Treatment of Advanced Neuroblastoma with EBV-Specific T-Lymphocytes Expressing a Chimeric Anti-GD2 Single Chain Antibody

Helen Heslop MD

Heidi Russell MD Pediatric Oncologist

Claudia Rossig MD Preclinical data

Martin Pule MD Preclinical data

Bambi Grilley RPh Regulatory Affairs

Neuroblastoma

- Cancer of childhood
- Approximately 550 new cases in the US each year
- Tumor cells are primitive neural crest cells that populate the sympathetic nervous system

Neuroblastoma

- Approximately ½ of children with neuroblastoma will have widespread disease
- Treatment with multi-agent chemotherapy, surgery, radiation, and autologous transplant only results in 30% EFS at 3 years
- Long term sequelae in survivors including secondary cancers (epithelial)

Neuroblastoma

- Currently no standard therapy for children who have recurrent or refractory neuroblastoma
- 21 open or developing NIH sponsored clinical trials including investigational drugs, immunotherapeutics, transplant

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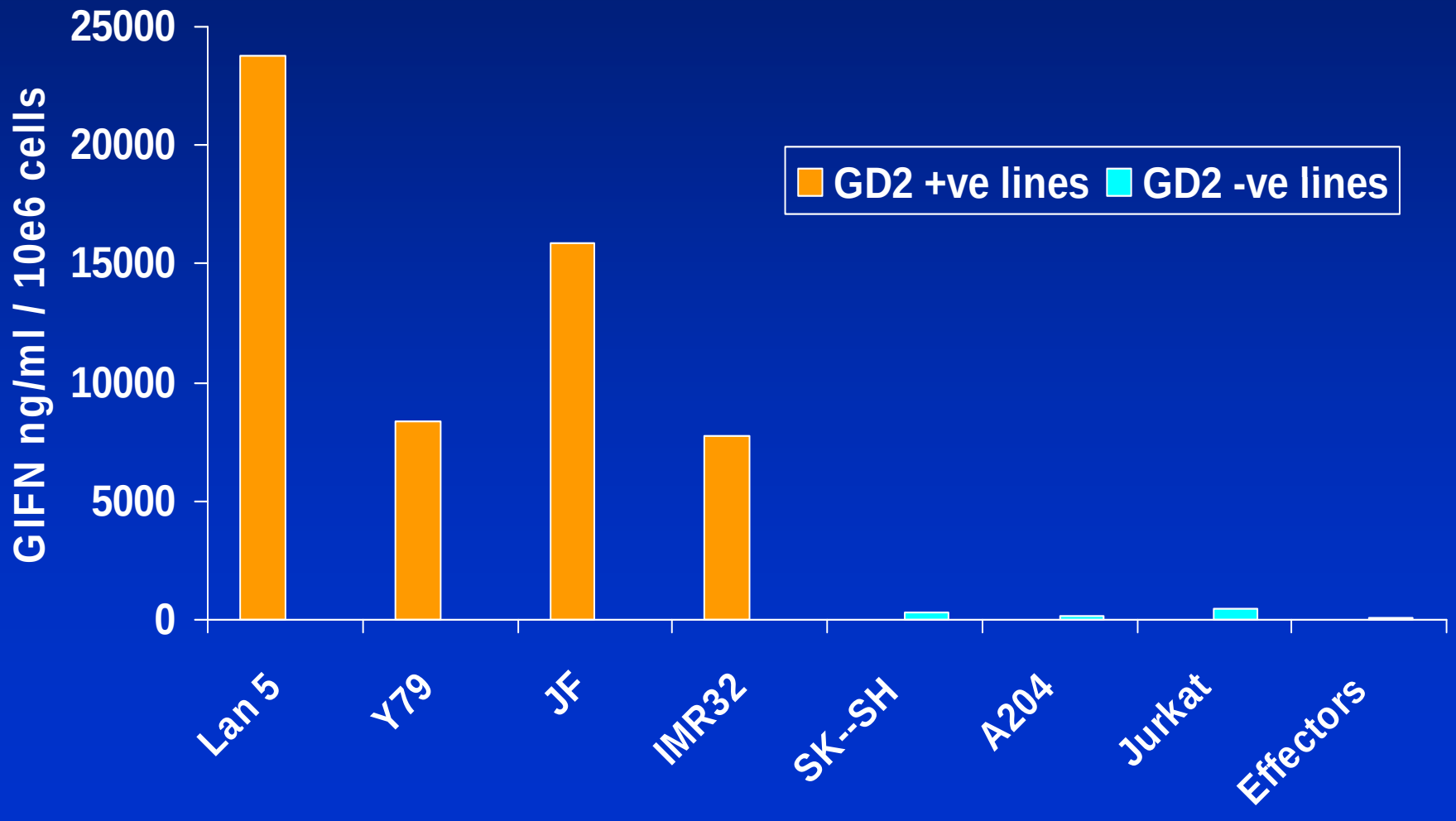
IFN-γ Release of 14.G2a-ζ Transduced PBMC Upon Coincubation With G_{D2}^{+} and G_{D2}^{-} Target Cells



