

Supplementary Materials: Glutamic Acid Residues in HIV-1 p6 Regulate Virus Budding and Membrane Association of Gag

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Table S1. ¹H chemical shifts of 2 mM p6 (1-52) E0A in TFE-H₂O (1:1; v/v) at 300 K.

	HN	H α	H β	H γ	H δ	H ϵ	NH ₂	Ar H
Leu-1		4.07	1.79	1.73	1.23/1.01			
Gln-2	8.66	4.52	2.18/2.08	2.44			7.50/6.72	
Ser-3	8.31	4.51	3.91/3.87					
Arg-4	8.10	4.74	1.93	1.79/1.73	3.25		7.19	
Pro-5		4.41						
Ala-6	8.03	4.67	1.40					
Pro-7		4.52	2.37/2.04	2.13	3.78/3.68			
Thr-8	7.82	4.37	4.25	1.25				
Ala-9	7.97	4.67	1.40					
Pro-10		4.69	2.40/1.96	2.12	3.89/3.61			
Pro-11		4.46	2.30/1.97	2.09	3.78/3.69			
Ala-12	8.03	4.28	1.43					
Ala-13	7.95	4.22	1.49					
Ser-14	7.92	4.28	3.92/3.86					
Phe-15	7.84	4.53	3.19/3.15					2/6 7.31
Arg-16	7.80	4.16	1.76	1.50	3.13		7.10	
Phe-17	7.94	4.55	3.26/3.08					2/6 7.28
Gly-18	8.10	3.99/3.87						
Ala-19	8.03	4.28	1.43					
Ala-20	8.00	4.28	1.49					
Thr-21	7.79	4.41	4.35	1.21				
Thr-22	7.80	4.51	4.32	1.27				
Thr-23	8.01	4.48	4.33	1.32				
Pro-24		4.46			3.91/3.78			
Ser-25	8.09	4.42	3.99/3.92					
Gln-26	7.98	4.41	2.18/2.06	2.42			7.36/6.65	
Lys-27	8.06	4.33	1.93/1.83	1.56/1.48	1.73	3.03	7.59	
Gln-28	8.25	4.35	2.22/2.14	2.45			7.40/6.69	
Ala-29	8.07	4.53	1.50					
Pro-30		4.46	2.33/2.02	2.11	3.83/3.72			
Ile-31	7.43	4.11	1.95	1.56/1.26/ 0.97/0.93				
Asp-32	8.09	4.57	2.95					
Lys-33	8.01	4.11	1.91	1.58/1.56	1.73	3.02	7.61	
Ala-34	7.77	4.29	1.51					
Leu-35	7.73	4.43	1.77	1.65	0.95/0.89			
Tyr-36	7.88	4.57	3.27/3.17					2/6 7.12 3/5 6.85
Pro-37		4.28	2.39/1.90	2.19/2.04	3.87			
Leu-38	7.23	4.23	1.82	1.73	1.01/0.94			

Table S1. Cont.

	HN	H α	H β	H γ	H δ	H ϵ	NH ₂	Ar H
Ala-39	8.27	4.08	1.47					
Ser-40	8.20	4.18	3.94/3.76					
Leu-41	7.88	4.19	1.92/1.83	1.71	0.98/0.93			
Arg-42	8.18	4.08	1.98/1.84	1.75	3.25/3.18		7.20	
Ser-43	7.95	4.33	4.06/3.96					
Leu-44	7.83	4.19	1.75	1.63/1.57	0.86/0.78			
Phe-45	8.03	4.67	3.36/3.04					2/6 7.33
Gly-46	8.04	4.04						
Ser-47	7.96	4.53	3.96					
Asp-48	8.28	5.09	3.09/2.88					
Pro-49		4.47	2.35	2.07	3.84/3.72			
Ser-50	8.00	4.50	4.00/3.93					
Ser-51	8.06	4.53	4.00/3.95					
Gln-52	8.05	4.47	2.27/2.06	2.40			7.39/6.62	

