outcome measure. Outcomes on the maternal level will be assessed by using a
48 260 binomial regression model with log-link function. When a statistically significant difference in
49 261 primary outcome is found between both groups, we will calculate the number needed to treat
50 262 (NNT). Time to delivery will be evaluated by Kaplan-Meier estimates and Cox proportional
51 263 hazard analysis, taking into account the different durations of gestation at study entry, and
52 264 will be tested with the log rank test. A p-value of less than 0.05 will be considered to indicate
53 265 statistical significance.

266 **Subgroup analyses**

267 The following subgroup analyses are planned:

268 1) singleton versus multiple pregnancy,

269 2) cervical length < 15 mm, versus cervical length 15 - 30 mm and a positive fibronectin test
270 (or no cervical length measurement and a positive Fibronectin test or Partus test),

271 3) ruptured or unruptured membranes at entry

272 5) previous preterm birth.

273 **Sensitivity analysis**

274 A sensitivity analysis will be performed excluding multiple pregnancies and pregnancies
275 complicated by preterm premature rupture of membranes.

276 To assess whether a subgroup effect is present we will add an interaction term between the
277 subgrouping variables and the treatment allocation to the regression model. When an
278 interaction term is statistically significant ($p < 0.05$), we will estimate the treatment effect within
279 strata of the subgrouping variable.

280 Details of the statistical analysis will be describes in separate statistical analysis plan that will
281 be completed before data lock.

282 **Cost-effectiveness analysis**

283 The cost-effectiveness analysis will be done according to the intention-to treat principle.

284 Missing cost and effect data will be imputed using multiple imputation according to the MICE
285 algorithm developed by van Buuren.²⁹ Rubin's rules will be used to pool the results from the
286 different multiply imputed datasets. Bivariate regression analyses will be used to estimate
287 cost and effect differences between atosiban and placebo while adjusting for confounders if
288 necessary. Incremental cost-effectiveness ratios (ICERs) will be calculated by dividing the
289 difference in the mean total costs between the treatment groups by the difference in mean
290 effect between the treatment groups. Bias-corrected and accelerated bootstrapping with
291 5000 replications will be used to estimate statistical uncertainty. Uncertainty surrounding
292 ICERs will be graphically presented on cost-effectiveness planes. Cost-effectiveness
293 acceptability curves will be estimated showing the probability that atosiban is cost-effective in
294 comparison with placebo for a range of different ceiling ratios thereby showing decision
295 uncertainty. Sensitivity analyses will be performed to assess the robustness of the results
296 using different assumptions regarding costs and effects.

297 **Patient and Public Involvement**

298 The preterm birth research line of the Dutch Consortium is in close collaboration with two
299 Dutch patient associations, the VOC (Vereniging van Ouders van Couveusekinderen, freely
300 translated to society of parents of children admitted to NICU) and the NVOM (Nederlandse
301 Vereniging van Ouders van Meerlingen, freely translated to Dutch society of parents of
302 multiples). They are involved in the design of new studies, updated on progress of running
303 trials, and informed of study results. Project members are invited speakers at yearly
304 conferences of these societies to present on the progress of our preterm birth research line.
305 At these conferences, surveys are being performed on patient preferences on study ideas.
306 Tocolysis was deemed an important research issue. Both associations have written support
307 letters to the funding agency ZonMw (The Netherlands organization for health research and
308 development) for the APOSTEL 8 study.
309 A project panel of parents that experienced a spontaneous preterm birth consisting of 6
310 couples was involved in the design of our study. A survey was performed during the design
311 of the study amongst members of the closed Facebook group of the VOC, to address
312 questions on whether they would be interested in participation in the APOSTEL 8 study.
313 The Dutch consortium has a website where it publishes all results of completed studies, and
314 publishes the protocols of currently recruiting studies.
315 Presentations will be held at yearly conferences at patient organizations and updates on
316 research are being published in the journal of the VOC.

317 **Ethics and dissemination**

318 The Research Ethics Committee (REC) at the Amsterdam University Medical Centres,
319 location AMC, approved this study. Additional regional approval was obtained for the
320 remaining participating hospitals in The Netherlands. The study is currently under review by
321 the local REC in Dublin, and the REC in the United Kingdom.
322 Protocol amendments will be communicated to a relevant parties.
323 This trial is registered with the Netherlands Trial Register, NTR6646.
324 A manuscript with the results of the primary study will be published in a peer-reviewed
325 journal. A separate manuscript will be written on the cost effectiveness analysis.
326 The results of this clinical trial will be presented at conferences and disseminated through
327 publication in a peer-reviewed journal.

328 **Acknowledgements**

1
2
3 329 We would like to thank the collaborators of the study group; the gynecologists of the
4 330 participating centres for their help as local investigators for the APOSTEL 8 study.
5
6 331 We also thank the patient associations VOC and NVOW for their input in this study.
7
8

9 332 **Author contributions**

10
11 333 CAN, JAMP, JEB, AHvK, ES, BWM, JHB, FMcA, JGT, MAO, MK were involved in conception
12
13 334 and design of the study.
14

15 335 JK and WB drafted the manuscript.
16

17
18 336 CR, CAN, JAMP, JEB, AHvK, ES, BWM, JHB, FMcA, JGT, MAO, MK reviewed and edited
19
20 337 the manuscript. All authors mentioned in the manuscript are member of the APOSTEL study
21
22 338 group or collaborators. They participated in the design of the study during several meetings
23
24 339 and are local investigators in the participating centres. All authors edited the manuscript and
25
26 340 read and approved the final manuscript.
27

28 341 **Funding**

29
30 342 This study is funded by ZonMw (The Netherlands organization for health research and
31
32 343 development), grant number 848041004 and the United Kingdom National Institute for Health
33
34 344 Research, Clinical Research Network.
35

36 345 **Competing interests statement**

37
38
39 346 JGT received a number of lectures and medical advisory fees from Ferring pharmaceuticals
40
41 347 between 2000 and 2016.
42

43 348 **References**

- 44
45
46 349 1. Howson CP, Kinney MV, Lawn JE (editors). Born Too Soon: The Global Action
47
48 350 Report on Preterm Birth. Geneva: World Health Organization, 2012
49
50 351 2. Liu L, Johnson HL, Cousens S, Perin J, Scott S, Lawn JE, et al. Global, regional, and
51
52 352 national causes of child mortality: an updated systematic analysis for 2010 with time
53
54 353 trends since 2000. *Lancet* 2012;379(9832):2151–61
55
56 354 3. Matthews TJ, Macdorman MF, Thoma ME. Infant Mortality Statistics From the 2013
57
58 355 Period Linked Birth/Infant Death Data Set. *Natl Vital Stat Rep.* 2015;64(9):1-30
59
60 356 4. Hack M, Fanaroff AA. Outcomes of children of extremely low birthweight and
357
gestational age in the 1990's. *Early Hum Dev.* 1999;53(3):193-218.

