

# **Strong Inhibition of Wild-Type and Integrase Inhibitor (INI)-Resistant HIV Integrase (IN) Strand Transfer Reaction by the Novel INI S/GSK1349572**

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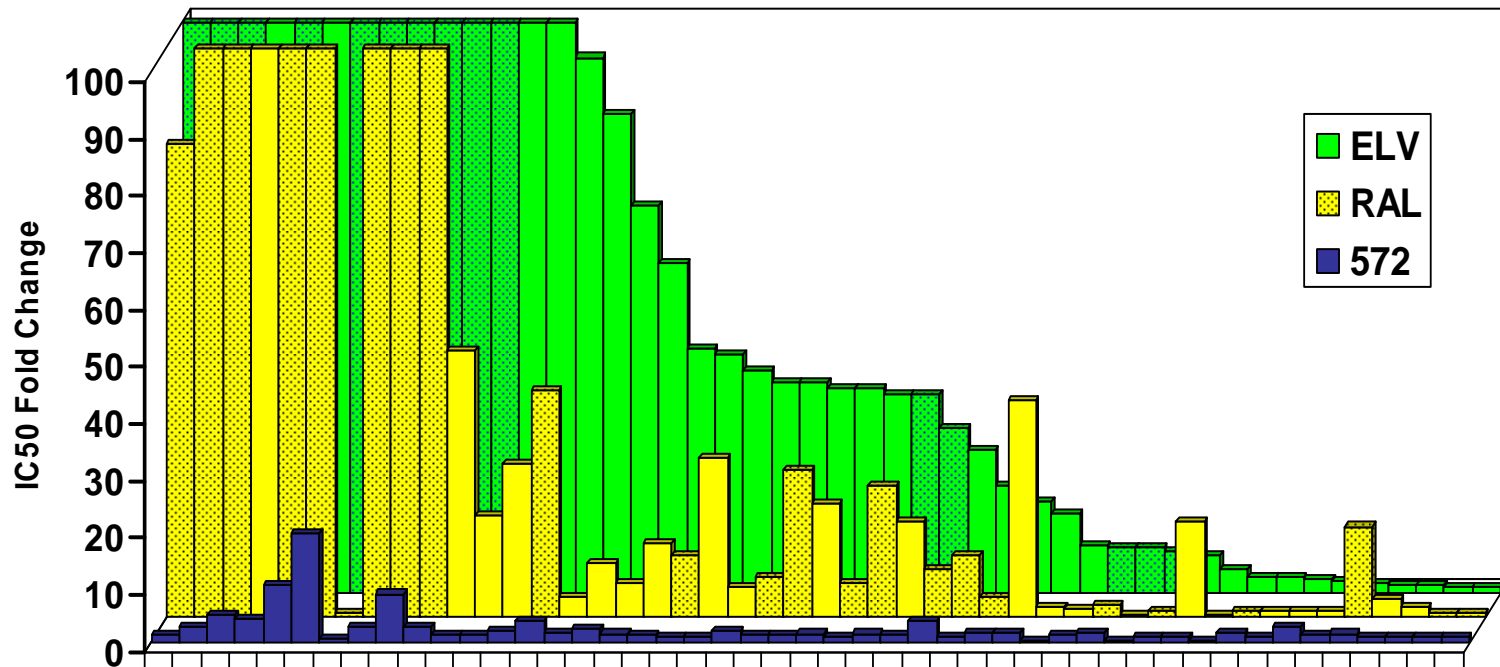
# **S/GSK1349572: Attributes of a Next Generation Integrase Inhibitor (INI)**

- **Once daily, unboosted INI in clinical development, currently in Ph2b studies**
- **Low PK variability and predictable exposure-response relationship with a low mg dose<sup>1,2</sup>**
- **Very strong antiviral activity in a Ph2a study**
- **Markedly different in vitro resistance profile with potential for higher genetic barrier to resistance<sup>3,4,5</sup>**

1. Min S, et al. IAS 2009, Cape Town, abstract WEPEA099.
2. Song I, et al. IAS 2009, Cape Town, abstract WEPEB250.
3. Sato A, et al. IAS 2009, Cape Town, abstract WEPEA097.
4. Underwood M, et al. IAS 2009, Cape Town, abstract WEPEA098.
5. Seki T, et al. CROI 2010, Poster abstract 555.



# Most Raltegravir- and Elvitegravir-Resistant Mutants are Susceptible to S/GSK1349572



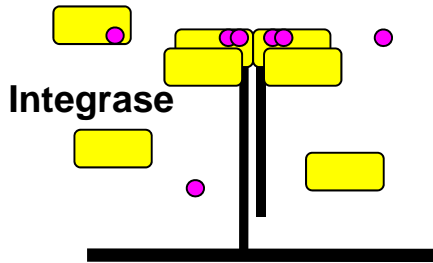
**Integrase Mutants**  
(shaded mutants observed in clinic with RAL or ELV failure)

Johns B, et al. The Discovery of S/GSK1349572: A Once Daily Next Generation Integrase Inhibitor with a Superior Resistance Profile. 17th CROI, San Francisco. Oral Session #55.