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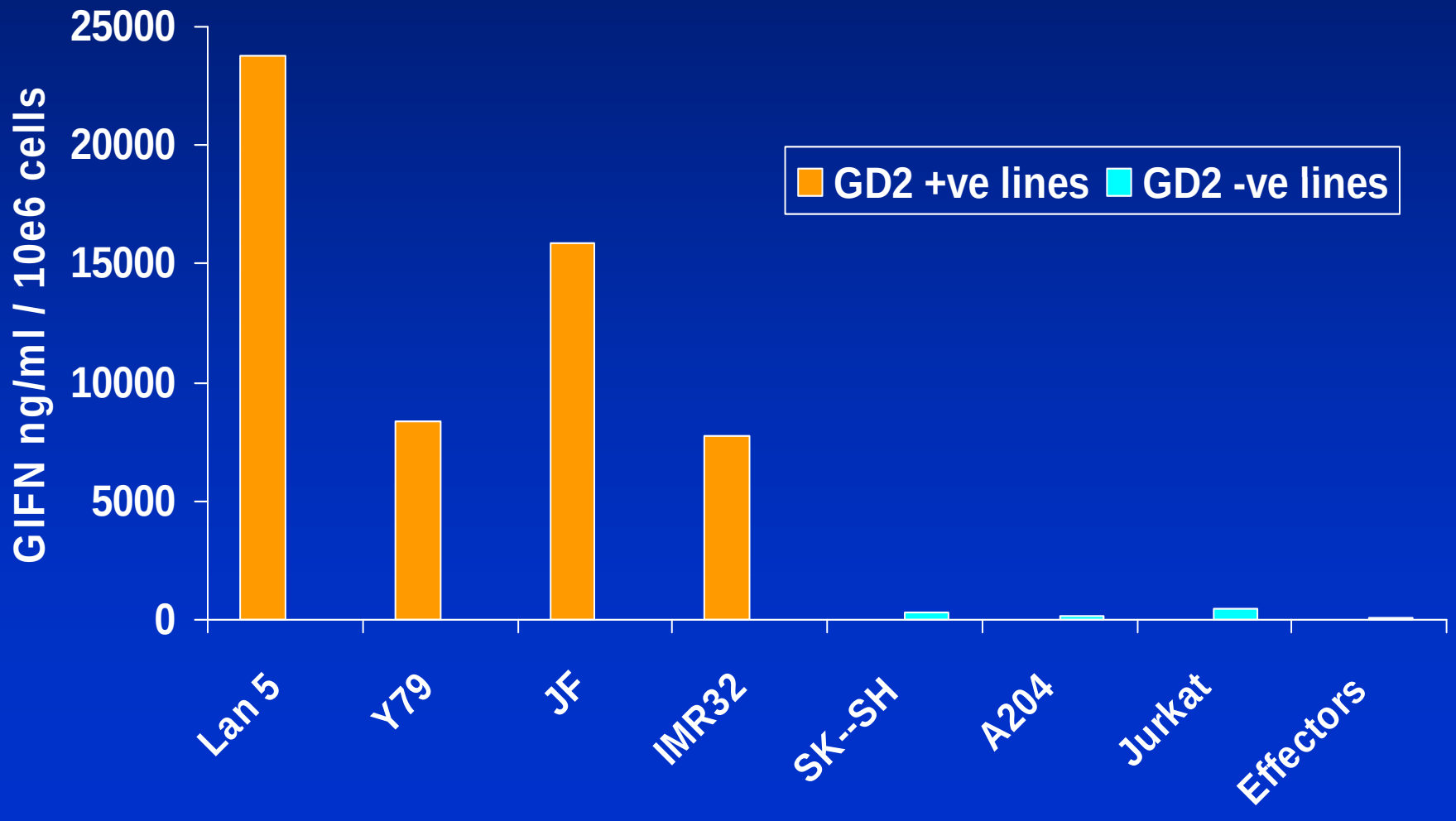