- 1
2
3 358 5. Moore T, Hennessy EM, Myles J, et al. Neurological and developmental outcome in
4 359 extremely preterm children born in England in 1995 and 2006: the EPICure studies.
5 360 BMJ. 2012;345:e7961.
- 6 361 6. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235
7 362 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the
8 363 Global Burden of Disease Study 2010. Lancet. 2012;380(9859):2095-128.
- 9 364 7. Roberts D, Brown J, Medley N, Dalziel SR. Antenatal corticosteroids for accelerating
10 365 fetal lung maturation for women at risk of preterm birth. Cochrane Database Syst
11 366 Rev. 2017;3:CD004454.
- 12 367 8. Haas DM, Imperiale TF, Kirkpatrick PR, Klein RW, Zollinger TW, Golichowski AM.
13 368 Tocolytic therapy: a meta-analysis and decision analysis. Obstet Gynecol 2009; 113:
14 369 585–94.
- 15 370 9. Haas DM, Caldwell DM, Kirkpatrick P, McIntosh JJ, Welton NJ. Tocolytic therapy for
16 371 preterm delivery: systematic review and network meta-analysis. BMJ 2012; 345:
17 372 e6226.
- 18 373 10. Van Vliet EO, Nijman TA, Schuit E, et al. Nifedipine versus atosiban for threatened
19 374 preterm birth (APOSTEL III): a multicentre, randomised controlled trial. Lancet.
20 375 2016;387(10033):2117-24.
- 21 376 11. De Heus R, Mol BW, Erwich JJ, et al. Adverse drug reactions to tocolytic treatment
22 377 for preterm labour: prospective cohort study. BMJ. 2009;338:b744.
- 23 378 12. Effectiveness and safety of the oxytocin antagonist atosiban versus beta-adrenergic
24 379 agonists in the treatment of preterm labour. The Worldwide Atosiban versus Beta-
25 380 agonists Study Group. BJOG. 2001;108(2):133-42.
- 26 381 13. Flenady V, Reinebrant HE, Liley HG, Tambimuttu EG, Papatsonis DNM. Oxytocin
27 382 receptor antagonists for inhibiting preterm labour. Cochrane Database of Systematic
28 383 Reviews 2014, Issue 6. Art. No.: CD004452.
- 29 384 14. Lyndrup J, and Lamont RF The choice of a tocolytic for the treatment of preterm
30 385 labor: a critical evaluation of nifedipine versus atosiban. Expert Opin. Investig. Drugs
31 386 (2007) 16(6):843-853.
- 32 387 15. Tita AT, Andrews WW. Diagnosis and management of clinical chorioamnionitis. Clin
33 388 Perinatol. 2010;37(2):339-54.
- 34 389 16. Romero R, Dey SK, Fisher SJ. Preterm labor: one syndrome, many causes. Science.
35 390 2014;345(6198):760-5.
- 36 391 17. Soll R, Ozek E. Prophylactic protein free synthetic surfactant for preventing morbidity
37 392 and mortality in preterm infants. Cochrane Database Syst Rev. 2010;(1):CD001079.
- 38 393 18. WHO (2015, November 17). WHO recommendation on the use of tocolytic treatment
39 394 for inhibiting preterm labour. Retrieved from:

- 1
2
3 395 [https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
4 396 [care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
5 397 [treatment-inhibiting-preterm-labour](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
6
7
8 398 19. Walker KF, Thornton JG. Tocolysis and preterm labour. *Lancet*.
9 399 2016;387(10033):2068-2070.
10
11 400 20. Van Baaren GJ, Vis JY, Wilms FF, et al. Predictive value of cervical length
12 401 measurement and fibronectin testing in threatened preterm labor. *Obstet Gynecol*.
13 402 2014;123(6):1185-92.
14
15 403 21. Castor: Amsterdam, Ciwit BV the N. Castor electronic data capture. 2017.
16
17 404 22. van 't Hooft J, Duffy JM, Daly M, Williamson P, Meher S, Thom E, et al. A core
18 405 outcome Set for evaluation of interventions to prevent preterm birth. *Obstet Gynecol*.
19 406 2016;127(1):49–58.
20
21 407 23. Jobe AH, Bancalari E: Bronchopulmonary dysplasia. *Am J Respir Crit Care Med*
22 408 2001, 163:1723–1729.
23
24 409 24. de Vries LS, Eken P, Dubowitz LM: The spectrum of leukomalacia using cranial
25 410 ultrasound. *Behav Brain Res* 1992, 49:1–6.
26
27 411 25. Ment LR, Bada HS, Barnes P, Grant PE, Hirtz D, Papile LA, Pinto-Martin J, Rivkin M,
28 412 Slovis TL: Practice parameter: neuroimaging of the neonate: report of the Quality
29 413 Standards Subcommittee of the American Academy of Neurology and the Practice
30 414 Committee of the Child Neurology Society. *Neurology* 2002, 58(12):1726–1738.
31
32 415 26. Bell MJ: Neonatal necrotizing enterocolitis. *Ann Surg* 1978, 187:1–7.
33
34 416 27. Kahan BC. Accounting for centre-effects in multicentre trials with a binary outcome -
35 417 when, why, and how?. *BMC Med Res Methodol*. 2014;14:20.
36
37 418 28. Gates S, Brocklehurst P. How should randomised trials including multiple
38 419 pregnancies be analysed?. *BJOG*. 2004;111(3):213-9.
39
40 420 29. Van Buuren, S., and Groothuis-Oudshoorn, K. (2011). MICE: Multivariate Imputation
41 421 by Chained Equations in R. *Journal of Statistical Software* 45(3), 1-68
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

422 Word count

423 2984 words



STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	<u>1</u>
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	<u>2</u>
	2b	All items from the World Health Organization Trial Registration Data Set	<u>2</u>
Protocol version	3	Date and version identifier	<u>2</u>
Funding	4	Sources and types of financial, material, and other support	<u>11</u>
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	<u>11</u>
	5b	Name and contact information for the trial sponsor	<u>1</u>
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	<u>11</u>
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	<u>11</u>

1 **Introduction**

2

3 Background and rationale 6a Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention 3

4

5

6 6b Explanation for choice of comparators 4

7

8 Objectives 7 Specific objectives or hypotheses 4

9

10 Trial design 8 Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory) 5

11

12

13

14 **Methods: Participants, interventions, and outcomes**

15

16 Study setting 9 Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained 5

17

18

19 Eligibility criteria 10 Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists) 5

20

21

22 Interventions 11a Interventions for each group with sufficient detail to allow replication, including how and when they will be administered 6

23

24

25 11b Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease) 7

26

27

28 11c Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests) NA

29

30

31 11d Relevant concomitant care and interventions that are permitted or prohibited during the trial 6

32

33

34 Outcomes 12 Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended 6

35

36

37

38

39

40 Participant timeline 13 Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure) 6

41

42

43

44

45

46

1 Sample size 14 Estimated number of participants needed to achieve study objectives and how it was determined, including 8
 2 clinical and statistical assumptions supporting any sample size calculations

