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1 **Serological reactivity and bacterial genotypes in *Chlamydia trachomatis***
2 **urogenital infections in Guadeloupe, French West Indies**

3

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19

20 **SUMMARY**

21

22 **Key-words:** *Chlamydia trachomatis*, epidemiology, serology, prevalence, genotyping,
23 Guadeloupe.

24 **Running title:** *Chlamydia trachomatis* in Guadeloupe

25

26 **KEY MESSAGES**

27

28 The prevalence of *Chlamydia trachomatis* urogenital infection was 16.9% in men and
29 9.8% in women in 2000 in Guadeloupe, French West Indies.

30

31 The distribution of *Chlamydia trachomatis* genotypes responsible for urogenital
32 infections in Guadeloupe, French West Indies, differed from that in mainland France.

33

34 The SERO-CT assay was unable to detect antibodies in the serum samples of patients
35 infected with *Chlamydia trachomatis* genotype Ia strains.

36

37 **STATEMENT**

38

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45

46 **COMPETING INTEREST**

47 None declared.

48

49 **ABSTRACT**

50

51 **Objectives:** To determine the prevalence and genotypes of *Chlamydia trachomatis*
52 urogenital infection in Guadeloupe, French West Indies. To compare *C. trachomatis*
53 direct detection to serological testing.

54 **Methods:** From March to November 2000, 971 consecutive patients (888 women and 83
55 men) who had been referred to clinical laboratory of the *Institut Pasteur de la*
56 *Guadeloupe* for routine testing for genital infection, were recruited. Samples were
57 subjected to a nucleic acid amplification assay (AMP CT, Gen-Probe). Genotypes were
58 determined by *omp1* PCR-RFLP analysis. Serological testing was carried out with the
59 commercially available peptide-based ELISA assay (SERO-CT IgG/IgA,
60 Savyon/BMD).

61 **Results:** Positive AMP CT test results were obtained for 102 (10.5%) of the 971
62 samples. The prevalence of infection was 16.9% in men and 9.8% in women. The most
63 common genotypes were E (34.3%), F (23.9%), Da (13.4%), I (9%), and Ia (7.5%). No
64 relationship was found between genogroups and age, sex or clinical symptoms. With
65 AMP CT used as a reference, the sensitivity, specificity, positive, and negative
66 predictive values of SERO-CT were 81.1%, 56%, 34.5% and 91.2 %, respectively, for
67 IgG and 55.4%, 76.8%, 59.4 %, and 85.8 %, respectively, for IgA. IgG seroprevalence
68 rates were very low (1/5, 20%) in patients infected with genotype Ia strains.

69 **Conclusions:** The prevalence found in Guadeloupe did not differ not significantly from
70 that which was found in mainland France. The genotypes Da, F, I , and Ia were more
71 prevalent in Guadeloupe; however, the SERO-CT assay was unable to detect serum
72 antibodies in 80% of the patients infected with genotype Ia strains.

73 INTRODUCTION

74

75 *Chlamydia trachomatis* is considered to be the most common bacterial cause of
76 sexually transmitted infections (STIs) worldwide. It is currently classified into 19
77 different serovars on the basis of immunogenic epitope analysis of the major outer
78 membrane protein (MOMP). Serovars D to K, and related variants, Da, Ia, and Ga, are
79 generally found to be associated with urogenital infections.^{1,2} Over the last two decades,
80 genotyping based on the *omp1* gene, encoding MOMP, has become a convenient
81 alternative to the laborious serotyping method. The single-copy *omp1* gene displays
82 extensive sequence variations in four regions (VS1-VS4), interspersed with five regions
83 of conserved sequence. The diversity of the DNA can be analyzed by restriction
84 fragment length (RFLP) analysis and/or by sequencing of the PCR-amplified *omp1*
85 gene.¹⁻¹¹ Genotyping was originally carried out on cell cultures, but direct genotyping of
86 clinical specimens is now performed by sensitive nested PCR of the *omp1* gene.¹²
87 Phylogenetic analysis of *omp1* subdivides strains into three distinct genogroups : B
88 (serovars B, Ba, D, Da, E, L1 and L2), intermediate (serovars F, G, and Ga), and C
89 (serovars A, C, H, I, Ia, J, K, and L3).^{1,2}

