

1 **Age-specific prevalence of anti-Pgp3 antibodies and severe conjunctival**
2 **scarring in the Solomon Islands**

3 *Robert Butcher*^{1*}, *Oliver Sokana*², *Kelvin Jack*², *Diana L Martin*³, *Matthew J Burton*¹, *Anthony W*
4 *Solomon*¹, *David CW Mabey*¹, *Chrissy h. Roberts*¹.

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6 1. Clinical Research Department, London School of Hygiene & Tropical Medicine, Keppel Street,
7 London, WC1E 7HT, UK

8 2. Eye Department, Solomon Islands Ministry of Health and Medical Services, P.O. Box 349,
9 Honiara, Solomon Islands

10 3. Division of Parasitic Diseases and Malaria, US Centers for Disease Control and Prevention,
11 Atlanta, GA, USA

12

13 * Corresponding author: robert.butcher@lshtm.ac.uk, +44 207 927 2419

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15 Keywords

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19 Running header

20 Chlamydial serology and scarring in the Solomon Islands

21 Abstract

22 **Background**

23 Trachomatous trichiasis (TT) and ocular *Chlamydia trachomatis* (Ct) infection in the Solomon
24 Islands are scarce, whereas trachomatous inflammation–follicular (TF) is prevalent.

25 **Methods**

26 We enrolled 1511 people aged ≥ 1 year from randomly selected households in 13 villages in which
27 $>10\%$ of the population had TF prior to a single round of azithromycin MDA undertaken six months
28 previously. Blood was collected from people of all ages to be screened for anti-Pgp3 antibodies.
29 Photographs were collected from people of all ages for analysis of scarring severity.

30 **Results**

31 Conjunctival scars were visible in 13.1% of photographs. Mild ($p < 0.0001$) but not severe ($p = 0.149$)
32 scars increased in prevalence with age. Anti-Pgp3 antibody seroprevalence was 18% in 1–9 year
33 olds, increased sharply around the age of sexual debut, and reached 69% in those over 25 years.
34 Anti-Pgp3 seropositivity did not increase significantly between the ages of 1–9 years, and was not
35 associated with scarring in children ($p = 0.472$) or TF in children ($p = 0.581$).

36 **Conclusions**

37 Signs of trachoma are common in the Solomon Islands but occur frequently in individuals who
38 have no serological evidence of prior ocular infection with Ct. WHO recommendations for directing
39 MDA provision based on signs alone may not be suitable in this context.

40 Introduction

41 Trachoma is responsible for approximately 1.9 million cases of visual impairment or blindness
42 globally.¹ International partners have committed to elimination of trachoma as a public health
43 problem by the year 2020. Repeated infections with *Chlamydia trachomatis* (*Ct*) and the
44 immunological response to them can cause a gradual accumulation of scar tissue in the tarsal
45 conjunctivae². Scarring typically begins to develop in late childhood³ and can reach a prevalence
46 of 25% in 10-year-olds in hyperendemic populations⁴. Scarring progresses throughout a lifetime
47 and, in some cases, can lead to entropion, trichiasis, abrasion of the cornea, corneal opacity and
48 blindness⁵. Infection with *Ct* induces the polyclonal production of antibodies to *Ct* antigens^{6–8}.
49 Measuring population-level accrual of antibodies to *Ct* is under investigation as a tool to monitor
50 transmission of both urogenital⁹ and ocular¹⁰ infections.

51 Global trachoma elimination strategies are guided by the signs trachomatous trichiasis (TT) and
52 trachomatous inflammation—follicular (TF). The World Health Organization (WHO) recommends at
53 least three years of mass drug administration (MDA) with azithromycin in districts with >10% TF
54 prevalence in 1–9 year-olds¹¹. As trachoma control reduces prevalence, the positive predictive
55 value of TF as a *Ct* infection marker drops and phenotypically similar diseases with other
56 aetiologies will be unmasked.¹² We have previously reported data from a 2013 population-based
57 prevalence survey covering the two provinces of Temotu and Rennell & Bellona of the Solomon
58 Islands, which showed the prevalence of TF in those aged 1–9 years was moderately high (26.1%
59 of those examined), but TT (0.1% of those examined), trachomatous inflammation—intense (TI;
60 0.2% of 1–9 years-olds examined) and ocular infection with *Ct* (1.3% of 1–9 year-olds examined)
61 were rare¹³. In accordance with WHO guidelines, MDA took place throughout the Solomon Islands
62 in 2014. The program administered approximately 24,000 doses of azithromycin and achieved
63 coverage of approximately 80% in Rennell & Bellona, and 85% in Temotu. After the baseline
64 trachoma mapping in the Solomon Islands, the National Program also used billboard posters and
65 regular radio spots to promote facial cleanliness and raise awareness of trachoma elimination.

