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Point of care testing of C-reactive protein and procalcitonin to diagnose urinary tract infections in nursing homes: PROGRESS study protocol

Journal:	<i>BMJ Open</i>
Manuscript ID	bmjopen-2019-031269
Article Type:	Protocol
Date Submitted by the Author:	25-Apr-2019
Complete List of Authors:	Kuil, Sacha; Amsterdam UMC, Medical Microbiology; Hidad, Soemeja; Amsterdam UMC, Medical Microbiology Fischer, Johan; Amsterdam UMC, Clinical Chemistry Harting, Janneke; Amsterdam UMC, Public Health Hertogh, C; VU University Medical Center , General Practice and Elderly Care Medicine Prins, Jan; Amsterdam UMC, Internal Medicine van Leth, Frank; AIGHD, Global Health de Jong, Menno; Amsterdam UMC, Medical Microbiology Schneeberger, Caroline; Amsterdam UMC, Medical Microbiology
Keywords:	Point of care testing, C-Reactive protein, Procalcitonin, Nursing homes, Urinary tract infections < UROLOGY, Antimicrobial resistance

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Manuscripts

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3 1 **Point of care testing of C-reactive protein and procalcitonin to diagnose urinary tract**
4 **infections in nursing homes: PROGRESS study protocol**

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48 33

49 34 **Word count abstract: 304 Word count full text: 4166**
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35 **ABSTRACT**

36 **Introduction:**

37 Suspected urinary tract infection (UTI) ranks among the most common reasons for antibiotic use
38 in nursing homes. However, diagnosing UTI in this setting is challenging because UTI often
39 present with non-specific symptomatology. Moreover asymptomatic bacteriuria is common in
40 elderly, which complicates attribution of causality to detection of bacteria in urine. These
41 diagnostic challenges contribute to overuse of antibiotics and emergence of antimicrobial
42 resistance (AMR) in nursing homes. Given the diagnostic challenges there is a need for point-
43 of-care (POC) diagnostic tests to support clinical rules for diagnosing UTI. Procalcitonin (PCT)
44 and C-reactive protein (CRP) are inflammatory blood markers that have been proven useful to
45 support diagnosis and monitoring of (bacterial) respiratory tract infections and sepsis. While
46 limited studies suggest their usefulness in supporting UTI diagnosis, their utility has not been
47 studied in elderly populations for this purpose.

49 **Methods and analysis:**

50 In an 24-month matched diagnostic accuracy study 'PROGRESS' will assess and compare the
51 sensitivity of rapid POC measurements of blood CRP and PCT levels to support clinical rules for
52 diagnosing UTI in nursing home residents. The primary outcome measure is sensitivity of the
53 POC tests to identify patients with true UTI based on the predefined definition, as derived from
54 Receiving Operating Curves (ROC).

56 **Discussion:**

57 This study will show the sensitivity of CRP and PCT in the diagnostic process of a UTI in elderly.
58 When effective, the potential impact of CRP and/or PCT measured by POC diagnostic tests on
59 antibiotic prescription needs to be established in a consecutive study.

61 **Ethics and dissemination:**

62 This study will be conducted in accordance with Good Clinical Practice guidelines and the
63 principles of the Declaration of Helsinki. The study protocol is approved by the Medical Ethical
64 Committee (METc) of AmsterdamUMC location VUmc with reference number 2017.350 and
65 National Central Committee on Research involving Human Subjects (CCMO) with reference
66 number NL62067.029.17.

68 **Registration details:** Dutch trial registry: NTR6467

69 **Keywords:** Point of care testing, C-Reactive protein, Procalcitonin, Nursing homes, Urinary
70 tract infections, Antimicrobial resistance

71

72 **Strengths and limitations**

73 Strengths

74 - Stringent post-hoc UTI criteria incorporating microbiology result and clinical response to
75 adequate antibiotic therapy

76 - Diagnostic accuracy study in relevant study population, namely nursing home residents

77 Limitations

78 - No on-site POCT, however POCT is performed within 4 hours

79 - Single country study

80

81 **Protocol version 4 March 2019, version 5**

82 *1 September 2017 Original*

83 *21 October 2017 Amendment 1:* Addition of information brochures for nursing home staff and
84 patients

85 *20 November 2017 Amendment 2:* Change of nomenclature for part of nursing home residents
86 to temporary rehabilitation patients (at rehabilitation wards)

87 *7 March 2018 Amendment 3:* Change exclusion criterion prior inclusion to prior inclusion in the
88 past 30 days. Subsequently to account for non-independence of observation for a limited
89 number of participants, the sample size is adjusted with a design effect of 1.1

90 *28 June 2018 Amendment 4:* (1) Change in blood sample collection method (venipuncture to
91 finger prick collection) due to availability of new procalcitonin POCT with small input volume, as
92 finger prick blood sample collection is a preferred collection method in the elderly nursing home
93 population. (2) Change sample size retaining stringent criteria (p-value 0.05 and power 90%) (3)
94 Prolongation study duration: 12 to 18 months (4) Expansion of number of nursing homes from
95 11 to 12 nursing homes

96 *3 March 2019 Amendment 5:* (1) Expansion of number of nursing homes from 12 to 13 nursing
97 homes (2) Addition of extra urine test for detection of gram-negative bacteria (3) Adjusted
98 informed consent procedure for capacitated residents

99

100 Introduction

101 Antimicrobial resistance (AMR) is mainly driven by inappropriate antibiotic use in both humans
102 and animals. AMR is a problem worldwide. Nursing homes are increasingly regarded as an
103 important reservoir for the emergence of AMR^{1 2 3 4}.

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106 The most frequently reported infections in elderly residents are urinary tract infections (UTI)⁵. In
107 Dutch nursing homes, an average weekly incidence of 10.3 (95% CI 9.8 – 10.8) per 1000
108 elderly residents was found⁶. UTI is the most common reason for prescribing antibiotics in Dutch
109 nursing homes. However, not all diagnosed UTIs are “true” UTIs since recognition and
110 diagnosing UTI in nursing homes is complex. Approximately one third of elderly residents UTIs
111 are misdiagnosed, leading to inappropriate antibiotic use^{7 8}. This is mainly due to the facts that
112 non-specific, non-urinary tract symptoms such as altered mental status are often attributed to
113 UTIs whilst asymptomatic bacteriuria (ASB), possibly resulting in positive urine tests, is also
114 common in the elderly residents.⁹

117 *Cognitive impairments and urinary tract infections*

118 The majority of nursing homes in the Netherlands consist of psychogeriatric wards (57%), for
119 elderly residents suffering from cognitive impairments, mainly Alzheimer disease^{10 11 12 13}. Their
120 ability to verbally communicate or express classical symptoms of UTI, such as dysuria, urgency
121 or frequency, is often limited¹⁴. The most frequently presented symptom in elderly residents
122 leading to antibiotic prescription for a suspected UTI is an altered mental status (43.3%), while
123 classical symptoms as dysuria, urgency and frequency are present in the minority of cases (0 –
124 3.8%)¹⁵. Confusion or an altered mental state are nonspecific symptoms and can result from
125 other infectious and non-infectious diseases in the elderly residents.

128 *Asymptomatic bacteriuria*

129 Asymptomatic bacteriuria (ASB) is defined by the presence of significant bacteriuria without
130 symptoms of UTI. ASB is thus regarded as colonization of the urinary tract rather than infection.
131 ASB is highly prevalent in healthy elderly persons with reported prevalence rates as high as 40-
132 50%^{16 17}. In the presence of ASB, frequently used urine tests based on detection of bacteria are
133 less applicable to diagnose UTI, because detection of bacteria does not discriminate between

134 ASB and UTI. In combination with above described non-specific symptomatology, it is difficult to
135 distinguish ASB from “true” UTI in the elderly.

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137

138 In conclusion, decisions about who to treat and who not to treat are challenging in elderly
139 residents with suspected UTI, resulting in potential antibiotic overuse in those who do not need
140 treatment. Availability of a simple and rapid point-of-care (POC) test to distinguish true UTI from
141 ASB represents an unmet need which would greatly assist clinical management of these
142 vulnerable patients, not only by improving appropriateness of antibiotic treatment but also by
143 enabling consideration of alternative causes of presenting non-specific symptomatology when
144 UTI is excluded.

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147 *C-reactive protein and procalcitonin*

148 In the diagnosis of respiratory tract infections and monitoring of bacterial sepsis, inflammatory
149 markers such as C-reactive protein (CRP) and procalcitonin (PCT) in blood have proven useful
150 to guide antibiotic therapy and reduce antibiotic use^{18 19}. Currently, point-of-care (POC) CRP
151 measurements are recommended by Dutch guidelines for general practitioners to guide
152 antibiotic treatment for acute respiratory tract infections. CRP and PCT represent potential
153 candidates for rapid POC testing to support UTI diagnosis.