3
 4 Recruitment 15 Strategies for achieving adequate participant enrolment to reach target sample size 5
 5

6 **Methods: Assignment of interventions (for controlled trials)**

7 Allocation:

8
 9
 10 Sequence 16a Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any 5
 11 generation factors for stratification. To reduce predictability of a random sequence, details of any planned restriction
 12 (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants
 13 or assign interventions
 14

15
 16 Allocation 16b Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, 5
 17 concealment opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned
 18 mechanism

19
 20 Implementation 16c Who will generate the allocation sequence, who will enrol participants, and who will assign participants to 5
 21 interventions
 22

23
 24 Blinding (masking) 17a Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome 5
 25 assessors, data analysts), and how
 26

27 17b If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's 5
 28 allocated intervention during the trial
 29
 30

31 **Methods: Data collection, management, and analysis**

32
 33 Data collection 18a Plans for assessment and collection of outcome, baseline, and other trial data, including any related 5
 34 methods processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of
 35 study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known.
 36 Reference to where data collection forms can be found, if not in the protocol
 37

38
 39 18b Plans to promote participant retention and complete follow-up, including list of any outcome data to be 7
 40 collected for participants who discontinue or deviate from intervention protocols
 41
 42

1	Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	<u>5, 6</u>
2				
3				
4				
5	Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	<u>8</u>
6				
7				
8		20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	<u>9</u>
9				
10		20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	<u>8, 9</u>
11				
12				
13				
14	Methods: Monitoring			
15				
16	Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	<u>7, 8</u>
17				
18				
19				
20				
21				
22		21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	<u>7, 8</u>
23				
24				
25	Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	<u>7</u>
26				
27				
28	Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	<u>7</u>
29				
30				
31				
32	Ethics and dissemination			
33				
34	Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	<u>10</u>
35				
36				
37	Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	<u>10</u>
38				
39				
40				
41				
42				
43				
44				
45				
46				

1	Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	<u>5</u>
2				
3				
4		26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	<u>NA</u>
5				
6				
7	Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	<u>5, 6</u>
8				
9				
10	Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	<u>11</u>
11				
12				
13	Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	<u>6</u>
14				
15				
16	Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	<u>Not named</u>
17				
18				
19				
20	Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	<u>10</u>
21				
22				
23				
24		31b	Authorship eligibility guidelines and any intended use of professional writers	<u>NA</u>
25				
26		31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	<u>NA</u>
27				
28				
29	Appendices			
30				
31	Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	<u>NA</u>
32				
33				
34	Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	<u>NA</u>
35				
36				

37 *It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items.
 38 Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons
 39 "[Attribution-NonCommercial-NoDerivs 3.0 Unported](https://creativecommons.org/licenses/by-nc-nd/3.0/)" license.
 40

BMJ Open

Study protocol for a randomised trial for atosiban versus placebo in threatened preterm birth: the APOSTEL 8 study

Journal:	<i>BMJ Open</i>
Manuscript ID	bmjopen-2019-029101.R2
Article Type:	Protocol
Date Submitted by the Author:	07-Oct-2019
Complete List of Authors:	<p>Klumper, Job; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Breebaart, Wouter; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Roos, Carolien; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Naaktgeboren, CA ; Julius Centrum voor Gezondheidswetenschappen en Eerstelijns Geneeskunde, University Medical Centre Utrecht van der Post, Joris; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Bosmans, J; Vrije Universiteit Amsterdam, Department of Health Sciences, Amsterdam Public Health research institute van Kaam, Anton; Amsterdam UMC, location AMC and VUmc, Department of neonatology Schuit, Ewoud; Julius Center for Health Sciences and Primary Care, University Medical Centre Utrecht; Stanford University, Stanford Prevention Research Center Mol, Ben; School of Medicine, Monash University, Melbourne, Australia, Department of obstetrics & gynaecology Baalman, Jelle; National Maternity Hospital, Department of obstetrics & gynaecology McAuliffe, Fionnuala; University College Dublin, UCD Perinatal Research Centre; National Maternity Hospital, Department of obstetrics & gynaecology Thornton, Jim; University of Nottingham, Department of obstetrics & gynaecology Kok, Marjolein; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology Oudijk, Martijn; Amsterdam UMC, location AMC, Department of obstetrics & gynaecology</p>
Primary Subject Heading:	Obstetrics and gynaecology
Secondary Subject Heading:	Obstetrics and gynaecology
Keywords:	preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60



1 Study protocol for a randomised trial for atosiban versus 2 placebo in threatened preterm birth: the APOSTEL 8 study

3 J. Klumper,¹ W. Breebaart,¹ C. Roos,¹ C.A. Naaktgeboren,² J.A.M van der Post,¹ J.E.
4 Bosmans,³ A.H. van Kaam,⁴ E. Schuit,² B.W. Mol,⁵ J.H. Baalman,⁶ F.M. McAuliffe,⁶ J.G.
5 Thornton,⁷ M. Kok¹ and M.A. Oudijk.¹

6 Affiliations

7 ¹Department of Obstetrics and Gynaecology, Amsterdam UMC, University of Amsterdam, The Netherlands

8 ²Julius Centre for Health Sciences and Primary Care, University Medical Centre Utrecht, Utrecht University, the
9 Netherlands

10 ³Department of Health Sciences, Faculty of Science, Vrije Universiteit Amsterdam, Amsterdam Public Health
11 research institute, The Netherlands

12 ⁴Department of Neonatology, Amsterdam UMC, Vrije Universiteit van Amsterdam, University of Amsterdam, the
13 Netherlands.

14 ⁵The Robinson Institute, School of Paediatrics and Reproductive Health, University of Adelaide, Australia.

15 ⁶UCD Perinatal Research Centre, School of Medicine, University College Dublin, National Maternity Hospital,
16 Dublin, Ireland

17 ⁷Department of Child Health Obstetrics and Gynaecology, Nottingham University Hospitals NHS Trust, University
18 of Nottingham, United Kingdom

19 Corresponding author

20 M.A. Oudijk, MD, PhD

21 Email: m.a.oudijk@amsterdamumc.nl

22 Amsterdam University Medical Centres, location AMC

23 PO box 22660 | 1100 DD Amsterdam

24 Telephone: +31 20 5667204

25
26 J. Klumper: j.klumper@amc.uva.nl

27 W. Breebaart: breebaart@gmail.com

28 C. Roos: c.dijkstra@amc.nl

29 C.A. Naaktgeboren: c.naaktgeboren@umcutrecht.nl

30 J.A.M van der Post: j.a.vanderpost@amc.uva.nl

31 J.E. Bosmans: j.e.bosmans@vu.nl

32 A.H. van Kaam: a.h.vankaam@amc.uva.nl

33 E. Schuit: e.schuit@umcutrecht.nl

34 B.W. Mol: ben.mol@monash.edu

35 J.H. Baalman: jelle.baalman@gmail.com

36 F.M. McAuliffe: fionnuala.mcauliffe@ucd.ie

37 J.G. Thornton: jim.thornton@nottingham.ac.uk

38 M. Kok: m.kok@amc.uva.nl

39 Abstract

40 **Introduction:** Preterm birth complicates more than 15 million pregnancies annually
41 worldwide. In many countries, women who present with signs of preterm labour are treated
42 with tocolytics for 48 hours. Although this delays birth, it has never been shown to improve
43 neonatal outcome. In 2015 the WHO stated that the use of tocolytics should be reconsidered
44 and that large placebo controlled studies to evaluate the effectiveness of tocolytics are
45 urgently needed.