66 The discordance between TF prevalence and *Ct* infection, TI and TT prevalence led us to question
67 whether the observed TF was of chlamydial aetiology. Antibodies to *Ct* and trachomatous scarring

68 (TS) are less susceptible to temporal variation than transient markers such as TF or infection and
69 may be more informative of an individual's cumulative trachoma history. We set out to determine
70 whether the high TF prevalence in Rennell & Bellona and Temotu provinces was concurrent with
71 significant burden of TS and whether TF occurred exclusively in those who had previously been
72 infected with *Ct*. To address these questions, villages that previously had high proportions of
73 children with TF were revisited 6 months after MDA and specimens were collected for assessment
74 of anti-Pgp3 antibodies and scars.

75 Methods

76 **Ethics**

77 Study approval was from the London School of Hygiene & Tropical Medicine (LSHTM; 8402) and
78 Solomon Islands National Health Research Ethics Committee (HRC15/03). Subjects aged 18+
79 years gave written informed consent to participate. A parent/guardian provided consent for those
80 aged under 18 years.

81 **Study design**

82 This study took place in June–July 2015, 6 months after a single round of azithromycin MDA had
83 been delivered by the Solomon Islands National Trachoma Elimination Program. To enable
84 comparison to pre-MDA data, only villages in Temotu and Rennell & Bellona provinces where
85 baseline mapping had been conducted were eligible for inclusion. Thirteen villages where over
86 10% of the community (all ages) had signs of TF¹³ were selected. Due to their small respective
87 populations (Temotu: 21,362; Rennell & Bellona: 3041), the two provinces were combined into one
88 evaluation unit during baseline mapping. The number of villages in each province surveyed as part
89 of this study were selected to reflect the relative population proportion (Temotu: 11 villages;
90 Rennell & Bellona: 2 villages).

91 This survey was powered to estimate the prevalence of anti-*Ct* antibody seropositivity in children
92 aged 1–9 years. Based on the low prevalence of ocular *Ct* infection prior to MDA (1.3%), we
93 expected the seroprevalence to be approximately 10%, in line with other communities with low *Ct*

94 prevalence¹⁴. To estimate seroprevalence with $\pm 5\%$ precision at the 95% confidence level, 367
95 children were required¹⁵. In our 2013 survey, we examined a mean of 1.1 children per household
96 and therefore needed 25 households in each of 13 clusters to reach our sample size. All residents
97 aged 1 year or above living in households drawn at random from a list of all households in a study
98 cluster were eligible to participate.

99 **Trachoma grading**

100 Grading using the WHO simplified system¹⁶ for TF, TI and TT was performed in the field by one of
101 two Global Trachoma Mapping Project (GTMP)-certified graders, wearing 2.5× binocular
102 magnifying loupes¹⁷. High-resolution digital photographs of the right tarsal conjunctivae were
103 graded for TS using the modified WHO trachoma grading system (Table 1)¹⁸. Photographs were
104 graded by two photo-graders who had previously achieved kappa scores for inter-observer
105 agreement of >0.8 for F, P and C (follicles, papillae and cicatricae) grades, compared to a highly
106 experienced trachoma grader. Photograph grading was undertaken masked to field grading,
107 laboratory results, and the other photograph grader's assessment. Discrepant grades were
108 arbitrated by a third highly experienced grader.

109 **Specimens**

110 Dried blood spots were collected for assessment of anti-Pgp3 antibody titre. Participants' fingers
111 were pricked with sterile lancets (BD Life Sciences, Oxford, UK) and 10 μ L of blood was collected
112 onto filter paper (CellLabs, Sydney, Australia). Filter wheels were air-dried for 4–12 hours before
113 being sealed in ziplock bags with desiccant sachets. These were refrigerated for up to one week
114 and then stored at -20°C before shipping at ambient temperature to LSHTM.