154 Ageing and frailty are associated with changes in serum inflammation protein levels, such as
155 CRP and PCT^{20 21 22}. Therefore, it is important to determine cut-off values for CRP and PCT
156 specifically in the elderly nursing home resident population.

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158

159 *Inflammatory markers CRP and PCT in UTI*

160 Studies in adults showed that CRP and PCT levels in parenchymatous infections (acute
161 pyelonephritis, prostatitis and epididymitis) are increased^{23 24}. Using a PCT-based algorithm in
162 UTI treatment was shown to reduce antibiotic exposure in adults²⁵. In a subgroup analysis
163 including elderly (> 70 years of age) with lower UTI antibiotic exposure was reduced as well,
164 suggesting a potential role for PCT.

165 A study in children showed that CRP or PCT can help to distinguish renal scarring due to a UTI
166 from renal scarring due to another reason and between pyelonephritis (upper UTI) and cystitis
167 (lower UTI)^{26 27 28}. However studies are small or were performed retrospectively. The specificity

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3 168 and positive predictive values were low because CRP and PCT were also increased in children
4 169 with lower UTI. This suggests a possible role for inflammatory markers in distinguishing a UTI
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6 170 (local inflammation of the bladder) from no inflammation.
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8 171 In the elderly population only two studies have been performed evaluating CRP and PCT and
9
10 172 their possible role in UTI^{29 30}. However these two studies focused on hospitalized and more
11 173 severely ill elderly populations. Therefore these results are not applicable to the nursing home
12 174 population. However, in 13% of the healthy elderly controls CRP values were increased, again
13 175 suggesting increased inflammation in ageing and frailty, indicating the need to determine
14 176 specific cutoffs in elderly. Studies on lower UTIs and CRP or PCT in elderly are scarce.
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21 179 *Distinguishing UTI and ASB*

22 180 The majority of UTIs in elderly residents are lower UTIs without fever or other signs of systemic
23 181 illness^{31 32} while most studies on CRP and PCT in UTI focus on diagnostic values in upper UTI
24 182 or focus on distinguishing upper and lower UTI. The only small study on distinguishing lower
25 183 UTI (infection) from ASB (colonization) in adults, shows a high negative predictive value (NPV)
26 184 for UTI of PCT levels at a cutoff of 0.25 ng/mL³³, suggesting that low PCT levels can rule out
27 185 UTI and contribute to reducing antibiotic use.
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35 188 **Methods and analysis**

36 189 *Aim*

37
38 190 To assess the utility of point-of-care measurements of blood CRP and PCT levels to support
39 191 clinical rules for diagnosing urinary tract infections UTI in elderly nursing home residents.
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44 194 *Outcome*

45 195 The primary outcome is the sensitivity of the point-of-care (POC) test to identify patients with a
46 196 true UTI based on the predefined definition, as derived from Receiving Operating Curves
47 197 (ROC).
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53 200 *Design and setting*

54 201 In a prospective matched diagnostic accuracy study we will assess and compare the sensitivity
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202 of rapid POC measurements of blood CRP and PCT levels to support clinical rules for
203 diagnosing UTI in elderly residents, with a post-hoc definition of UTI with stringent criteria
204 including microbiology results as gold standard.

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207 In this study a true UTI is present when the following five criteria are met: presence of at least
208 two urinary or non-specific symptoms (1), positive urine leucocyte esterase test (2), presence of
209 uropathogens in bacterial culture at $10^4 \geq$ CFU/mL (3), maximum of two uropathogens present
210 (4) and symptom resolution in the course of adequate antibiotic treatment, where adequate
211 treatment is defined by proven susceptibility of isolated uropathogens to the administered
212 antibiotic (5).

213
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215 The matching refers to the assessment of blood CRP and PCT levels in the same study
216 participants. The study will be performed in nursing homes of the University Network for
217 Organizations of Elderly Care of the VUmc University Medical Center (UNO-VUmc). The study
218 duration is 18 months.

219 The nursing home population in this study consists mostly of psychogeriatric and somatic (long-
220 stay) wards and some rehabilitation (short-stay) wards.

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223 *Informed consent procedure*
224 Most nursing home patients are incapacitated. In case of incapacity legal representatives will be
225 asked for informed consent. Capacitated patients will be asked for informed consent
226 themselves. When nursing home staff suspect a UTI, it is not considered practical to obtain
227 written informed consent from the representatives because preferably the blood sample should
228 be drawn as soon as possible. Therefore informed consent will be obtained pre-emptively at the
229 start of the study or when admitted to the nursing home. This means that patients or their legal
230 representatives provide consent a priori to participate in the study once a UTI is clinically
231 suspected during the study period. This procedure will greatly enhance feasibility of enrolment
232 in psychogeriatric nursing home wards.

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236 *Inclusion and exclusion criteria*

237 Eligible for study participation are elderly nursing home residents clinically suspected of UTI by
238 the attending physician or nurse. Exclusion criteria are suspected respiratory tract infection,
239 suspected other infection requiring antibiotic therapy, previous study inclusion in the past 30
240 days or lack of written informed consent.

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242

243 *Study procedures*

244 *Study enrolment*

245 The study physician will be notified by the nursing home staff when there is potentially eligible
246 patient and will visit the nursing home as soon as possible. The research physician will verify if
247 eligibility criteria are met and will complete study enrolment.

248

249 *Data collection*

250 To meet our post-hoc UTI criteria data on signs and symptoms, type and duration of antibiotic
251 used, urine leucocyte esterase, bacterial culture and antimicrobial susceptibility and clinical
252 response are collected. Demographic, clinical and laboratory data are collected through an
253 electronic data capture system using software of Open Data Kit³⁴ with which case report forms
254 (CRFs) are designed that incorporate consistency checks to minimize incompatible data points.
255 Data will be handled encoded, working with barcodes scanned by the database APP. Data of
256 paper registration forms is directly entered into online CRFs and uploaded to a pre-defined
257 database.

258 Data on culture results (species, susceptibility patterns by Minimal Inhibitory Concentrations
259 MIC's) from the laboratory system will be collected in the currently used laboratory systems
260 (Labtrain and Kiestra)

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262

263 *Data on signs and symptoms, clinical response and antibiotic use*

264 Demographic and clinical data are collected at the day of study enrolment by the attending
265 physician or nurse.

266 The research physician will visit the nursing homes regularly for monitoring of follow-up. The
267 attending physician or nurse will evaluate clinical response at day 5 and 10 after study
268 enrolment. Improvement of clinical symptoms, compared to symptoms at enrolment is
269 evaluated. Data on antibiotic use (timing of initial antimicrobial therapy, type of antimicrobial

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3 270 agent and possible switch in antimicrobial therapy) will be collected by the research physician
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5 271 10 days after study enrolment.

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9 274 *Urine sample collection, dipstick analysis and bacterial culture*

10 275 The attending nurse will collect a urine sample for purpose of this study. Urine samples are
11
12 276 collected spontaneously voiding either directly in a sterile urine container or in chamber pot
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14 277 (insert pan). When participants have an indwelling urine catheter, urine will be collected when
15
16 278 the urine bag is changed. However when participants suffer from urinary incontinence or are not
17
18 279 able to urinate on the toilet or chamber pot, diapers will be used to obtain urine for bacterial
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20 280 culture and dipstick analysis^{35 36} Urine extraction from diapers can provide a reliable diagnostic
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22 281 specimen, if fecal contamination can reasonably excluded³⁷. Date, time and way of urine
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24 282 collection method are registered by the attending nurse. The urine sample is stored at 4°C.
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26 283 For dipstick analysis Combur 2 (Roche Diagnostics) will be used (nitrite and leucocyte
27
28 284 esterase). Urines will be used for semi-quantitative bacterial culture. Inoculation of 10 µL urine
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30 285 to CHROMID CPS Elite agar (Biomerieux) and Columbia CNA agar with 5% sheep blood
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32 286 (Biomerieux) will be streaked using a four quadrant pattern. Bacterial growth will be interpreted
33
34 287 after overnight incubation at 35°C in aerobic (CPSE) and CO₂ enriched (CNA) environment.
35
36 288 Uropathogens will be identified by Maldi-tof mass spectrometry (Microflex, Bruker Daltonic).
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38 289 When bacterial growth of ≥10⁴ CFU/mL is found, antibiotic susceptibility testing will be
39
40 290 performed using the VITEK2 platform (BioMérieux).