46 **Methods and analysis:** We designed an international, multicentre, randomised, double
47 blinded, placebo-controlled clinical trial. Women with threatened preterm birth (gestational
48 age 30 – 34 weeks), defined as uterine contractions with
49 1) a cervical length of ≤ 15 mm or 2) a cervical length of 15-30 mm and a positive fibronectin
50 test or 3) in centres where cervical length measurement is not part of the local protocol: a
51 positive fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or
52 4) ruptured membranes, will be randomly allocated to treatment with atosiban or placebo for
53 48 hours. The primary outcome is a composite of perinatal mortality and severe neonatal
54 morbidity. Analysis will be by intention to treat. A sample size of 1514 participants (757 per
55 group) will detect a reduction in adverse neonatal outcome from 10% to 6% (alpha 0.05, beta
56 0.2). A cost-effectiveness analysis will be performed from a societal perspective.

57 **Ethics and dissemination:** This study has been approved by the Research Ethics
58 Committee (REC) of the Amsterdam University Medical Centres, location AMC, as well as
59 the REC's in Dublin and the United Kingdom. The results will be presented at conferences
60 and published in a peer-reviewed journal. Participants will be informed about the results.

61
62 **Trial registration number:** NTR6646 (date registration 24-aug-2017)

63 **Protocol version:** 2.0, dated 27-02-2019

64 **Keywords:** preterm birth, preterm labour, tocolysis, atosiban, perinatal outcome

65 Article summary

66 Strengths and limitations of this study:

- 67 • The primary outcome is perinatal mortality and neonatal morbidity, not prolongation of
68 pregnancy.
- 69 • This is the largest randomised trial comparing atosiban to placebo for women with
70 threatened preterm birth.

- 71 • Over 40 hospitals in Europe will participate.
- 72 • Tocolysis is incorporated in daily routine as it has been the recommendation in many
- 73 guidelines.
- 74 • It will prove to be a challenge in counselling patients to participate in a placebo
- 75 controlled trial, especially in an acute setting.

76 Introduction

77 Preterm birth, defined as birth before 37 weeks gestation, is a major contributor to perinatal
78 mortality and morbidity, complicating over 15 million pregnancies worldwide.^{1,2} Of all infant
79 deaths before the age of 5 years, more than one third can be attributed to preterm birth.³ In
80 addition, spontaneous preterm birth is the leading cause of neonatal morbidity, mostly due to
81 respiratory immaturity, intracranial haemorrhage and infections.^{4,5} These conditions can have
82 long-term neurodevelopmental sequelae such as cognitive impairment, cerebral palsy and
83 visual and hearing deficiencies. Preterm birth is one of the largest single contributors to the
84 Global Burden of Disease because of the high mortality early in life and the morbidity of
85 lifelong impairment.⁶

86 Maternal administration of corticosteroids to accelerate fetal lung maturation is an effective
87 treatment for women with threatened preterm birth.⁷ Since steroids have their maximum
88 effect if birth is delayed by 48 hours, many obstetricians administer a tocolytic drug alongside
89 the steroids to allow maximal steroid effect and facilitate transport of the mother to a centre
90 with neonatal intensive care unit facilities if needed. Several tocolytics are used, including β
91 adrenoceptor agonists, cyclooxygenase inhibitors (COX), magnesium sulphate, calcium-
92 channel blockers and oxytocin receptor antagonists. Though more or less effective in
93 delaying delivery, no tocolytics used in obstetrical practice are proven effective in reducing
94 neonatal morbidity and mortality.^{8,9} None of the studies so far have been powered to show
95 such an effect.

96 The two most commonly used tocolytic drugs, atosiban and nifedipine, showed comparable
97 perinatal outcome in the APOSTEL III study.¹⁰ However, neonatal mortality was higher in the
98 nifedipine group, although not significant (5.4% vs. 2.4% RR 2.20; 95% CI 0.91-5.33).

99 The oxytocin receptor antagonist atosiban has fewer maternal side effects in head to head
100 comparison with alternative drugs,¹¹ and showed similar effectiveness in delaying birth
101 compared to ritodrine.¹² In placebo controlled trials, a Cochrane review showed that atosiban
102 did not reduce perinatal mortality (RR 2.25, 95% CI 0.79 to 6.38; two studies with 729
103 infants) or major neonatal morbidity¹³, although the quality of this review has been
104 questioned.¹⁴

1
2
3 105 One explanation might be that since spontaneous preterm birth is associated in 40-70% of
4 106 cases with chorioamnionitis,^{15,16} tocolysis may prolong fetal exposure to an infectious
5 107 environment, which may worsen neonatal outcome.

8 108 Perinatal outcome has also markedly improved over the last few decades, in part due to
9 109 postnatal interventions such as exogenous surfactant treatment which reduces mortality and
10 110 respiratory morbidity in preterm infants.¹⁷ This might also limit the potential benefit of
11 111 tocolytics.

12 112 Worldwide, practice varies widely. Several large institutions in countries like Canada,
13 113 Scotland and Ireland, rarely use tocolytics, while in the USA, cyclooxygenase inhibitors
14 114 (indomethacin) and calcium channel blockers (nifedipine) are popular. In Europe, nifedipine
15 115 and the oxytocin antagonist, atosiban, are both widely used.

16 116 In conclusion, current widespread use of tocolytic drugs for this indication is not supported by
17 117 the available evidence. The primary goal of tocolysis should not be prolongation of
18 118 pregnancy, but improvement of neonatal outcome. This view is supported by the WHO, as
19 119 they state in their 2015 guidelines on preterm birth that the effectiveness of tocolytics is not
20 120 proven, and that placebo controlled studies are urgently needed.¹⁸ Based on the results of
21 121 the APOSTEL III study,¹⁰ the associated editorial,¹⁹ and its safety profile we chose to
22 122 evaluate atosiban in the APOSTEL 8 study.

23 123 **Objective**

24 124 To test the hypothesis that tocolysis with atosiban in late preterm birth (30 to 34 weeks)
25 125 reduces neonatal mortality and morbidity and is cost-effective compared with placebo.

26 126

27 127 **Methods and analysis**

28 128 **Design and setting**

29 129 We will conduct an international, multicentre, double-blind, randomised, placebo-controlled
30 130 clinical trial, performed in The Netherlands, United Kingdom and Ireland.