90 In Guadeloupe, French West Indies, there is no data in the literature concerning
91 *C. trachomatis* urogenital infection. In mainland France, commercially available PCR
92 methods have showed that the prevalence of such infection in symptomatic populations
93 was 8-10.2% in women and 16.4% in men (1994-1998).^{13, 14} In the Caribbean region,
94 the prevalence ranges from 5.4% to 21% in asymptomatic women from Curaçao,
95 Barbados, and Trinidad, to 55% in STI clinic patients from Jamaica.¹⁵⁻¹⁹ Despite the use
96 of different methods and populations, which made the results difficult to compare, these

97 studies provide useful data to national health authorities. A piece of valuable information
98 that has not yet been reported in the Caribbean region are the genotypes or serovars of
99 circulating *C. trachomatis* strains.

100 We report here the first study on *C. trachomatis* urogenital infection in
101 Guadeloupe. We established the prevalence of this infection by carrying out nucleic acid
102 amplification tests (NAATs) on a population of 971 consecutive patients referred for
103 routine testing for urogenital infection, and determined the genotypes by PCR-RFLP.
104 We searched for relationships between genogroups and particular demographic, clinical,
105 serological, and microbiological characteristics of the infected population.

106

107 **MATERIALS AND METHODS**

108

109 **Study population**

110

111 Guadeloupe is an overseas French *département* covering 1705 km² and located in
112 the Caribbean region. The archipelago had a total population of 422,000 inhabitants
113 (1999 census data).

114 The selected population consisted of 971 consecutive patients (888 women and
115 83 men), who were referred to the clinical laboratory of the *Institut Pasteur de la*
116 *Guadeloupe*, Pointe à Pitre, Guadeloupe, by general practitioners, dermatologists or
117 gynaecologists during the period from March to November 2000 for routine testing for
118 urogenital infection. All the specimens were collected and all assays other than
119 genotyping were carried out in the laboratory.

120 Data, including demographic characteristics such as age and sex, clinical

121 symptoms and results of all microbiological investigations were recorded for two subsets
122 of patients. The first subset was comprised of patients with a positive NAAT (n=102).
123 The second subset comprised of those with an available serological test (n=333), was
124 classified as follows: one CT+ group (n=74; median age: 24.6±7.4 years; range: 13-44)
125 comprised of patients, 65 of which were women and 9 of which were men, with AMP
126 CT-positive assays; and one CT- group (n=259; median age: 29.2±9.9 years; range: 14-
127 70) comprised of 238 women and 21 men. Ethical approval was not required under local
128 law for this study due to its retrospective design and use of data obtained through routine
129 testing.

130

131 ***C. trachomatis* nucleic acid detection**

132

133 *C. trachomatis* direct detection was performed with the Amplified *Chlamydia*
134 *trachomatis* assay (AMP CT; Gen-Probe, San Diego, USA). This NAAT targets a
135 specific 23S rRNA of *C. trachomatis* in a transcription-mediated amplification and
136 hybridisation protection assay. Male urethral (n=72) and female endocervical (n=867)
137 swabs or first-void urine specimens from men (n=11) or women (n=21) were collected
138 and tested according to the manufacturer's instructions.

139

140 ***C. trachomatis* antibody detection**

141

142 *C. trachomatis* antibody status was determined on 333 sera from 333 patients
143 with the SERO-CT IgG/IgA test (Savyon/BMD, Marne-La-Vallée, France), according to

144 the manufacturer's instructions. This microtiter-based ELISA uses a mixture of species-
145 specific peptides from the *C. trachomatis* MOMP for the detection of specific IgG and
146 IgA antibody. Samples with index values (OD sample/OD negative control x 2) \leq 1.1
147 were considered negative, whereas those with index values $>$ 1.1 were considered
148 positive.

149

150 **Genotyping of *C. trachomatis***

151

152 Of the 100 AMP CT-positive non-urine specimens, stored in a Gen-Probe
153 transport tube at -70°C , 99 were transported frozen, overnight, to the French National
154 Reference Centre for *C. trachomatis*. Samples were diluted 1/10 with lysis buffer and
155 nested PCR analyses were performed by using primers NLO and NRO in the first step
156 and PCTM3 and SERO2A in the second step, as described previously.^{5, 6} The RFLP
157 analysis of nested PCR products was carried out by using *AluI*, *HpaII-EcoRI-HinfI*,
158 *CfoI*, *DdeI*, and *FokI*, as described previously.^{5, 6}

159

160 **Statistical analysis**

161

162 Univariate comparisons were based on the Pearson's chi-squared statistic or two-
163 tailed Fisher's exact tests for categorical variables, and on Student's t tests or analysis of
164 variance for continuous variables. Analyses were performed with Stat software (Stata
165 7.0; Stata, College Station, USA). A p value of 0.05 was considered statistically
166 significant.

