



# Spectacular Shigellas: how to tame a host

Dasha Abuzova, Dima Zvezdin, Inna Shapovalenko, Liza Kaminskaya

Natalia Dranenko, Aygul Minnegalieva, Yulia Yakovleva, Olga Bochkareva



### Why Shigella have T3SS effectors on the chromosome?



- 1. Are there common or unique types of *ipaH* genes for the plasmid and the chromosome?
- 2. Is ipaH regulation different on the chromosome and the plasmid? What does the conservative upstream before *ipaH* encode?
- 3. Are there specific sites for the integration of different ipaH into the chromosome and into the plasmid?
- 4. What mobile elements bring *ipaH* to the chromosome?





## Clusterization of *ipaH* sequences does not match with the tree topology

#### All types of ligases present in all species of Shigella

Cluster number 0: 223 Shigella flexneri: 95 Shigella boydii: 30 Shigella sonnei: 58 Shigella dysenteriae: 40

Cluster number 1: 145 Shigella flexneri: 51 Shigella boydii: 21 Shigella sonnei: 35 Shigella dysenteriae: 39 Cluster number 2: 54 Cluster number 4: 57 Shigella flexneri: 22 Shigella boydii: 12 Shigella sonnei: 7 Shigella dysenteriae: 13

Cluster number 3: 113 Shigella flexneri: 38 Shigella boydii: 20 Shigella sonnei: 35 Shigella dysenteriae: 22

Shigella flexneri: 24 Shigella bovdii: 12 Shigella sonnei: 7 Shigella dysenteriae: 14 Cluster number 5: 132

Shigella flexneri: 42 Shigella boydii: 22 Shigella sonnei: 35 Shigella dysenteriae: 33

Cluster number 6: 94 Shigella flexneri: 54 Shigella boydii: 21 Shigella sonnei: 7 Shigella dysenteriae: 12

Cluster number 7: 168 Shigella flexneri: 83 Shigella boydii: 25 Shigella sonnei: 35 Shigella dysenteriae: 25

Cluster number 8: 67 Shigella flexneri: 27 Shigella boydii: 14 Shigella sonnei: 7 Shigella dysenteriae: 19

#### Ligases vary by leucine-rich domain, no transfers between plasmids and chromosomes were found.





*ipaH* pseudogenes are presented both in plasmids and chromosomes.

## *ipaH*, why do you need 400 conservative letters?

**Conserved fragment: 1...1184** organism = '*Shigella boydii* CDC3083-94'

gene: 406 ... >1184 /product = 'ubiquitin ligase'

Sites of transcription One more factors?? protein?!

### location of MxiE box site in reference *Shigella* strain

		MyiE hoy	-	τάτδ	ΔΤ			Δ	TG
VP genes				INIA				1	U O
ospB	ttttt	GTtcCGTTTTTTTAAAt	catagtatacaaagaagc	TtgcAT	actatat	-	57	F.	ATG
ospC1	agaaa	GTATCGTTTTTTALAG	taaaattcattgctgttcaa	TAaAAT	tgatat <u>A</u>	-	229	F.	ATG
ospE1	aaaaa	GTATCGTTTTTTACAG	taaacttcatttageegae.	TATAAT	gtaaaaa	-	38	F.	ATG
ospE2	aaaaa	GTATCGTTTTTTTACAG	taaacttcatttagccgac.	TATAAT	gtaaaaa	-	36	F.	ATG
ospF	ataat	GTATCGTTTTaTaAaAG	atgataaacaatcaatataa	gATAAT	atatcta	-	20	F.	ATG
virA	gaaat	GTATCGTTTTCTTAAAG	agaagaataacattccattt	TATTAT	gttccc <u>A</u>	-	56	F.	ATG
ipaH4	ctcta	GGATtGTTTTTTAAAG	actttctcgttttatttgc.	atTAAT	agaccaa	-	25	⊢	ATG
ipaH9	aaaca	GTATCGTTTTTTACAG	ccaattttgtttatccttat	TATAAT	aaaaaa <u>G</u>	-	96	F	ATG
ipaH7	aatgt	GTATCGTTTTTTACAG	taatttttaatttgttattc	TATAAT	aggaata	-	271	ŀ	ATG
Chromosomal genes									
ipaHa	ttaca	GgcgCGTTTTTTAaAG	aateeteacteeatttgeac	gAatAT	attaata	-	25	⊢	ATG
ipaHb	agaat	GTACtGTTTTTTTAAAG	aaaaaaacagtacattgaga	TATGAT	tttgatc	-	357	F.	ATĠ
ipaHc	aaaca	GTATCGTTTTTTTACAG	ccaattttgtttgtccttt.	TATAAT	aaaaaaa	-	88	Ŀ.	ATG
ipaHd	aaaaa	GTATCGTTTTTTTACAL	taaaccgatccagtttagga	TAgtgT	aaagcag	-	163	⊢	ATG
ipaHe	aaata	GTATCGTTTTTTACAG	ttaaatcaacatcactteet	TAaAAT	gaaaaca	-	85	ŀ	ATG
MxiE box		GTATCGTTTTTTANAG	20 n	TATAAT				1	

TL Gall et al., Microbiology (2005)







# Chromosomal ligases have specific locus of genomic island integration



Distribution of neighbour blocks across the Shigella genomes



Block names

Distribution of neighbour blocks across the Shigella genomes



Distribution of neighbour blocks across the Shigella genomes



## Conclusions

- I. Chromosomal and plasmid ligases are different; we have not identified significant cases of transfer.
- II. Ligases of all types, including chromosomal and plasmid, are regulated by MxiE
- III. Only in one cluster (# 3) we did not identify a single pseudogene, perhaps this indicates the importance of this type of ligases (*maybe we could not find them?*)
- IV. In the upstreams of two types of chromosomal ligases, we found ncRNA overlapping the site of the transcription factor. In the upstream of one of the plasmid types, we also found ncRNA, but shorter (*perhaps ncRNA regulates ipaH?*)
- V. We found that ligases surrounded by many phage genes, but their strong diversity does not allow us to unambiguously reconstruct the genomic island.
- VI. The same bacterial genes of *E. coli* was found with ligases of the same type, that indicates the specificity of the genomic island insertion loci.