115 The proportion of children with infection but without active trachoma was low (0.4%) during pre-
116 MDA mapping, therefore swabs were only collected from those with TF during this survey. Swabs
117 were passed three times with a 120° -turn between each pass over the right conjunctiva of children
118 aged 1–9 years who had signs of TF and/or TI in the right eye. Swabs were refrigerated for up to
119 one week and then stored at -20°C before shipping to LSHTM on dry ice for processing. The

120 proportion of active trachoma cases in study villages before MDA was extracted from the full
121 baseline dataset and presented here for comparison.

122 **Serological and nucleic acid testing**

123 Anti-*Ct* Pgp3 antibody titre was assessed using enzyme-linked immunosorbent assay (ELISA)¹⁹²⁰.
124 Optical density (OD) at 450 nm was measured using SpectraMax M3 photometric plate reader
125 (Molecular Devices, Sunnyvale, USA) then normalised to a 20% dilution of presumed-positive
126 standard in presumed-negative standard.

127 DNA was extracted from swabs with the QIAamp DNA mini kit (Qiagen, Manchester, UK). We
128 tested samples for *Homo sapiens* ribonuclease subunit (RPP30 endogenous control) and open
129 reading frame 2 of the *Ct* plasmid (diagnostic target) using a droplet digital PCR assay²¹ with minor
130 modifications²².

131 **Data analysis**

132 All data analyses were conducted using R 3.2.3²³. Pre- and post-MDA proportions were compared
133 using Wilcoxon's rank sum test. ddPCR tests for current ocular *Ct* infection were classified into
134 negative and positive populations according to methods described previously²¹. ELISAs for
135 antibodies to *Ct* were classified as negative or positive using an expectation-maximisation finite
136 mixture model²⁴. Using this method, the threshold normalised OD value for positivity was 0.7997.

137 **Results**

138 **Study demographics**

139 In total, 1511 people (46.3% male; 466 1–9 year-olds) aged 1 year and over were examined in 382
140 households from the 13 selected study villages. By comparison, the pre-MDA survey of the same
141 villages yielded 1534 people (490 1–9 year-olds) in 394 households. Data on non-participation
142 were not collected in June 2015, but the number enrolled was similar to that for the pre-MDA
143 survey, suggesting a similar participation rate (~90%) on both occasions. In this study, there was a
144 mean of 4 people per household aged 1 year and over, and a mean of 1.2 children per household
145 aged 1–9 years, which, after accounting for non-participants, are similar to the means in the 2009
146 Solomon Islands National Census (4.9 people of any age and 1.4 children aged 1–9 years per
147 household in Temotu, 4.4 people of any age and 1.1 people aged 1–9 years per household in
148 Rennell & Bellona).²⁵

149 **Active trachoma and TT**

150 Prior to MDA, there were 165/489 (33.7%) cases of TF in either eye and 1/489 (0.2%) case of TI in
151 those aged 1–9 years in study villages¹³. Following MDA, we observed 66/466 (14.2%) cases of TF
152 and no cases of TI in either eye, representing a decrease in TF of 58% ($p < 0.0001$). A similar
153 pattern was observed in right eyes considered alone – the eyes from which swabs were collected if
154 indicated (Table 2). 56% of TF cases following MDA were bilateral. No cases of TT were identified
155 during this study.

156 In the two enrolled villages in Rennell & Bellona, a slight increase in the prevalence of TF in either
157 eye in those aged 1–9 years following MDA was noted, but it was not statistically significant (11/60
158 [17.9%] before MDA to 14/78 [18.3%] after MDA; $p = 0.956$). In contrast, in the 11 enrolled villages
159 of Temotu, a substantial decrease in TF (from 155/430 [36.0%] before MDA to 52/388 [13.4%] after
160 MDA; $p < 0.0001$) was observed.

