

Genetic Variability in *Mycoplasma genitalium*: A mechanism for persistence of infection?



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Pathobiology

Mycoplasma genitalium

- Isolated in 1981
- Associated with reproductive tract disease
- Fastidious organism
- Minimal genome
- Alternative genetic code
- Flask-shaped morphology:
 - Attachment organelle
 - Location of MgpB & MgpC





The diagram shows a DNA strand with several regulatory elements represented by colored boxes: a yellow box, a red box, a dark red box, a purple box, and a red box. A yellow arrow points to the right from the first yellow box.

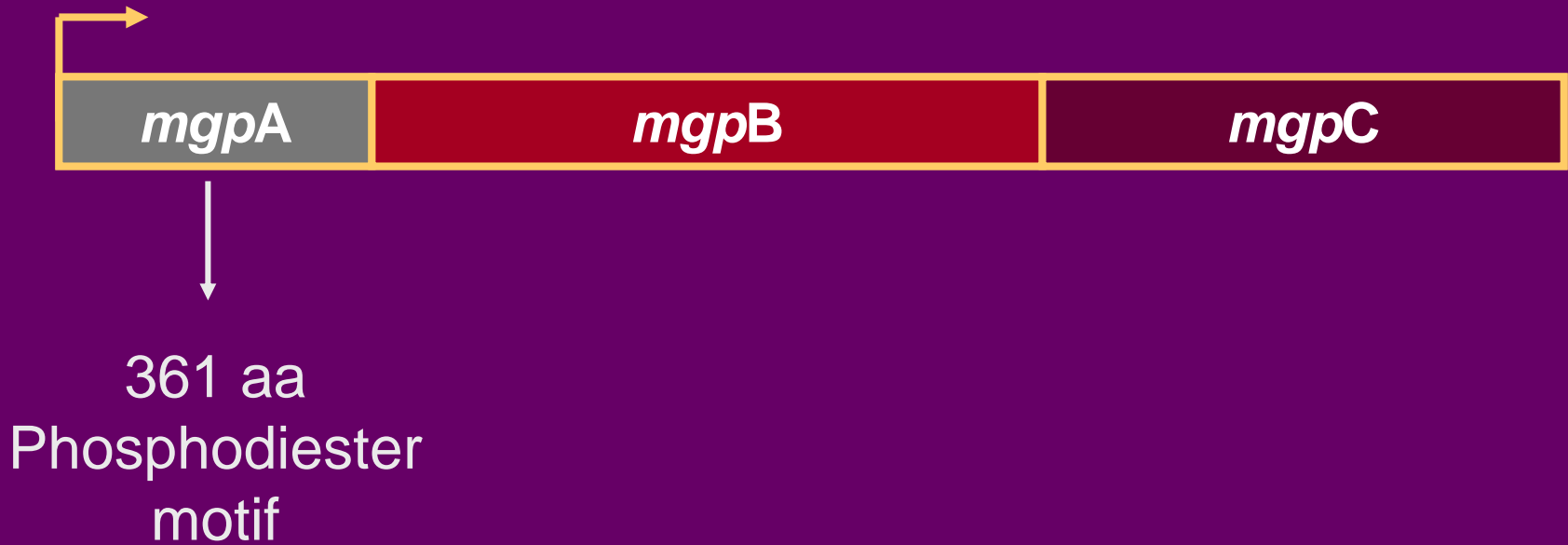
MgPa Operon





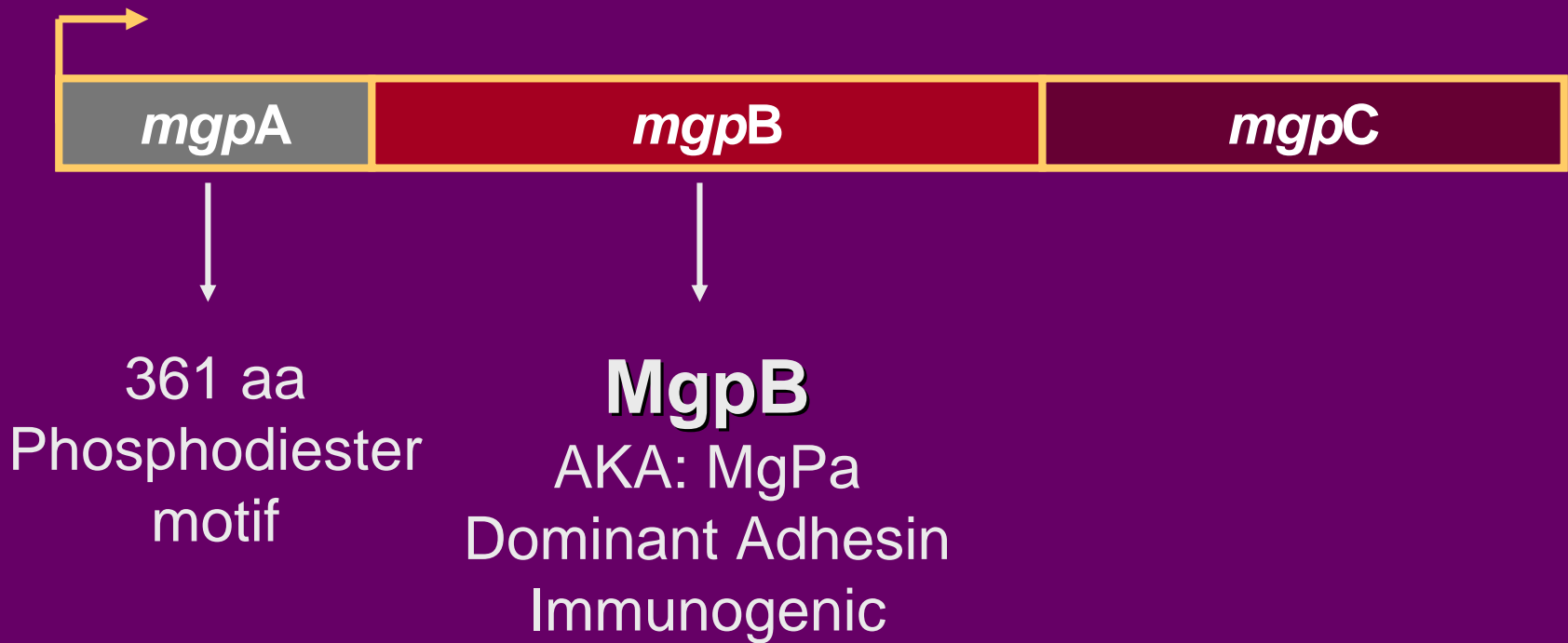
The diagram shows a DNA double helix with several regulatory elements represented by colored boxes: a red box, a dark red box, a purple box, and a red box. Below this, the title 'MgPa Operon' is displayed in large white font.