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43 293 *Blood sample collection and point of care testing*

44 294 For POC testing the research physician will collect a blood sample by capillary fingerprick.
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46 295 Capillary blood sample collection is a preferred collection method in the elderly nursing home
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48 296 population. For CRP testing the Afinion AS100 point-of-care (POC) platform (Alere Health B.V.,
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50 297 Tilburg) is used. For PCT testing the Afias1 PCT Plus (Avant Medical B.V., Geffen) is used.
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52 298 Both, CRP and PCT platforms enable capillary blood sample collection to better facilitate future
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54 299 implementation in nursing homes. POC testing are performed according to the manufacturer's
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56 300 protocol. For logistic reasons the POC testing is performed centrally at the Medical Microbiology
57
58 301 laboratory of our hospital. POC tests are performed within 4 hours after blood sample collection
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60 302 to ensure stability of CRP and PCT.
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304 Participants, attending physicians and nurses will not be informed of POC-test and urinary

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3 304 culture results.
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8 307 *Biological specimen storage and molecular analysis In future*

9 308 Aliquots of urine samples will be stored (-80°C) when consent is obtained specifically for genetic
10 309 studies. In future studies metagenomic sequencing will be performed on bacterial DNA, to
11 310 identify uropathogens and resistance genes.
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16 313 *Sample size*

17 314 For the sample size calculation, we used a two-sided p-value of 0.05 and a power of 90%. We
18 315 assume a difference in sensitivity between the two POC tests of at least 10% as clinically
19 316 relevant. This difference is the net result of discordant test outcomes between the two tests. The
20 317 magnitude of discordance is the main parameter in the McNemar test used to calculate the
21 318 sample size for the matched design.

22 319 With a proposed sample size of 440 enrolled participants, we are able to adequately assess an
23 320 increased sensitivity of 10% or more, when the prevalence of UTI in the study population is 40%
24 321 or more. If the prevalence of UTI is lower, the difference in prevalence that is statistically
25 322 significant (at the 5%) level increases to 11 or 12%.

26 323 Based on data of the national sentinel surveillance network for infectious diseases in nursing
27 324 homes (SNIV) in 2015, the prevalence of UTI in Dutch nursing homes is 10.3 in 1000 patient
28 325 weeks. UTI is defined by SNIV as patients with nonspecific or urinary tract complaints and a
29 326 positive result on nitrite or leucocyte esterase in dipstick analysis and an urine culture positive
30 327 for a uropathogen.

31 328 In order to reach the target, the PROGRESS study will take place in 13 nursing homes with
32 329 around 1350 elderly residents in total. Recruitment of a manageable number of approximately
33 330 12 participants per week in total is anticipated.
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38 333 *Data and statistical analysis*

39 334 The sensitivity of both POC tests to diagnose UTIs defined by the post-hoc definition is derived
40 335 from the optimal point of a Receiver Operating Curve (ROC). Sensitivities of CRP and PCT tests
41 336 are compared by a matched analysis approach using a two-tailed McNemar test assessing the
42 337 statistical significance of differences between the sensitivities of each pair of tests (performed in
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338 each study participant).

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340 This study protocol is written using the SPIRIT reporting guidelines³⁸

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342 **Ethics and dissemination:**

343 This study will be conducted in accordance with Good Clinical Practice guidelines and the
344 principles of the Declaration of Helsinki. The study protocol is approved by the Medical Ethical
345 Committee (METc) of AmsterdamUMC location VUmc with reference number 2017.350 and
346 National Central Committee on Research involving Human Subjects (CCMO) with reference
347 number NL62067.029.17. Written informed consent from nursing home residents or legal
348 representatives (when incapacitated) will be obtained prior to study enrolment. Informed
349 consent from residents will be obtained directly by researchers or via mail. Informed consent
350 from legal representatives will be obtained via mail. Treating physicians decide which residents
351 are capacitated.

352

353 **Data safety monitoring board**

354 No data safety monitoring board has been appointed for this study as the risks of this study are
355 assumed negligible.

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357

358 **Discussion**

359 To our knowledge there is no other (ongoing) trial evaluating the diagnostic value of C-reactive
360 protein and procalcitonin in urinary tract infections in elderly³⁹. This trial will hopefully provide
361 evidence for supporting the diagnosis of UTI in elderly by increased levels of inflammatory
362 markers, to guide diagnosis and treatment and aiming at reduction of antibiotic prescription
363 rates and resistance rates in nursing homes. Since effectiveness of a new test does not just
364 depend on its proven efficacy in research studies, but also on successful implementation after
365 the study, qualitative research on identifying the barriers and facilitators for implementation are
366 needed and will be performed in parallel to the described study. The potential impact of CRP
367 and/or PCT measured by POC diagnostic tests on antibiotic prescription for UTI in elderly
368 nursing home residents needs to be established in a consecutive study once evidence of their
369 utility for diagnosing UTI has been obtained from the current planned study.

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3 3714 372 **List of abbreviations**

5 373 Amsterdam UMC Amsterdam Universitair Medische Centra

6 374 AMR Antimicrobial resistance

7 375 ASB Asymptomatic bacteriuria

8 376 CCMO National Central Committee on Research involving Human Subjects

9 377 CFU Colony forming units

10 378 CRP C-reactive protein

11 379 LQAS Lot Quality Assurance Sampling

12 380 MIC Minimal inhibitory concentration

13 381 NPV Negative predictive value

14 382 ODK Open data kit

15 383 PCT Procalcitonin

16 384 POC Point of care

17 385 SNIV Surveillance network for infectious diseases in nursing homes

18 386 UNO Universitair netwerk ouderenzorg

19 387 UTI urinary tract infection

20 388 VUmc Vrije Universiteit medisch centrum

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22 390

23 391 **Declarations**

24 392

25 393 *Dissemination policy*

26 394 According to the CCMO statement on publication policy, the results of this study will be

27 395 disclosed unreservedly.

28 396

29 397 *Competing interests*

30 398 This study is funded by The Netherlands Organization for Health Research and Development

31 399 (ZonMW).

32 400

33 401

34 402 *Funding*

35 403 This study is funded by The Netherlands Organization for Health Research and Development

1
2
3 404 (ZonMW) grant 541001003. ZonMW, Laan van Nieuw Oost Indië 334, 2593 CE Den Haag, The
4
5 405 Netherlands. The study protocol is being peer reviewed by the funder. The funder has no role in
6
7 406 collection, management, analysis, and interpretation of data, writing of the report or the decision
8
9 407 to submit the report for publication.
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11 408
12 409

13 410 *Roles and responsibilities*

14 411 Research physician/PhD student (SDK): coordinating PROGRESS, patient recruiting, data
15 412 collection, specimen handling, POC and urine testing, preparation protocols, CRFs and
16 413 publication of study reports including annual ethical committee report.

17 414 Research assistant (SH): patient recruiting, data collection, specimen handling, POC and urine
18 415 testing

19 416 Principal investigator (MdJ), senior researcher (CS) and lead epidemiologist (FvL): Study
20 417 planning, agreement of final protocols, reviewing progress of study

21 418 Lead epidemiologist (FvL): design of the electronic report form (eCRF), database design and
22 419 maintenance, data validation and data management plans, user account maintenance, design
23 420 of automated data validation scripts, preparation of the final data analysis sets, supervising of
24 421 data analysis.

25 422 Laboratory chemist (JCF): responsible for verification of POCT, contractual issues with
26 423 manufacturers

27 424 In each participating center a lead investigator (elderly care physician) will be identified, to be
28 425 responsible for progress monitoring and assisting with study set-up per site.
29
30 426

31 427 *Author contributions*

32 428 SDK recruits patients, coordinates and performs the clinical study and the wrote the manuscript.
33 429 SH recruits patients and performs the clinical study. FvL, JCF, JH, NM, CMPMH, JMP, MDdJ
34 430 and CS developed the protocol and secured funding for this project. CS, FvL and MdJ
35 431 supervised the design of the study and writing this manuscript. FvL provided the statistical
36 432 analysis plan, database set-up. All authors have read and approved the manuscript.
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39 434 *Patient and public involvement*

40 435 Patients or public are not involved in designing this study.
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Reporting checklist for protocol of a clinical trial.

Based on the SPIRIT guidelines.

Instructions to authors

Complete this checklist by entering the page numbers from your manuscript where readers will find each of the items listed below.

Your article may not currently address all the items on the checklist. Please modify your text to include the missing information. If you are certain that an item does not apply, please write "n/a" and provide a short explanation.

Upload your completed checklist as an extra file when you submit to a journal.

