

**Figure S1 : Nucleotide sequence of the IAPE-D1 provirus.**

The nucleotide sequence of IAPE-D1, together with the amino acid sequence of the encoded retroviral proteins (Gag, Pro, Pol and Env), are indicated. The 5' and 3' LTRs are underlined, the splice donor site (SD) and the two alternative splice acceptor sites (SA<sub>1</sub> and SA<sub>2</sub>) are positioned and the domain with CTE activity is indicated in bold.

**Figure S1** : Nucleotide sequence of the IAPE-D1 provirus

1 tgtaaagggctgcaatctacgtgccgccactagatagtgctaaacctctt  
51 tgtgcgagctgagagacagccagccaaagtcaccagtcctgtctccg  
101 aggctttttgcccaccccctgggggctttaacctatccccagggtgcatga  
151 ggatcatgggtgttgctaccagtcctagtttagacacgtcaccaaaagta  
201 tttaagccacacacttttctgagcttttctttgtctacctcaatatgatata  
251 gaagtaaaagttaaccgtggaagaatctgggtgtgtcctgtgtttatttcc  
301 cgcgggacggggataagctcgggatacttggtgctagaaacccgggaaaa  
351 aacgccttaacaacgcggcacctagaggagaccctccgaccaaccggagc  
401 agatttcagaaccacggtggatttttgaattccgagagcatgctgaatcc  
451 ttgta<sup>SD</sup>gtaactctgatccttcttaatccctccccttaactggaaggtgg  
501 cactaaaagccttttgttctcatgctggctgtgttaaccctattttctccc  
551 ggtttgatcctgcactcaccagaagtggcgctgaaagaaaaagttttgaa  
601 tgctgtgaagcacgatcagacggacgtatataagccgcagtgctcgtctc  
651 atatttaagcctctcgtagataagggagcagtatgggaaattcacattct  
701 gtaactacagctctacactctgtttttaa<sup>M G N S H S</sup>acagcgagagataaaagtttc  
751 <sup>V T T A L H S V L K Q R E I K V S</sup>tacaagaaccttagaaacgttcattaaggaaatagaacgtatatcacctt  
801 <sup>T R T L E T F I K E I E R I S P W</sup>ggtagcgtgttcagggtcgttaactctttcctcttgggagaaattaagg  
851 <sup>Y A C S G S L T L S S W E K L R</sup>gaagacttagctaaggaacaacagaatgggaaattaaaggcagggaccat  
901 <sup>E D L A K E Q Q N G K L K A G T M</sup>gccattatggaaactggtgagatcgtgcctggaggatgagagatgccgtc  
951 <sup>P L W K L V R S C L E D E R C R P</sup>cagcaataataacagggcaggcaatattagaagaagcacaggacagcatg  
1001 <sup>A I I T G Q A I L E E A Q D S M</sup>gcagaaacggaatggtgtgaaagggttaggagctccaaaaaaggaaaatgt  
1051 <sup>A E T E W C E R L G A P K K E N V</sup>gcacaaagtaaaaagcccttccagagaccttgagtcagaggaagtgaaaa  
1101 <sup>H K V K S P S R D L E S E E V K N</sup>atttgaggataagtcctcagggagagaaaaaggaaaaggagaaagttcag  
1151 <sup>L R I S P Q G E K K E K E K V Q</sup>aaaaggaaaagccttttatccggtaaaggaactagaggctttgaagcttga  
<sup>K R K S L Y P V K E L E A L K L D</sup>