Seki, T, et al. S/GSK1349572 is a potent next generation HIV integrase inhibitor and demonstrates a superior resistance profile substantiated with 60 integrase mutant molecular clones. 17th CROI, San Francisco. Poster #555.

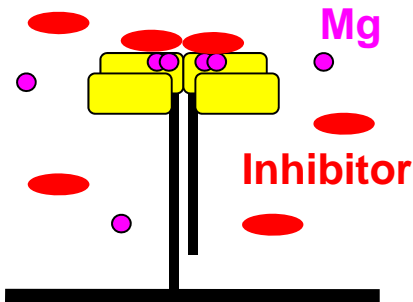
# Schematic of Two Assay Methods

## 1. IN-DNA complex formation

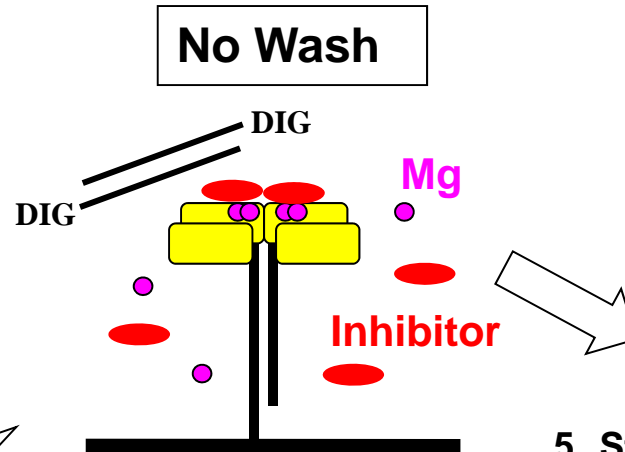


## 2. Wash free IN

## 3. Add inhibitor

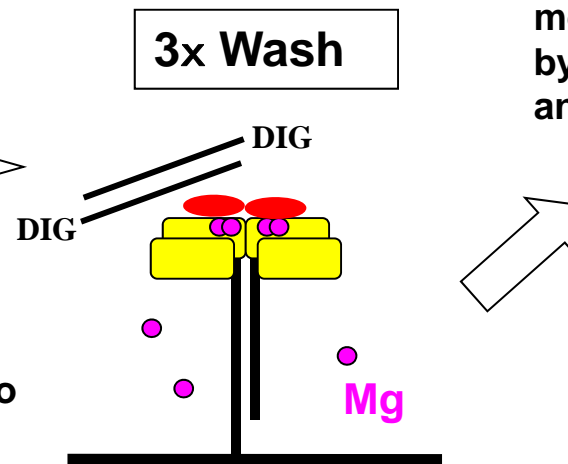


## 4. Add target DNA to strand transfer



5. Strand transfer products were measured by ELISA with anti-DIG antibody

## 4'. Wash IN-DNA-inhibitor ternary complex three times and add target DNA to strand transfer



# IC50 Values of Three INIs Under Less Stringent (No Wash) and Stringent (3xWash) Conditions

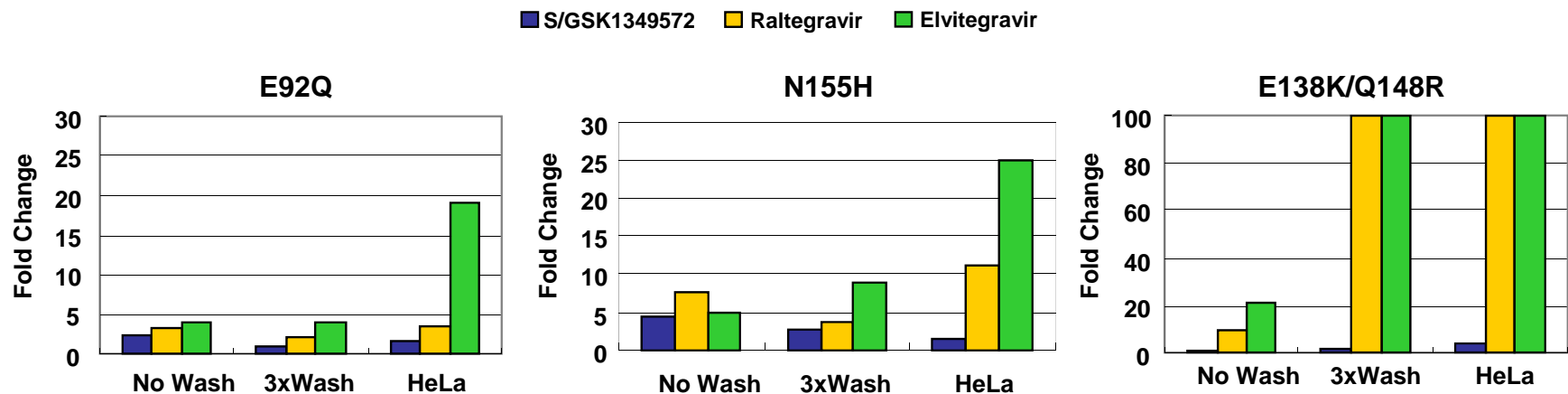
IC50 (nM) values of S/GSK1349572, RAL and ELV in No Wash assay and 3xWash assay