In your methods section, say that you used the SPIRIT reporting guidelines, and cite them as:

Chan A-W, Tetzlaff JM, Altman DG, Laupacis A, Gøtzsche PC, Krleža-Jerić K, Hróbjartsson A, Mann H, Dickersin K, Berlin J, Doré C, Parulekar W, Summerskill W, Groves T, Schulz K, Sox H, Rockhold FW, Rennie D, Moher D. SPIRIT 2013 Statement: Defining standard protocol items for clinical trials. *Ann Intern Med.* 2013;158(3):200-207

		Reporting Item	Page Number
Title	#1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	#2a	Trial identifier and registry name. If not yet registered, name of intended registry	2
Trial registration: data set	#2b	All items from the World Health Organization Trial Registration Data Set https://www.who.int/ictrp/network/trds_1.2.1/en/	First enrollment: 23-11-2017. Recruitment status: recruiting Other items in manuscript
Protocol version	#3	Date and version identifier	3
Funding	#4	Sources and types of financial, material, and other support	12

1	Roles and	#5a	Names, affiliations, and roles of protocol contributors	13
2	responsibilities:			
3	contributorship			
4				
5				
6	Roles and	#5b	Name and contact information for the trial sponsor	12
7	responsibilities:			
8	sponsor contact		Contact information medical microbiologie	
9	information		AmsterdamUMC: Prof. Dr. M.D. de Jong University of	
10			Amsterdam, Department of Medical Microbiology,	
11			Amsterdam Infection & Immunity Institute,	
12			Meibergdreef 9, Amsterdam, The Netherlands	
13				
14				
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17				
18	Roles and	#5c	Role of study sponsor and funders, if any, in study	12, 13
19	responsibilities:		design; collection, management, analysis, and	
20	sponsor and		interpretation of data; writing of the report; and the	
21	funder		decision to submit the report for publication, including	
22			whether they will have ultimate authority over any of	
23			these activities	
24				
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26				
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28	Roles and	#5d	Composition, roles, and responsibilities of the	13
29	responsibilities:		coordinating centre, steering committee, endpoint	
30	committees		adjudication committee, data management team, and	
31			other individuals or groups overseeing the trial, if	
32			applicable (see Item 21a for data monitoring	
33			committee)	
34				
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36				
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38	Background and	#6a	Description of research question and justification for	4-6
39	rationale		undertaking the trial, including summary of relevant	
40			studies (published and unpublished) examining	
41			benefits and harms for each intervention	
42				
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45	Background and	#6b	Explanation for choice of comparators	4-6
46	rationale: choice			
47	of comparators			
48				
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50	Objectives	#7	Specific objectives or hypotheses	6
51				
52	Trial design	#8	Description of trial design including type of trial (eg,	6, 7
53			parallel group, crossover, factorial, single group),	
54			allocation ratio, and framework (eg, superiority,	
55			equivalence, non-inferiority, exploratory)	
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1	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	6, 7
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8	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)	8
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14	Interventions: description	#11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	n/a observational study
15				
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20	Interventions: modifications	#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)	n/a observational study
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27	Interventions: adherence	#11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return; laboratory tests)	n/a observational study
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32	Interventions: concomitant care	#11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	n/a
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37	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
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48	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)	8, 9
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55	Sample size	#14	Estimated number of participants needed to achieve study objectives and how it was determined, including	10
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		clinical and statistical assumptions supporting any sample size calculations	
Recruitment	#15	Strategies for achieving adequate participant enrolment to reach target sample size	10
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	n/a observational study
Allocation concealment mechanism	#16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	n/a observational study
Allocation: implementation	#16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	n/a
Blinding (masking)	#17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	n/a observational study
Blinding (masking): emergency unblinding	#17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	n/a observational study
Data collection plan	#18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	8
Data collection plan: retention	#18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be	8

1			collected for participants who discontinue or deviate	
2			from intervention protocols	
3				
4	Data	#19	Plans for data entry, coding, security, and storage,	8
5	management		including any related processes to promote data	
6			quality (eg, double data entry; range checks for data	
7			values). Reference to where details of data	
8			management procedures can be found, if not in the	
9			protocol	
10				
11	Statistics:	#20a	Statistical methods for analysing primary and	10
12	outcomes		secondary outcomes. Reference to where other	
13			details of the statistical analysis plan can be found, if	
14			not in the protocol	
15				
16	Statistics:	#20b	Methods for any additional analyses (eg, subgroup	n/a
17	additional		and adjusted analyses)	
18	analyses			
19				
20	Statistics:	#20c	Definition of analysis population relating to protocol	10
21	analysis		non-adherence (eg, as randomised analysis), and any	
22	population and		statistical methods to handle missing data (eg,	
23	missing data		multiple imputation)	
24				
25	Data	#21a	Composition of data monitoring committee (DMC);	No data safety
26	monitoring:		summary of its role and reporting structure; statement	monitoring board
27	formal		of whether it is independent from the sponsor and	has been
28	committee		competing interests; and reference to where further	appointed for this
29			details about its charter can be found, if not in the	study as the risks
30			protocol. Alternatively, an explanation of why a DMC	of this study are
31			is not needed	assumed
32				negligible.
33	Data	#21b	Description of any interim analyses and stopping	n/a No interim
34	monitoring:		guidelines, including who will have access to these	analysis will be
35	interim analysis		interim results and make the final decision to	performed
36			terminate the trial	
37				
38	Harms	#22	Plans for collecting, assessing, reporting, and	n/a No (serious)
39			managing solicited and spontaneously reported	adverse events
40			adverse events and other unintended effects of trial	are expected from
41			interventions or trial conduct	this study
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4	Auditing	#23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	N/a, no audit trial will be conducted
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11	Research ethics approval	#24	Plans for seeking research ethics committee / institutional review board (REC / IRB) approval	12
12				
13				
14	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)	3, Protocol modifications will be communicated to the ethical committee, trial registry and funding body
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27	Consent or assent	#26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	11
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33	Consent or assent: ancillary studies	#26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	10, (specific subsection in informed consent form)
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40	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	8, 9
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51	Declaration of interests	#28	Financial and other competing interests for principal investigators for the overall trial and each study site	12
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55	Data access	#29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	12
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1	Ancillary and	#30	Provisions, if any, for ancillary and post-trial care, and	n/a observational
2	post trial care		for compensation to those who suffer harm from trial	study
3			participation	
4				
5				
6	Dissemination	#31a	Plans for investigators and sponsor to communicate	12
7	policy: trial		trial results to participants, healthcare professionals,	
8	results		the public, and other relevant groups (eg, via	
9			publication, reporting in results databases, or other	
10			data sharing arrangements), including any publication	
11			restrictions	
12				
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16	Dissemination	#31b	Authorship eligibility guidelines and any intended use	13
17	policy:		of professional writers	
18	authorship			
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22	Dissemination	#31c	Plans, if any, for granting public access to the full	No plans for
23	policy:		protocol, participant-level dataset, and statistical code	public access.
24	reproducible			Data use can be
25	research			requested in
26				order to control
27				access and fair
28				use.
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33	Informed	#32	Model consent form and other related documentation	14-26
34	consent		given to participants and authorised surrogates	(Appendices)
35	materials			
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40	Biological	#33	Plans for collection, laboratory evaluation, and storage	10
41	specimens		of biological specimens for genetic or molecular	
42			analysis in the current trial and for future use in	
43			ancillary studies, if applicable	
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BMJ Open

Sensitivity of point of care testing C-reactive protein and procalcitonin to diagnose urinary tract infections in Dutch nursing homes: PROGRESS study protocol

Journal:	<i>BMJ Open</i>
Manuscript ID	bmjopen-2019-031269.R1
Article Type:	Protocol
Date Submitted by the Author:	03-Jul-2019
Complete List of Authors:	Kuil, Sacha; Amsterdam UMC, Medical Microbiology; Hidad, Soemeja; Amsterdam UMC, Medical Microbiology Fischer, Johan; Amsterdam UMC, Clinical Chemistry Harting, Janneke; Amsterdam UMC, Public Health Hertogh, C; VU University Medical Center , General Practice and Elderly Care Medicine Prins, Jan; Amsterdam UMC, Internal Medicine van Leth, Frank; AIGHD, Global Health de Jong, Menno; Amsterdam UMC, Medical Microbiology Schneeberger, Caroline; Amsterdam UMC, Medical Microbiology
Primary Subject Heading:	Infectious diseases
Secondary Subject Heading:	Geriatric medicine, Urology
Keywords:	Point of care testing, C-Reactive protein, Procalcitonin, Nursing homes, Urinary tract infections < UROLOGY, Antimicrobial resistance

SCHOLARONE™
Manuscripts

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3 1 **Sensitivity of point of care testing C-reactive protein and procalcitonin to diagnose**
4 2 **urinary tract infections in Dutch nursing homes: PROGRESS study protocol**

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39 34 **Word count abstract: 295 Word count full text: 4243**
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35 **ABSTRACT**

36 **Introduction:**

37 Suspected urinary tract infection (UTI) ranks among the most common reasons for antibiotic use
38 in nursing homes. However, diagnosing UTI in this setting is challenging because UTI often
39 present with non-specific symptomatology. Moreover asymptomatic bacteriuria is common in
40 elderly, which complicates attribution of causality to detection of bacteria in urine. These
41 diagnostic challenges contribute to overuse of antibiotics and emergence of antimicrobial
42 resistance (AMR) in nursing homes. Given the diagnostic challenges there is a need for point-
43 of-care (POC) diagnostic tests to support clinical rules for diagnosing UTI. Procalcitonin (PCT)
44 and C-reactive protein (CRP) are inflammatory blood markers that have been proven useful to
45 support diagnosis and monitoring of (bacterial) respiratory tract infections and sepsis. While
46 limited studies suggest their usefulness in supporting UTI diagnosis, their utility has not been
47 studied in elderly populations for this purpose.