167

168

169 **RESULTS**

170

171 ***C. trachomatis* prevalence and characteristics of the infected population**

172

173 Positive results in the NAAT were obtained for 102 of the 971 samples (10.5%).

174 The prevalence of *C. trachomatis* was 16.9% in men (15/83) and 9.8% (87/888) in

175 women. The mean age of infected men and women was 29.4±6.9 years (range 20-44)

176 and 24.4±7.2 years (range 13-44), respectively. Clinical symptoms were present in

177 89.3% (75/84) of infected women and in 92.9% (13/14) of infected men. The prevalence

178 in women of co-infection with HIV, *Neisseria gonorrhoeae*, *Trichomonas vaginalis* and

179 *Candida albicans* was 5.9% (5/84), 2.3% (2/87), 3.4% (3/87), 19.5% (17/87), and 5.7%

180 (5/87), respectively. Bacterial vaginosis was seen in 58.6% (51/87) of the *C.*

181 *trachomatis*-infected women. Infected men were co-infected with only one other agent,

182 *N. gonorrhoeae*, in 3 of 14 (21.4%) patients.

183

184 **Distribution of the *C. trachomatis* genotypes**

185

186 The *omp1* gene was successfully amplified in 67/99 (67.7%) non-urine positive

187 samples. The distribution of the genotypes is shown in Table 1. The genotypes E

188 (34.3%), F (23.9%), Da (13.4%), I (9%), and Ia (7.5%) were the most common. The

189 genotypes were evenly distributed over the study period (data not shown). Due to the

190 small number of non-E and non-F genotypes, the genotypes were grouped together into

191 the B, intermediate, and C genogroups for further analysis.¹

192

193 **Association of genogroups with demographic characteristics and clinical signs**

194

195 Table 2 shows the association between demographic characteristics, clinical
196 signs, laboratory data and genogroup. No relationships were found between genogroup
197 and age, sex, clinical symptoms or other associated genital infections other than *N.*
198 *gonorrhoeae* and the C genogroup ($p = 0.005$). These co-infections (two caused by Ia
199 genotype strains and two by I genotype strains) occurred between July and September.

200

201 **Relationships between chlamydial infection and serological data**

202

203 We evaluated the potential usefulness of serological testing, by analysing a
204 subpopulation of 333 patients with an available serological test result with the
205 commercially available peptide-based ELISA, SERO-CT IgG/IgA. A comparison of the
206 serological results with the results of the NAAT is shown in Table 3. The IgG and IgA
207 seroprevalence rates and the mean IgG and IgA ratios in the CT+ group were
208 significantly higher than those in the CT- group.

209 The sensitivity of SERO-CT for antibody detection (using the NAAT as the
210 reference) was 81.1% for IgG and 55.4% for IgA. The specificity of detection was 56%
211 for IgG and 76.8% for IgA. The specificity was higher (81.5%) for tests for the
212 simultaneous presence of IgG and IgA. Positive predictive values (PPV) were 34.5% for
213 IgG and 59.4% for IgA and negative predictive values (NPV) were 91.2% for IgG and
214 85.8% for IgA. IgG antibodies were found in 55.6% (35/63) of patients suffering from

215 lower abdominal pain. They were found in 81.2% (13/16) in such patients in the CT+
216 group and 46.8% (22/47) of such patients in the CT- group; whereas the occurrence of
217 abdominal pain was similar in the two groups: 22.2% (16/72) and 19.1% (47/246),
218 respectively.

219

220 **Relationships between genogroup and serological results**

221

222 Serological results were available for 46 of the 67 patients whose strains were
223 genotyped. Table 4 shows IgA and IgA seroprevalence rates as a function of genogroup.
224 Statistically significant higher IgG seroprevalence rates were found in patients infected
225 with genotypes of the intermediate (100%) and B (92.3%) genogroups than in those
226 infected with genotypes of the C genogroup (40%). Five of the 10 C genogroup strains
227 were of genotype I and the other five were of genotype Ia. IgG was detected in 60%
228 (3/5) of patients infected with strains of genotype I but in only 20% (1/5) of patients
229 infected with strains of genotype Ia.