MgPa Operon



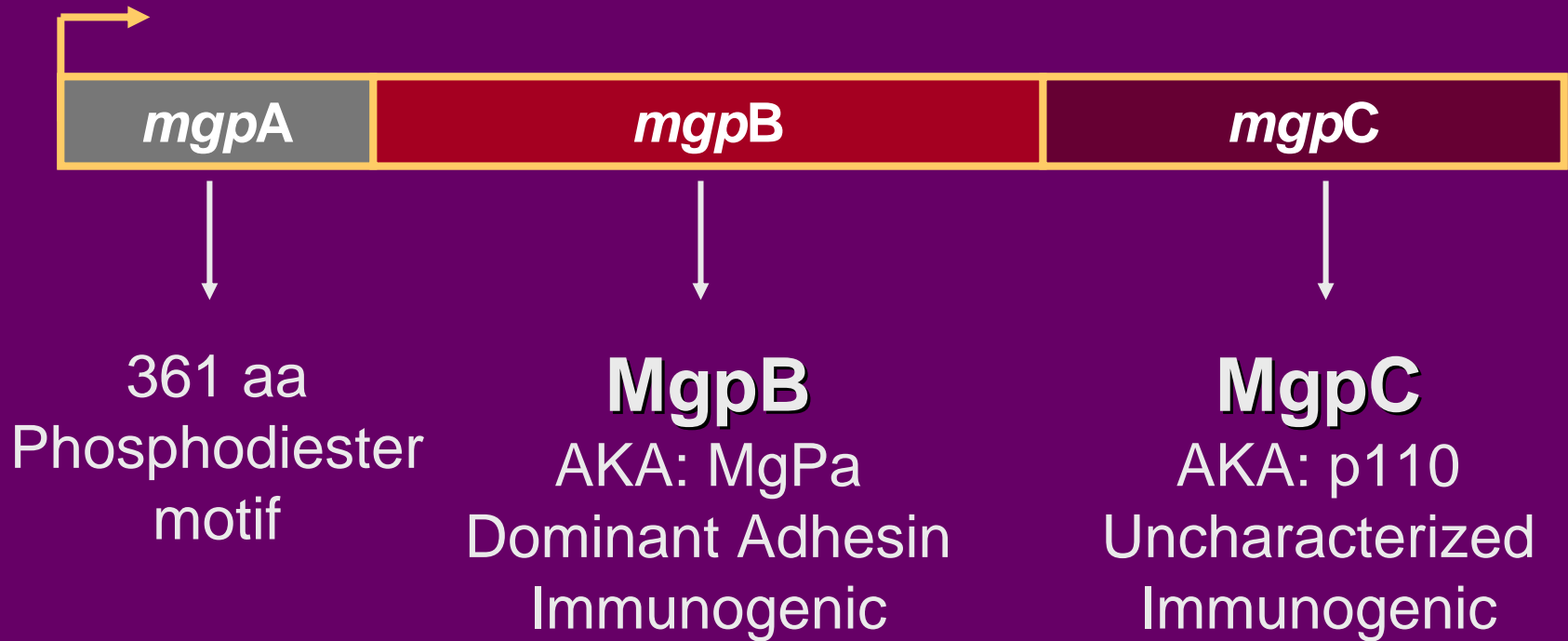


MgPa Operon





MgPa Operon





The diagram shows a DNA strand with several regulatory elements represented by colored boxes: a red box, a dark red box, a purple box, and a red box. Below this, the MgPa Operon is shown as a horizontal bar with three segments: a grey segment labeled *mgpA*, a red segment labeled *mgpB*, and a dark red segment labeled *mgpC*. An arrow on the left indicates the direction of transcription. Below each segment, arrows point to descriptive text.