Enzyme	WT		E92Q		G140S		N155H		E138K/Q148R		G140S/Q148H		E92Q/N155H	
	No Wash	3xWash	No Wash	3xWash	No Wash	3xWash	No Wash	3xWash	No Wash	3xWash	No Wash	3xWash	No Wash	3xWash
S/GSK1349572	4.2	4.8	10	7	9.8	9.6	17	8	6.2	7.5	9.9	20	9.3	656
Raltegravir	6.4	7.5	20	11	11	15	50	21	67	>10 $\mu$ M	134	>10 $\mu$ M	82	>10 $\mu$ M
Elvitegravir	4.9	5.1	19	23	5.1	9.1	24	36	105	>10 $\mu$ M	230	>10 $\mu$ M	189	>10 $\mu$ M

- The IC50 (4.2 and 4.8 nM) of S/GSK1349572 against WT IN was equivalent to RAL (6.4 and 7.5 nM) and ELV (4.9 and 5.1 nM) for both less stringent and stringent assay conditions
- Under more stringent assay conditions, the IC50 of RAL and ELV double mutants increased considerably
  - This might suggest that RAL and ELV had weaker affinity to the double mutant INs, consistent with the dissociation experiment data\*

\*Hightower K., et. al., IDRW 2010, Dubrovnik, Croatia.

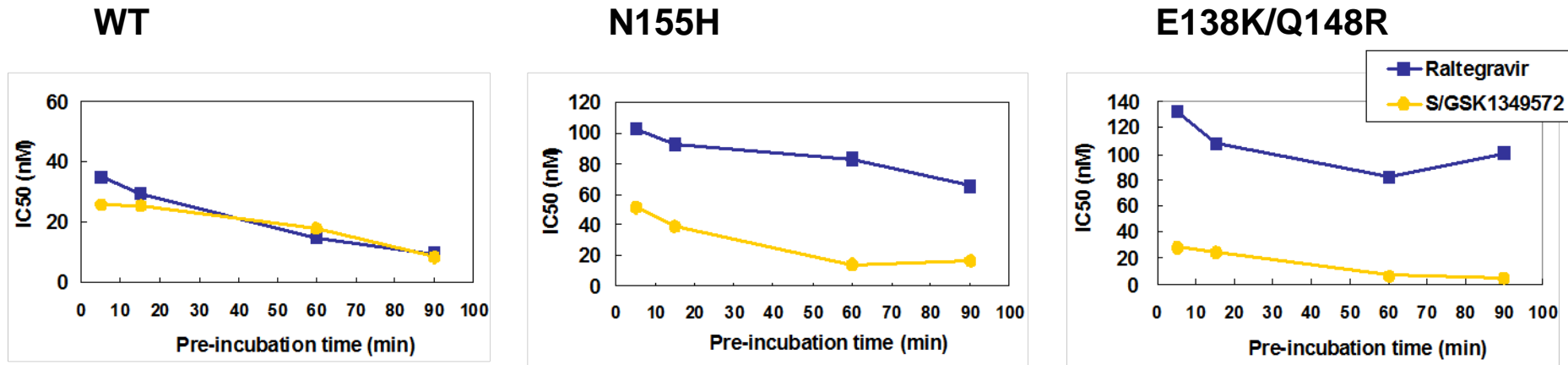
# Fold Changes of Mutants Under Various Assay Conditions were Roughly Correlated Each Other



- Additional mutants G140S, G140S/Q148H and E92Q/N155H demonstrated a similar trend
- Fold change of the more stringent assay was better correlated with those of HeLa-CD4 assay\* than those of the less stringent assay

