

# Comparison of three commercial platforms for Hepatitis C Virus Genotyping and NS5B sequencing. GEHEP-007 study.

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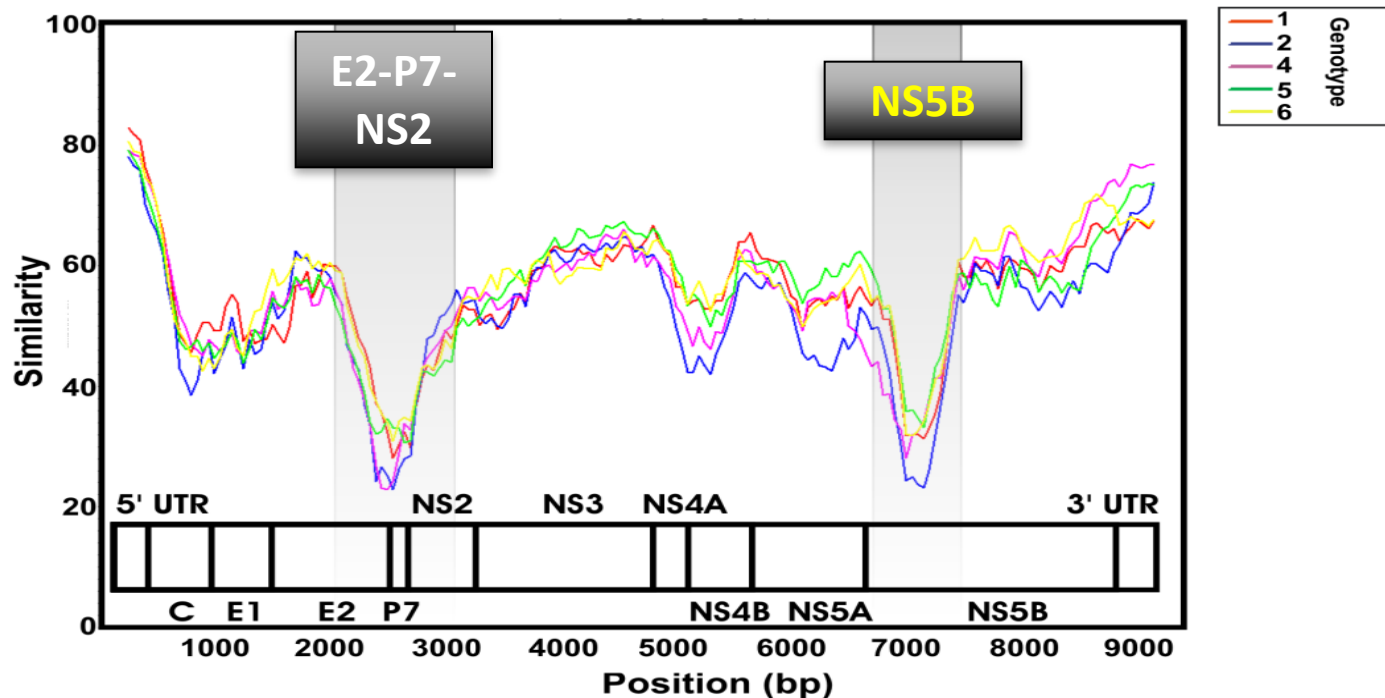
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# Background

- Genotyping plays a critical role for HCV antiviral treatment
- “The HCV genotype and genotype 1 subtype (1a/1b) must be assessed prior to treatment initiation and will determine the choice of therapy (A1)”