49 **Methods and analysis:**

50 In a 24-month matched prospective study 'PROGRESS' will assess and compare the sensitivity
51 of rapid POC measurements of blood CRP and PCT levels to support clinical rules for
52 diagnosing UTI in nursing home residents. The primary outcome measure is sensitivity of the
53 POC tests to identify patients with true UTI based on the predefined definition, as derived from
54 Receiving Operating Curves (ROC).

56 **Ethics and dissemination:**

57 This study will be conducted in accordance with Good Clinical Practice guidelines and the
58 principles of the Declaration of Helsinki. The study protocol is approved by the Medical Ethical
59 Committee (METc) of AmsterdamUMC location VUmc with reference number 2017.350 and
60 National Central Committee on Research involving Human Subjects (CCMO) with reference
61 number NL62067.029.17.

63 **Registration details:** Dutch trial registry: NTR6467

64 **Keywords:** Point of care testing, C-Reactive protein, Procalcitonin, Nursing homes, Urinary
65 tract infections, Antimicrobial resistance

67 **Strengths and limitations**

68 Strengths

69 - Stringent post-hoc UTI criteria incorporating microbiology result and clinical response to
adequate antibiotic therapy

71 - Sensitivity in relevant study population, namely nursing home residents

72 Limitations

73 - No on-site point of care test (POCT), however POCT is performed within 4 hours

74 - Single country study

76 **Protocol version 4** March 2019, version 5

77 *1 September 2017 Original*

78 *21 October 2017 Amendment 1:* Addition of information brochures for nursing home staff and
patients

80 *20 November 2017 Amendment 2:* Change of nomenclature for part of nursing home residents
to temporary rehabilitation patients (at rehabilitation wards)

82 *7 March 2018 Amendment 3:* Change exclusion criterion prior inclusion to prior inclusion in the
past 30 days. Subsequently to account for non-independence of observation for a limited
number of participants, the sample size is adjusted with a design effect of 1.1

85 *28 June 2018 Amendment 4:* (1) Change in blood sample collection method (venipuncture to
finger prick collection) due to availability of new procalcitonin POCT with small input volume, as
finger prick blood sample collection is a preferred collection method in the elderly nursing home
population. (2) Change sample size retaining stringent criteria (p-value 0.05 and power 90%) (3)
Prolongation study duration: 12 to 18 months (4) Expansion of number of nursing homes from
11 to 12 nursing homes

91 *3 March 2019 Amendment 5:* (1) Expansion of number of nursing homes from 12 to 13 nursing
homes (2) Adjusted informed consent procedure for capacitated residents

93 *1 July 2019 Amendment 6:* (1) Expansion of number of nursing homes from 13 to 14 nursing
homes (2) Addition of post-hoc analysis using different UTI definitions

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98 **Introduction**

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100 Antimicrobial resistance (AMR) is mainly driven by inappropriate antibiotic use in both humans
101 and animals. AMR is a problem worldwide. Nursing homes are increasingly regarded as an
102 important reservoir for the emergence of AMR^{1 2 3 4}.

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6 105 The most frequently reported infections in elderly residents are urinary tract infections (UTI)⁵. In
7 106 Dutch nursing homes, an average weekly incidence of 10.3 (95% CI 9.8 – 10.8) per 1000
8 107 elderly residents was found⁶. UTI is the most common reason for prescribing antibiotics in Dutch
9 108 nursing homes. However, not all diagnosed UTIs are “true” UTIs since recognition and
10 109 diagnosing UTI in nursing homes is complex. Approximately one third of elderly residents UTIs
11 110 are misdiagnosed, leading to inappropriate antibiotic use^{7 8}. This is mainly due to the facts that
12 111 non-specific, non-urinary tract symptoms such as altered mental status are often attributed to
13 112 UTIs whilst asymptomatic bacteriuria (ASB), possibly resulting in positive urine tests, is also
14 113 common in the elderly residents.⁹
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24 116 *Cognitive impairments and urinary tract infections*

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26 117 The majority of nursing homes in the Netherlands consist of psychogeriatric wards (57%), for
27 118 elderly residents suffering from cognitive impairments, mainly Alzheimer disease^{10 11 12 13}. Their
28 119 ability to verbally communicate or express classical symptoms of UTI, such as dysuria, urgency
29 120 or frequency, is often limited¹⁴. The most frequently presented symptom in elderly residents
30 121 leading to antibiotic prescription for a suspected UTI is an altered mental status (43.3%), while
31 122 classical symptoms as dysuria, urgency and frequency are present in the minority of cases (0 –
32 123 3.8%)¹⁵. Confusion or an altered mental state are nonspecific symptoms and can result from
33 124 other infectious and non-infectious diseases in the elderly residents.
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41 127 *Asymptomatic bacteriuria*

42 128 Asymptomatic bacteriuria (ASB) is defined by the presence of significant bacteriuria without
43 129 symptoms of UTI. ASB is thus regarded as colonization of the urinary tract rather than infection.
44 130 ASB is highly prevalent in healthy elderly persons with reported prevalence rates as high as 40-
45 131 50%^{16 17}. In the presence of ASB, frequently used urine tests based on detection of bacteria are
46 132 less applicable to diagnose UTI, because detection of bacteria does not discriminate between
47 133 ASB and UTI. In combination with above described non-specific symptomatology, it is difficult to
48 134 distinguish ASB from “true” UTI in the elderly.
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140 *C-reactive protein and procalcitonin*

141 In the diagnosis of respiratory tract infections and monitoring of bacterial sepsis, inflammatory
142 markers such as C-reactive protein (CRP) and procalcitonin (PCT) in blood have proven useful
143 to guide antibiotic therapy and reduce antibiotic use^{18 19 20 21}. Currently, point-of-care (POC)
144 CRP measurements are recommended by Dutch guidelines for general practitioners to guide
145 antibiotic treatment for acute respiratory tract infections. CRP and PCT represent potential
146 candidates for rapid POC testing to support UTI diagnosis.

147 Ageing and frailty are associated with changes in serum inflammation protein levels, such as
148 CRP and PCT^{22 23 24}. Therefore, it is important to determine cut-off values for CRP and PCT
149 specifically in the elderly nursing home resident population.

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152 *Inflammatory markers CRP and PCT in UTI*

153 Studies in adults showed that CRP and PCT levels in parenchymatous infections (acute
154 pyelonephritis, prostatitis and epididymitis) are increased^{25 26}. Using a PCT-based algorithm in
155 UTI treatment was shown to reduce antibiotic exposure in adults²⁷. In a subgroup analysis
156 including elderly (> 70 years of age) with lower UTI antibiotic exposure was reduced as well,
157 suggesting a potential role for PCT.

158 In children with UTI, sensitivity of CRP (cut-off 20 mg/L) and PCT (cut-off 0.5 ng/mL) in
159 predicting pyelonephritis is high (94% and 86% respectively), but specificity varies (39% and
160 74% respectively)²⁸. However the number of studies in this systematic review was limited and
161 the heterogeneity substantial. The specificity and positive predictive values varied because CRP
162 and PCT were also increased in children with lower UTI. This suggests a possible role for
163 inflammatory markers in distinguishing UTI (upper and lower UTI) from no inflammation (ASB),
164 where studies are lacking.

165 In the elderly population only three studies have been performed evaluating CRP and PCT and
166 their possible role in UTI^{29 30 31}. These studies focused on hospitalized and more severely ill
167 elderly populations, making the results not applicable to the nursing home population. In 13% of
168 the healthy elderly controls CRP values were increased³⁰, again suggesting increased
169 inflammation in ageing and frailty, indicating the need to determine specific cutoffs in elderly.
170 Studies on lower UTIs and CRP or PCT in elderly are scarce.