31 131 **Participants/eligibility criteria**

32 132 Women, aged ≥ 18 years, with threatened preterm birth and a gestational age between 30⁺⁰
33 133 and 33⁺⁶ weeks are eligible. Threatened preterm birth is defined as uterine contractions with

34 134 1) a cervical length of ≤ 15 mm or

- 1
2
3 135 2) a cervical length of 15-30 mm and a positive fibronectin test or
4 136 3) Or in centres where cervical length measurement is not part of the local protocol: a
5 137 positive fibronectin test or insulin-like growth factor binding protein-1 (Actim-Partus test) or
6 138 4) ruptured membranes.
7
8
9

10 139 These inclusion criteria are based on the results and conclusions of the APOSTEL I study²⁰
11 140 and current guidelines within the Netherlands and the United Kingdom. Moreover, our
12 141 previous APOSTEL III study, with resembling inclusion criteria, showed that half of the
13 142 women with these criteria deliver within seven days,¹⁰ validating this definition of women at
14 143 high risk for preterm birth. In addition, the sample size of expected adverse neonatal
15 144 outcome in the gestational age group of 30-34 weeks, was calculated from the APOSTEL III
16 145 study.
17
18
19
20
21

22 146 This study was designed in a pragmatic fashion, in order for the results to be applicable in
23 147 the current clinical practice. As most national guidelines and local protocols propose
24 148 treatment for threatened preterm birth in both singleton and multiple pregnancies, as well as
25 149 women with ruptured membranes, all these categories of patients are eligible for the study.
26
27
28

29 150 Women with a contra-indication for tocolysis, signs of fetal distress, clinical signs of intra-
30 151 uterine infection, previous treatment for threatened preterm birth with corticosteroids in the
31 152 current pregnancy and known fetal chromosomal or severe structural abnormalities are not
32 153 eligible.
33
34
35

36 154 **Procedures, recruitment, randomization and collection of data**

37
38

39 155 Potential participants will be identified by the local research co-ordinators and/or the staff of
40 156 participating hospitals. Women eligible for the trial will be counselled by doctors, midwives or
41 157 research nurses trained in 'good clinical practice', and will be given a patient information form
42 158 to read. Those who wish to participate, will be asked to give for written informed consent, and
43 159 are registered within the central trial database. Randomisation will be performed by using
44 160 sequentially numbered medication packs available in each centre. Only the independent data
45 161 manager has access to the computer-generated randomization list in which the medication
46 162 numbers are linked with atosiban or placebo. Treatment allocation is blinded to investigators,
47 163 participants, clinicians and research coordinators. Randomisation will be balanced with
48 164 varying block sizes of 2 and 4, and stratified by centre.
49
50
51
52
53
54

55 165 At study entry, baseline demographic, past obstetric and medical history will be recorded into
56 166 the web-based Case Report Form (CRF) accessible through a secure central website
57 167 (Castor Electronic Data Capture, Ciwit B.V.)²¹ Details of delivery, maternal and neonatal
58
59
60

1
2
3 168 assessments during pregnancy and post-partum period will be recorded on the same
4
5 169 system. All data will be coded, processed and stored with adequate precautions to ensure
6
7 170 patient confidentiality. This is described in a separate data management plan.

8 9 171 **Interventions**

10
11 172 Participants are allocated to atosiban or matching placebo (0.9% saline) for 48 hours. The
12
13 173 medication will be administered by a bolus injection of 6.75 mg/0.9 ml in one minute followed
14
15 174 by a continuous infusion of 18 mg/hour for 3 hours followed by a continuous infusion of 6
16
17 175 mg/hour for the remaining 45 hours. Participating women will otherwise be treated according
18
19 176 to local protocol based on national guidelines, including corticosteroids MgSO₄ for
20
21 177 neuroprotection and antibiotics if needed.

22 178 **Outcome measures**

23
24 179 Outcome parameters are in line with the core outcome set for studies on prevention of
25
26 180 preterm birth defined by members of GONet and the Core Outcomes in Women's health
27
28 181 (CROWN) initiative (www.crown-initiative.org).²²

29
30 182 The primary outcome measure is a composite of adverse perinatal outcome composed of
31
32 183 perinatal in-hospital mortality and six severe perinatal morbidities: bronchopulmonary (BPD),
33
34 184 periventricular leucomalacia (PVL) > grade 1, intraventricular haemorrhage > grade 2,
35
36 185 necrotizing enterocolitis (NEC) ≥ stage 2, retinopathy of prematurity > grade 2 or needing
37
38 186 laser therapy, and culture proven sepsis.

39 187 The diagnosis of BPD will be made according to the international consensus guideline as
40
41 188 described by Jobe and Bancalari.²³ PVL > grade 1 and intraventricular haemorrhage > grade
42
43 189 2 will be diagnosed by repeated cranial ultrasound according to the guidelines on neuro
44
45 190 imaging described by de Vries et al.²⁴ and Ment et al.²⁵ NEC ≥ stage 2 will be diagnosed
46
47 191 according to Bell.²⁶ Culture proven sepsis is diagnosed on the combination of clinical signs
48
49 192 and positive blood cultures. The components of the composite adverse perinatal outcome will
50
51 193 also be assessed separately.

52
53 194 Secondary infant outcomes will be birth within 48 hours, time to birth, gestational age at birth,
54
55 195 birth weight, number of days on invasive mechanical ventilation, length of NICU stay,
56
57 196 convulsions, asphyxia, meningitis, pneumothorax until hospital discharge.

58
59 197 Maternal outcomes will be mortality, infection of inflammation and harm to mother from
60
198 interventions (side effects). Side effects are defined as admission to intensive care,
199
200 199 anaphylactic shock, dyspnoea, hypotension (leading to CTG abnormalities), liver test

1
2
3 200 abnormalities (elevated ASAT or ALAT), general side effects (nausea, vomiting, headache),
4 201 post-partum haemorrhage defined as > 500 ml blood loss and maternal mortality.

6
7 202 We will ask informed consent to approach the parents for long-term follow-up of the children.
8 203 We intend, subject to funding, to use standardized questionnaires at 2 years and 5 years of
9 204 age.

11
12 205 Maternal quality of life will be assessed at randomisation and at three months baby corrected
13 206 age using the EuroQol (EQ-5D-5L) questionnaire. This consists of five dimensions (mobility,
14 207 self-care, usual activities, pain/discomfort, anxiety/depression) that are rated using five levels
15 208 (no problems, slight problems, moderate problems, severe problems, extreme problems).

17
18 209 Societal costs will be assessed using adapted versions of the iMTA Medical and Productivity
19 210 Cost Questionnaires at three months corrected baby age. Cost data include costs of the
20 211 intervention, other health care utilization, patient and family costs and costs of productivity
21 212 losses.

