Lack of demonstrated Clinical Value in Pre-Treatment Determination of the Presence of *parC* Mutations in *Mycoplasma genitalium*.

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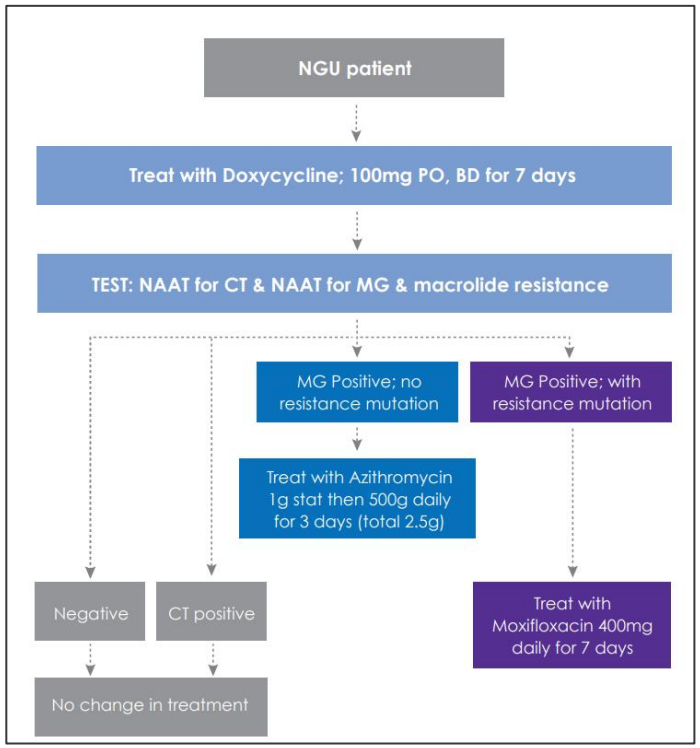
Introduction

- British, European and international guidelines recommend treating *Mycoplasma genitalium (Mgen)* infections according to their macrolide resistance-mediating mutation (MRMM) status[1-4] (Figure 1).
- This is now referred to as Resistance Guided Therapy (RGT) and has been demonstrated to dramatically increase *Mgen* cure rates in high resistant populations.[5,6]
- During implementation of RGT, *Mgen* infections identified with MRMMs are recommended to be treated with extended spectrum fluoroquinolones (FQ), most commonly moxifloxacin.[1-4]
- Mutations in the quinolone resistance determining region (QRDR) of the topoisomerase IV gene (*parC*) of *Mgen* (predominantly resulting in amino acid changes in positions Ser83 and Asp87) have been linked to high minimum inhibitory concentrations[7,8] and have been more commonly identified in patients who have failed FQ treatment[9-12].

Figure 1.
Resistance Guided Therapy for management of non-gonococcal urethritis (NGU)[1-4]



NGU = Non-gonococcal urethritis. PO = per os (oral administration) . BD = bis in die (twice daily). NAAT = Nucleic acid amplification test. CT = Chlamydia trachomatis

Study Aim

- We assessed whether the data reported in treatment studies, supports pre-treatment determination of *parC* mutations to implement RGT of macrolide-resistant *Mgen*.

Methods

- A literature review was undertaken to identify studies meeting the following criteria:
    - Peer-reviewed patient outcome study of treatment of macrolide-resistant *Mgen*
    - Presence of *parC* QRDR mutations determined at enrolment
    - Clinical and microbiological tests of cure performed
    - Number of subjects with *parC* QRDR mutants reported irrespective of outcome

Results

- 6 studies met the criteria outlined for inclusion in this review:
    - 1 study reported treatment of *Mgen* infection following RGT.
    - The remaining 5 studies reported treatment of *Mgen* infection which was not based on MRMM status (non-RGT). Predominantly this involved 1st line treatment with a macrolide (Azithromycin), followed by 2nd line treatment with FQ (Moxifloxacin or Sitafloxacin).
- 119 subjects in total had *parC* mutation-positive *Mgen* infection, with FQ treatment outcome data available:
    - 82 subjects (68.9%) were successfully treated with a FQ
    - 36 subjects (30.2%) failed treatment with a FQ
- Full results are shown in Table 1.