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6 173 *Distinguishing UTI and ASB*
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8 174 The majority of UTIs in elderly residents are lower UTIs without fever or other signs of systemic
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10 175 illness^{32 33} while most studies on CRP and PCT in UTI focus on diagnostic values in upper UTI
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12 176 or focus on distinguishing upper and lower UTI. The only small study on distinguishing lower
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14 177 UTI (infection) from ASB (colonization) in adults, shows a high negative predictive value (NPV)
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16 178 for UTI of PCT levels at a cutoff of 0.25 ng/mL³⁴, suggesting that low PCT levels can rule out
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18 179 UTI and contribute to reducing antibiotic use.
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20 181 In conclusion, decisions about who to treat and who not to treat are challenging in elderly
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22 182 residents with suspected UTI, resulting in potential antibiotic overuse in those who do not need
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24 183 treatment. Availability of a simple and rapid point-of-care (POC) test to distinguish true UTI from
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26 184 ASB represents an unmet need which would greatly assist clinical management of these
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28 185 vulnerable patients, not only by improving appropriateness of antibiotic treatment but also by
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30 186 enabling consideration of alternative causes of presenting non-specific symptomatology when
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32 187 UTI is unlikely.
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34 190 **Methods and analysis**

35 191 36 192 *Aim*

37 193 To assess the sensitivity of point-of-care measurements of blood CRP and PCT levels to
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39 194 support clinical rules for diagnosing urinary tract infections UTI in elderly nursing home
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41 195 residents.
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43 196 44 197 45 198 *Outcome*

46 199 The primary outcome is the sensitivity of the point-of-care (POC) test to identify patients with a
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48 200 true UTI based on the predefined definition, as derived from Receiving Operating Curves
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50 201 (ROC).
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52 202 53 203 54 204 *Design and setting*

205 In a prospective matched study we will assess and compare the sensitivity of rapid POC
206 measurements of blood CRP and PCT levels to support clinical rules for diagnosing UTI in
207 elderly residents, with a post-hoc definition of UTI with stringent criteria including microbiology
208 results as gold standard. The start date of this study is November 2017 and the planned end
209 date is December 2019.

212 In this study a true UTI is present when the following five criteria are met: presence of at least
213 two urinary or non-specific symptoms (1), positive urine leucocyte esterase test (2), presence of
214 uropathogens in bacterial culture at $10^4 \geq$ CFU/mL (3), maximum of two uropathogens present
215 (4) and symptom resolution in the course of adequate antibiotic treatment, where adequate
216 treatment is defined by proven susceptibility of isolated uropathogens to the administered
217 antibiotic (5).

220 The matching refers to the assessment of blood CRP and PCT levels in the same study
221 participants. The study will be performed in nursing homes of the University Network for
222 Organizations of Elderly Care of the VUmc University Medical Center (UNO-VUmc). The
223 expected study duration is 24 months.

224 The nursing home population in this study consists mostly of psychogeriatric and somatic (long-
225 stay) wards and some rehabilitation (short-stay) wards.

228 *Informed consent procedure*

229 Most nursing home patients are incapacitated. In case of incapacity legal representatives will be
230 asked for informed consent. Capacitated patients will be asked for informed consent
231 themselves. When nursing home staff suspect a UTI, it is not considered practical to obtain
232 written informed consent from the representatives because preferably the blood sample should
233 be drawn as soon as possible. Therefore informed consent will be obtained pre-emptively at the
234 start of the study or when admitted to the nursing home. This means that patients or their legal
235 representatives provide consent *a priori* to participate in the study once a UTI is clinically
236 suspected during the study period. This procedure will greatly enhance feasibility of enrolment
237 in psychogeriatric nursing home wards.

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Inclusion and exclusion criteria

241 Eligible for study participation are elderly nursing home residents clinically suspected of UTI by
242 the attending physician or nurse. Exclusion criteria are suspected respiratory tract infection,
243 suspected other infection requiring antibiotic therapy, previous study inclusion in the past 30
244 days or lack of written informed consent.

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Study procedures

Study enrolment

249 The study physician will be notified by the nursing home staff when there is potentially eligible
250 patient and will visits the nursing home as soon as possible. The research physician will verify if
251 eligibility criteria are met and will complete study enrolment.

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Data collection

255 To meet our post-hoc UTI criteria data on signs and symptoms, type and duration of antibiotic
256 used, urine leucocyte esterase, bacterial culture and antimicrobial susceptibility and clinical
257 response are collected. Demographic, clinical and laboratory data are collected through an
258 electronic data capture system using software of Open Data Kit³⁵ with which case report forms
259 (CRFs) are designed that incorporate consistency checks to minimize incompatible data points.
260 Data will be handled encoded, working with barcodes scanned by the database APP. Data of
261 paper registration forms is directly entered into online CRFs and uploaded to a pre-defined
262 database.

263 Data on culture results (species, susceptibility patterns by Minimal Inhibitory Concentrations
264 MIC's) from the laboratory system will be collected in the currently used laboratory systems
265 (Labtrain and Kiestra)

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Data on signs and symptoms, clinical response and antibiotic use

269 The attending physician or nurse collects demographic data and data on signs and symptoms of
270 UTI at the day of study enrolment (day 0), see Figure 1. The attending physician or nurse will
271 evaluate improvement of signs and symptoms at day 5 and 10 and hospitalization and/or
272 mortality at day 10. The research physician will visit the nursing homes regularly for monitoring

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3 273 of follow-up. Data on antibiotic use (timing of initial antimicrobial therapy, type of antimicrobial
4 274 agent and possible switch in antimicrobial therapy) will be collected by the research physician
5 275 10 days after study enrolment.
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11 278 *Urine sample collection, dipstick analysis and bacterial culture*

12 279 The attending nurse will collect an urine sample for purpose of this study. Urine samples are
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14 280 collected spontaneously voiding either directly in a sterile urine container or in chamber pot
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16 281 (insert pan). When participants have an indwelling urine catheter, urine will be collected when
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18 282 the urine bag is changed. When participants suffer from urinary incontinence or are not able to
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20 283 urinate on the toilet or chamber pot, diapers will be used to obtain urine for bacterial culture and
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22 284 dipstick analysis^{36 37} Urine extraction from diapers can provide a reliable diagnostic specimen, if
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24 285 fecal contamination can reasonably excluded³⁸. Date, time and way of urine collection method
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26 286 are registered by the attending nurse. The urine sample is stored at 4°C.

27 287 For dipstick analysis Combur 2 (Roche Diagnostics) will be used (nitrite and leucocyte esterase)
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29 288 by the research physician. Urine will be used for semi-quantitative bacterial culture. Inoculation
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31 289 of 10 µL urine to CHROMID CPS Elite agar (Biomerieux) and Columbia CNA agar with 5%
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33 290 sheep blood (Biomerieux) will be streaked using a four quadrant pattern. Bacterial growth will be
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35 291 interpreted after overnight incubation at 35°C in aerobic (CPSE) and CO₂ enriched (CNA)
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37 292 environment. Uropathogens will be identified by Maldi-tof mass spectrometry (Microflex, Bruker
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39 293 Daltonic). All primary and secondary uropathogens in the European Consensus Guideline are
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41 294 considered as uropathogens in this study. Doubtful isolates are considered as non-
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43 295 uropathogens³⁹. When bacterial growth of $\geq 10^4$ CFU/mL is found, antibiotic susceptibility testing
44
45 296 will be performed using the VITEK2 platform (BioMérieux). Unlike the European Consensus
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47 297 Guideline, for all uropathogens $\geq 10^4$ CFU/mL growth will be used as cut-off, as urine collection
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49 298 methods will differ and the presence or absence of specific urinary symptoms. Participants,
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51 299 attending physicians and nurses will not be informed of urine dipstick and bacterial culture
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53 300 results (blinded).
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51 303 *Blood sample collection and point of care testing*

52 304 For POC testing the research physician will collect a blood sample by capillary fingerprick at day
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54 305 0. Capillary blood sample collection is a preferred collection method in the elderly nursing home
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56 306 population. For CRP testing the Afinion AS100 point-of-care (POC) platform (Alere Health B.V.,

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3 307 Tilburg) is used. For PCT testing the Afias1 PCT Plus (Avant Medical B.V., Geffen) is used.
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5 308 Both, CRP and PCT platforms enable capillary blood sample collection to better facilitate future
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7 309 implementation in nursing homes. POC testing is performed according to the manufacturer's
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9 310 protocol. For logistic reasons the POC testing is performed centrally at the Medical Microbiology
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11 311 laboratory of our hospital. POC tests are performed within 4 hours after blood sample collection
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13 312 to ensure stability of CRP and PCT.

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313 Participants, attending physicians and nurses will not be informed of POC-test results (blinded).

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316 *Biological specimen storage and molecular analysis In future*

317 Aliquots of urine samples will be stored (-80°C) when consent is obtained specifically for genetic
318 studies. In future studies metagenomic sequencing will be performed on bacterial DNA, to
319 identify uropathogens and resistance genes.