230

231 **DISCUSSION**

232

233 We found the prevalence of *C. trachomatis* urogenital infection to be 10.7%
234 (10% in women and 18.1% in men) in our study population. This prevalence was lower
235 than those observed in symptomatic patients from the Caribbean region but did not differ
236 significantly from that of mainland France.

237 The failure rate (33%) for determination of genotypes by nested *omp1* PCR on
238 clinical samples in this study was higher than generally reported (< 20%).^{10, 20, 21} There
239 may be several reasons for this. It is more difficult to amplify the 1.2 kb *omp1* gene,
240 which is present as a single copy in the bacterium, than the smaller rRNAs present in
241 many thousands of copies. High failure rates are therefore likely for samples with small
242 numbers of micro-organisms or samples containing small number of infected cells. This
243 may account for the higher failure rate in men (50%) than in women (30.2%), due to
244 milder urethral scraping in men. Other hypothesis are the lack of a DNA extraction
245 procedure before PCR (Vanduyhoven *et al.* reported a nested PCR failure rate of
246 22.9%, which decreased to 4.9% following the use of DNA extraction methods) and the
247 quality of the DNA matrix stored in the Gen-Probe transport medium, which was
248 developed for the stabilisation of RNA molecules.¹

249 This study is the first to describe the *C. trachomatis* genotypes circulating in
250 Guadeloupe and in the Caribbean region. The two most prevalent genotypes in
251 Guadeloupe, E and F, have also been reported to be the most prevalent around the
252 world.^{9, 11, 20, 22} However, the distribution of some other genotypes differed considerably
253 from that in mainland France: in particular, genotypes Da (13.4% vs 4.5%), F (23.8% vs
254 13.5%), I (9% vs 3.5%) and Ia (7.4% vs 0.3%) were more prevalent, whereas genotypes

255 D (5.9% vs 11.2%), G (3% vs 11.8%), H (0% vs 5.1%), and K (0% vs 5.1%) were less
256 prevalent.²³ Genotype Da infections have rarely been described since the first
257 description of this serovar in 1991.²⁴ No Da genotypes were found among the 507 *ompI*
258 sequences obtained for samples from a large US nationwide study conducted between
259 1995 and 1997.⁹ A genotyping study by PCR-RFLP (*CfoI*) in 2001 in Cameroon found
260 a prevalence of the Da genotype of 8.6% (3/35), whereas in Senegal in 1996-1997, *ompI*
261 sequencing revealed a prevalence of D/Da genotypes of 19% (4/21).^{6, 11} The D/Da
262 strains belong to two distinct phylogenetic lineages.⁸ The Da strains from each lineage
263 have a specific single nucleotide polymorphism (SNP) within *ompI* resulting in the loss
264 of a *CfoI* restriction site and in amino acid substitution alanine to threonine at position
265 311/312 modifying the epitope recognised by the monoclonal antibodies used for the
266 serotyping of group D strains.^{7,8} BLAST analysis can group *ompI* sequences into the
267 appropriate lineage, but it is necessary to identify the SNP encoding the Ala311/312Thr
268 substitution precisely for Da assignment. This may explain why it is more difficult to
269 detect Da variants by *ompI* sequencing than by serotyping or PCR-RFLP, leading to
270 possible underestimation. The PCR-RFLP profiles obtained with the Da and D strains
271 from Guadeloupe were similar to those predicted from the *ompI* sequences of Da/TW-
272 448 (first lineage), and D1 or D2 group (second lineage) strains, respectively.