5' LTR

Gag

1201 tagttttaaagcagatgagcttagctcctctgaggaagaggagtcgcatt  
       S L K A D E L S S S E E E E S H Y  
 1251 atgaggcagctcattataaaaaagaaagataccatccagaggaaaggcga  
       E A A H Y K K E R Y H P E E R R  
 1301 gtgaagaaatcagagaaaaaattaaaagttactggagacacagaacaggc  
       V K K S E K K L K V T G D T E Q A  
 1351 cacctctgggccttctagtctcaatacacctccaccttatgtggagaaat  
       T S G P S S L N T P P P Y V E K F  
 1401 ttatttctgattcttttctttcaaaggaggaaaagaaaaattatatcag  
       Y S D S F L S K E E K K K L Y Q  
 1451 gcatttcctgtctttgaagctgcagatggaggcgagtacacgccccgt  
       A F P V F E A A D G G R V H A P V  
 1501 ggagtatactcagataaaagaacttgctgagtcggttcgcaattatggag  
       E Y T Q I K E L A E S V R N Y G V  
 1551 ttagtgccaacttcactatctcccaaattgaaagatttgctacttttagcc  
       S A N F T I S Q I E R F A T L A  
 1601 atgacccttgagactggcagacaacagtgaaggctgcactccctaatat  
       M T P G D W Q T T V K A A L P N M  
 1651 ggggcaatatatggagtggaaagcgttggtggcatgatgcttctcagacac  
       G Q Y M E W K A L W H D A S Q T Q  
 1701 aggccaaagtcaatgccactgctgaaggcaatcagagaaattggacattt  
       A K V N A T A E G N Q R N W T F  
 1751 gaattattaacaggacaagggcaatatgctaataatcagacgaattatga  
       E L L T G Q G Q Y A N N Q T N Y D  
 1801 ttggggagcatacaatcaaactctccactgctgccattaaggcgtggaagg  
       W G A Y N Q I S T A A I K A W K A  
 1851 cactttccaaaaaggaggagctctggaagatacttgacaaaaattatacaa  
       L S K K G E S G R Y L T K I I Q  
 1901 aatcctcaggagtcattctcagactttgtagctagaatgacagaggcagc  
       N P Q E S F S D F V A R M T E A A  
 1951 aggcagaatttttggagactctgaacaggcaatgccttttagtggaacagt  
       G R I F G D S E Q A M P L V E Q L  
 2001 tgatttatgagcaagccactcaagaatgcagagcagcaattacaccaga  
       I Y E Q A T Q E C R A A I T P R  
 2051 aagagcaaaggactgcaagattggttaagggtatgccgtgagcttgagg  
       K S K G L Q D W L R V C R E L G G  
 2101 ccccttatctaatgcagggttagcggtgccatcctgcaaggccaaaggc  
       P L S N A G L A A A I L Q G Q R H  
 2151 attcagatacaaatagtaataacagggtctgctataactgtggcaaacca  
       S D T N S N N R V C Y N C G K P  
 2201 gggcatatgagaagagactgccaggcactcgtaaaagggaagtgttagg  
       G H M R R D C Q A L V K G K V L G  
 2251 gctatgtaccaggtgtggaaaaggctatcatcgagccagtgagtgtcggt  
       L C T R C G K G Y H R A S E C R S  
 2301 caattaaagacgtcagaggcagattttattcagtctggacctcaggcagaa  
       I K D V R G R F I Q S G P Q A E  
 2351 agaagtgaagacaattcaaaaaacgagtttctgggccccagggtcccagg  
       R S E D N S K N E F L G P R S Q G  
 2401 ccccaaacatatgggatcccagctcacaacaggcggaactacagtcca  
       Q N I W D P S S Q Q A D T T V H  
       P K T Y G I P A H N R R T L Q S T