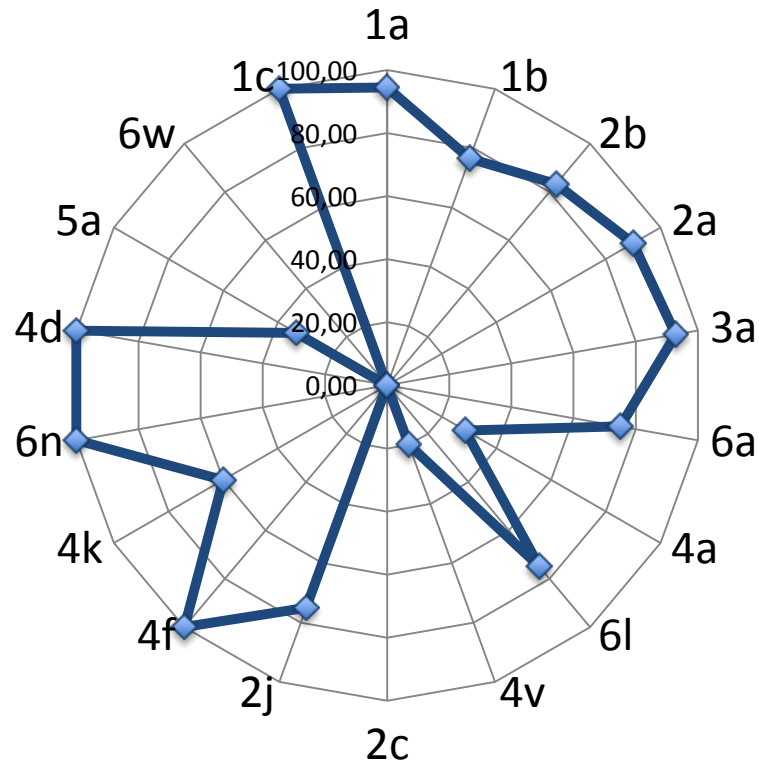
# Background

- HCV genotyping can be done interrogating different genomic regions



# Background

- 5'-UTR may not be the best region for HCV genotyping

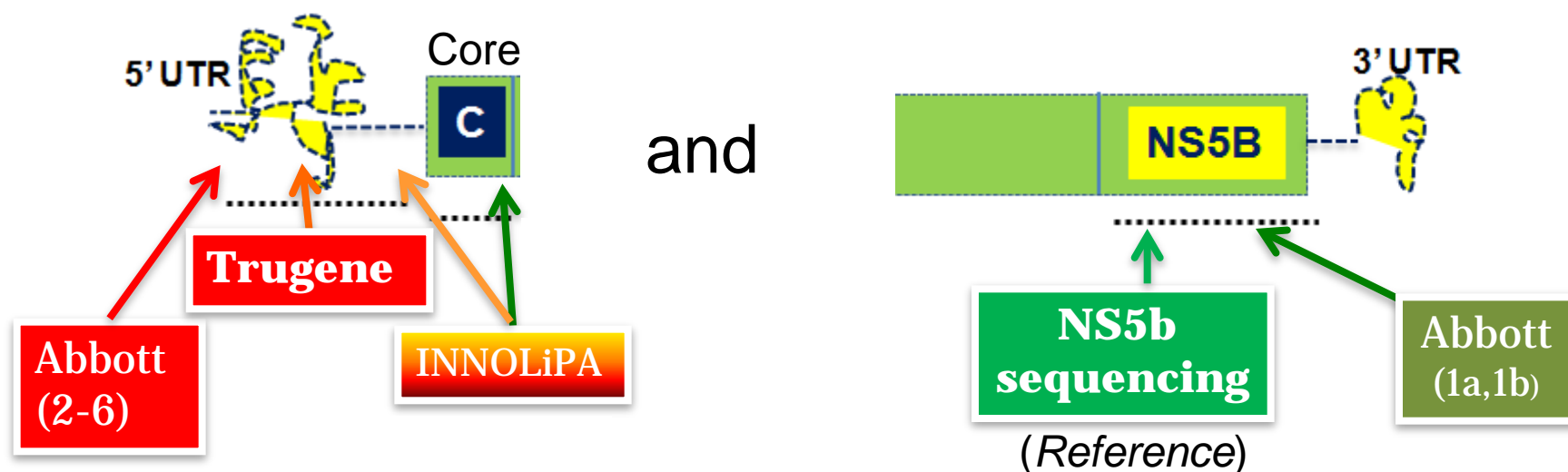


GenBank: 5' -UTR & NS5B correlation (n=303)

# GEHEP-007 Study

In the setting of the Spanish Group for Viral Hepatitis (GEHEP), we aimed to evaluate the correct assignment of HCV genotypes by three commercial methods, compared to **NS5B sequencing**:

- a) **Trugene HCV** genotyping kit (Siemens),
- b) **VERSANT HCV** Genotype 2.0 assay (Siemens),
- c) **Real Time HCV** genotype II (Abbott),



# Methods

## Proficiency Panel 10 samples



Geno/subtype	Log VL
2a	5,65
4d	5,81
1b	6,21
4a	5,33
5a	5,43
3a	5,69
1a_clade1	6,62
1b	6,59
1a_interclade	5,56
1a_clade 2	6,45

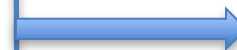
## 135 samples: Trugene HCV



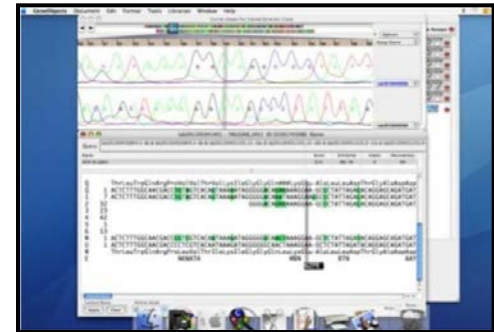
## 85 samples: Real Time HCV



## 74 samples: Versant HCV



## NS5B DNA sequencing



<http://hcv.bioinf.mpi-inf.mpg.de>

[http://hcv.lanl.gov/content/sequence/BASIC\\_BLAST/basic\\_blast.html](http://hcv.lanl.gov/content/sequence/BASIC_BLAST/basic_blast.html)

# Genotype distribution

Genotype	Subtype	Trugene	RealTime	Versant	LIPA	Total	
1	1a	33	48	34		115	209
	1b	47	17	17		81	(71%)
	No	6	-	6		12	(69%)
2	2a	1	-	1*		2	
	2c	1	-	1*		2	
	No	-	6	-		6	10 (3%)
3	3a	23	-	8		31	
	3d	1	-	-		1	44 (15%)
	No	2	9	1		12	(20%)
4	4a	8	-	1		9	
	4c	12	-	1		13	29 (10%)
	No	-	3	4		7	(8%)
5	5a	1	-	-		1	
	No	-	2	-		2	3
<b>Total</b>		135	85	74		294	

# Major Discordance

(Differences in genotype & subtype 1a & 1b)

	Trugene	Versant LIPA	RealTime
<b>1b→1a</b>	<b>13</b>	<b>8</b>	<b>-</b>
<b>1a→1b</b>	<b>5</b>	<b>-</b>	<b>-</b>
<b>1b→3a</b>	<b>1</b>	<b>-</b>	<b>-</b>
<b>3a→1b</b>	<b>1</b>	<b>-</b>	<b>-</b>
<b>4c→1b</b>	<b>1</b>	<b>-</b>	<b>-</b>
<b>TOTAL</b>	<b>21 (16%)</b>	<b>8 (11%)</b>	<b>0</b>



# Minor Discordance

(Differences in subtype assignment)

	Trugene	Versant LIPA	RealTime
<b>1→1a</b>	2	1	-
<b>1→1b</b>	4	1	-
<b>2*→2a/b/c</b>	2	-	6
<b>3*→3a</b>	3	1	9
<b>4*→4a/c/d</b>	13	3	3
<b>5→5a</b>	-	-	2
<b>TOTAL</b>	<b>24 (18%)</b>	<b>6 (8%)</b>	<b>20 (24%)</b>

# Summary and Conclusion

When **NS5B sequencing** is used as reference:

- ✓ **Trugene** failed to correctly assign HCV genotype in up to a third of cases, with more than 15% of errors having a high impact on clinical practice.
- ✓ With the **Versant LIPA** HCV Genotype 2.0 assay, up to 11% of the samples showed discordant results with a great impact on clinical practice.
- ✓ **Abbott Real Time** HCV genotype II assay was not able to discriminate subtype for genotypes 2, 3, 4 and 5.

DNA sequencing of **high discriminating regions**, such as NS5b, should be considered **prior to DAA** treatment

# Acknowledgments



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