Table 1. Outcomes of patients with *parC* mutation-positive *Mgen* infection

Study	No. *Mgen* positive patients with baseline *parC* data	Treatment/ management pathway	No. of patients with confirmed *parC* mutation and FQ treatment outcome	Outcome of *Mgen* Infection	
				FQ Cure (%)*	FQ Failure (%)*
Couldwell et al. (2013)[13]	33	Non RGT	3	0 (0%)	3 (100%)
Kikuchi et al. (2014)[11]	51	Non RGT	9	9 (100%)	0 (0%)
Murray et al. (2017)[14]	140	Non RGT	10	4 (40%)	6 (60%)
Chambers et al. (2019)[15]	20	Non RGT	3	3 (100%)	0 (0%)
Murray et al. (2020)[16]	326	RGT	81	57 (70.4%)	24 (29.6%)
Conway et al. (2020)[17]	96	Non RGT	3	2 (66.7%)	1 (33.3%)
TOTAL			**119**	**82 (68.9%)**	**36 (30.2%)**

***Pre-treatment *parC* mutations identified**
Ref 13 – FQ failure: 3x S83I
Ref 11 – FQ cure: 12x S83N, 3x S83I, 2x D87N & 1x A119E
Ref 14 – FQ cure: 3x S83I, 1x D87N. FQ failure: 4x S83I, 2x S83R
Ref 15 – FQ cure: 2x S83I, 1x S83N
Ref 16 – FQ cure: 1x G81S, 1x S83C, 33x S83I, 4x S83N, 3x S83R, 13x D87N, 1x D87Y, 1x D87G. FQ failure: 1x S83C, 22x S83I, 1x D87N, 1x D87Y
Ref 17 – FQ cure: 1x S83R or S83I, 1x D87N. FQ failure: 1x S83R or S83I. Study utilised qPCR test which did not distinguish between S38R and S83I

Discussion

- The majority of patients with pre-treatment *parC* QRDR mutations were cured with a FQ containing regimen. This includes the *parC* mutation causing amino acid change S83I which has more commonly been linked to treatment failure.[9,12,16]

- The FQ treatment successes outlined in this study may indicate the potential role of other factors, including fitness and organism load which can contribute to spontaneous clearance.[14,15,18]

- The use of RGT, which involves 7-day pre-treatment with Doxycycline, has also been demonstrated to reduce *Mgen* organism load,[5,6] likely contributing to successful treatment of *Mgen* infections with FQ in the presence of *parC* mutations.

- Interactions between *parC* and *gyraseA* (*gyrA*) mutations may also play a potential role, but this is yet to be determined. *gyrA* mutations have been reported to provide an additive effect to *parC* mutations when contributing to FQ resistance,[16] however they are yet to be associated with FQ treatment failure in the absence of *parC* mutations.[19]

- Despite the high FQ success rate demonstrated in this study, the European Medicines Agency recommends that FQ should not be used when an alternative antibiotic therapy is available due to potentially harmful side effects.[20] This indicates that RGT is critical to minimise use of FQ for treatment only when macrolide resistant *Mgen* infections have been identified.

Conclusions

- Limited data is currently available concerning factors influencing the outcome of *Mgen* infections treated with FQ.
- There is currently little evidence that pre-treatment testing for *par*C QRDR mutations can positively influence therapeutic decision making.

References

1. Soni et al. Int J STD AIDS. 2019 Sep;30(10):938-950
2. Jensen JS et al. J Eur Acad Dermatol Venereol 2016 Oct;30(10):1650-1656.
3. Horner PJ et al. Int J STD AIDS 2016 Oct;27(11):928-37.
4. Australian STI Management Guidelines – Mycoplasma genitalium 2018. http://www.sti.guidelines.org.au/sexually-transmissible-infections/mycoplasma-genitalium
5. Read et al. Clin Infect Dis. 2019 Feb 1;68(4):554-560
6. Durukan et al. Clin Infect Dis. 2019 Oct 20 [Epub ahead of print]
7. Fookes et al. BMC Genomics 2017 Dec 28;18(1):993.
8. Hamusura et al. PLoS One. 2018 Jun 8;13(6):e0198355.
9. Bissessor et al. Clin Infect Dis 2015 Apr 15;60(8):1228-36.
10. Shimada et al. Int J Antimicrob Agents 2010 Sep;36(3):255-8.
11. Kikuchi et al. J Antimicrob Chemother 2014 Sep;69(9):2376-82.
12. Deguchi et al. J Antimicrob Chemother 2014 Sep;69(9):2376-82.
13. Couldwell et al. Int J STD AIDS 2013 Oct;24(10):822-8.
14. Murray et al. Emerg Infect Dis 2017 May;23(5):809-812.
15. Chambers et al. Sex Transm Dis 2019 Dec;46(12):805-809.
16. Murray et al. J Infect Dis 2020 Mar 2;221(6):1017-1024.
17. Conway et al. Sex Transm Dis 2020 Mar;47(3):197-198.
18. Walker et al. Clin Infect Dis 2013 Apr;56(8):1094-100.
19. Unemo M, Jensen JS. Nat Rev Urol 2017 Mar;14(3):139-152
20. European Medicines Agency. Disabling and potentially permanent side effects lead to suspension or restrictions of quinolone and fluoroquinolone antibiotics. 2018. https://www.ema.europa.eu/documents/referral/quinolone-fluoroquinolone-article-31-referral-disablingpotentially-permanent-side-effects-lead_en.pdf.