22 213 **Withdrawal of subjects**

23 214 Participants can cease study treatment at any time for any reason if they wish to do so.
24 215 Unless they refuse to allow further data collection, such participants will continue to be
25 216 followed-up and will be analysed in the group to which they were originally allocated.
26 217 Participants who decline follow-up will have no further trial data collected. Any results
27 218 collected up to the point at which they decline follow up will be analysed. Study medication
28 219 will be discontinued in patients with signs of intra-uterine infections or signs of fetal distress
29 220 (abnormal CTG, meconium stained amniotic fluid). Data of such participants will continue to
30 221 be analysed. Further management will be left to the expertise of the responsible clinician.
31 222 The responsible clinician can contact a perinatologist from the project group in case of
32 223 suspected side effects or other medical problems. If necessary, treatment will be
33 224 discontinued.

34 225 **Monitoring and Safety**

35 226 An independent data safety monitoring board (DSMB) will focus on both effectiveness and
36 227 safety. Serious Adverse Events (SAEs) will be collected from the first study-related
37 228 procedure until 3 months after delivery. All SAEs will be reported to the DSMB within 7 days.
38 229 Whenever there is proof of effectiveness (at interim analysis) or safety issues (increased
39 230 (serious) adverse events in one of the two treatment arms) the DSMB will advise whether the
40 231 trial should be stopped or continued. The data safety monitoring board will be blinded when
41 232 first analysing the data, but unblinded before reaching a decision.

1
2
3 233 The advice(s) of the DSMB will only be sent to the sponsor of the study. Should the sponsor
4 234 decide not to fully implement the advice of the DSMB, the sponsor will send the advice to the
5 235 reviewing Medical Ethics Committee, including a note to substantiate why (part of) the advice
6 236 of the DSMB will not be followed.
7
8
9

10 237 A formal interim analysis is planned after data collection of 500 and of 1000 women. At these
11 238 interim analyses, the Haybittle-Peto alpha spending function will be used, which means that
12 239 an effect at interim with a p-value <0.001 is considered statistically significant.
13
14
15

16 240 **Sample size**

17
18 241 Based on the APOSTEL III data, the proportion of adverse perinatal outcome in women
19 242 randomized between 30 and 34 weeks gestation and treated with atosiban was 6%.¹⁰ To
20 243 show a 40% reduction (10% in the placebo group to 6% in the atosiban group) we need to
21 244 randomise 1438 women (beta-error 0.2; alpha error 0.05). Assuming a 5% drop-out or loss-to
22 245 follow up rate, we will randomize 1514 women (757 in each arm).
23
24
25
26

27 246 **Statistical analysis**

28 29 247 **Data analysis**

30 248 Data analysis will be performed according to the intention-to-treat principle. In the baseline
31 249 table, categorical variables will be expressed as a number with the percentage of the total
32 250 allocation arm. Continuous variables will be presented as mean with standard deviation, as
33 251 geometric mean with 95% confidence interval (CI) or as median with interquartile range,
34 252 whichever appropriate.
35
36
37
38
39

40 253 The main outcome 'adverse neonatal outcome' will be assessed on the infant level, using a
41 254 log-binomial generalized estimating equations model (GEEs) to take into account the
42 255 correlation of outcomes in multiples, resulting in a relative risk (RR) with accompanying 95%
43 256 confidence interval (CI). To account for stratified randomization by centre, we will also take
44 257 centre into the model if the model converges.²⁷ We will account for interdependence between
45 258 outcomes in multiple pregnancies by considering the mother as a cluster variable.²⁸
46
47
48
49

50 259 The other secondary outcome measures on the child level will be analyzed similarly to the
51 260 primary outcome measure. Outcomes on the maternal level will be assessed by using a
52 261 binomial regression model with log-link function. When a statistically significant difference in
53 262 primary outcome is found between both groups, we will calculate the number needed to treat
54 263 (NNT). Time to delivery will be evaluated by Kaplan-Meier estimates and Cox proportional
55 264 hazard analysis, taking into account the different durations of gestation at study entry, and
56
57
58
59
60

1
2
3 265 will be tested with the log rank test. A p-value of less than 0.05 will be considered to indicate
4 266 statistical significance.

7 267 **Subgroup analyses**

8 268 The following subgroup analyses are planned:

10 269 1) singleton versus multiple pregnancy,

11 270 2) cervical length < 15 mm, versus cervical length 15 - 30 mm and a positive fibronectin test
12 271 (or no cervical length measurement and a positive Fibronectin test or Partus test),

13 272 3) ruptured or unruptured membranes at entry

14 273 5) previous preterm birth.

15 274 **Sensitivity analysis**

16 275 A sensitivity analysis will be performed excluding multiple pregnancies and pregnancies
17 276 complicated by preterm premature rupture of membranes.

18 277 To assess whether a subgroup effect is present we will add an interaction term between the
19 278 subgrouping variables and the treatment allocation to the regression model. When an
20 279 interaction term is statistically significant ($p < 0.05$), we will estimate the treatment effect within
21 280 strata of the subgrouping variable.

22 281 Details of the statistical analysis will be describes in separate statistical analysis plan that will
23 282 be completed before data lock.

24 283 **Cost-effectiveness analysis**

25 284 The cost-effectiveness analysis will be done according to the intention-to treat principle.

26 285 Missing cost and effect data will be imputed using multiple imputation according to the MICE
27 286 algorithm developed by van Buuren.²⁹ Rubin's rules will be used to pool the results from the

28 287 different multiply imputed datasets. Bivariate regression analyses will be used to estimate

29 288 cost and effect differences between atosiban and placebo while adjusting for confounders if

30 289 necessary. Incremental cost-effectiveness ratios (ICERs) will be calculated by dividing the

31 290 difference in the mean total costs between the treatment groups by the difference in mean

32 291 effect between the treatment groups. Bias-corrected and accelerated bootstrapping with

33 292 5000 replications will be used to estimate statistical uncertainty. Uncertainty surrounding

34 293 ICERs will be graphically presented on cost-effectiveness planes. Cost-effectiveness

35 294 acceptability curves will be estimated showing the probability that atosiban is cost-effective in

36 295 comparison with placebo for a range of different ceiling ratios thereby showing decision

1
2
3 296 uncertainty. Sensitivity analyses will be performed to assess the robustness of the results
4 297 using different assumptions regarding costs and effects.

7 298 **Patient and Public Involvement**

8 299 The preterm birth research line of the Dutch Consortium is in close collaboration with two
9 300 Dutch patient associations, the VOC (Vereniging van Ouders van Couveusekinderen, freely
10 301 translated to society of parents of children admitted to NICU) and the NVOM (Nederlandse
11 302 Vereniging van Ouders van Meerlingen, freely translated to Dutch society of parents of
12 303 multiples). They are involved in the design of new studies, updated on progress of running
13 304 trials, and informed of study results. Project members are invited speakers at yearly
14 305 conferences of these societies to present on the progress of our preterm birth research line.
15 306 At these conferences, surveys are being performed on patient preferences on study ideas.
16 307 Tocolysis was deemed an important research issue. Both associations have written support
17 308 letters to the funding agency ZonMw (The Netherlands organization for health research and
18 309 development) for the APOSTEL 8 study.

