

## SUPPLEMENTARY MATERIAL

Supplementary Table 1. Genital coinfections with <i>Mycoplasma genitalium</i> <sup>ab</sup>						
	MG negative n= 1220 (%, 95% CI)		MG positive n=83 (%, 95% CI)		Adjusted Odds Ratio (95% CI) <sup>c</sup>	p-value <sup>d</sup>
<b><i>Chlamydia trachomatis</i><sup>e</sup></b>						
Negative	1118	(92, 91–94)	75	(90, 82–96)	1	
Positive	92	(8, 6–9)	8	(10, 4–18)	1.28	(0.60–2.74) 0.525
<b><i>Neisseria gonorrhoeae</i><sup>f</sup></b>						
Negative	1199	(99, 98–100)	82	(99, 93–100)	1	
Positive	11	(1, 0–2)	1	(1, 0–7)	1.27	(0.16–10.01) 0.818
<b>Bacterial vaginosis<sup>g</sup></b>						
Negative	813	(70, 67–73)	51	(64, 52–74)	1	
Positive	349	(30, 27–33)	29	(36, 26–48)	1.29	(0.80–2.07) 0.295
<b>Vulvovaginal candidiasis<sup>h</sup></b>						
Negative	918	(77, 74–79)	61	(74, 64–83)	1	
Positive	281	(23, 21–26)	21	(26, 17–36)	1.12	(0.67–1.88) 0.658

Abbreviations: MG=*Mycoplasma genitalium*, n=number, CI=confidence interval

<sup>a</sup>Women with an unassessable MG result (n=15) were excluded from the analysis

<sup>b</sup>*Trichomonas vaginalis* is extremely uncommon at Melbourne Sexual Health Centre and present in <1% of attendees. 41 women with MG were assessed for *T. vaginalis* and only 1 (2%) was positive for *T. vaginalis*.

<sup>c</sup>Adjusted for number of male sexual partners in the last three months as a risk factor.

<sup>d</sup>p-value calculated using logistic regression

<sup>e</sup>*C. trachomatis* result was unassessable in 9 women and not tested for in 1 woman.

<sup>f</sup>*N. gonorrhoeae* result was unassessable in 8 women and not tested for in 2 women.

<sup>g</sup>Bacterial vaginosis was not assessed in 61 women.

<sup>h</sup>Vulvovaginal candidiasis was not assessed in 22 women.

**Supplementary Table 2. Associations between *Chlamydia trachomatis* and demographics, past sexual practices and self-reported symptoms in the prior week<sup>a</sup>**

	CT negative N=1133 median (range) or n (%), 95% CI)	CT positive N=92 [median (range) or n (%), 95% CI]	Adjusted Odds Ratio (95% CI) <sup>b</sup>	p- value <sup>c</sup>
<b>Median age</b>	26 (23-29)	24 (22-29)	0.97 (0.92–1.01)	0.123
<b>Median number of male partners in past 12m</b>	4 (2-6)	4 (3-6)	1.02 (0.98–1.06)	0.341
<b>Condom use in past 12m<sup>d</sup></b>				
Always	82 (7, 6-9)	3 (3, 1-9)	1	
Not Always	1032 (93, 91-94)	88 (97, 91-99)	2.90 (0.69–12.09)	0.144
<b>STI in the past 6m<sup>de</sup></b>				
No	966 (87, 85-89)	77 (87, 78-93)	1	
Yes	146 (13, 11-15)	12 (13, 7-22)	0.65 (0.29-1.45)	0.295
<b><u>Self-reported symptoms (in the week prior to recruitment)</u></b>				
<b>Abdominal Pain</b>				
No	967 (86, 84-88)	79 (86, 77-92)	1	
Yes	161 (14, 12-16)	13 (14, 8-23)	0.86 (0.44-1.66)	0.647
Missing	5	0		
<b>Dyspareunia</b>				
<b>Women with VVC</b>				
No	256 (93, 89-96)	8 (73, 39-94)	1	
Yes	20 (7, 4-11)	3 (27, 6-61)	8.66 (1.90-39.54)	<b>0.005</b>
<b>Women without VVC</b>				
No	741 (90, 87-91)	71 (93, 85-98)	1	
Yes	87 (10, 9-13)	5 (7, 2-15)	0.48 (0.17-1.35)	0.164
<b>Vaginal discharge</b>				
No	755 (67, 64-70)	48 (53, 43-64)	1	
Yes	369 (33, 30-36)	42 (47, 36-57)	2.12 (1.32-3.42)	<b>0.002</b>
Missing	9	2		
<b>Abnormal vaginal odour<sup>f</sup></b>				
No	859 (76, 74-79)	65 (71, 61-80)	1	
Yes	264 (24, 21-26)	26 (29, 20-39)	1.36 (0.80-2.32)	0.257
Missing	10	1		
<b>Vaginal itch</b>				
No	886 (79, 76-81)	77 (85, 76-91)	1	
Yes	241 (21, 19-24)	14 (15, 9-24)	0.74 (0.37-1.48)	0.399
Missing	6	1		
<b>Post-coital bleeding</b>				
No	1033 (92, 90-94)	81 (89, 81-95)	1	
Yes	89 (8, 6-10)	10 (11, 5-19)	1.22 (0.56-2.62)	0.620
Missing	11	1		
<b>Intermenstrual bleeding</b>				
No	1013 (90, 88-92)	82 (92, 84-97)	1	
Yes	111 (10, 8-12)	7 (8, 3-16)	0.78 (0.35-1.77)	0.557
Missing	9	3		