Gag

Pro

2451 cagagctgtagaagtctcagcaagagtggatctctgtgccatcactcgat  
       R A V E V S A R V D L C A I T R F  
       E L  
 2501 tcatgctgacaccctcaatggggatgcagcccattcccactgacttcaaa  
       M L T P S M G M Q P I P T D F K  
 2551 gggcctctgccatccagaagtgttggcctaatactgggccaggcctcctt  
       G P L P S R S V G L I L G Q A S L  
 2601 ggctttacaaggcctcattatccaccctggagtcataaatcaagactatg  
       A L Q G L I I H P G V I N Q D Y E  
 2651 agggggaacttcaagttctttgttcctgtcctcaaggtgttttttctata  
       G E L Q V L C S C P Q G V F S I  
 2701 gcacaaggagataaaaatagctcagctaatagtcttgccaagcctacatgg  
       A Q G D K I A Q L I V L P S L H G  
 2751 ctactccccctcccctgggtgtccttcaagctgctagagaaattggctctg  
       Y S P S P G V L Q A A R E I G S A  
 2801 ctgaaaatgattctgcttacttaataatgcctcttgattccaggccaact  
       E N D S A Y L I M P L D S R P T  
 2851 ttagagttagttatagaagggaaacacttcaaagggattctagatacagg  
       L E L V I E G K H F K G I L D T G  
 2901 agcagataaaaagtatcatctcttcccactgggtggccaaaatcttggcccg  
       A D K S I I S S H W W P K S W P V  
 2951 ttactcaatcatcacattctttgcaagggttaggctatcagtcctgcccc  
       T Q S S H S L Q G L G Y Q S C P  
 3001 actatcagctctcgcactctgagctggcaagcacctgaaggtcaaacggg  
       T I S S R T L S W Q A P E G Q T G  
 3051 aaggtttactccccatgtcttaccactcccagttaatctttgggggagag  
       R F T P H V L P L P V N L W G R D  
 3101 acatttttacaggcacttaggaataaccttgactaatgagtactcaccacaa  
       I L Q A L G I T L T N E Y S P Q  
 3151 gctgtttcaaattgatgaagaaaatgggctataaagaggggaaagggttagg  
       A V Q M M K K M G Y K E G K G L G  
 3201 gaaaagagagcagggttagacttgaacctattcctcaagagggttaataaag  
       K R E Q G R L E P I P Q E G N K G  
 3251 gaagacaaggcttgggttttttctagaatcagctgttgagggttccatgc  
                           F F L E S A V E G S M P  
       R Q G L G F F  
 3301 ccataccttggattacagaggagaatgtatgggttcctcaatggcctcta  
       I P W I T E E N V W V P Q W P L  
 3351 tcctctgaaaaaattagaagcagccacaaaactgggtctctgaacagctacg  
       S S E K L E A A T K L V S E Q L R  
 3401 cttaggtcattttagaaccctctacctcaccctggaatacacctatttttg  
       L G H L E P S T S P W N T P I F V  
 3451 taattaagaaaaaatctggcaaattggcgtttattacatgatcttagagca  
       I K K K S G K W R L L H D L R A  
 3501 attaatgcacaaatgcgcctgtttggatcagttcagcgagggtcccatt  
       I N A Q M R L F G S V Q R G L P L  
 3551 gctctccgccctacctaataaattgggaaattataattatagatattaag  
       L S A L P K N W E I I I I D I K D  
 3601 attgttttttctctatacctttatgtcctcaagataggcaaagatttgca  
       C F F S I P L C P Q D R Q R F A  
 3651 ttaccatcccagccattaatcactcagagcctgatcataggtaccaatg  
       F T I P A I N H S E P D H R Y Q W

P10

P01

3701 gaaggtcttgccctcaggggatggcaaatagccccactatgtgtcagctgt  
       K V L P Q G M A N S P T M C Q L Y  
 3751 atgtgcaattggcacttaagtcagttaaaaatcattttccatcgattctt  
       V Q L A L K S V K N H F P S I L  
 3801 ctgataaattatatggatgacatactgatatgtcacagagatttacagtt  
       L I N Y M D D I L I C H R D L Q L  
 3851 attacaagaagcatatcctatatattaataaaaacgttagggaatggggac  
       L Q E A Y P I L I K T L G K W G L  
 3901 tgcaggtagccactgaaaaggtgcaagttgctcaaataaggagccttctta  
       Q V A T E K V Q V A Q I G A F L  
 3951 gggtcactcattttatcctgacaaaattgttcctcaaagattagagattcg  
       G S L I Y P D K I V P Q R L E I R  
 4001 cagagatcattttacacaccttaaatgattttcaaaaattattaggagata  
       R D H L H T L N D F Q K L L G D I  
 4051 taaattggctgagaccatttttaagatttcctctgctgagttaaaacct  
       N W L R P F L K I S S A E L K P  
 4101 ttatttgacatttttgggaaggagagtctcatatctcctcccctagagccct  
       L F D I L E G E S H I S S P R A L  
 4151 tactcctgctgcaaatacaggccttgcgagacagtagaaaaagctttgcaa  
       T P A A N Q A L Q T V E K A L Q K  
 4201 aggctcaattacaacgcattgatgagtcacaaccttttgatttgtgtgtc  
       A Q L Q R I D E S Q P F D L C V  
 4251 tttaagacagcccagttgccaaaccgcagtcttgtggcaaatggaccttt  
       F K T A Q L P T A V L W Q N G P L  
 4301 attatgggttcatccaaatgcatctcctgctaaaataatagattggtatc  
       L W V H P N A S P A K I I D W Y P  
 4351 ctaatgcagttgcacaacttgcacttcgaggaataaaaagcagctattact  
       N A V A Q L A L R G I K A A I T  
 4401 tattttggacaagatcctaaaatcctaattgtaccttatactgctgcaca  
       Y F G Q D P K I L I V P Y T A A Q  
 4451 gggtcaaaccttggcagctacatctgatgattgggcagtttttagtcacct  
       V Q T L A A T S D D W A V L V T S  
 4501 ccttttcaggacaaattgacaaccattatcctaatacatccaattttacag  
       F S G Q I D N H Y P K H P I L Q  
 4551 tttgctctaaatcaggctatagtgtttccacaagtaacagctaaagaccc  
       F A L N Q A I V F P Q V T A K D P  
 4601 acttacagatgggattgtggtgtacactgatggatcaaaaactggtatag  
       L T D G I V V Y T D G S K T G I G  
 4651 gtgcttatgtggttaataatgagatagtaactaagcaatataatgagact  
       A Y V V N N E I V T K Q Y N E T  
 4701 tcacctcaagttgtggaatgttttagtagtagtagaggttcttaaggcatt  
       S P Q V V E C L V V L E V L K A F  
 4751 cccaggaccgcttaacattgtatcagattcctcctatgtagtaaatgcag  
       P G P L N I V S D S S Y V V N A V  
 4801 ttaacatttcttgaaacagctggggtaataaaaacctccagcagagttgcc  
       N I L E T A G V I K P S S R V A  
 4851 aatatttttcgacaaatacagtttgttttattaaatagaaaattccctgt  
       N I F R Q I Q F V L L N R K F P V  
 4901 ttatattactcatgttagggcacactcaggcctccctggtcctatgtctc  
       Y I T H V R A H S G L P G P M S L  
 4951 tgggaaatgatttagcagacaaagccactaagggttatagctgtttgctctg  
       G N D L A D K A T K V I A V A L

