**Table S4.** **Selection pressure acting upon codons (individual genes) of Hu-WNV strains collected in the US (1999-2011).**

|  |  |  |
| --- | --- | --- |
| **Genea** | **Codon** | **Methodsc** |
|  |  | **FEL** | **IFEL** | **SLAC** | **MEME** | **REL** |
|  |  | ***P* value** | **BF** |
| E (n=54), codons: 501 , ωb: 0.11039 negatively selected sites |  |  |  |  |  |  |
| 1 | N8S | 0.07 | 1 | 0.47 | 0.07 | 1 |
|  |  |  |  |  |  |  |
| NS1 (n=49), codons: 352 , ωb: 0.15617 negatively selected sites |  |  |  |  |  |  |
| 1 | D208E/N | 0.08 | 1 | 0.49 | 0.08 | 1 |
|  |  |  |  |  |  |  |
| NS3 (n=54), codons: 619, ωb: 0.09539 negatively selected sites |  |  |  |  |  |  |
| 1 | L336S | 0.09 | 1 | *0.15* | 0.09 | 1 |
|  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |
| NS4A (n=39), codons: 149 , ωb: 0.0978 negatively selected sites |  |  |  |  |  |  |
| 1 | A85T/Id  | *0.12* | 0.03 | 0.29 | *0.12* | 3781 |
|  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |
| NS5 (n=58), codons: 905, ωb: 0.13762 negatively selected sites |  |  |  |  |  |  |
| 1 | K194R | 0.30 | *0.19* | 0.52 | 0.30 | 439 |
| 2 | K314R | *0.14* | 0.02 | 0.26 | *0.13* | 49528 |
| 3 | **S604R** | **0.06** | **1** | 0.49 | **0.06** | **388** |
| 4 | P712Q | 0.33 | 1 | 0.30 | 0.004 | 2540 |
| 5 | **A860T** | *0.16* | **0.10** | 0.44 | **0.06** | **156** |

Table represent the analysis of the individual genes of WNV for the H dataset (n=61), detected by the methods employed in HyPhy (Datamonkey server).
a number of sequences differ from the input (n=61), after removal of identical sequences by the server.

b ω = dN/dS ratio

c FEL = Fixed effects likelihood, IFEL = Internal Fixed effects likelihood, SLAC = Single-likelihood ancestor counting, MEME = Mixed Effects Model of Evolution, REL = Random Effects Likelihood, BF = Bayes Factor. All codons present in the table are recognized by at least two methods. In bold, codons recognized by three or four methods, with statistical significance. *p* values in italics represent codons detected to be under positive selection, not significantly, but close to *p* threshold (0.1).

d Site NS4A85 detected also under positive selection pressure by the Bayesian empirical method employed by Selecton (Stern A, Doron-Faigenboim A, Erez E, Martz E, Bacharach E, et al. (2007). Selecton 2007: advanced models for detecting positive and purifying selection using a Bayesian inference approach. Nucleic Acids Res 35(Web Server issue):W506–11).