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322 *Sample size*

323 For the sample size calculation, we used a two-sided p-value of 0.05 and a power of 90%. We
324 assume a difference in sensitivity between the two POC tests of at least 10% as clinically
325 relevant. This difference is the net result of discordant test outcomes between the two tests. The
326 magnitude of discordance is the main parameter in the McNemar test used to calculate the
327 sample size for the matched design.

328 With a proposed sample size of 440 enrolled participants, we are able to adequately assess an
329 increased sensitivity of 10% or more, when the prevalence of UTI in the study population is 40%
330 or more. If the prevalence of UTI is lower, the difference in prevalence that is statistically
331 significant (at the 5%) level increases to 11 or 12%.

332 Based on data of the national sentinel surveillance network for infectious diseases in nursing
333 homes (SNIV) in 2015, the prevalence of UTI in Dutch nursing homes is 10.3 in 1000 patient
334 weeks. UTI definition by SNIV is broader where response to adequate antibiotic treatment is not
335 taken into account.

336 In order to reach the target, the PROGRESS study will take place in 13 nursing homes with
337 around 1350 elderly residents in total. Recruitment of a manageable number of approximately
338 12 participants per week in total is anticipated.

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3 341 *Data and statistical analysis*

4 342 The sensitivity of both POC tests to diagnose UTIs defined by the post-hoc definition is derived
5 343 from the optimal point of a Receiver Operating Curve (ROC). Sensitivities of CRP and PCT tests
6 344 are compared by a matched analysis approach using a two-tailed McNemar test assessing the
7 345 statistical significance of differences between the sensitivities of each pair of tests (performed in
8 346 each study participant).

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11 349 *Patient and public involvement*

12 350 Patients or public are not involved in designing this study.

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15 353 This study protocol is written using the SPIRIT reporting guidelines⁴⁰

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17 355

18 356 **Ethics and dissemination:**

19 357 This study will be conducted in accordance with Good Clinical Practice guidelines and the
20 358 principles of the Declaration of Helsinki. The study protocol is approved by the Medical Ethical
21 359 Committee (METc) of AmsterdamUMC location VUmc with reference number 2017.350 and
22 360 National Central Committee on Research involving Human Subjects (CCMO) with reference
23 361 number NL62067.029.17. Written informed consent from nursing home residents or legal
24 362 representatives (when incapacitated) will be obtained prior to study enrolment. Informed
25 363 consent from residents will be obtained directly by researchers or via mail. Informed consent
26 364 from legal representatives will be obtained via mail. Treating physicians decide which residents
27 365 are capacitated.

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30 368 **Data safety monitoring board**

31 369 No data safety monitoring board has been appointed for this study as the risks of this study are
32 370 assumed negligible.

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35 373 **Discussion**

36 374 This trial will test the hypothesis that increased levels of inflammatory markers support the

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3 375 diagnosis of UTI in elderly and can guide empirical antibiotic treatment in nursing homes.
4 376 Although our study involves the target population (elderly nursing home residents), found CRP
5 377 and/or PCT cut-off values need to be confirmed by prospective trials. Besides test sensitivity the
6 378 potential impact on antibiotic prescription needs to be established in a consecutive, randomized
7 379 controlled, study⁴¹. The aim of this study is assessing the sensitivity of POCT CRP and PCT in
8 380 current clinical practice. This clinical setting suboptimal for urine collection, where urine
9 381 incontinence complicates collection methods. Chamber pots and diapers are frequently used,
10 382 which introduces the risk of UTI overestimation by positive dipstick urinalysis and bacterial
11 383 cultures. Although we have shown in our add-on laboratory study that diapers can be used for
12 384 UTI diagnosis, the outcome can be affected by suboptimal urine collection.
13 385 In this population a reference test for UTIs does not exist, this is the actual gap we are trying to
14 386 address. To reduce classification bias when a reference test is lacking we use a post-hoc
15 387 definition of 'true' UTI⁴². We defined a stringent definition of UTI that in our opinion makes a
16 388 clear distinction between UTI and ASB. We will perform post-hoc analysis in which UTI
17 389 definition is less stringent to assess the effects of potential misclassification of the outcome. UTI
18 390 definition in this post-hoc analysis will include classical UTI symptoms irrespective of resolution
19 391 with adequate antibiotics, in accordance with the Dutch national guidelines⁴³. We will include
20 392 this procedure in the current amendment and data analysis plan that is drawn up before the
21 393 completion of the data collection.
22 394 Since effectiveness of a new test does not just depend on its proven efficacy in research
23 395 studies, but also on successful implementation after the study, qualitative research on
24 396 identifying the barriers and facilitators for implementation are needed and will be performed in
25 397 parallel to the described study.
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400 **Figure legend**

401 Figure 1 Data collection PROGRESS study

402 * Participants and attending physicians/nurses will not be informed of results (blinded)

403 POCT – Point of Care Testing; CRP – C-reactive protein; PCT – Procalcitonin

406 **List of abbreviations**

407 Amsterdam UMC Amsterdam Universitair Medische Centra

408 AMR Antimicrobial resistance

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3 409 ASB Asymptomatic bacteriuria
4 410 CCMO National Central Committee on Research involving Human Subjects
5 411 CFU Colony forming units
6 412 CRP C-reactive protein
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10 414 MIC Minimal inhibitory concentration
11 415 NPV Negative predictive value
12 416 ODK Open data kit
13 417 PCT Procalcitonin
14 418 POC Point of care
15 419 SNIV Surveillance network for infectious diseases in nursing homes
16 420 UNO Universitair netwerk ouderenzorg
17 421 UTI urinary tract infection
18 422 VUmc Vrije Universiteit medisch centrum
19 423
20 424

425 **Declarations**

426 *Dissemination policy*

427 According to the CCMO statement on publication policy, the results of this study will be
428 disclosed unreservedly.

431 *Competing interests*

432 No competing interests.

434 *Funding*

435 This study is funded by The Netherlands Organization for Health Research and Development
436 (ZonMW) grant 541001003. ZonMW, Laan van Nieuw Oost Indië 334, 2593 CE Den Haag, The
437 Netherlands. The study protocol is being peer reviewed by the funder. The funder has no role in
438 collection, management, analysis, and interpretation of data, writing of the report or the decision
439 to submit the report for publication.

440 *Roles and responsibilities*

441 Research physician/PhD student (SDK): coordinating PROGRESS, patient recruiting, data
442 collection, specimen handling, POC and urine testing, preparation protocols, CRFs and

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3 443 publication of study reports including annual ethical committee report.
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5 444 Research assistant (SH): patient recruiting, data collection, specimen handling, POC and urine
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7 445 testing
8 446 Principal investigator (MdJ), senior researcher (CS) and lead epidemiologist (FvL): Study
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10 447 planning, agreement of final protocols, reviewing progress of study
11 448 Lead epidemiologist (FvL): design of the electronic report form (eCRF), database design and
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13 449 maintenance, data validation and data management plans, user account maintenance, design
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15 450 of automated data validation scripts, preparation of the final data analysis sets, supervising of
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17 451 data analysis.
18 452 Laboratory chemist (JCF): responsible for verification of POCT, contractual issues with
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20 453 manufacturers
21 454 In each participating center a lead investigator (elderly care physician) will be identified, to be
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23 455 responsible for progress monitoring and assisting with study set-up per site.
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25 456
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27 458 *Author contributions*

28 459 SDK recruits patients, coordinates and performs the clinical study and the wrote the manuscript.
29
30 460 SH recruits patients and performs the clinical study. FvL, JCF, JH, CMPMH, JMP, MDdJ and
31
32 461 CS developed the protocol and secured funding for this project. CS, FvL and MdJ supervised
33
34 462 the design of the study and writing this manuscript. FvL provided the statistical analysis plan,
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36 463 database set-up. All authors have read and approved the manuscript.
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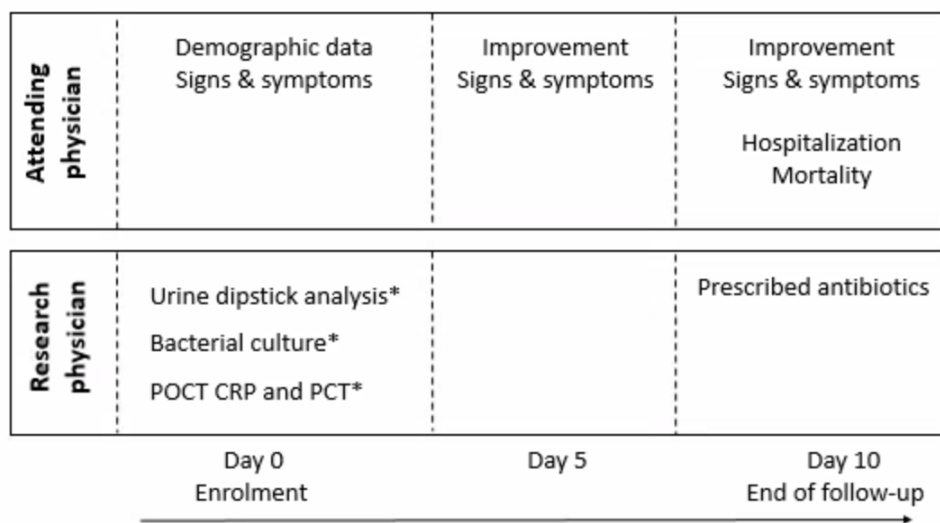


Figure 1 Data collection PROGRESS study

* Participants and attending physicians/nurses will not be informed of results (blinded)
POCT – Point of Care Testing; CRP – C-reactive protein; PCT – Procalcitonin

Reporting checklist for protocol of a clinical trial.