Pol

5001 tcttcccaggtaaaagcagcaaaagaatttcataagcgctttcatgtgac  
 S S Q V K A A K E F H K R F H V T  
 5051 agctgagacttttacgtcgccgctttgcattaactagaaaagaagctagag  
 A E T L R R R F A L T R K E A R E  
 5101 agattgttactcaatgtcacaattgttgtgaatttttacctgtccctcat  
 I V T Q C H N C C E F L P V P H  
 5151 gtggggataaacctcgtggcgtcaggccattacaggtctggcaaattgga  
 V G I N P R G V R P L Q V W Q M D  
 5201 tgtcacgcatttttccctcctttgggagaaatcaatatttacatgtttcta  
 V T H F S S F G R N Q Y L H V S I  
 5251 ttgacacctgctctgggtgtaatgtttgccacacctttaacaggtgaaaaa  
 D T C S G V M F A T P L T G E K  
 5301 gcctctcatgtttataacaacattgtctcgaggcatggagtgcttggggaaa  
 A S H V I Q H C L E A W S A W G K  
 5351 acccaaactccttaagacagacaatggaccagcttacacctctcaaaaat  
 P K L L K T D N G P A Y T S Q K F  
 5401 ttcaacagttctgtcgccagatggaagtaactcatctgactgggtcttccg  
 Q Q F C R Q M E V T H L T G L P  
 5451 tataatcctcaagggcagggcattgtagaacgtgcccacgcaccctcaa  
 Y N P Q G Q G I V E R A H R T L K  
 5501 gtccttccttattaaacaaaaggggggaatccaggaagttctaccacag  
 S F L I K Q K G G I Q E V L P T V  
 5551 tgccaagagtagctgtttctatggcactcttcactcttaattttttaaac  
 P R V A V S M A L F T L N F L N  
 5601 cttgatgctcaaggccataactgcggtgaacgtcactgttcagagcctga  
 L D A Q G H T A A E R H C S E P D  
 5651 caggccaaaagagatggtaaaaatggaaagatgttttgactgggtctctgga  
 R P K E M V K W K D V L T G L W K  
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 P Q E E D N P F W L P E R L T R R  
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 I P T P E D D S Q M D P L E T T S  
 M T P R W I P W K L P  
 5851 ctaatcctcatgctacctctgtggatttgggttcctagtatatatttgaga  
 N P H A T S V D L G S  
 L I L M L P L W I W V P S I F G E  
 5901 gtatagatgggcaattctgtctgtctttccctaagcctatgcctgttcgtc  
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 5951 ataatacagctgtttttcccaagttttttacgaccaataaaaacttttaggc  
 N T A V F P K F F T T N K T L G  
 6001 ctgccatacctaccatttgaccctatctgggcaccattgggagaaaaacg  
 L P Y L P F D P I W A P L G E K R  
 6051 ctctttaaggggaacgaggttctctctgtttccaaatttatgaattaggag  
 S L R E R G S L C F Q I Y E L G G  
 6101 gttgtatcagactcaccagccaagctttgggaatgttttttaagtatcga  
 C I R L T S Q A L G M F F K Y R  
 6151 ggaggcggtgtgaaaataacccaagacacttccaacagagatatataaccct  
 G G V V K I T Q D T S N R D I T L