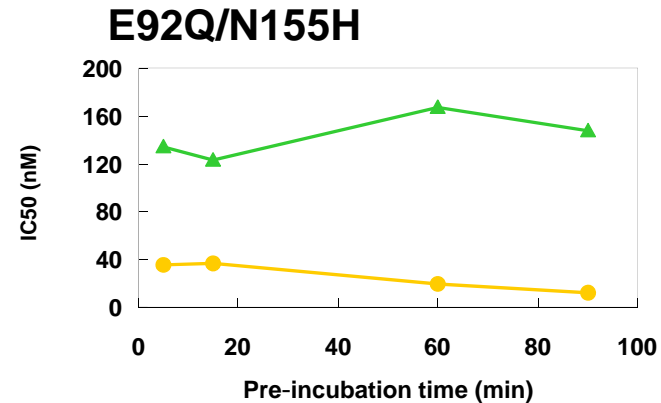
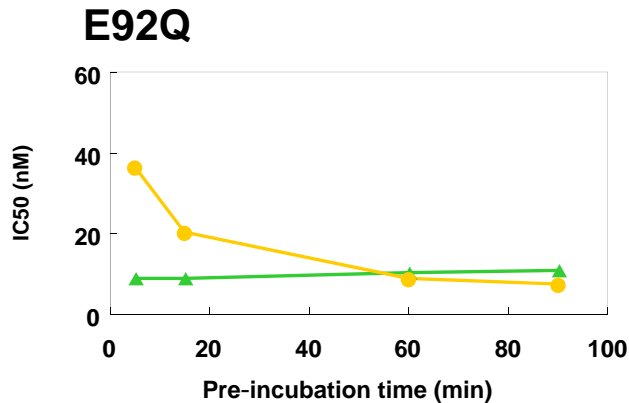
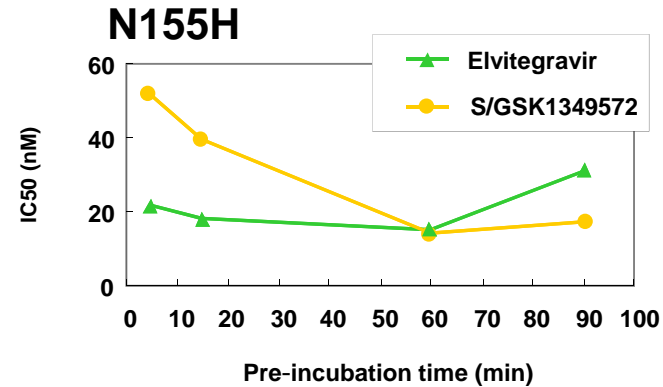
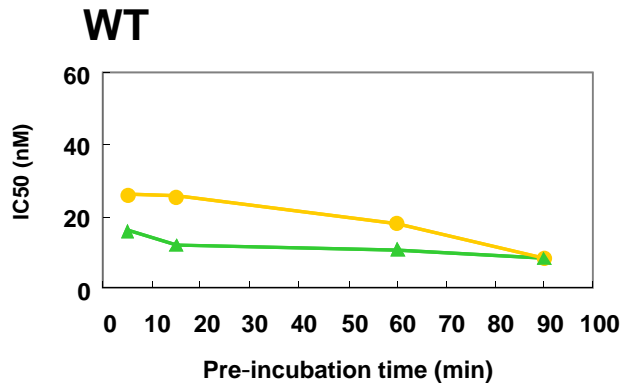
\*Seki T., et al. CROI 2010, San Francisco, CA, USA.

# Comparison of the Effect of Pre-Incubation Time of IN-DNA-INI Complex on Strand Transfer IC50 for S/GSK1349572 or RAL



- **S/GSK1349572 and RAL bound to integrase slowly and reached to the steady state by 90 minutes**
- **572 bound to N155H and E138K/Q148R as similar time course to WT, and inhibited N155H and E138K/Q148R virus proliferation as well as WT virus**
- **RAL could not fully bind to N155H and E138K/Q148R as to wild-type even when pre-incubation time was prolonged, and therefore N155H and E138K/Q148R virus showed resistant to RAL in HeLa-CD4 assay (FC = 11, 110, respectively)**

# Comparison of the Effect of Pre-Incubation Time of IN-DNA-INI Complex on Strand Transfer IC50 for S/GSK1349572 or ELV



- ELV bound to WT faster than 572 in our experiments
- ELV bound to E92Q and N155H as similar time course to WT, although these viruses showed resistant to ELV in HeLa-CD4 assay (FC = 19, 25, respectively)
- ELV reduced binding affinity to E92Q/N155H double mutant than each single mutant

# Conclusions

- **The next generation HIV-1 integrase inhibitor S/GSK1349572 has strong inhibitory activity not only against WT IN enzyme but also against the signature RAL/ELV resistant pathway mutants**
- **S/GSK1349572 had stronger and faster binding to mutant IN than RAL**
- **The in vitro enzyme assay data supports the different binding characteristics of S/GSK1349572 and potentially explains its better resistance profile**

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**All patients enrolled in the clinical trials**

