

## SUPPLEMENTARY DATA

### **Epstein Barr Virus Nuclear Antigen 1 (EBNA-1) peptide recognized by adult multiple sclerosis patient sera induces neurologic symptoms in a murine model**

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**Short title:** EBNA-1 cross-reactivity in multiple sclerosis

#### **Footnote:**

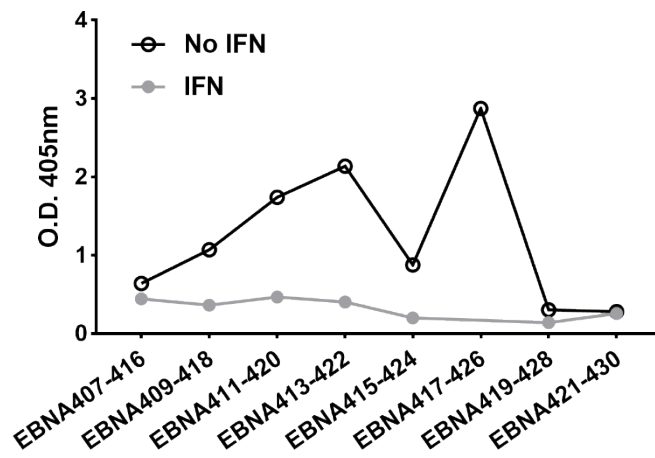
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**Supplementary Table 1.****EBNA411-426 EADYFEYHQEGGPDGE**

Peptide Number	Peptide sequence	EBNA-1 amino acid region	% patients with binding		
			No IFN	IFN	MS pts
200	PPGRRPFFHP	398-408	8.3	27.3	17.4
201	GRRPFFHPVG	401-410	8.3	9.1	8.7
202	RPFHPVGEA	403-412	16.7	9.1	13.0
203	FFHPVGEADY	405-414	25.0	9.1	17.4
204	HPVGEADYFE	407-416	66.7	66.7	69.6
205	VGEADYFEYH	409-418	58.3	27.3	43.5
206	EADYFEYHQE	411-420	50.0	45.5	47.8
207	DYFEYHQEGG	413-422	25.0	18.2	21.7
208	FEYHQEGGPD	415-424	41.7	18.2	30.4
209	YHQEGGPDGE	417-426	33.3	0.0	17.4
210	QEGGPDGEPD	419-428	58.3	36.4	47.8
211	GGPDGEPDVP	421-430	66.7	27.3	47.8
212	PDGEPDVPPG	423-432	16.7	9.1	13.0
213	GEPDVPPGAI	425-434	25.0	9.1	17.4