Based on the SPIRIT guidelines.

Instructions to authors

Complete this checklist by entering the page numbers from your manuscript where readers will find each of the items listed below.

Your article may not currently address all the items on the checklist. Please modify your text to include the missing information. If you are certain that an item does not apply, please write "n/a" and provide a short explanation.

Upload your completed checklist as an extra file when you submit to a journal.

In your methods section, say that you used the SPIRIT reporting guidelines, and cite them as:

Chan A-W, Tetzlaff JM, Altman DG, Laupacis A, Gøtzsche PC, Krleža-Jerić K, Hróbjartsson A, Mann H, Dickersin K, Berlin J, Doré C, Parulekar W, Summerskill W, Groves T, Schulz K, Sox H, Rockhold FW, Rennie D, Moher D. SPIRIT 2013 Statement: Defining standard protocol items for clinical trials. *Ann Intern Med.* 2013;158(3):200-207

		Reporting Item	Page Number
Title	#1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	#2a	Trial identifier and registry name. If not yet registered, name of intended registry	2
Trial registration: data set	#2b	All items from the World Health Organization Trial Registration Data Set https://www.who.int/ictrp/network/trds_1.2.1/en/	First enrollment: 23-11-2017. Recruitment status: recruiting Other items in manuscript
Protocol version	#3	Date and version identifier	3
Funding	#4	Sources and types of financial, material, and other support	12

1	Roles and	#5a	Names, affiliations, and roles of protocol contributors	13
2	responsibilities:			
3	contributorship			
4				
5				
6	Roles and	#5b	Name and contact information for the trial sponsor	12
7	responsibilities:			
8	sponsor contact		Contact information medical microbiologie	
9	information		AmsterdamUMC: Prof. Dr. M.D. de Jong University of	
10			Amsterdam, Department of Medical Microbiology,	
11			Amsterdam Infection & Immunity Institute,	
12			Meibergdreef 9, Amsterdam, The Netherlands	
13				
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17				
18	Roles and	#5c	Role of study sponsor and funders, if any, in study	12, 13
19	responsibilities:		design; collection, management, analysis, and	
20	sponsor and		interpretation of data; writing of the report; and the	
21	funder		decision to submit the report for publication, including	
22			whether they will have ultimate authority over any of	
23			these activities	
24				
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28	Roles and	#5d	Composition, roles, and responsibilities of the	13
29	responsibilities:		coordinating centre, steering committee, endpoint	
30	committees		adjudication committee, data management team, and	
31			other individuals or groups overseeing the trial, if	
32			applicable (see Item 21a for data monitoring	
33			committee)	
34				
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38	Background and	#6a	Description of research question and justification for	4-6
39	rationale		undertaking the trial, including summary of relevant	
40			studies (published and unpublished) examining	
41			benefits and harms for each intervention	
42				
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44				
45	Background and	#6b	Explanation for choice of comparators	4-6
46	rationale: choice			
47	of comparators			
48				
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50	Objectives	#7	Specific objectives or hypotheses	6
51				
52	Trial design	#8	Description of trial design including type of trial (eg,	6, 7
53			parallel group, crossover, factorial, single group),	
54			allocation ratio, and framework (eg, superiority,	
55			equivalence, non-inferiority, exploratory)	
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1	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	6, 7
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8	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)	8
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14	Interventions: description	#11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	n/a observational study
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20	Interventions: modifications	#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)	n/a observational study
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27	Interventions: adherence	#11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return; laboratory tests)	n/a observational study
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32	Interventions: concomitant care	#11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	n/a
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37	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
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48	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)	8, 9
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55	Sample size	#14	Estimated number of participants needed to achieve study objectives and how it was determined, including	10
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		clinical and statistical assumptions supporting any sample size calculations	
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4	Recruitment	#15 Strategies for achieving adequate participant enrolment to reach target sample size	10
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8	Allocation:	#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	n/a observational study
9	sequence generation		
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19	Allocation concealment mechanism	#16b Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	n/a observational study
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26	Allocation: implementation	#16c Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	n/a
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31	Blinding (masking)	#17a Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	n/a observational study
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37	Blinding (masking): emergency unblinding	#17b If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	n/a observational study
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43	Data collection plan	#18a Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	8
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56	Data collection plan: retention	#18b Plans to promote participant retention and complete follow-up, including list of any outcome data to be	8
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1			collected for participants who discontinue or deviate	
2			from intervention protocols	
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4	Data	#19	Plans for data entry, coding, security, and storage,	8
5	management		including any related processes to promote data	
6			quality (eg, double data entry; range checks for data	
7			values). Reference to where details of data	
8			management procedures can be found, if not in the	
9			protocol	
10				
11	Statistics:	#20a	Statistical methods for analysing primary and	10
12	outcomes		secondary outcomes. Reference to where other	
13			details of the statistical analysis plan can be found, if	
14			not in the protocol	
15				
16	Statistics:	#20b	Methods for any additional analyses (eg, subgroup	n/a
17	additional		and adjusted analyses)	
18	analyses			
19				
20	Statistics:	#20c	Definition of analysis population relating to protocol	10
21	analysis		non-adherence (eg, as randomised analysis), and any	
22	population and		statistical methods to handle missing data (eg,	
23	missing data		multiple imputation)	
24				
25	Data	#21a	Composition of data monitoring committee (DMC);	No data safety
26	monitoring:		summary of its role and reporting structure; statement	monitoring board
27	formal		of whether it is independent from the sponsor and	has been
28	committee		competing interests; and reference to where further	appointed for this
29			details about its charter can be found, if not in the	study as the risks
30			protocol. Alternatively, an explanation of why a DMC	of this study are
31			is not needed	assumed
32				negligible.
33	Data	#21b	Description of any interim analyses and stopping	n/a No interim
34	monitoring:		guidelines, including who will have access to these	analysis will be
35	interim analysis		interim results and make the final decision to	performed
36			terminate the trial	
37				
38	Harms	#22	Plans for collecting, assessing, reporting, and	n/a No (serious)
39			managing solicited and spontaneously reported	adverse events
40			adverse events and other unintended effects of trial	are expected from
41			interventions or trial conduct	this study
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Auditing	#23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	N/a, no audit trial will be conducted
Research ethics approval	#24	Plans for seeking research ethics committee / institutional review board (REC / IRB) approval	12
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)	3, Protocol modifications will be communicated to the ethical committee, trial registry and funding body
Consent or assent	#26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	11
Consent or assent: ancillary studies	#26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	10, (specific subsection in informed consent form)
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	8, 9
Declaration of interests	#28	Financial and other competing interests for principal investigators for the overall trial and each study site	12
Data access	#29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	12

1	Ancillary and	#30	Provisions, if any, for ancillary and post-trial care, and	n/a observational
2	post trial care		for compensation to those who suffer harm from trial	study
3			participation	
4				
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6	Dissemination	#31a	Plans for investigators and sponsor to communicate	12
7	policy: trial		trial results to participants, healthcare professionals,	
8	results		the public, and other relevant groups (eg, via	
9			publication, reporting in results databases, or other	
10			data sharing arrangements), including any publication	
11			restrictions	
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16	Dissemination	#31b	Authorship eligibility guidelines and any intended use	13
17	policy:		of professional writers	
18	authorship			
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22	Dissemination	#31c	Plans, if any, for granting public access to the full	No plans for
23	policy:		protocol, participant-level dataset, and statistical code	public access.
24	reproducible			Data use can be
25	research			requested in
26				order to control
27				access and fair
28				use.
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33	Informed	#32	Model consent form and other related documentation	14-26
34	consent		given to participants and authorised surrogates	(Appendices)
35	materials			
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40	Biological	#33	Plans for collection, laboratory evaluation, and storage	10
41	specimens		of biological specimens for genetic or molecular	
42			analysis in the current trial and for future use in	
43			ancillary studies, if applicable	
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