**Dysuria**

No	978	(87, 85-89)	83	(92, 85-97)	1		
Yes	148	(13, 11-15)	7	(8, 3-15)	0.55	(0.23-1.30)	0.173
Missing	7		2				

**Urinary frequency**

No	935	(83, 81-85)	71	(79, 69-87)	1		
Yes	191	(17, 15-19)	19	(21, 13-31)	1.22	(0.68-2.18)	0.508
Missing	7		2				

Abbreviations: CT=*Chlamydia trachomatis*, n=number, CI=confidence interval, STI=sexually transmitted infection; m=month, VVC = vulvovaginal candidiasis

<sup>a</sup>Women with an unassessable CT result (n=10) or with *M. genitalium* (n= 83; includes 8 women co-infected with MG and *C. trachomatis*) were excluded from the analysis

<sup>b</sup>All analyses were adjusted for number of male partners, VVC, *N. gonorrhoea* and concurrent BV.

<sup>c</sup>p-value calculated using logistic regression and bold indicates significant findings p<0.05.

<sup>d</sup>Data missing for up to 3% of participants.

<sup>e</sup>STI in the past six months referred to bacterial STI only, however some women may have misinterpreted this question and answered with regards to warts or other nonbacterial STI, and therefore this should be interpreted with caution.

<sup>§</sup> We tested for interaction terms between CT and genital co-infections and the only significant interaction was between CT and VVC for dyspareunia (p=0.016). Therefore, the association between dyspareunia and CT could not be adjusted for VVC. To account for potential confounding by VVC on the relationship between CT and dyspareunia, data were then stratified by VVC status, and the association between CT and dyspareunia was investigated within each stratum.

<sup>f</sup>Abnormal vaginal odour refers to any self-reported odour, not specifically a fishy odour