161 **Trachomatous scarring**

162 Of the right eye photographs collected, 1440/1511 (95.3%) were suitable for grading. 188/1440
163 (13.1%) photographs were graded as C>0, of which 127 were C1, 53 were C2 and 8 were C3.
164 Exemplars of scarring that resembled normal trachoma phenotypes are shown in Figures 3A and
165 3B. The photo-graders noted that some conjunctivae met the criteria for C3, with clear bands of
166 scarring, but also showed clear features not typically associated with trachomatous pathology. In
167 some cases, these were characterised by boundaries demarcating heavily scarred from apparently
168 healthy conjunctiva (Figure 3C and 3D). Photogrades noted atypical scars in 4/53 (7.5%) C2
169 cases and 3/8 (37.5%) C3 cases. Of the scarred eyelids classed as typical for trachoma, 36/54
170 (67%) were seropositive, whereas 2/7 (29%) of those classed as 'atypical' were seropositive,
171 although the difference in proportions was not significant (chi-squared test $p=0.123$).

172 The age-specific prevalence of scarring is shown in Figure 4. Of 435 photographs graded from
173 children aged 1–9 years, 25 (5.7%) were graded as C>0. In 311 adults aged >40 years who were
174 examined, 74 (23.8%) had C>0 (65 cases of C1, 9 cases of C2, 0 cases of C3). Of 8 cases of C3
175 in the population, 4 (50%) were in children aged 1–9 years, although 2 of these were classed as
176 'atypical'.

177 The proportion of people with C1 increased with age (logistic regression $p<0.0001$) but the
178 proportion of people with more severe scarring (C2 or C3) did not increase with age (logistic
179 regression $p=0.149$). There was no significant association between having C>0 and gender (chi-
180 squared test $p=0.80$). In Rennell & Bellona, 25/225 (11.1%) of photos were graded C>0, whereas
181 in Temotu, 163/1215 (13.4%) of photos were graded C>0; the difference in scarring between
182 provinces was not significant (Wilcoxon Rank Sum test $p=0.289$).

183

184 **Anti-Pgp3 serology**

185 Dried blood spots were collected from 1499/1511 (99.2%) people of all ages during the post-MDA
186 survey; the other 12 people declined finger-prick. Overall, 633/1499 (42.2%) people were
187 considered to be seropositive. In children aged 1–9 years, the prevalence of anti-Pgp3 antibodies
188 was 83/462 (18.0%); in 1-year-olds alone, it was 5/47 (10.6%). The mean seroprevalence in those

189 aged 6–10 years was not significantly higher than in those aged 1–5 years (20.3% compared to
190 16.6%, Wilcoxon rank sum $p=0.276$). The largest increase in seroprevalence was observed
191 between those aged 16–20 years and 21–25 years when the seroprevalence rose significantly
192 from 30.4% to 71.6% (Wilcoxon rank sum $p<0.0001$). Of those aged over 25 years, 67.4% were
193 seropositive (Figure 1). In the 16–20-year-old age group, the prevalence of seropositivity amongst
194 females was higher than in males (13.9% versus 41.1%, Wilcoxon rank sum test $p<0.0001$). The
195 seroprevalence among children in Rennell & Bellona was significantly higher than that in Temotu
196 (38.5% versus 13.8%; chi-squared $p<0.0001$).

197 There was no association between seropositivity and signs of TF in children aged 1–9 years
198 (19.7% seropositive in those with TF in either eye compared to 17.7% seropositive in those without
199 TF, $p=0.581$). In those younger than the median self-reported age of sexual debut (18 years²⁶),
200 there was no association between C grade and anti-Pgp3 OD (linear regression adjusted for age
201 and gender $p=0.453$) or anti-Pgp3 positivity (logistic regression adjusted for age and gender
202 $p=0.472$).

203 **Ocular *Chlamydia trachomatis* infection**

204 Swabs from all 61 children aged 1–9 years with TF in the right eye that were tested for *Ct* had a
205 positive endogenous control result; the median load of the human RPP30 target was 83,000
206 copies, equivalent to over 40,000 conjunctival cells. In this study, 6/61 (9.8%) of children with
207 active trachoma had *Ct* infection. Of the 6 specimens from children positive for *Ct*, the median load
208 was 104,100 plasmid copies/swab. We previously showed that before MDA, 5/160 (3.1%) of those
209 with active trachoma in study villages had evidence of infection with *Ct*. The median pre-MDA load
210 of *Ct* infections in these villages was 51,880 plasmid copies/swab.¹³ Neither the difference between
211 the pre- and post-MDA *Ct* prevalence nor pre- and post-MDA *Ct* load was statistically significant
212 (Wilcoxon rank sum test $p=0.08$ and $p=0.22$, respectively). The relationship between *Ct* infection,
213 signs of trachoma and seropositivity was examined in children aged 1–9 years and is summarised
214 in Table 3. All six cases of active trachoma in which infection was also detected were in
215 seropositive individuals (Figure 2). All study villages had at least one case of TF, but infections

216 were limited to three of the 13 villages studied. Two villages in Rennell & Bellona housed 5/6 *Ct*
217 infections identified during this study.