PoI

Env

6201 tacaaatcggaccttctggcatgaggcaacttgggttaatggtacatttc  
       T N R T F W H E A T W V N G T F L  
 6251 taccacctaacttttcagataaagaacgtcccaatcaaccaaaaatggct  
       P P N F S D K E R P N Q P K M A  
 6301 cctcattgtagcttggaagatgaagggtgatcctgccttggtctgattg  
       P H C S L E D E G L I L P W S D C  
 6351 tcaatcctctgtcactcgttgggcagatcagagtaaaactttttcctttt  
       Q S S V T R W A D Q S K T F S F S  
 6401 ctcccaacatgatagatgacccagagcagaaatatgttatgaaaaaggga  
       P N M I D D P E Q K Y V M K K G  
 6451 cttttcatacaggacttttagaatgcatccttttcataaatgggtactttg  
       L F I Q D F R M H P F H K W V L C  
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       G I N G S C T D L N P L V F L Q G  
 6551 gaggagctgctggaaaagcgatttttaatggtattttcaaaatttgctcag  
       G A A G K A I F N G I S K F A Q  
 6601 tttcatcaagttctgttacctgatagaaccatttatcaaaattctactac  
       F H Q V L L P D R T I Y Q N S T T  
 6651 tgaaatcactggattttaataagacattgataaagcagactaattatttac  
       E I T G F N K T L I K Q T N Y L P  
 6701 ctactccagtttgtgtgtacacccttttcttgtttattctttctaattggt  
       T P V C V Y T P F L F I L S N G  
 6751 tcctttgaatcctgcactaatgaaacctgctggatgtcacaatgctggag  
       S F E S C T N E T C W M S Q C W S  
 6801 ccttaagtgggccagccgtgcaatgcttgccaagattccacgatgggttc  
       L K W A S R A M L A K I P R W V P  
 6851 ctgtccccgtggagaccccttccaccataaccttacacaggcaaaagcga  
       V P V E T P S T I T L H R Q K R  
 6901 gactttggtatcactgcagccgttgtggtggcaatggctgccagtgccggc  
       D F G I T A A V V V A M A A S A A  
 6951 ggcagctacagctgctggaattgctatggcaacatcagtccagtcagca  
       A A T A A G I A M A T S V Q S S T  
 7001 caactgtcgagcagttgtcctcctccgtggcgaggcgatcgatcaacat  
       T V E Q L S S S V A E A I D Q H  
 7051 tcagtgtcagtgcccaattaaaagggtgggctaattgatagtgaaccagcg  
       S V L S A Q L K G G L M I V N Q R  
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       I D L V E E R L E I L L Q L A Q L  
 7151 ttggctgtgataagaaatcagtagccttatgtattactagtgtccaatat  
       G C D K K S V A L C I T S V Q Y  
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       E N W T H A A N L S K E L S L F L  
       SA<sub>2</sub>  
 7251 tacagTgaaactggtctgaaggatttgacgaaaaacttgaggcgctacgta  
       T G N W S E G F D E K L E A L R T  
 7301 cagcagtgatgacgattaattctacgcgcgtggacccatcattgatcgat  
       A V M T I N S T R V D P S L I D  
 7351 ggaatcaaaggacttttcttcctggatgtcctctgcattctcacattttaa  
       G I K G L S S W M S S A F S H F K  
 7401 agagtgggtgggagtgggcctgtttggtgcaaccctgtgttggtgactag  
       E W V G V G L F G A T L C C G L V

Env

7451 tgtttcttctatggctggtatgcaaactcagatctcaacagaaacgtgac  
       F L L W L V C K L R S Q Q K R D  
 7501 aaggtggtaattgctcaagcacttgctgccatagagcaaggcacctcccc  
       K V V I A Q A L A A I E Q G T S P  
 7551 tgaaatttggctatctattctcaagaattagttggcatttgagttctctg  
       E I W L S I L K N  
 7601 tccttgcacatcccacaaaattgtggatacattgcattgagatgagagaaac  
  