MgPa Operon

mgpA

mgpB

mgpC

361 aa

Phosphodiester
motif

MgpB

AKA: MgPa

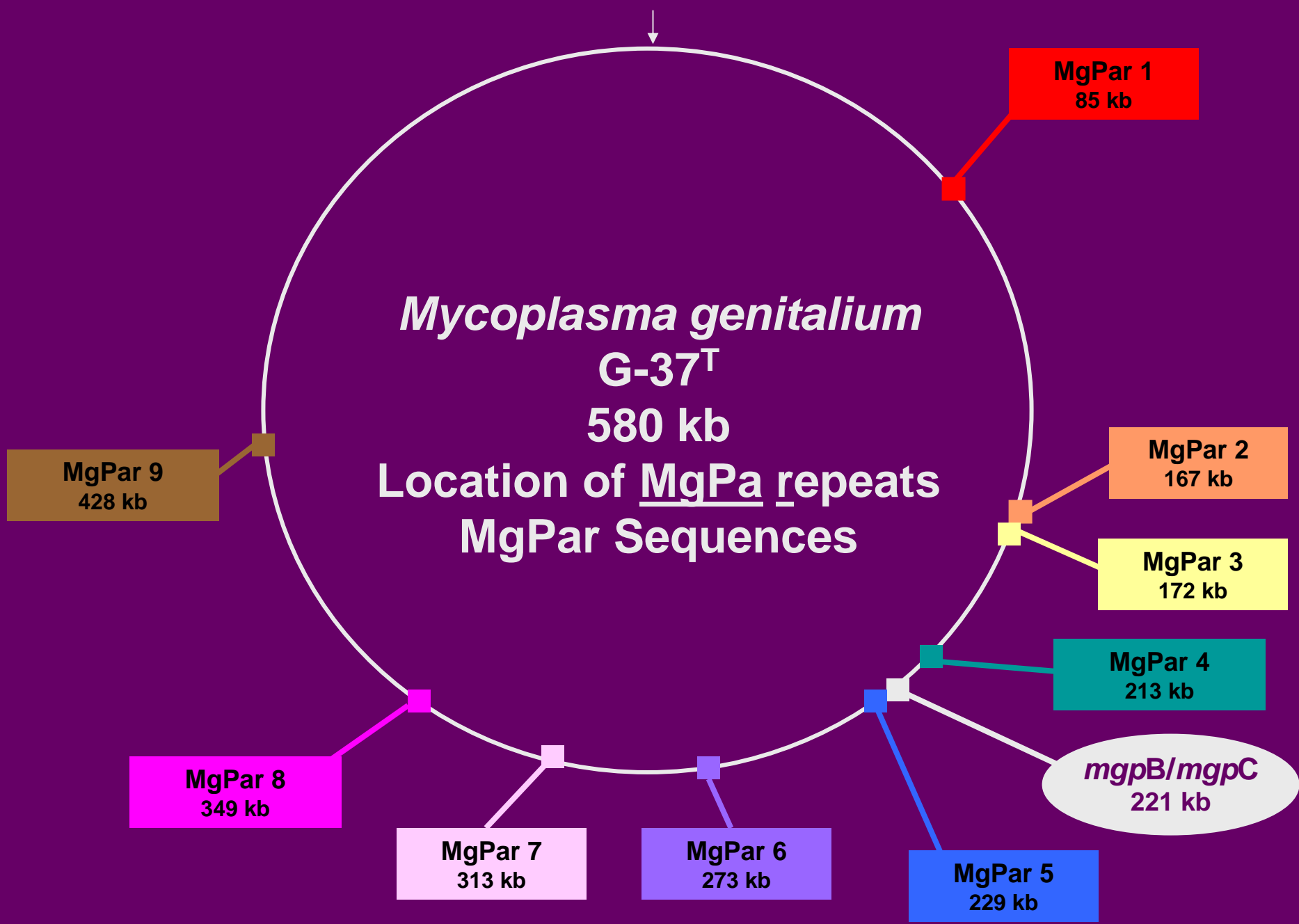
Dominant Adhesin
Immunogenic

MgpC

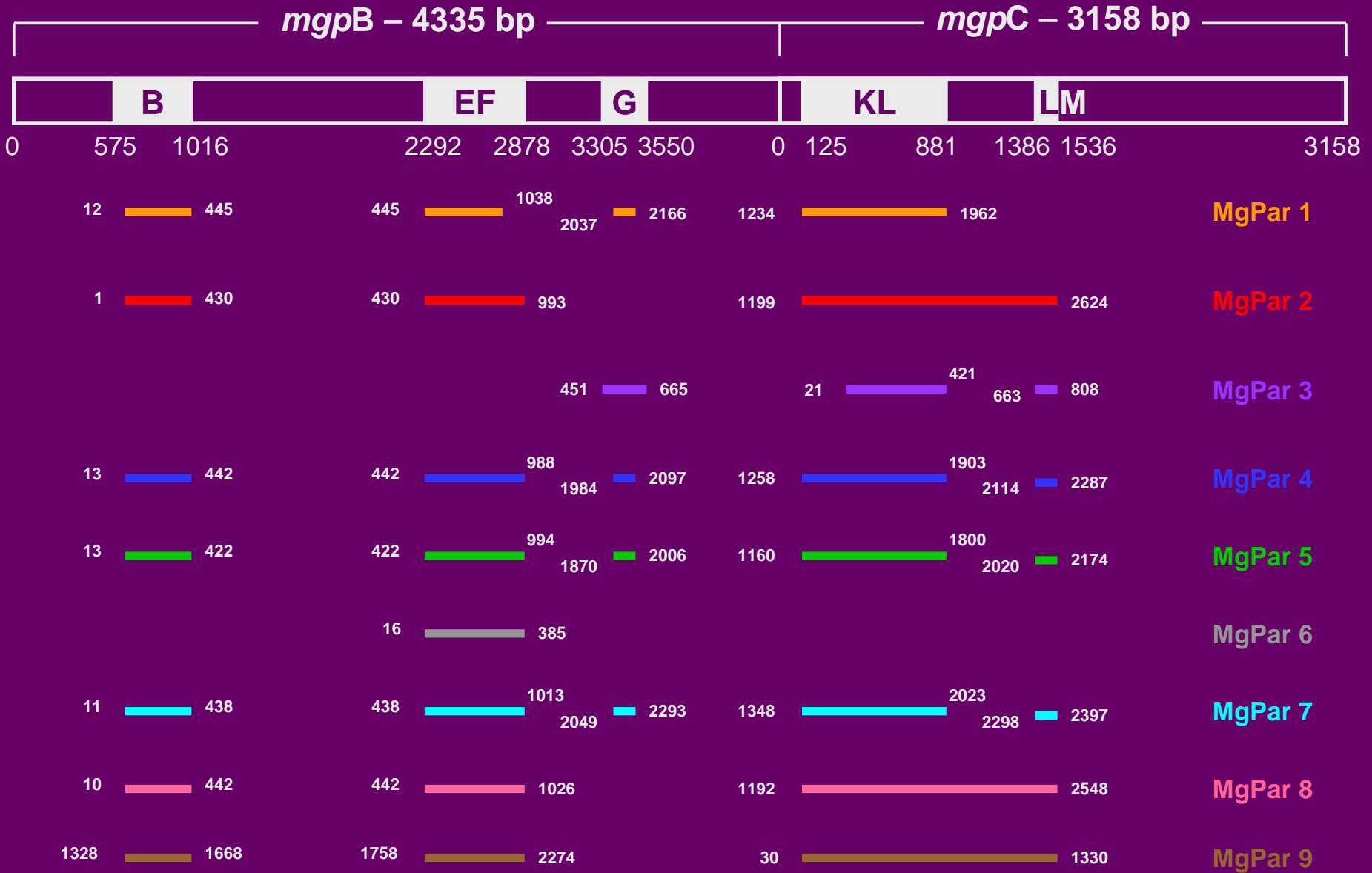
AKA: p110

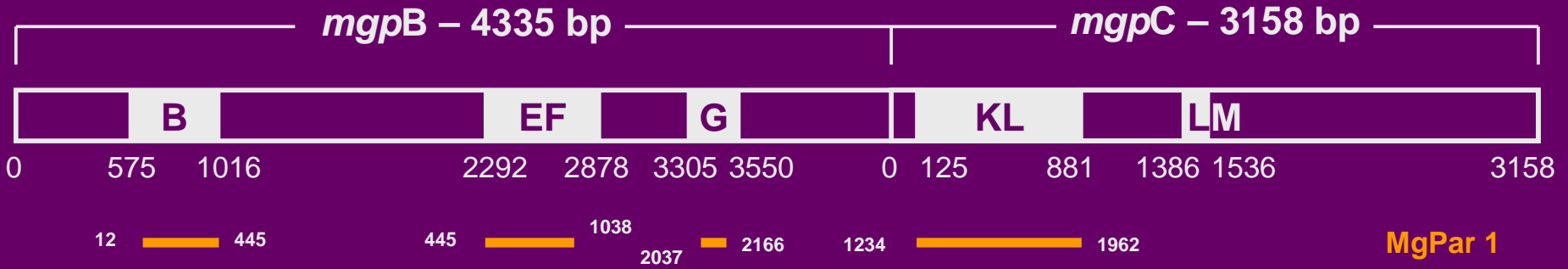
Uncharacterized
Immunogenic

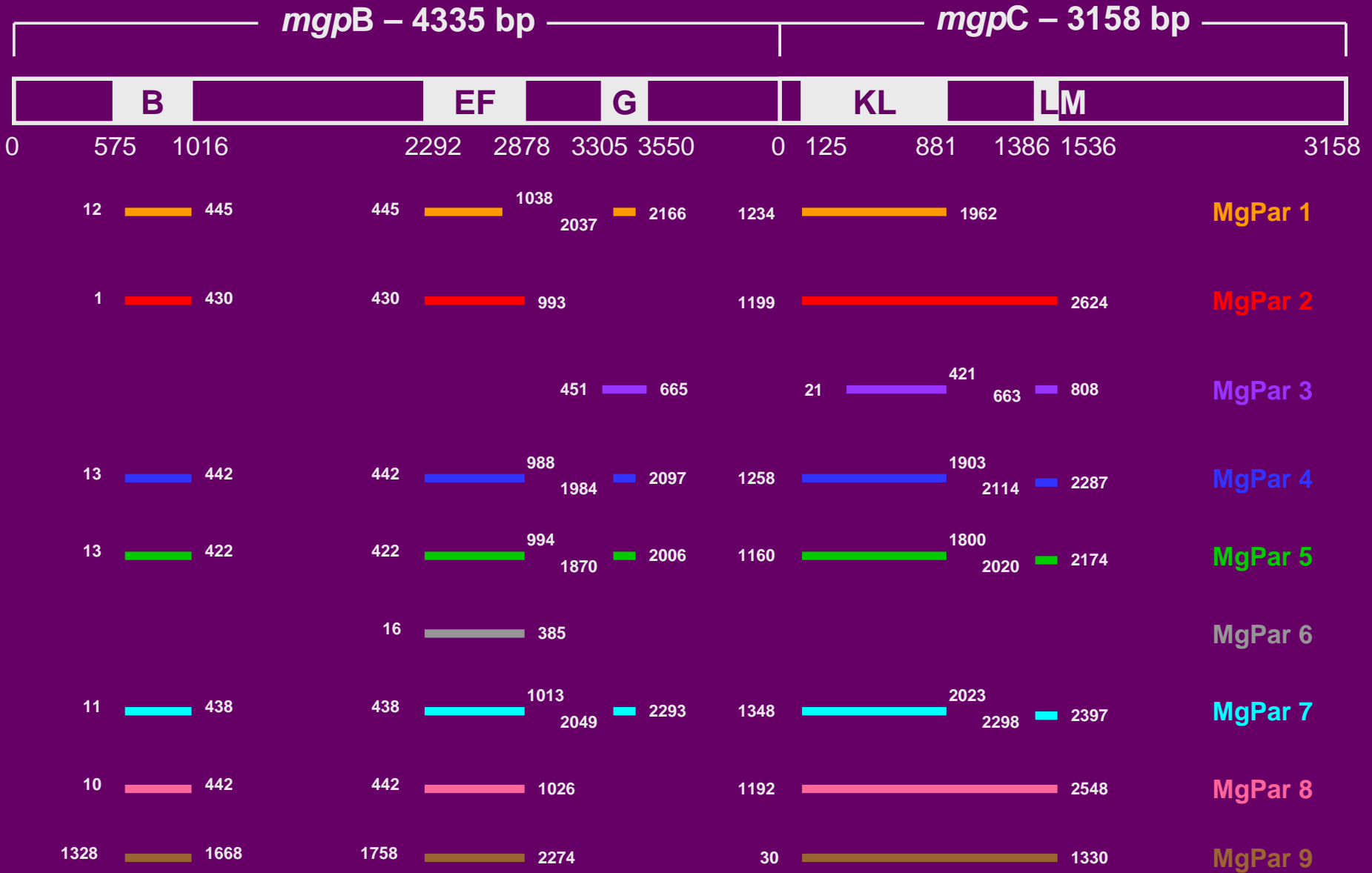
Sequences repeated
throughout genome














Hypotheses

- MgPar sequences serve as “donor” sequences