19 310 A project panel of parents that experienced a spontaneous preterm birth consisting of 6
20 311 couples was involved in the design of our study. A survey was performed during the design
21 312 of the study amongst members of the closed Facebook group of the VOC, to address
22 313 questions on whether they would be interested in participation in the APOSTEL 8 study.
23 314 The Dutch consortium has a website where it publishes all results of completed studies, and
24 315 publishes the protocols of currently recruiting studies.
25 316 Presentations will be held at yearly conferences at patient organizations and updates on
26 317 research are being published in the journal of the VOC.

39 318 **Ethics and dissemination**

40 319 The Research Ethics Committee (REC) at the Amsterdam University Medical Centres,
41 320 location AMC, approved this study. Additional regional approval was obtained for the
42 321 remaining participating hospitals in The Netherlands. Furthermore, the study was approved
43 322 by the REC of the National Maternity Hospital in Dublin, Ireland, and the REC of East
44 323 Midlands - Derby in the United Kingdom.

45 324 Protocol amendments will be communicated to a relevant parties.

46 325 This trial is registered with the Netherlands Trial Register, NTR6646.

47 326 A manuscript with the results of the primary study will be published in a peer-reviewed
48 327 journal. A separate manuscript will be written on the cost effectiveness analysis.

49 328 The results of this clinical trial will be presented at conferences and disseminated through
50 329 publication in a peer-reviewed journal.
51
52
53
54
55
56
57
58
59
60

330 **Acknowledgements**

331 We would like to thank the collaborators of the study group; the gynecologists of the
332 participating centres for their help as local investigators for the APOSTEL 8 study.

333 We also thank the patient associations VOC and NVOW for their input in this study.

334 **Author contributions**

335 CAN, JAMP, JEB, AHvK, ES, BWM, JHB, FMcA, JGT, MAO, MK were involved in conception
336 and design of the study.

337 JK and WB drafted the manuscript.

338 CR, CAN, JAMP, JEB, AHvK, ES, BWM, JHB, FMcA, JGT, MAO, MK reviewed and edited
339 the manuscript. All authors mentioned in the manuscript are member of the APOSTEL study
340 group or collaborators. They participated in the design of the study during several meetings
341 and are local investigators in the participating centres. All authors edited the manuscript and
342 read and approved the final manuscript.

343 **Funding**

344 This study is funded by ZonMw (The Netherlands organization for health research and
345 development), grant number 848041004 and the United Kingdom National Institute for Health
346 Research, Clinical Research Network.

347 **Competing interests statement**

348 JGT received a number of lectures and medical advisory fees from Ferring pharmaceuticals
349 between 2000 and 2016.

350 **References**

- 351 1. Howson CP, Kinney MV, Lawn JE (editors). Born Too Soon: The Global Action
352 Report on Preterm Birth. Geneva: World Health Organization, 2012
- 353 2. Liu L, Johnson HL, Cousens S, Perin J, Scott S, Lawn JE, et al. Global, regional, and
354 national causes of child mortality: an updated systematic analysis for 2010 with time
355 trends since 2000. *Lancet* 2012;379(9832):2151–61
- 356 3. Matthews TJ, Macdorman MF, Thoma ME. Infant Mortality Statistics From the 2013
357 Period Linked Birth/Infant Death Data Set. *Natl Vital Stat Rep.* 2015;64(9):1-30

- 1
2
3 358 4. Hack M, Fanaroff AA. Outcomes of children of extremely low birthweight and
4 359 gestational age in the 1990's. *Early Hum Dev.* 1999;53(3):193-218.
- 5 360 5. Moore T, Hennessy EM, Myles J, et al. Neurological and developmental outcome in
6 361 extremely preterm children born in England in 1995 and 2006: the EPICure studies.
7 362 *BMJ.* 2012;345:e7961.
- 8 363 6. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235
9 364 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the
10 365 Global Burden of Disease Study 2010. *Lancet.* 2012;380(9859):2095-128.
- 11 366 7. Roberts D, Brown J, Medley N, Dalziel SR. Antenatal corticosteroids for accelerating
12 367 fetal lung maturation for women at risk of preterm birth. *Cochrane Database Syst*
13 368 *Rev.* 2017;3:CD004454.
- 14 369 8. Haas DM, Imperiale TF, Kirkpatrick PR, Klein RW, Zollinger TW, Golichowski AM.
15 370 Tocolytic therapy: a meta-analysis and decision analysis. *Obstet Gynecol* 2009; 113:
16 371 585–94.
- 17 372 9. Haas DM, Caldwell DM, Kirkpatrick P, McIntosh JJ, Welton NJ. Tocolytic therapy for
18 373 preterm delivery: systematic review and network meta-analysis. *BMJ* 2012; 345:
19 374 e6226.
- 20 375 10. Van Vliet EO, Nijman TA, Schuit E, et al. Nifedipine versus atosiban for threatened
21 376 preterm birth (APOSTEL III): a multicentre, randomised controlled trial. *Lancet.*
22 377 2016;387(10033):2117-24.
- 23 378 11. De Heus R, Mol BW, Erwich JJ, et al. Adverse drug reactions to tocolytic treatment
24 379 for preterm labour: prospective cohort study. *BMJ.* 2009;338:b744.
- 25 380 12. Effectiveness and safety of the oxytocin antagonist atosiban versus beta-adrenergic
26 381 agonists in the treatment of preterm labour. The Worldwide Atosiban versus Beta-
27 382 agonists Study Group. *BJOG.* 2001;108(2):133-42.
- 28 383 13. Flenady V, Reinebrant HE, Liley HG, Tambimuttu EG, Papatsonis DNM. Oxytocin
29 384 receptor antagonists for inhibiting preterm labour. *Cochrane Database of Systematic*
30 385 *Reviews* 2014, Issue 6. Art. No.: CD004452.
- 31 386 14. Lyndrup J, and Lamont RF The choice of a tocolytic for the treatment of preterm
32 387 labor: a critical evaluation of nifedipine versus atosiban. *Expert Opin. Investig. Drugs*
33 388 (2007) 16(6):843-853.
- 34 389 15. Tita AT, Andrews WW. Diagnosis and management of clinical chorioamnionitis. *Clin*
35 390 *Perinatol.* 2010;37(2):339-54.
- 36 391 16. Romero R, Dey SK, Fisher SJ. Preterm labor: one syndrome, many causes. *Science.*
37 392 2014;345(6198):760-5.
- 38 393 17. Soll R, Ozek E. Prophylactic protein free synthetic surfactant for preventing morbidity
39 394 and mortality in preterm infants. *Cochrane Database Syst Rev.* 2010;(1):CD001079.