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 7701 tgtgataaggtgatcat**gatttccccactaattcatttttggctggcctc**  
 7751 **ttttgtagagttcacctaggccatttagcagttatagtcacacagcact**  
 7801 **gtggtatccaaagacgggcaactttcctcatgcacagaccaacctaagac**  
 7851 **aatggggcccgggtggtgatagggttaccgataacgggtaaggctgtgac**  
 7901 **attggaggaacgacctaataacaggggccacagcagatggacgtaactaca**  
 7951 **taggcctagaccagcctcattttaataaattatagagggggagatgtaaa**  
 8001 gggctgcaatctacgtgccgccactagatagtgctaaacctctttgtgcg  
 8051 cagctgcgagagcacagccagccaaagtcaccagtcctgtctccgaggctt  
 8101 ttgccaacccctgggggctttaacctatccccaggggtgcatgaggatca  
 8151 tgggtgttgctaccagtcctagtttagacacgtcaccaaagatatttaag  
 8201 ccacacactttctgagctttctttgtctacctcaatatgatatagaagta  
 8251 aaagttaaccgtggaagaatctgggtgtgtcctgtgtttatttcccgcggg  
 8301 acggggataagctcgggata

Env

CTE

3' LTR

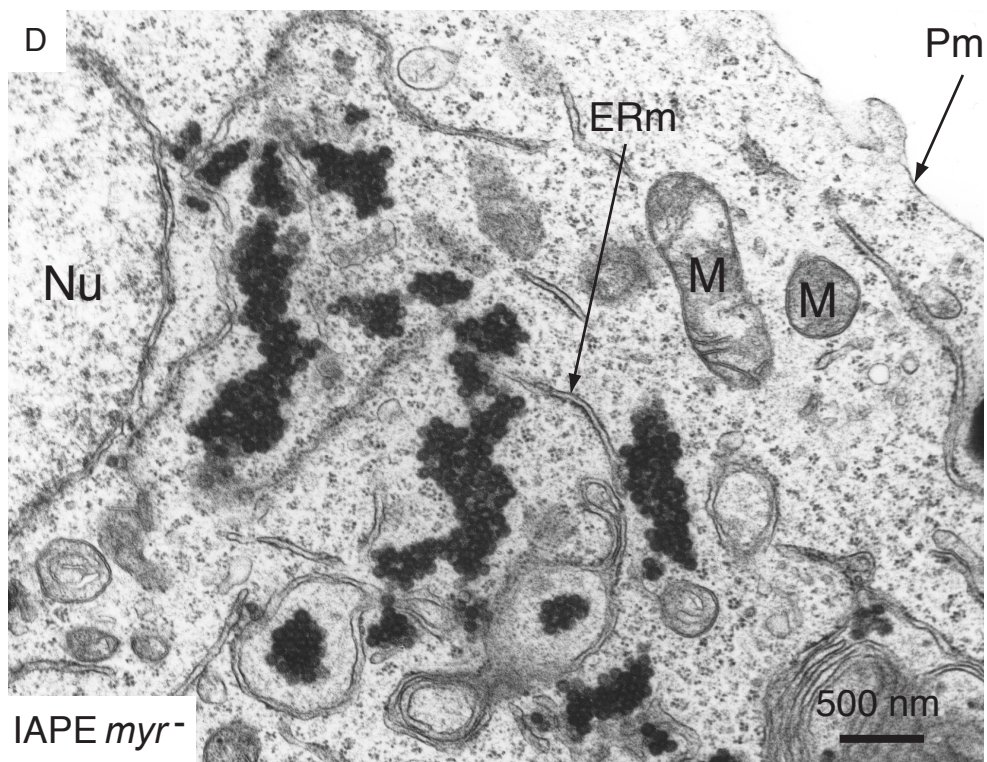
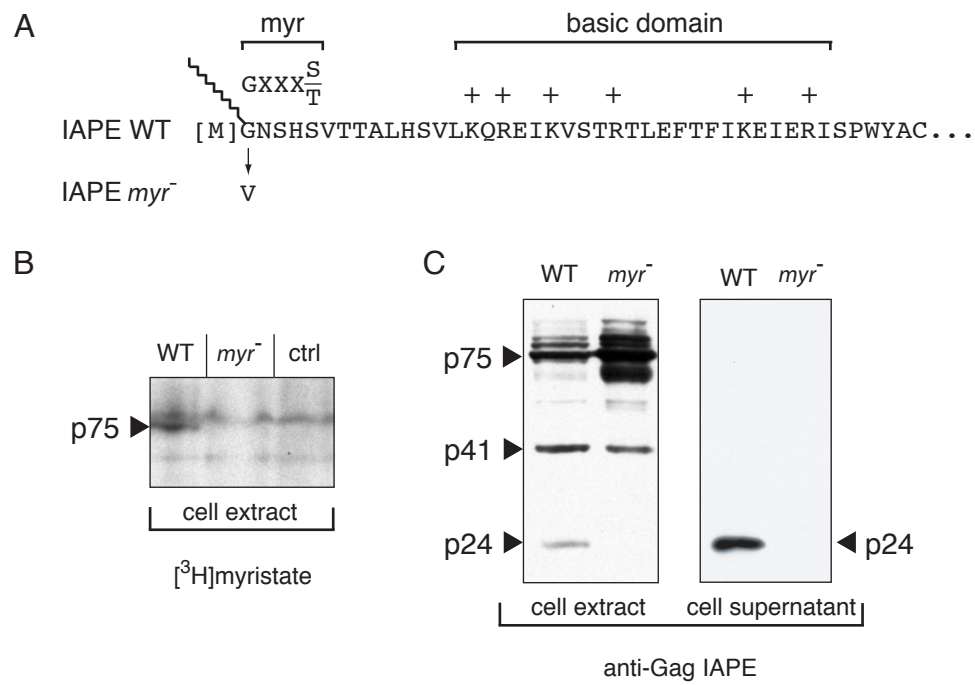
**Figure S2 : Rationale of the retrotransposition assay with *neo*<sup>TNF</sup>-marked elements.**