 - MgPar recombination with the *mgpB* and/or *mgpC* expression sites
 - Repeat regions = variable regions
 - Sequence heterogeneity occurs within a single strain
 - MgpB & MgpC antigenic variation contributes to *M. genitalium* survival and persistence of infection
- 



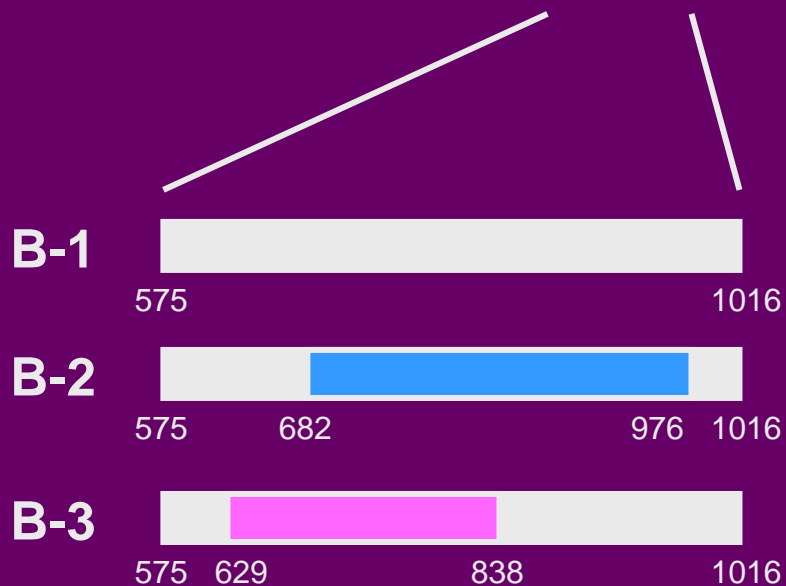
mgpB Heterogeneity in vitro

- Amplified *mgpB* repeat region from G37-S:
 - All PCRs used the same template DNA
 - Clone into *E. coli* to separate sequences