Limitations of Chimeric T cells

- Poor activation through chimeric receptor pathway and signals through TCR-zeta chain alone are insufficient to prime resting T-cells *(Brocker T, Karjalainen K. J, Exp Med 1995; Brocker R, Blood 2001)*
- Lack of cognate help/co-stimulator molecules on tumor cells
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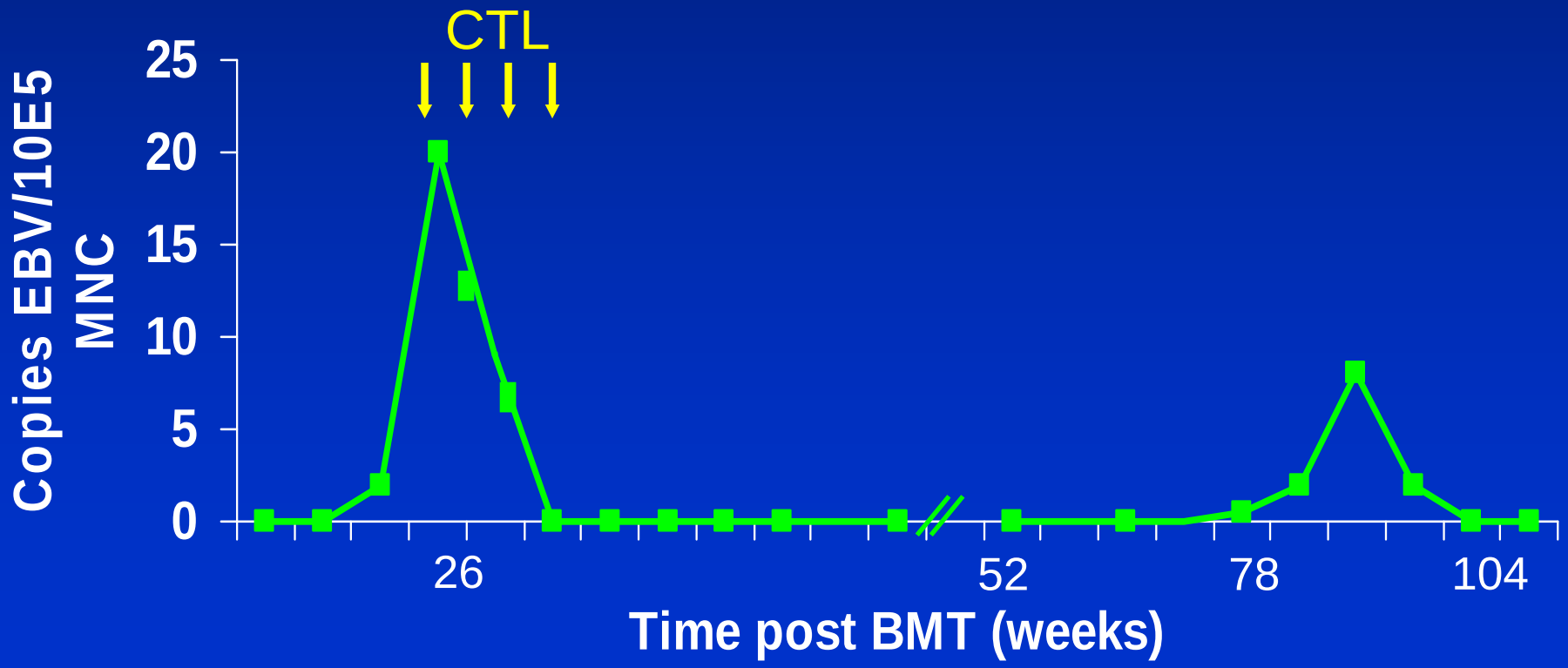
Experience with gene-marked EBV-specific CTLs