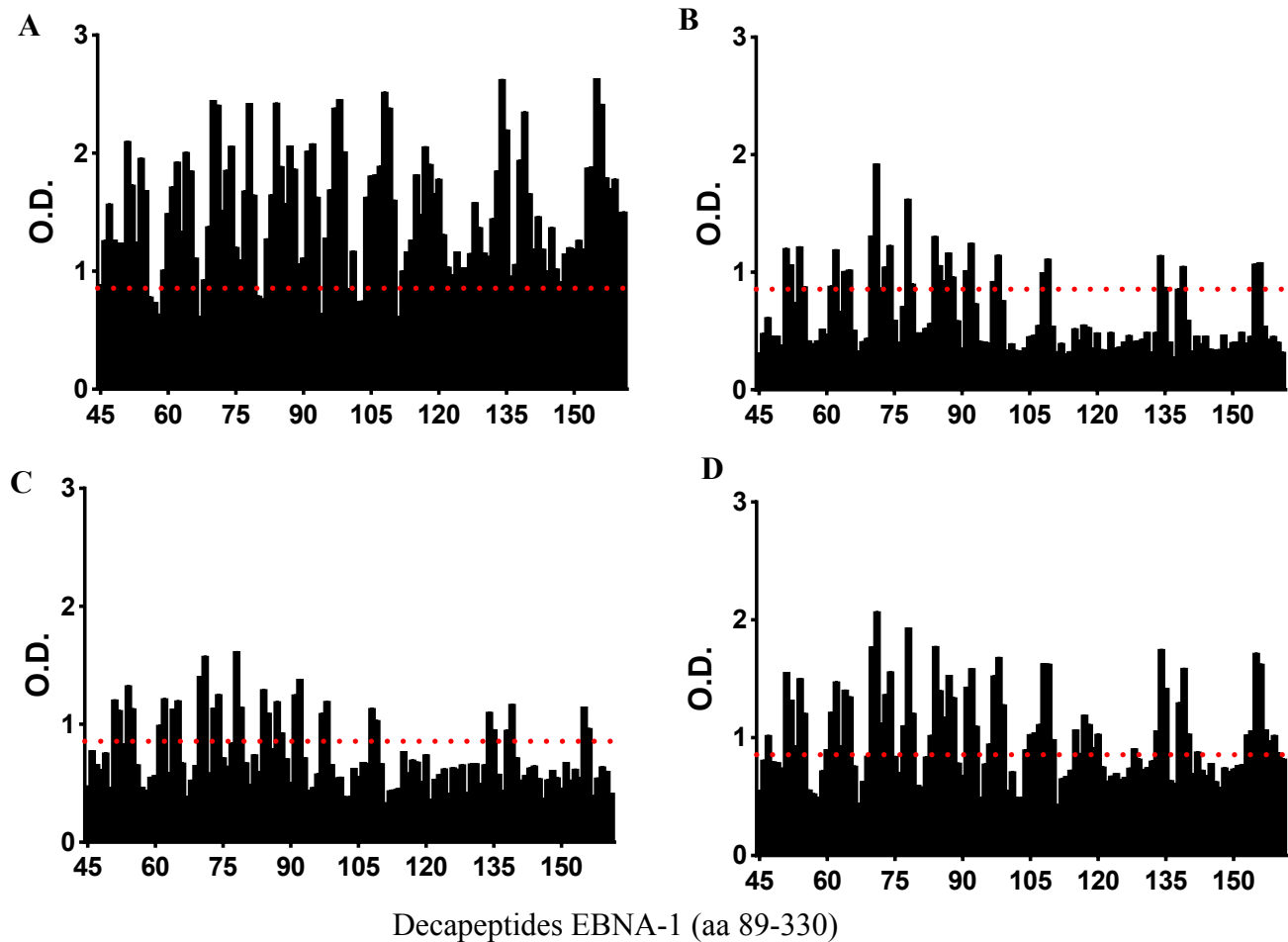
Decapeptides overlapping sequential peptide by eight amino acids spanning the EBNA-1 protein were constructed according to the primary amino acid sequence using solid phase peptide chemistry. The EBNA411-426 regions are highlighted in the sequential peptides in red. Anti-peptide assays were conducted with patient or control sera by a modified ELISA. The cutoff for each peptide binding was defined as the mean OD of the specific peptide in controls + (2\*Standard deviation of the peptide in controls). Percent of MS patients not on IFN therapy (No IFN), percent of MS patients on IFN therapy (IFN), and percent of total MS patients (MS pts) that bound each peptide are shown.

## Supplementary Figure S1



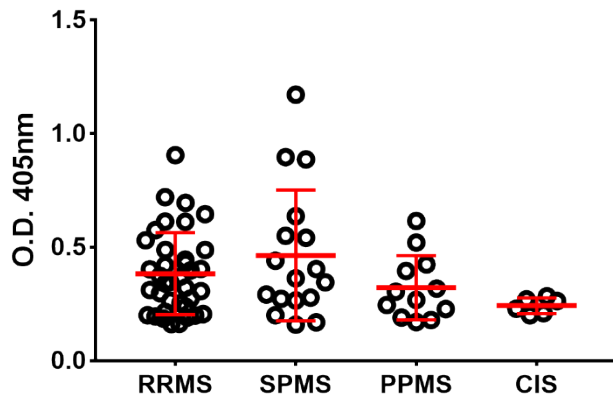
**MS patients not on IFN therapy showed increased binding to amino acid region 411-422 and 417-426.** The mean OD values for each peptide in patients that showed reactivity towards the peptide are presented. The reactivity towards each peptide was defined as in Table 1.

## Supplementary Figure S2



**Binding to glycine-alanine rich regions of EBNA-1** Average binding profiles of the untreated MS patients (A), MS patients treated with interferon  $\beta$  (B), healthy controls (C), all MS patients together (D) against unique overlapping decapeptides of EBNA-1 spanning the amino acid region 89-330 were determined using solid-phase ELISAs. Positive cutoff (dotted line) was defined as mean plus twice the standard deviation (SD) of healthy controls. The overlapping decapeptides are numbered from 45-161.

### Supplementary Figure S3



**Patients with MS tend to have higher IgG towards EBNA411-426 compared to CIS.**

Reactivity towards EBNA411-426 in MS patients was determined by ELISA as described in Methods. RRMS, relapsing remitting MS, SPMS, secondary progressive MS, PPMS, primary progressive MS, CIS, clinically isolated syndrome. Mean $\pm$ SD are shown.