mgpB

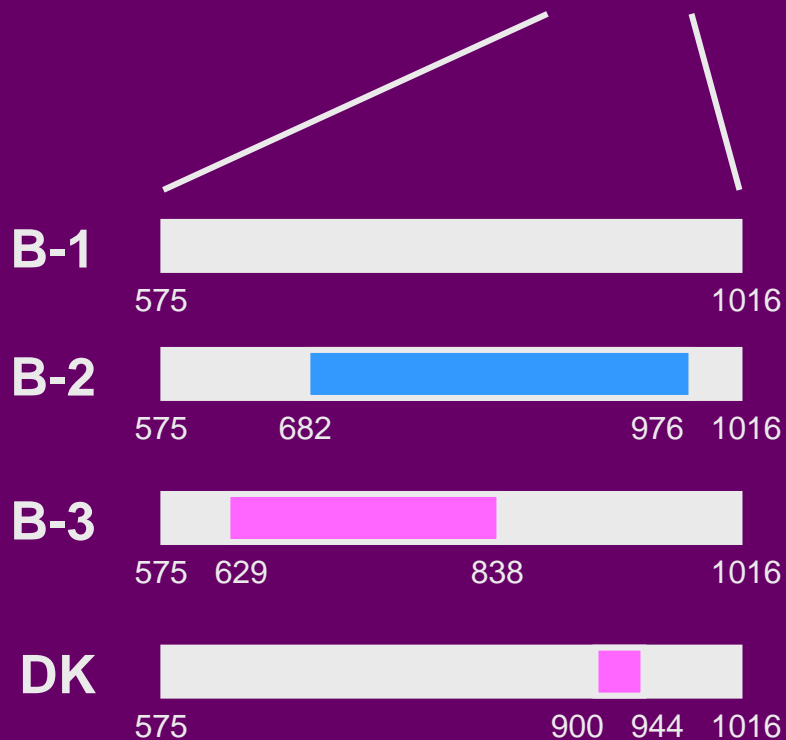


mgpB Heterogeneity in vitro



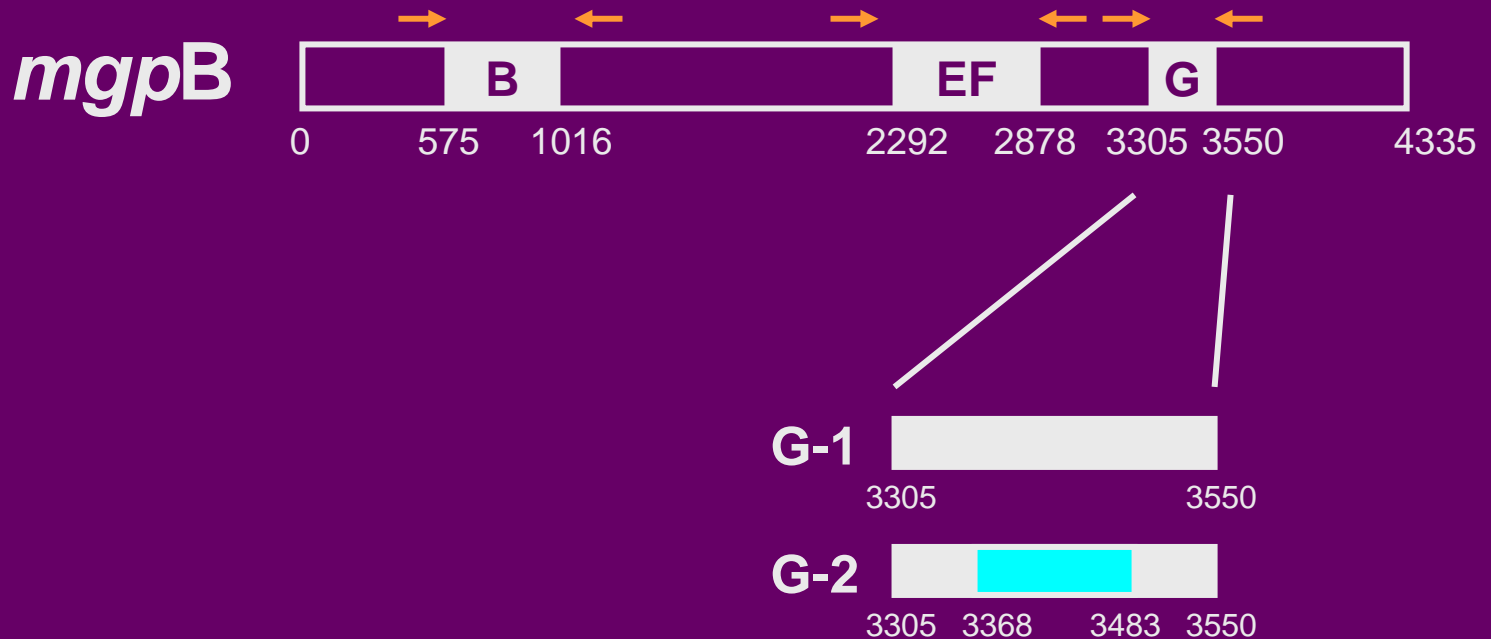
Key: MgPar 3 MgPar 7 MgPar 8 MgPar 8 or 9

mgpB Heterogeneity in vitro



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mgpB Heterogeneity in vitro



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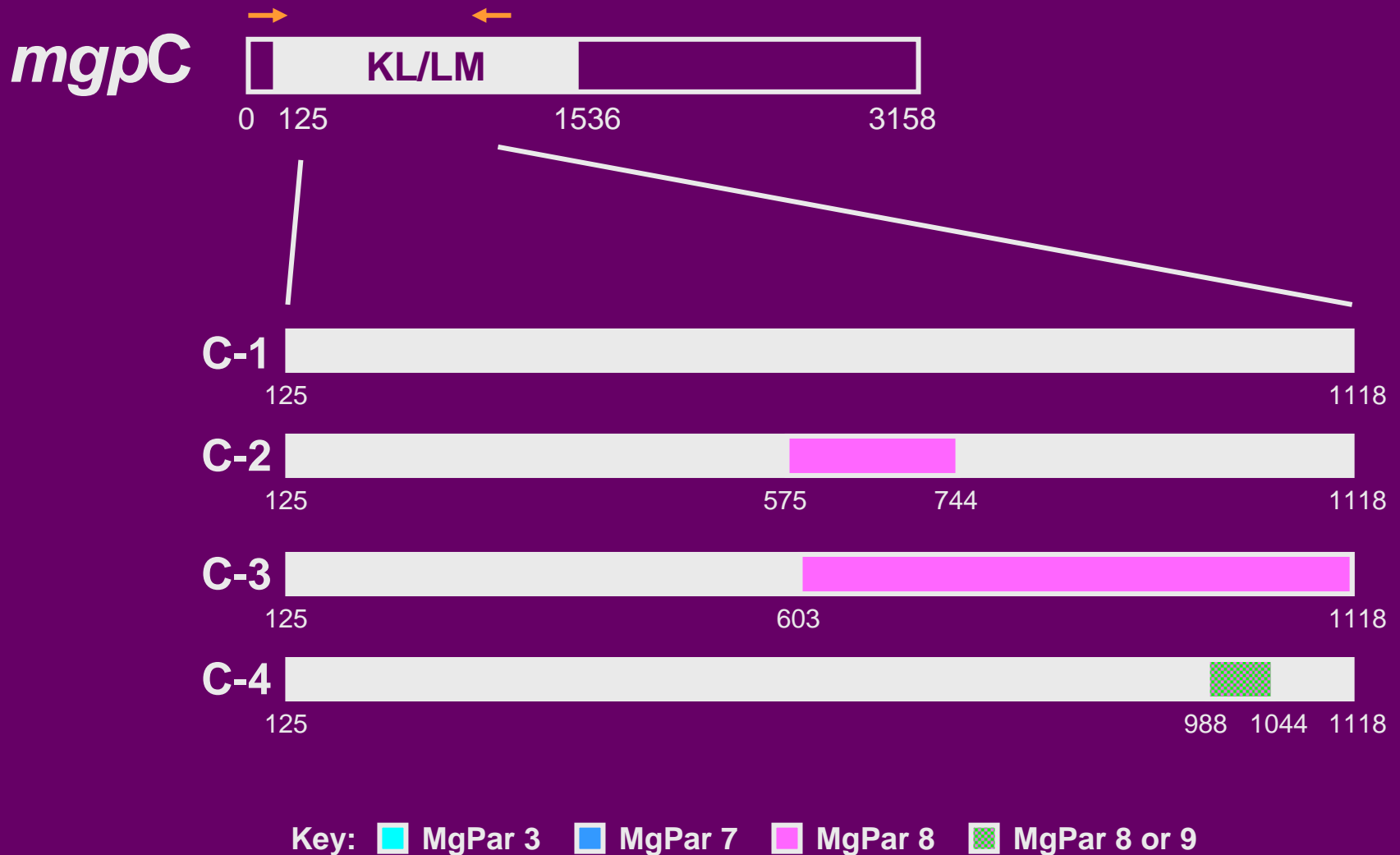