218 Discussion

219 The Solomon Islands, along with other Pacific Island states, has been identified as trachoma-
220 endemic based on moderately high province-level prevalences of TF. Whilst measures for
221 trachoma elimination have already been deployed in Temotu and Rennell & Bellona, we have
222 previously noted that TI, ocular *Ct* infection and late-stage disease (TT) are rare¹³. If the findings
223 from these study villages were replicated throughout the district, TF would still be sufficiently
224 prevalent to warrant intervention. However, using a suite of non-TF tools (clinical photography for
225 evaluation of conjunctival scarring, nucleic acid infection testing and serological testing), we have
226 demonstrated here that ocular *Ct* is scarce and is not being widely transmitted, and that TF is not
227 concurrent with prevalent severe scarring or TT in this population.

228 We would not expect to see large numbers of individuals with TF who have not previously been
229 infected with *Ct*, but in this population, the majority (80.3%) of individuals with TF were
230 seronegative, and participants with TF were no more likely to be seroreactive to Pgp3 than their
231 peers without TF. We found a small and non-significant increase in age-specific seroprevalence
232 between young children (0–5 years) and older children (6–10 years), which suggests that there is
233 limited horizontal transmission of *Ct* strains among children. This is concordant with our previous
234 data, which suggested that although ocular *Ct* strains are present in the Solomon Islands, they are
235 rare¹³. The increase in seropositivity with age in this group was modest compared with that seen in
236 hyperendemic villages of the United Republic of Tanzania, where seropositivity has been observed
237 to increase from approximately 25% to 94% between the ages of 1 and 6 years.¹⁹ In the current
238 dataset, there was a rapid increase in age-specific seroprevalence around the age of 18 years, the
239 self-reported median age of sexual debut in a nearby population²⁷. The prevalence of urogenital *Ct*
240 infection is known to be high in women attending antenatal clinics in the Solomon Islands²⁷, which
241 may account for the high seroprevalence in adults, and exposure during parturition may also be a
242 major contributor to the 10% of 1-year-olds in our study who had evidence of prior Pgp3
243 exposure.²⁸

244 Antibodies to Pgp3 have recently been suggested for monitoring *Ct* transmission in trachoma
245 programmes^{10,29}, but there is still much to learn about the dynamics of these responses. For

246 example, it is not clear whether anti-Pgp3 responses are detectable in all individuals previously
247 infected with *Ct*, or if multiple exposures are required to develop sustained anti-*Ct* responses. In
248 this study, only 19.7% of children with TF were positive for anti-Pgp3 antibodies, but all six children
249 with current infection were seropositive. There was also a high prevalence of Pgp3 reactivity in
250 adults living in Temotu and Rennell & Bellona, a proportion of whom are likely to have had a
251 previous urogenital *Ct* infection²⁷. While seroreversion due to clearance of infection by MDA is a
252 possible explanation for the low seroprevalence and absence of association of anti-Pgp3
253 antibodies with TF, there is currently no evidence for complete seroreversion for Pgp3-specific
254 antibodies^{9,19} after clearance of infection. We are therefore confident that Pgp3 is an appropriate
255 antigen for serosurveillance in this population.

256 Analysis of the age-specific prevalence of conjunctival scarring illustrated that, while the proportion
257 of people with mild scars increased with age, the proportion of those with more extensive or eyelid-
258 distorting scars did not increase with age. Contrary to what might be expected in a trachoma-
259 endemic community³⁰, no eyelid-distorting scars were found in 311 adults >40 years. There does
260 appear to be severe scarring among children, but some cases are atypical (Figure 3) and are in
261 children who lack Pgp3 reactivity (Table 3), so other causes of scarring may be contributing to this
262 (presumed) ongoing incidence. There are a number of inflammatory conditions (e.g. adenoviral,
263 acute haemorrhagic or membranous conjunctivitis) which may result in conjunctival scarring,
264 although the pathology, incidence and prevalence of these are poorly understood.³¹ It is currently
265 unclear whether the TF that we observed is directly linked to conjunctival scarring in this setting.
266 Currently in Temotu and Rennell & Bellona, the low prevalence of severe scars suggests that the
267 proportion of the population at risk of developing TT is very low, although we cannot determine
268 how this might change temporally.