- 1
2
3 395 18. WHO (2015, November 17). WHO recommendation on the use of tocolytic treatment
4 396 for inhibiting preterm labour. Retrieved from:
5 397 [https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
6 398 [care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
7 399 [treatment-inhibiting-preterm-labour](https://extranet.who.int/rhl/topics/preconception-pregnancy-childbirth-and-postpartum-care/pregnancy-complications/preterm-birth/who-recommendation-use-tocolytic-treatment-inhibiting-preterm-labour)
8 400
9 401 19. Walker KF, Thornton JG. Tocolysis and preterm labour. *Lancet*.
10 402 2016;387(10033):2068-2070.
11 403
12 404 20. Van Baaren GJ, Vis JY, Wilms FF, et al. Predictive value of cervical length
13 405 measurement and fibronectin testing in threatened preterm labor. *Obstet Gynecol*.
14 406 2014;123(6):1185-92.
15 407
16 408 21. Castor: Amsterdam, Ciwit BV the N. Castor electronic data capture. 2017.
17 409
18 410 22. van 't Hooft J, Duffy JM, Daly M, Williamson P, Meher S, Thom E, et al. A core
19 411 outcome Set for evaluation of interventions to prevent preterm birth. *Obstet Gynecol*.
20 412 2016;127(1):49–58.
21 413
22 414 23. Jobe AH, Bancalari E: Bronchopulmonary dysplasia. *Am J Respir Crit Care Med*
23 415 2001, 163:1723–1729.
24 416
25 417 24. de Vries LS, Eken P, Dubowitz LM: The spectrum of leukomalacia using cranial
26 418 ultrasound. *Behav Brain Res* 1992, 49:1–6.
27 419
28 420 25. Ment LR, Bada HS, Barnes P, Grant PE, Hirtz D, Papile LA, Pinto-Martin J, Rivkin M,
29 421 Slovis TL: Practice parameter: neuroimaging of the neonate: report of the Quality
30 422 Standards Subcommittee of the American Academy of Neurology and the Practice
31 423 Committee of the Child Neurology Society. *Neurology* 2002, 58(12):1726–1738.
32 424
33 425 26. Bell MJ: Neonatal necrotizing enterocolitis. *Ann Surg* 1978, 187:1–7.
34 426
35 427 27. Kahan BC. Accounting for centre-effects in multicentre trials with a binary outcome -
36 428 when, why, and how?. *BMC Med Res Methodol*. 2014;14:20.
37 429
38 430 28. Gates S, Brocklehurst P. How should randomised trials including multiple
39 431 pregnancies be analysed?. *BJOG*. 2004;111(3):213-9.
40 432
41 433 29. Van Buuren, S., and Groothuis-Oudshoorn, K. (2011). MICE: Multivariate Imputation
42 434 by Chained Equations in R. *Journal of Statistical Software* 45(3), 1-68
43 435
44 436
45 437
46 438
47 439
48 440
49 441

424 Word count

50 425 3074 words
51
52
53
54
55
56
57
58
59
60



STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	<u>1</u>
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	<u>2</u>
	2b	All items from the World Health Organization Trial Registration Data Set	<u>2</u>
Protocol version	3	Date and version identifier	<u>2</u>
Funding	4	Sources and types of financial, material, and other support	<u>11</u>
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	<u>11</u>
	5b	Name and contact information for the trial sponsor	<u>1</u>
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	<u>11</u>
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	<u>11</u>

1 **Introduction**

2

3 Background and rationale 6a Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention 3

4

5

6 6b Explanation for choice of comparators 4

7

8 Objectives 7 Specific objectives or hypotheses 4

9

10 Trial design 8 Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory) 5

11

12

13

14 **Methods: Participants, interventions, and outcomes**

15

16 Study setting 9 Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained 5

17

18

19 Eligibility criteria 10 Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists) 5

20

21

22 Interventions 11a Interventions for each group with sufficient detail to allow replication, including how and when they will be administered 6

23

24

25 11b Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease) 7

26

27

28 11c Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests) NA

29

30

31 11d Relevant concomitant care and interventions that are permitted or prohibited during the trial 6

32

33

34 Outcomes 12 Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended 6

35

36

37

38

39

40 Participant timeline 13 Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure) 6

41

42

43

44

45

46

1 Sample size 14 Estimated number of participants needed to achieve study objectives and how it was determined, including 8
 2 clinical and statistical assumptions supporting any sample size calculations

3
 4 Recruitment 15 Strategies for achieving adequate participant enrolment to reach target sample size 5
 5

6 **Methods: Assignment of interventions (for controlled trials)**

7 Allocation:

8
 9
 10 Sequence 16a Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any 5
 11 generation factors for stratification. To reduce predictability of a random sequence, details of any planned restriction
 12 (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants
 13 or assign interventions
 14

15
 16 Allocation 16b Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, 5
 17 concealment opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned
 18 mechanism
 19

20 Implementation 16c Who will generate the allocation sequence, who will enrol participants, and who will assign participants to 5
 21 interventions
 22

23 Blinding (masking) 17a Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome 5
 24 assessors, data analysts), and how
 25

26
 27 17b If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's 5
 28 allocated intervention during the trial
 29

30
 31 **Methods: Data collection, management, and analysis**

32
 33 Data collection 18a Plans for assessment and collection of outcome, baseline, and other trial data, including any related 5
 34 methods processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of
 35 study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known.
 36 Reference to where data collection forms can be found, if not in the protocol
 37

38
 39 18b Plans to promote participant retention and complete follow-up, including list of any outcome data to be 7
 40 collected for participants who discontinue or deviate from intervention protocols
 41

1	Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	<u>5, 6</u>
2				
3				
4				
5	Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	<u>8</u>
6				
7				
8		20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	<u>9</u>
9				
10		20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	<u>8, 9</u>
11				
12				
13				
14	Methods: Monitoring			
15				
16	Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	<u>7, 8</u>
17				
18				
19				
20				
21				
22		21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	<u>7, 8</u>
23				
24				
25	Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	<u>7</u>
26				
27				
28	Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	<u>7</u>
29				
30				
31				
32	Ethics and dissemination			
33				
34	Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	<u>10</u>
35				
36				
37	Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	<u>10</u>
38				
39				
40				
41				
42				
43				
44				
45				
46				

1	Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	<u>5</u>
2				
3				
4		26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	<u>NA</u>
5				
6				
7	Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	<u>5, 6</u>
8				
9				
10	Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	<u>11</u>
11				
12				
13	Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	<u>6</u>
14				
15				
16	Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	<u>Not named</u>
17				
18				
19				
20	Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	<u>10</u>
21				
22				
23				
24		31b	Authorship eligibility guidelines and any intended use of professional writers	<u>NA</u>
25				
26		31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	<u>NA</u>
27				
28				
29	Appendices			
30				
31	Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	<u>NA</u>
32				
33				
34	Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	<u>NA</u>
35				
36				

37 *It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items.
 38 Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons
 39 "[Attribution-NonCommercial-NoDerivs 3.0 Unported](https://creativecommons.org/licenses/by-nc-nd/3.0/)" license.
 40