<b>Supplementary Table 3. Self-reported symptoms and clinical signs identified in women with <i>Mycoplasma genitalium</i> compared to women with <i>Chlamydia trachomatis</i><sup>ab</sup></b>					
<b>Self-reported symptoms</b>					
	<b>MG positive, n=75</b>		<b>CT positive, n=92</b>		<b>p-value<sup>c</sup></b>
	<b>(%, 95% CI)</b>		<b>(%, 95% CI)</b>		
<b>Cohort</b>					
Asymptomatic	8	(11, 5-20)	13	(14, 8-23)	0.502
Symptomatic	67	(89, 80-95)	79	(86, 77-92)	
<b>Abdominal Pain</b>					
No	64	(86, 77-93)	79	(86, 77-92)	0.909
Yes	10	(14, 7-23)	13	(14, 8-23)	
Missing	1		0		
<b>Dyspareunia</b>					
No	66	(90, 81-96)	81	(90, 82-95)	0.930
Yes	7	(10, 4-19)	9	(10, 5-18)	
Missing	2		2		
<b>Vaginal discharge</b>					
No	48	(65, 53-76)	48	(53, 43-64)	0.136
Yes	26	(35, 24-47)	42	(47, 36-57)	
Missing	1		2		
<b>Abnormal odour<sup>c</sup></b>					
No	60	(81, 70-89)	65	(71, 61-80)	0.150
Yes	14	(19, 11-30)	26	(29, 20-39)	
Missing	1		1		
<b>Vaginal itch</b>					
No	55	(74, 63-84)	77	(85, 76-91)	0.100
Yes	19	(26, 16-37)	14	(15, 9-24)	
Missing	1		1		
<b>Post-coital bleeding</b>					
No	64	(89, 79-95)	81	(89, 81-95)	0.980
Yes	8	(11, 5-21)	10	(11, 5-19)	
Missing	3		1		
<b>Intermenstrual bleeding</b>					
No	65	(88, 78-94)	82	(92, 84-97)	0.359
Yes	9	(12, 6-22)	7	(8, 3-15)	
Missing	1		3		
<b>Dysuria</b>					
No	64	(85, 75-92)	83	(92, 85-97)	0.158
Yes	11	(15, 8-25)	7	(8, 3-15)	
Missing	0		1		
<b>Urinary frequency</b>					
No	62	(83, 72-90)	71	(79, 69-88)	0.541
Yes	13	(17, 10-28)	19	(21, 13-31)	
Missing	0		2		
<b>Fever</b>					
No	71	(96, 89-99)	87	(97, 91-99)	0.807
Yes	3	(4, 1-11)	3	(3, 1-9)	
Missing	1		2		

<b>Clinical signs on examination<sup>b</sup></b>					
	<b>MG positive, n=67</b>		<b>CT positive, n=79</b>		<b>p-value<sup>c</sup></b>
		<b>(%, 95% CI)</b>		<b>(%, 95% CI)</b>	
<b>Vaginal discharge</b>					
No	15	(25, 14-37)	17	(32, 18-48)	0.826
Yes	46	(75, 63-86)	57	(68, 52-82)	
Not assessed/missing	6		5		
<b>Abnormal odor<sup>c</sup></b>					
No	44	(72, 59-83)	50	(68, 56-78)	0.566
Yes	17	(28, 14-41)	24	(32, 22-44)	
Not assessed/missing	6		5		
<b>Vulval redness</b>					
No	46	(75, 63-86)	61	(82, 72-90)	0.317
Yes	15	(25, 14-37)	13	(18, 10-28)	
Not assessed/missing	6		5		
<b>Mucopurulent cervicitis</b>					
No	37	(82, 68-92)	46	(73, 60-83)	0.263
Yes	8	(18, 8-32)	17	(27, 17-40)	
Not assessed/missing	22		16		
<b>Cervical or adnexal motion tenderness</b>					
No	31	(86, 71-95)	37	(76, 61-87)	0.227
Yes	5	(14, 5-29)	12	(24, 13-39)	
Not assessed/missing	31		30		
<b>Cervical contact bleeding</b>					
No	36	(84, 69-93)	44	(75, 62-85)	0.267
Yes	7	(16, 7-31)	15	(25, 15-38)	
Not assessed/missing	24		20		
<b>Vaginal pH</b>					
≤4.5	37	(56, 43-68)	38	(51, 39-63)	0.577
>4.5	29	(44, 32-57)	36	(49, 37-61)	
Not assessed/missing					
<b>High vaginal polymorph count</b>					
<5	35	(54, 41-66)	31	(41, 30-53)	0.121
≥5	30	(46, 34-59)	45	(59, 47-70)	
Not assessed/missing	2		3		

**Notes:** Abbreviations: MG=*Mycoplasma genitalium*, n=number, CI=confidence interval, CT=*Chlamydia trachomatis*

<sup>a</sup> Analysis only includes women with CT or MG infection. Women co-infected with CT and MG have been excluded.

<sup>b</sup> Asymptomatic women were not clinically assessed and have been excluded from the analysis. Clinical signs were elicited only in women with clinical indications for examination and in particular, cervical assessment and bimanual examination was undertaken in women with specific indications for a speculum and bimanual exam..