269 Prior to intervention with MDA, more than 15% of children living in the selected villages had TF.
270 The 13 communities included here were the most highly endemic of those surveyed in Temotu and
271 Rennell & Bellona during the GTMP. In this study, we showed that the burden of TF in many of
272 these villages dropped significantly following a single round of MDA, but still remains above the
273 threshold for continued intervention. The drop in clinical disease was not reflected by a drop in

274 ocular *Ct* in children with TF, which increased, albeit statistically insignificantly. From interventions
275 in other settings, 6 months after a single round of MDA we might expect TF prevalence to drop by
276 approximately 50%, given 80% population coverage^{32,33}. Azithromycin has anti-inflammatory and
277 broad-spectrum antibiotic effects, which may help explain the observed decrease in clinical
278 disease. We observed regional variation across the study villages. Compared to Temotu, we noted
279 more children were seropositive, more children with TF had infection, and MDA did not have as
280 significant an impact on TF levels in Rennell & Bellona. Our survey was not prospectively designed
281 to assess these differences, and the subgroup size in Rennell & Bellona precludes more detailed
282 analysis. Temotu is much more similar to the rest of the Solomon Islands in terms of the geology of
283 the islands, and the lifestyle and ethnicity of the majority of the inhabitants. Further studies on the
284 localisation of trachoma in the islands are warranted.

285 There were limitations to our study. Firstly, due to the remoteness of the islands studied, both
286 blood and swab samples were stored for up to 24 hours at room temperature (commonly around
287 25–30°C in the Solomon Islands) and up to one week at 4°C prior to freezing. DNA and antibodies
288 may degrade if stored for prolonged periods at high temperatures, however, several studies have
289 now shown that short- and long-term storage of swabs at room temperature will not cause
290 significant loss of diagnostic signal in any but the lowest load (and therefore least relevant to
291 transmission) samples^{22,34–36}. Antibodies too are very stable even after short-term re Fridgeration³⁷.
292 Secondly, some commentators have criticised the sensitivity of the ddPCR diagnostic used in this
293 study³⁸. The technique was evaluated against Amplicor and demonstrated only to fail diagnostically
294 in the lowest load samples²¹. Similar protocols were used in other studies in the Pacific and
295 elsewhere in the world and were able to detect infections at high prevalence^{39–41}. Finally, only one
296 antigen was tested for during serological testing using an assay which is still under evaluation.
297 Although Pgp3 is emerging as the marker of choice for *Ct* serosurveillance^{10,42}, verification of these
298 data by testing for an independent immunodominant antigen such as major outer membrane
299 protein with microimmunofluorescence may increase our confidence in the findings.

300 The complex, multistage nature of trachoma makes it difficult to predict the outcome of a given
301 intervention⁴³. Data from cross-sectional surveillance tools used in isolation can be hard to

302 interpret, especially given the prolonged persistence of TF after clearance of infection⁴⁴. Some
303 features of conjunctivitis in the Solomon Islands resemble trachoma, particularly the prevalent
304 follicular inflammation and some of the severe conjunctival scarring. Crucially, these clinical
305 features were not co-endemic with TT at a prevalence that indicates a public health problem. In
306 this setting, tests for infection gave a better indication of the public health threat from trachoma
307 than TF. A combined approach in which various age-specific markers of trachoma are assessed
308 together across the complete age range of the population, may prove useful for prioritising areas
309 for intervention where the prevalence of TF alone does not coherently reflect trachoma's public
310 health importance.

311 Contrary to the WHO recommendation for treatment based solely on prevalence of TF, our data
312 provide compelling evidence that trachoma is not a public health problem in these villages. Whilst
313 there have been substantial collateral benefits to local residents from having received MDA (such
314 as on genital *Ct*⁴⁵ and yaws⁴⁶), further rounds of azithromycin MDA do not appear to be indicated
315 for the purposes of trachoma elimination. As the positive predictive value of TF decreases globally,
316 other countries may emerge where TF is not reflective of threat to vision. WHO recommendations
317 for implementation of MDA and the SAFE strategy should be reviewed in the light of this evidence.