mgpC Heterogeneity in vitro

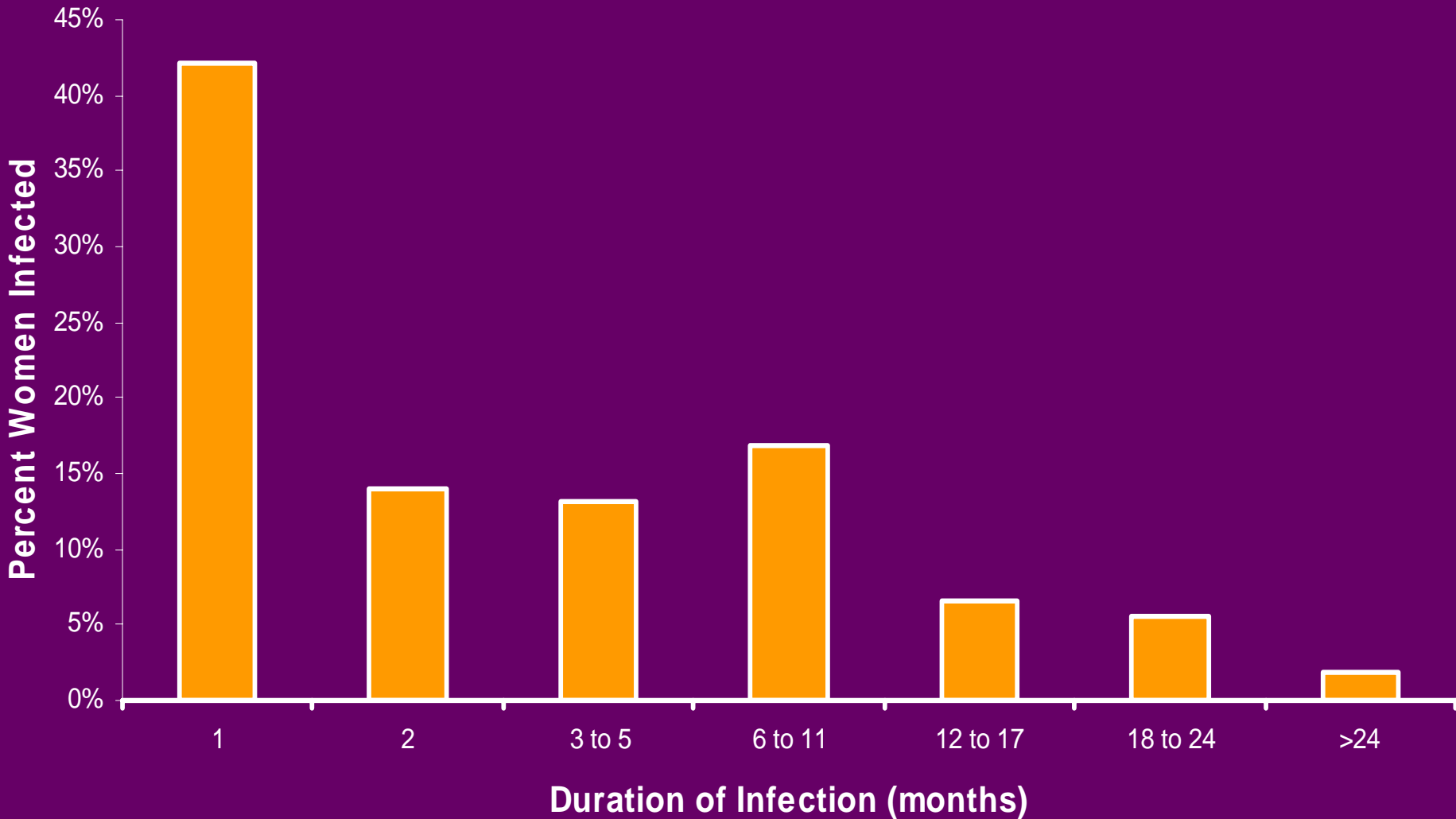
- Amplified *mgpB* repeat region from G37-S:
 - All PCRs used the same template DNA
 - Clone into *E. coli* to separate sequences



mgpC Heterogeneity in vitro



Persistent Infection in Women





Persistent *M. genitalium* Infection

- KOR Sample Set:
 - Participant #10139:
 - 15 Mg+ samples
 - 21 months

Date	Result
4-Oct-01	MG
14-Feb-02	MG
18-Jun-02	MG
5-Mar-02	MG
12-Apr-02	MG
25-Apr-02	MG
8-May-02	MG
10-Jun-02	MG
22-Jul-02	MG
21-Aug-02	MG
9-Aug-02	MG
17-Sep-02	MG
9-Oct-02	MG
13-Dec-02	MG
9-Jul-03	MG



Persistent *M. genitalium* Infection

- **KOR Sample Set:**
 - **Participant #10139:**
 - 15 Mg+ samples
 - 21 months
- ***mgpB* Sequence Analysis:**
 - Four time points
 - Strain typing
 - Repeat region B

Date	Result
4-Oct-01	MG
14-Feb-02	MG
18-Jun-02	MG
5-Mar-02	MG
12-Apr-02	MG
25-Apr-02	MG
8-May-02	MG
10-Jun-02	MG
22-Jul-02	MG
21-Aug-02	MG
9-Aug-02	MG
17-Sep-02	MG
9-Oct-02	MG
13-Dec-02	MG
9-Jul-03	MG

Collection
Date

mgpB Repeat Region B
(575-1016 bp)