Retrotransposition assays were performed as previously described in Dewannieux M, Dupressoir, A., Harper, F., Pierron, G., and Heidmann, T. 2004. *Nat. Genet.* **36**: 534-539, using IAP/IAPE elements marked with the *neo*<sup>TNF</sup> indicator gene for retrotransposition. Basically, the *neo*<sup>TNF</sup> indicator gene contains a backward neomycin resistance gene (with the *neo* ORF interrupted by a forward intron) becoming functional only after a complete retrotransposition cycle. Indeed, the intron is removed by splicing during transcription -in the forward direction- of the element expression vector and the proviruses newly generated by reverse transcription and integration into the host cell genome are now intron-less, allowing for expression of the reconstituted *neo* gene -in the backward direction- from its own promoter (A). Retrotransposition events are then detected upon G418 selection of the transfected cells (B). The structure of the plasmids used for the assays in Fig. 5 and 6 are indicated (C).



**Figure S3 : IAPE-D1 contains a myristoylation and a plasma membrane targeting signal.**

(A) Sequence of the N-terminal domain of the IAPE-D Gag protein disclosing a canonical bipartite signal for plasma membrane targeting, *i.e.* a consensus sequence required for myristoylation (myr, [M]GXXXS/T) with the first M corresponding to the Gag initiation codon (reviewed in Schultz, A.M., Henderson, L.E., and Oroszlan, S. 1988. *Annu. Rev. Cell Biol.* **4**: 611-647) and a domain rich in positively charged basic residues (basic domain, +) interacting with the membrane phospholipids (Yuan, X., Yu, X., Lee, T.H., and Essex, M. 1993. *J. Virol.* **67**: 6387-94; Saad, J.S., Miller, J., Tai, J., Kim, A., Ghanam, R.H., and Summers, M.F. 2006. *Proc. Natl. Acad. Sci. USA.* **103**: 11364-11369). The Glycine residue in the wild-type IAPE-D1 was replaced by the non-myristoylable Valine residue (Rhee, S.S., and Hunter, E. 1987. *J. Virol.* **61**: 1045-1053) in the IAPE *myr*<sup>-</sup> mutant using the QuickChange II XL Site-Directed Mutagenesis kit (Stratagene). (B) Myristoylation of the wild-type IAPE Gag. Whole-cell lysates were prepared from human 293T cells transfected with the wild-type and *myr*<sup>-</sup> mutant IAPE constructs and labeled with [<sup>3</sup>H]myristate. Myristoylated proteins were revealed by fluorography, after separation by SDS-PAGE. Myristoylation is no more observed in the *myr*<sup>-</sup> mutant. (C) Western blot analysis of whole-cell lysates or cell supernatants from 293T cells transfected as in (B) with the indicated IAPE constructs, using a rabbit anti-serum directed against the IAPE Gag polyprotein. Molecular weights of the Gag-specific bands are indicated, as in Fig. 3C. Lack of myristoylation in the mutant results in the absence of mature Gag in the supernatant. (D) Electron microscopy of 293T cells transfected with the IAPE *myr*<sup>-</sup> mutant disclosing absence of IAPE particle assembly at the plasma cell membrane, and particles –of an immature morphology– accumulating as aggregates in the cytoplasm (Nu : nucleus, M : mitochondria, Pm : plasma membrane, ERm : endoplasmic reticulum membrane).