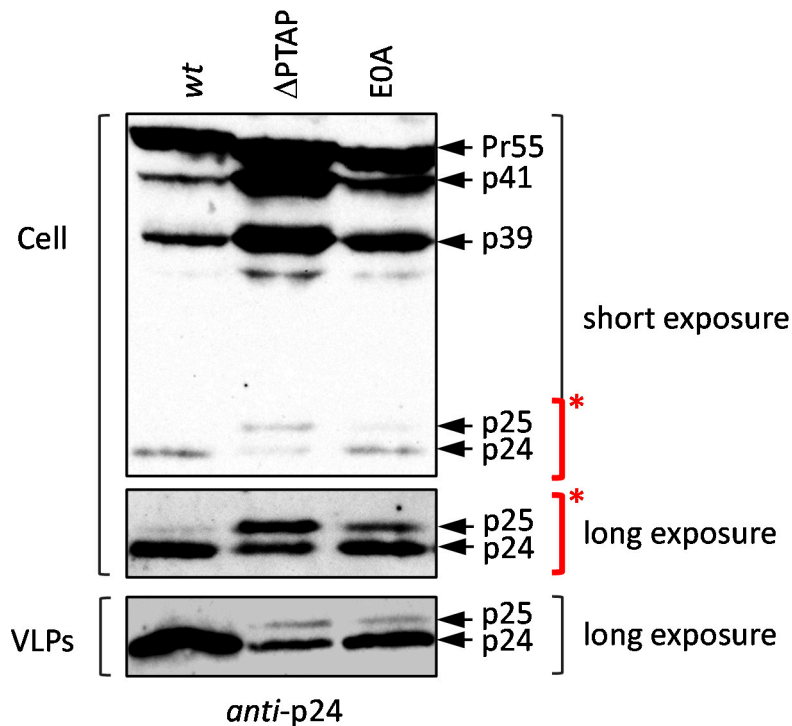


Figure S1. Western blot analysis of virus and cell fractions of *wt*, Δ PTAP and E0A expressing HeLa cells using a high performance chemiluminescence film. HeLa cells were transfected with pNLenv1 expression plasmids directing the expression of *wt*, Δ PTAP, and E0A. VLP fractions and whole cell lysates were separated by SDS PAGE and analyzed by western blot using an anti-CA antibody. Short time exposure: 30 s; long time exposure: 5 min; * same region of the western blot.

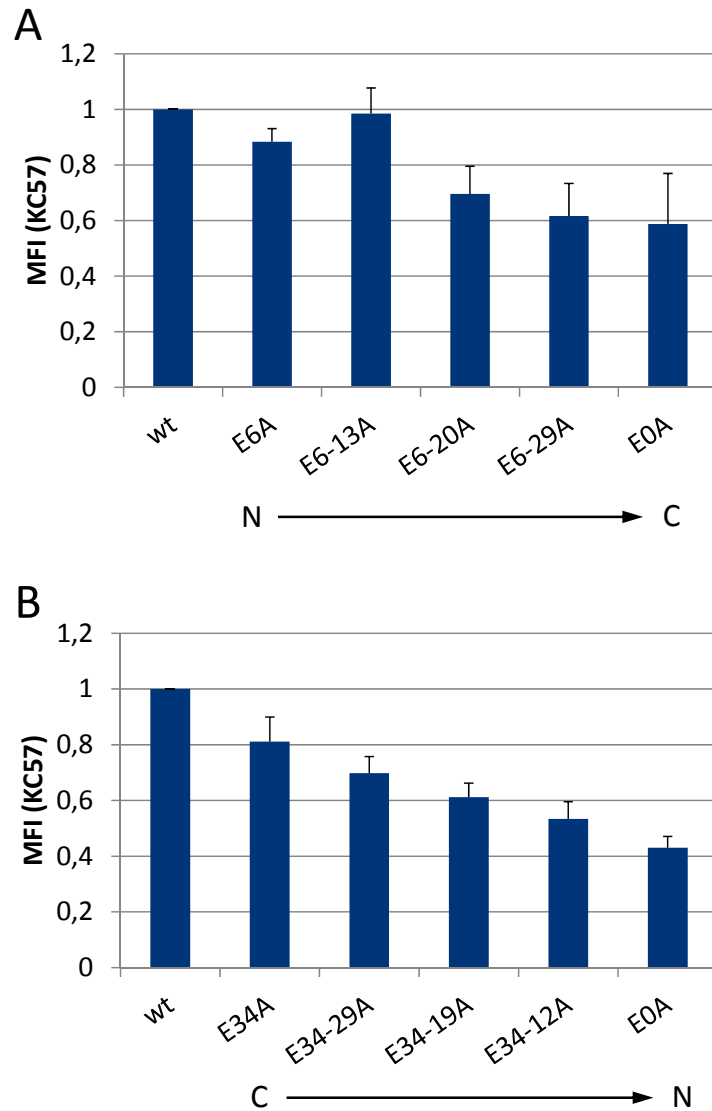


Figure S2. MFI of intracellular Gag staining corresponding to the experiments depicted in Figure 7. HeLa-K^b cells were transiently transfected with pNLenv1-SL expression plasmids coding for *wt* or the sequential Glu mutants of p6 from (A) the N- to the C-terminus; or (B) the C- to the N-terminus, respectively. Intracellular Gag was stained with *anti-Gag* Ab KC57-FITC.



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