273 The serovar variant Ia was described in 1991, following a study of 41 genital
274 isolates isolated worldwide, in which Ia isolates were much more frequent than I isolates
275 (41 vs 9).²⁴ In the United States, a high prevalence (14%) of genotype Ia was found
276 during the 1995-1997 study.⁹ Other studies in the US found that infection with serovar
277 Ia was significantly associated with being black.^{25, 26} In a study carried out in 1994 in
278 the Netherlands, male patients from Suriname were significantly more frequently

279 infected with genotype Ia strains than were men from the Netherlands, and women from
280 Suriname were more frequently infected with genogroup C strains than were women
281 from the Netherlands.¹ In Africa, one study conducted in Senegal, found a genotype Ia
282 prevalence of 4.8% (1/21).¹¹ In Asia, one studied reported a prevalence of genotype Ia
283 infection of 6.6% in pregnant women in Thailand, whereas another study reported a
284 significant increase in the prevalence of genotype I strains (Ia not differentiated from I)
285 in Japan between the mid 1990s (1/41, 2.1%) and 2003-2005 (10/81, 12.3%).^{9, 27} In
286 Guadeloupe, a large proportion of the population is Black and of African descent (about
287 80 %) and about 15% of the population is “Indian” and of Asian descent. That may
288 explain why genotypes I (9%) and Ia (7.5%) were well represented.

289 Serological results and direct evidence of infection have not been found to
290 correlate in many reports, even with an ELISA using specific *C. trachomatis* MOMP
291 peptides.^{28, 29} This has been partially attributed to the antibody kinetics (i.e., the lag
292 period between being infected and the antibody response and the persistence of
293 antibodies after a resolved infection). Rabenau *et al.* reported a lower specificity (37.3%)
294 and a much lower PPV (< 20%) for IgG detection with SERO-CT than for detection
295 with NAAT.²⁹ However, a high NPV (96.4%) was found to be useful for identifying
296 patients at high risk in whom *C. trachomatis* infection is unlikely to play a role. SERO-
297 CT was reported to outperform PCR for IgG detection in another study (specificity of
298 69% and PPV of 50%).²⁸ We also found that this assay performed well for IgG and IgA
299 detection in comparison with AMP CT. Our results, however, could be affected by
300 patient selection bias as the analyses were done on a subpopulation selected based on the
301 basis on available serology. The requests for serology might indicate a higher risk of *C.*
302 *trachomatis* infection, in particular upper genital tract infection or recurrent infections.

303 Indeed, the prevalence of this infection was found to be 21.5% (65/333) in the
304 subpopulation vs 10.7% within the whole population. For optimal serological results, the
305 selection or preparation of the microbial antigen is important. In a follow-up study, the
306 SERO-CT gave false-negative results and the authors suggested that this test might not
307 be suitable for all serovars.³⁰ We show here that SERO-CT detected no antibody
308 response in 6/10 patients infected with strains of the C genogroup, particularly genotype
309 Ia strains. However, further studies involving more cases are necessary to confirm this
310 finding. The exact composition of the SERO-CT antigen mixture has not been made
311 publicly available, but it seems likely that no Ia-derived peptide was included.
312 Therefore, in seroepidemiological studies using such high-specificity ELISA-based
313 assays, it is essential to establish, before beginning the study, that the ELISAs used are
314 capable of detecting an antibody response in patients infected with the various
315 circulating genotypes.

316

317 **AUTHOR CONTRIBUTIONS**

318

319 F-X.W. and B.D.B. designed the study; F-X.W. and S.L.H performed sampling and
320 microbiological analysis; M.C. performed genotyping; C.S. performed statistical
321 analysis; F-X.W. and B.D.B. analysed data; F-X.W., S.L.H, and B.D.B. wrote the
322 manuscript.

323

324 **WORD COUNT**

325

326 Abstract (254), text (2567), number of figures (0) and tables (4)

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Serovar		Male	Female	Total
		n	n (%)	n (%)
B genogroup	D	0	4 (6.7)	4 (6)
	Da	1	8 (13.4)	9 (13.4)
	E	3	20 (33.3)	23 (34.3)
	Subtotal	4	32 (53.4)	36 (53.7)
Intermediate genogroup	F	1	15 (25)	16 (23.9)
	G	0	2 (3.3)	2 (3)
	Subtotal	1	17 (28.3)	18 (26.9)
C genogroup	H	0	0	0
	I	2	4 (6.7)	6 (9)
	Ia	0	5 (8.3)	5 (7.5)
	J	0	2 (3.3)	2 (3)
	K	0	0	0
	Subtotal	2	11 (18.3)	13 (19.4)
Total		7	60 (100)	67 (100)

420

421 **Table 1: Distribution of *C. trachomatis* genotypes in women and men, as**
422 **determined by *omp1* nested PCR-based RFLP genotyping**