<sup>c</sup> p-value calculated using Chi-square test

<sup>c</sup> Abnormal odour refers to any odour, not specifically a fishy odour.

Indications for testing women for *M. genitalium*.

<b>Supplementary Table 4. Associations between <i>Chlamydia trachomatis</i> and clinical signs among symptomatic women<sup>ab</sup></b>						
	<b>Total women n=1037</b>	<b>CT negative n=958 (% , 95% CI)</b>	<b>CT positive n=79 (% , 95% CI)</b>	<b>Adjusted odds ratio (95% CI)<sup>c</sup></b>	<b>p-value<sup>d</sup></b>	
<b>Vaginal discharge</b>						
No	325	308 (35, 32-39)	17 (23, 14-34)	1		
Yes	623	566 (65, 61-68)	57 (77, 66-86)	2.13 (1.19-3.82)	<b>0.011</b>	
Not assessed/missing	89	84	5			
<b>Abnormal odour<sup>e</sup></b>						
No	717	667 (76, 73-79)	50 (68, 56-78)	1		
Yes	233	209 (24, 21-27)	24 (32, 22-44)	1.42 (0.84-2.41)	0.193	
Not assessed/missing	87	82	5			
<b>Vulval redness</b>						
No	690	629 (72, 69-75)	61 (82, 72-90)	1		
Yes	252	239 (28, 25-31)	13 (18, 10-28)	0.82 (0.40-1.67)	0.589	
Not assessed/missing	95	90	5			
<b>Mucopurulent cervicitis</b>						
No	657	611 (89, 86-91)	46 (73, 60-83)	1		
Yes	93	76 (11, 9-14)	17 (27, 17-40)	2.85 (1.49-5.44)	<b>0.002</b>	
Not assessed/missing	287	271	16			
<b>Cervical or adnexal motion tenderness</b>						
No	482	445 (78, 75-82)	37 (76, 61-87)	1		
Yes	135	123 (22, 18-25)	12 (24, 13-39)	0.99 (0.48-2.07)	0.986	
Not assessed/missing	420	390	30			
<b>Cervical contact bleeding</b>						
No	600	556 (86, 83-88)	44 (75, 62-85)	1		
Yes	107	92 (14, 12-17)	15 (25, 15-38)	1.93 (0.98-3.78)	0.056	
Not assessed/missing	330	310	20			
<b>Vaginal pH</b>						
≤4.5	602	564 (62, 59-65)	38 (51, 39-63)	1		
>4.5	384	348 (38, 25-41)	36 (49, 37-61)	1.42 (0.87-2.33)	0.161	
Not assessed/missing						
<b>High vaginal polymorph count</b>						
<5	587	556 (60, 57-63)	31 (41, 30-53)	1		
≥5	414	369 (40, 37-43)	45 (59, 47-70)	2.50 (1.49-4.20)	<b>0.001</b>	
Not assessed/missing	36	33	3			

Abbreviations: MG=*Mycoplasma genitalium*, n=number, CI=confidence interval

<sup>a</sup>Women with an unassessable CT result (n=10) or with *M. genitalium* (n= 83; including 8 women coinfecting with CT and *M. genitalium* en) were excluded from the analysis. In addition, asymptomatic women (n=188) were not clinically assessed and have been excluded from the analysis.

<sup>b</sup>Clinical signs were elicited only in women with clinical indications for examination and in particular, cervical assessment and bimanual examination was undertaken in women with specific indications for a speculum and bimanual exam.

<sup>c</sup>All analyses were adjusted number of male sexual partners, vulvovaginal candidiasis, *N. gonorrhoea* and concurrent BV, with the exception that we did not adjust for BV in models examining associations with individual Amsel criteria (i.e. vaginal discharge, abnormal vaginal odour and vaginal pH)

<sup>d</sup>p-value calculated using logistic regression and bold indicates significant findings p<0.05.

### Indications for testing women for *M.genitalium*.

<sup>e</sup>Abnormal vaginal odor refers to any clinician-recorded odour, not specifically a fishy odour