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334 Author Contributions

335 Conceived and designed the study: RMRB, OS, DLM, AWS, DCWM, ChR.

336 Performed the fieldwork: RMRB, OS, KJ

337 Provided training and reagents: DLM

338 Performed the experiments: RMRB, MJB, ChR

339 Analysed the data: RMRB, MJB, ChR

340 Wrote the manuscript: RMRB, ChR

341 Revised and approved the manuscript: RMRB, OS, KJ, DLM, MJB, AWS, DCWM, ChR

342 Tables

Table 1. Grading system for conjunctival scarring, reproduced from Dawson and colleagues¹⁸.

Grade	Classification	Definition
C0	Absent	No scarring on the conjunctiva
C1	Mild	Fine scattered scars on the upper tarsal conjunctiva, or scars on other parts of the conjunctiva
C2	Moderate	More severe scarring but without shortening or distortion of the upper tarsus
C3	Severe	Scarring with distortion of the upper tarsus

343

Table 2. Population characteristics of study populations before and after MDA, 13 selected communities of Temotu and Rennell & Bellona Provinces, Solomon Islands.

Characteristic	Pre-MDA (October– November 2013; ¹³)	Post-MDA (June–July 2015: this study)	p-value*
Number examined, all ages	1534	1511	-
Number examined aged 1–9 years	490	466	-
Number of households enrolled	394	382	-
% male of those examined	46.5	46.3	0.836
TF in either eye in 1–9 year olds	33.7%	14.2%	<0.0001
Active trachoma in swabbed eye (right eye field assessment)	160 (32.7%)	61 (13.1%)	<0.0001
<i>Ct</i> infection in those aged 1–9 years	5 (3.1%)	6 (9.8%)	0.08
Median <i>Ct</i> infection load in positive specimens (plasmid copies/swab)	51,880	104,100	0.219

Ct: Chlamydia trachomatis

* t-test

344

Table 3. Serological status compared to other tests for trachoma, 13 selected communities of Temotu and Rennell & Bellona Provinces, Solomon Islands, June-July 2015

Comparator		1–9 year-olds			≥10 year-olds		
		Seronegative	Seropositive	Total	Seronegative	Seropositive	Total
Ct infection by ddPCR*	Positive	0	6	6	-	-	-
	Negative	48	7	55	-	-	-
TF	Positive	53	13	66	13	9	22
	Negative	326	70	396	474	541	1015
TI	Positive	0	0	0	0	1	1
	Negative	379	83	462	487	549	1036
Scarring	C0	333	77	410	414	418	832
	C1	15	1	16	36	75	111
	C2	3	2	5	16	32	48
	C3	3	1	4	1	3	4

Ct: *Chlamydia trachomatis*; ddPCR: droplet digital polymerase chain reaction; TF: trachomatous inflammation—follicular;

TI: trachomatous inflammation—intense.

* presence or absence of infection only assessed in children with TF and/or TI.

345

346

347 Figure legends

348 **Figure 1:** Age-specific seroprevalence of anti-Pgp3 antibodies from dried blood spots collected
349 from 13 selected communities of Temotu and Rennell & Bellona Provinces, Solomon Islands,
350 June–July 2015.

351 **Figure 2:** Anti-Pgp3 antibody titre from children aged 1–9 years in 13 selected communities of
352 Temotu and Rennell & Bellona Provinces, Solomon Islands, June–July 2015. Those with TF and/or
353 TI but no infection are highlighted in blue, those with TF and/or TI and infection are red and those
354 with neither TF nor TI nor infection are grey.

355 **Figure 3.** Photographs taken of residents in 13 selected communities of Temotu and Rennell &
356 Bellona Provinces, Solomon Islands, June-July 2015. **(A and B)** Conjunctivae with features
357 meeting the criteria for C3 characteristic of trachoma. **(C and D)** Conjunctivae with features
358 meeting the criteria for C3 but thought not to be trachomatous in origin. Study IDs were SB113564,
359 SB108878, SB107613 and SB108669.

360 **Figure 4.** Age-specific prevalence of scarring (defined as $C > 0$), as identified by photograph
361 grading, in 13 selected communities of Temotu and Rennell & Bellona Provinces, Solomon Islands,
362 June–July 2015.

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