423

		B genogroup	Intermediate genogroup	C genogroup	P
		(n=36)	(n=18)	(n=13)	
		n (%)	n	n	
Sex	Male	4 (11.1)	1	2	0.6**
	Female	32 (88.9)	17	11	
Age	<=20	10 (27.8)	10	4	0.12**
]20-25]	13 (36.1)	3	6	
]25-30]	5 (13.9)	4	3	
	>30	8 (22.2)	1	0	
		Mean/Std.dev	25.1 ± 7.6	22.8 ± 6.3	22.1 ± 4.1
	Min-Max	15-44	16-42	13-30	
Clinical symptoms	Absent	1 (2.9)	1	1	0.76**
	Present	33 (97.1)	17	12	
Pruritis	Absent	21 (63.6)	10	4	0.19*
	Present	12 (36.4)	7	8	
Pelvic pain	Absent	23 (69.7)	10	8	0.72**
	Present	10 (30.3)	7	4	
Discharge	Absent	6 (18.2)	2	2	0.9**
	Present	27 (81.8)	15	10	
Urethritis	Absent	30 (90.9)	16	10	0.72**
	Present	3 (9.1)	1	2	
Genital infections					
N. gonorrhoeae	Absent	35 (97.2)	18	9	0.005**
	Present	1 (2.8)	0	4	
Vaginosis	Absent	18 (50.0)	10	5	0.64*
	Present	18 (50.0)	8	8	
T. vaginalis	Absent	34 (94.4)	18	12	0.58**
	Present	2 (5.6)	0	1	
Candida sp.	Absent	34 (94.4)	17	12	1**
	Present	2 (5.6)	1	1	
HIV	Absent	30 (88.2)	18	13	0.23**
	Present	4 (11.8)	0	0	

425 *Chi-squared test, ** Fisher's exact test, *** Analysis of variance test

426 **Table 2: Clinical and laboratory data as a function of *C. trachomatis* genogroup**

Serological data	N (%) of					
	Men			Women		
	CT+ (n=9)	CT- (n=21)	p	CT+ (n=65)	CT- (n=238)	p
IgA-negative	4	17	0.05*	29 (44.6)	182 (76.5)	<0.0001*
IgA-positive	5	4		36 (55.4)	56 (23.5)	
Mean IgA ratio	2.1±1.6	0.8±0.43	0.002***	1.6±1.4	1.0±1.1	0.0002***
Min-Max	0.5-5.32	0.39-2.13		0.27-8.02	0.14-12.34	
IgG-negative	1	14	0.005*	13 (20.0)	131 (55.0)	<0.0001*
IgG-positive	8	7		52 (80.0)	107 (45.0)	
Mean IgG ratio	3.0±2.1	1.3±1.5	0.02***	4.5±3.1	2.0±2.2	<0.0001***
Min-Max	0.7-7.56	0.38-5.48		0.41-11.6	0.22-11	
IgA+/IgG+	5	3	0.02**	35 (53.8)	45 (18.9)	<0.0001**
IgA-/IgG+	3	4		17 (26.2)	62 (26.0)	
IgA-/IgG-	1	13		12 (18.5)	120 (50.4)	
IgA+/IgG-	0	1		1 (1.5)	11 (26.1)	

428 * Chi-squared test, ** Fisher's exact test, *** Student's t test

429

430 **Table 3: Serological data in men and women as a function of positive and negative**
 431 **results for *C. trachomatis* detection in the Gen-Probe AMP CT assay**

432

	B genogroup (n=26)	Intermediate genogroup (n=10)	C genogroup (n=10)	P
Serological data	n	n	n	
IgA-negative	10	3	7	0.15*
IgA-positive	16	7	3	
Mean IgA ratio	1.44±0.93	1.6±0.74	1.87±2.47	0.70**
Min-Max	0.27-3.45	0.72-3.07	0.34-8.02	
IgG-negative	2	0	6	0.001*
IgG-positive	24	10	4	
Mean IgG ratio	5.5±2.9	6.5±3.8	2.3±2.6	0.008**
Min-Max	0.74-10.46	1.1-11.6	0.41-7.39	
IgA+/IgG+	16	7	3	0.007*
IgA-/IgG+	8	3	1	
IgA-/IgG-	2	0	6	

434 * Fisher's exact test, **Analysis of variance

435

436 **Table 4: Serological data as a function of *C. trachomatis* genogroup**

437