**Fig. S3**

**Table S1 : Primer list**

| Purpose                             | Primer name         | Primer sequence                                          |
|-------------------------------------|---------------------|----------------------------------------------------------|
| Cloning of IAPE-D Env               | IAPE-D Env f        | 5' -ATACACTCGAGCGGAACGACTGACACGGAGA-3'                   |
|                                     | IAPE-D Env r        | 5' -ATACACTCGAGGCGGGAAATAAACACAGGACA-3'                  |
| Mutagenesis of IAPE-D1 Env          | IAPE-D1 Env AQAR f  | 5' -CTTCCACCATAACCTTACACGCACAAGCACGAGACTTTGGTATCACTGC-3' |
|                                     | IAPE-D1 Env AQAR r  | 5' -GCAGTGATACCAAAGTCTCGTGCTTGTGCGTGTAAGGTTATGGTGGAAG-3' |
| Mutagenesis of IAPE-D2 Env          | IAPE-D2* Env f      | 5' -GGCAGCTACAGCTGCTGGAATTGCTATGGCAACAT-3'               |
|                                     | IAPE-D2* Env r      | 5' -ATGTTGCCATAGCAATTCAGCAGCTGTAGCTGCC-3'                |
| Cloning of IAPE-D proviruses        | IAPE-D R-U5 f       | 5' -ATACAGAGCTCGTTTAGTGAACCGTCTTTGTCTACCTCAATATG-3'      |
|                                     | IAPE-D R-U5 r       | 5' -ATACACTCGAGGCACCAAGTATCCCGAGCTTATC-3'                |
| Mutagenesis of IAPE-D1 Gag          | IAPE-D1 myr f       | 5' -TAGATAAGGGAGCAGTATGGTAAATTCACATTCTGTAACATA-3'        |
|                                     | IAPE-D1 myr r       | 5' -TAGTTACAGAATGTGAATTTACCATACTGCTCCCTTATCTA-3'         |
| Cloning of IAPE-D1 CTE              | IAPE-D1 CTE(7447) f | 5' -ATACAAGATCTACTAGTGTTCCTTCTATGGC-3'                   |
|                                     | IAPE-D1 CTE(8182) r | 5' -ATACAGGTACCACGTGTCTAAACTAGGACTG-3'                   |
|                                     | IAPE-D1 CTE(7684) f | 5' -ATACAAGATCTGAGGTTTCCTCATTGCATGT-3'                   |
|                                     | IAPE-D1 CTE(8050) r | 5' -ATACAGGTACCCGCACAAAGAGGTTTAGCAC-3'                   |
|                                     | IAPE-D1 CTE(7717) f | 5' -ATACAAGATCTTGATTTCCTCCACTAATTCAT-3'                  |
|                                     | IAPE-D1 CTE(8009) r | 5' -ATACAGGTACCTGGCAGCCCTTTACATCTCC-3'                   |
|                                     | IAPE-D1 CTE(7753) f | 5' -ATACAAGATCTTTGTAGAGTTCACCCTAGGC-3'                   |
|                                     | IAPE-D1 CTE(7973) r | 5' -ATACAGGTACCAAAATGAGGCTGGTCTAGGC-3'                   |
| Characterization of insertion sites | IAPE-D1 nested f1   | 5' -TTTCCTCATGCACAGACCAACCT-3'                           |
|                                     | IAPE-D1 nested r1   | 5' -GTCATCTCACCTTGCTCCTGCCGA-3'                          |
|                                     | IAPE-D1 nested f2   | 5' -TGGAGGAACGACCTAAACAGGG-3'                            |
|                                     | IAPE-D1 nested r2   | 5' -AAACATCGCATCGAGCGAGCACGTA-3'                         |
|                                     | Locus 8q12 f        | 5' -ACGTTTTGCAGCCTTTCTCC-3'                              |
|                                     | Locus 8q12 r        | 5' -AAAGGAAAGGAGGCCAGTGT-3'                              |
|                                     | Locus 2q37 f        | 5' -AGGGAGGAGGCAGGACAACA-3'                              |
|                                     | Locus 2q37 r        | 5' -AGAGAAAATGCAAACGCAAC-3'                              |
|                                     | Locus 1p22 f        | 5' -AGGACCAAAGCAAGGAATGA-3'                              |
|                                     | Locus 1p22 r        | 5' -GGAGAAAACATGGGAGCCAG-3'                              |