identified
analyzed

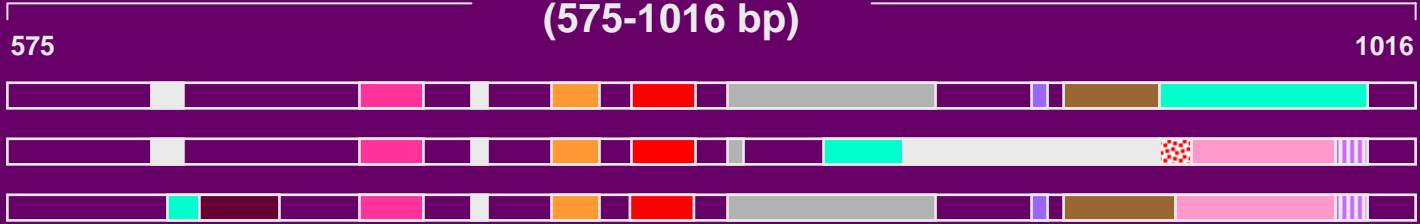
575

1016

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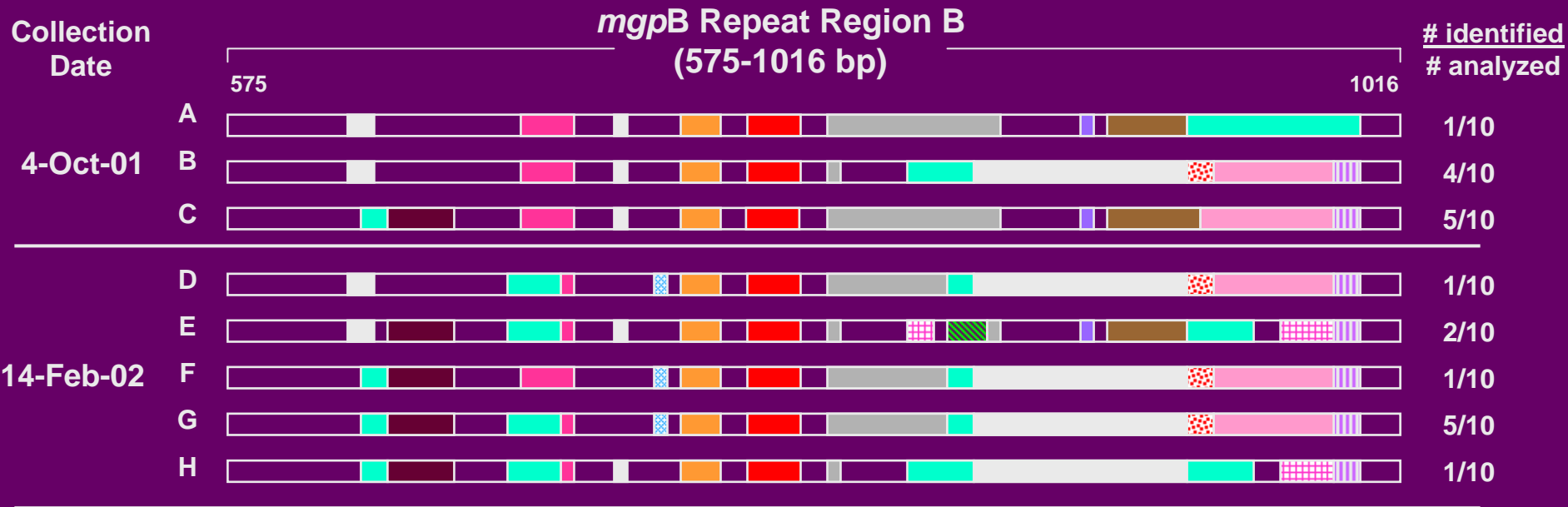


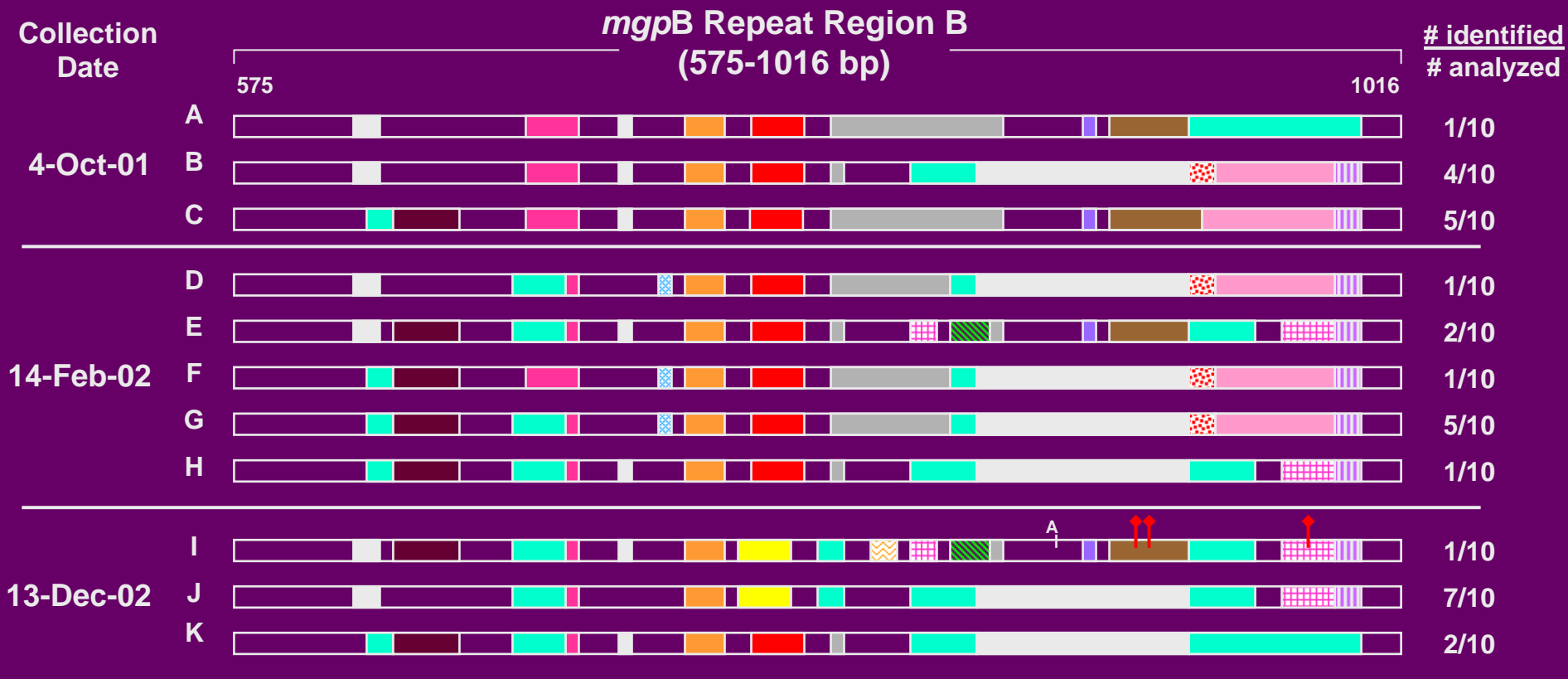
4-Oct-01

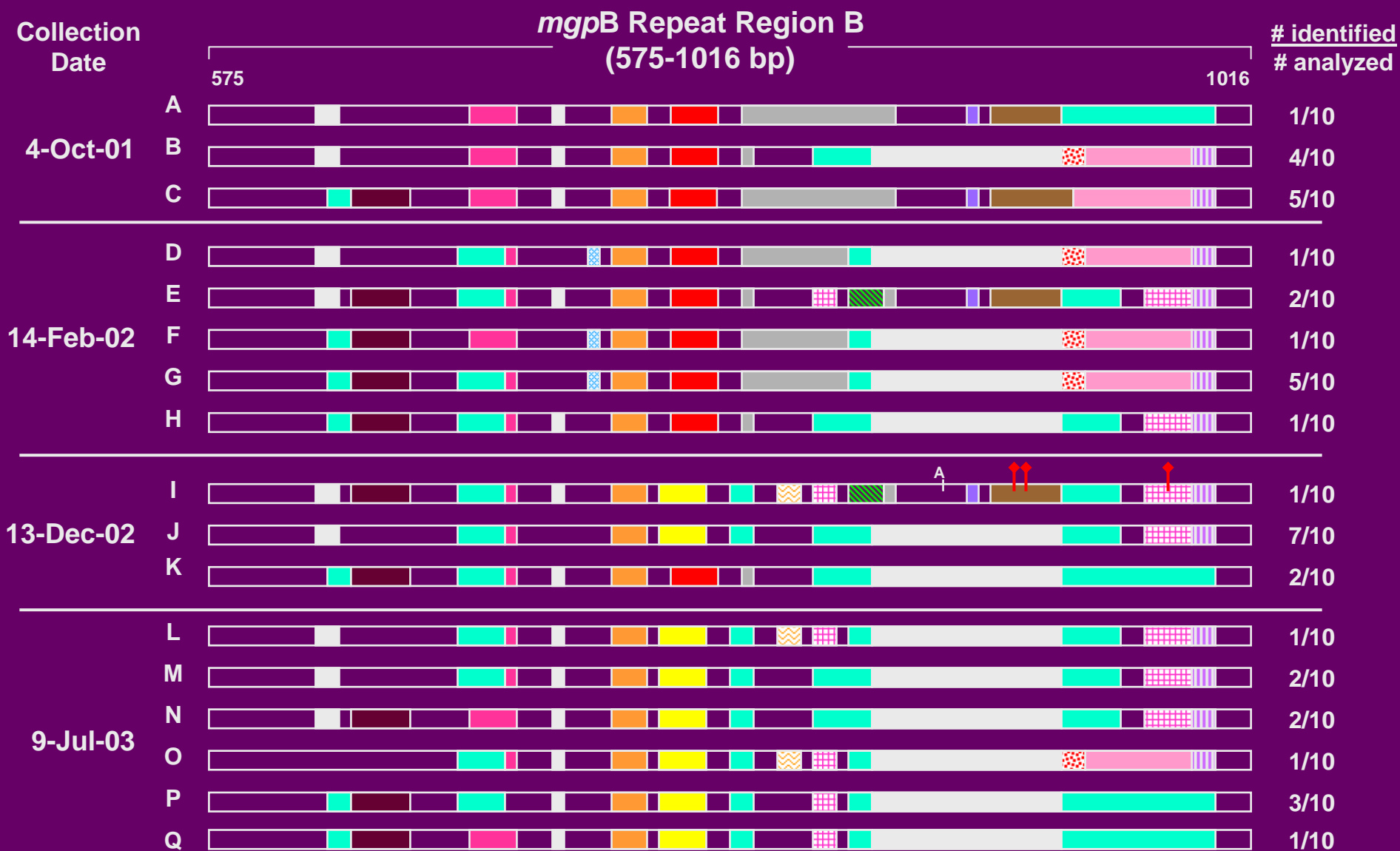
1/10

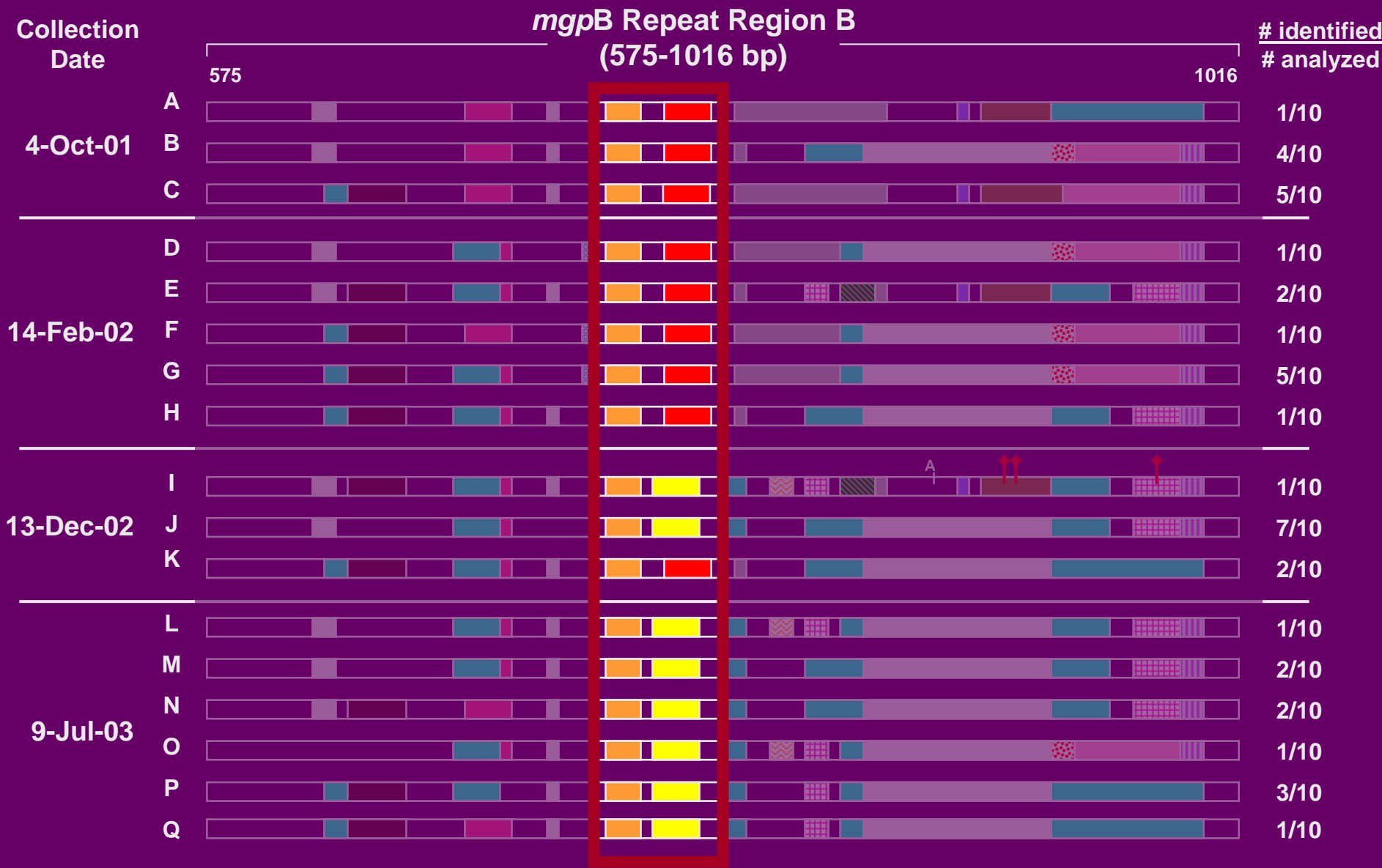
4/10

5/10









mgpB Heterogeneity in vivo

G-37 ^T	767	TCAGTGAAAC	AACTGCTTCA	TCCATGAGTA	GTGGTATGGC	TACATCCACT	816
10/01	A	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	B	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	C	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
2/02	D	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	E	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	F	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	G	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
	H	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
12/02	I	TC-----	-GAGGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	J	TC-----	-GAAGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	K	TCAGTTTGGC	AGATGGTGGG	TCAATGAGTA	GTGGTGGGGC	TACATCCACT	
7/03	L	TC-----	-GAAGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	M	TC-----	-GAAGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	N	TC-----	-GAAGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	O	TC-----	-GAAGAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
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	P	TC-----	-GAAGAAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	
	Q	TC-----	-GAAGAAATCA	TCCATGAGT-	-----GGGGC	TACATCCACT	



Conclusions

- Repetition of sequences within *mgpB* & *mgpC*
- Multiple sequences identified in G37-S
 - Apparent insertion of MgPar “donor” sequences
- Heterogeneity in women persistently infected w/ a single strain
 - 17 *mgpB*-B sequences
 - 8 *mgpC*-KL sequences





Future Directions

- The reciprocal nature of recombination
 - Are alternative *mgpB/mgpC* sequences associated with changes in donor MgPar sequences?
- Define the structure of MgpB & MgpC
 - What are the structural, functional, and/or antigenic epitopes?
- Heterogeneity of *mgpB/mgpC* contributes to long-term *M. genitalium* infection
 - What is the biologic activity of MgpB & MgpC antibodies?



Acknowledgements

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