- Prevention and treatment of EBV-LPD post transplant
  - 62 patients received CTLs
  - Marking with neomycin resistance gene in first 26 patients
- Relapsed EBV+ve Hodgkin's disease
  - 13 patients treated
  - Gene marking in 7

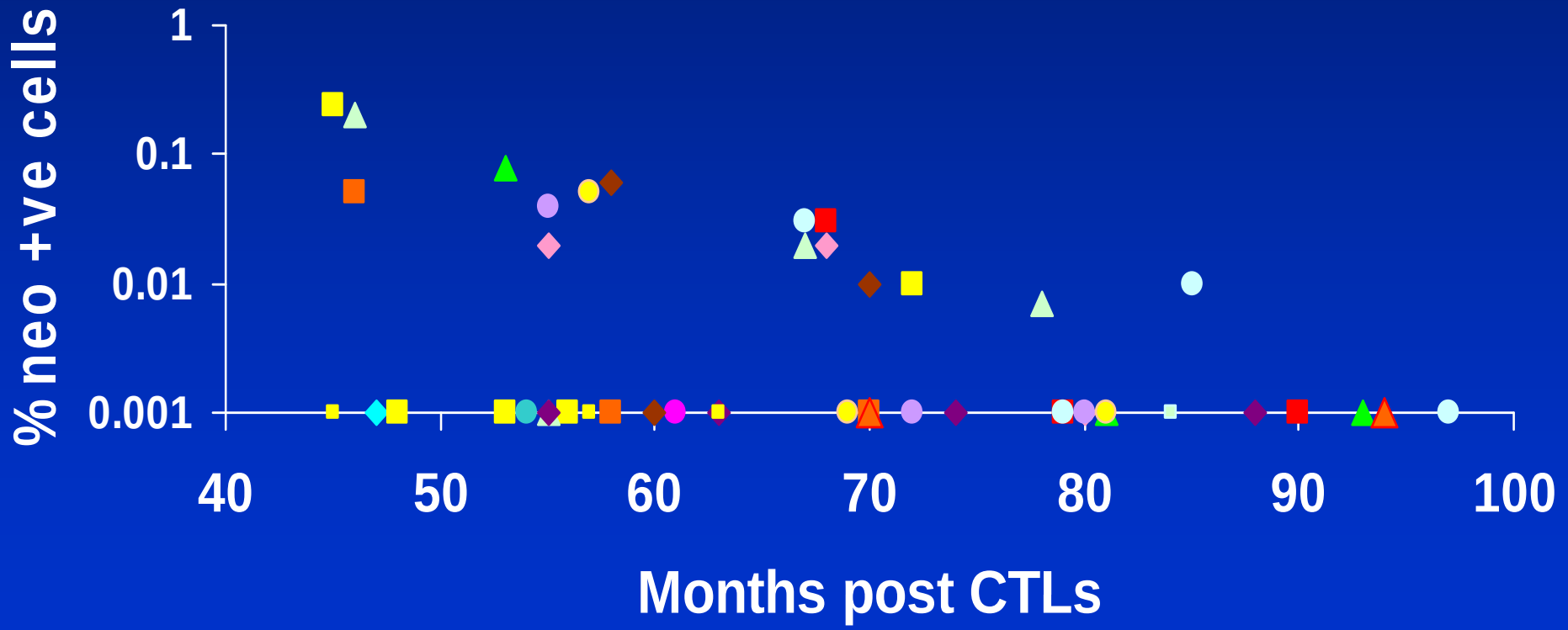
Late Reactivation of EBV

EBV DNA versus neo DNA

Neo DNA



Long Term Detection of Neo by Real Time PCR in PBM EBV CTL Study



Multiple Clones Persist

1) Multiple Vß subclasses on immunophenotyping

2) Multiple marked clones on inverse PCR

Markers

-ve control

+ve control

Patient 1

Patient 2

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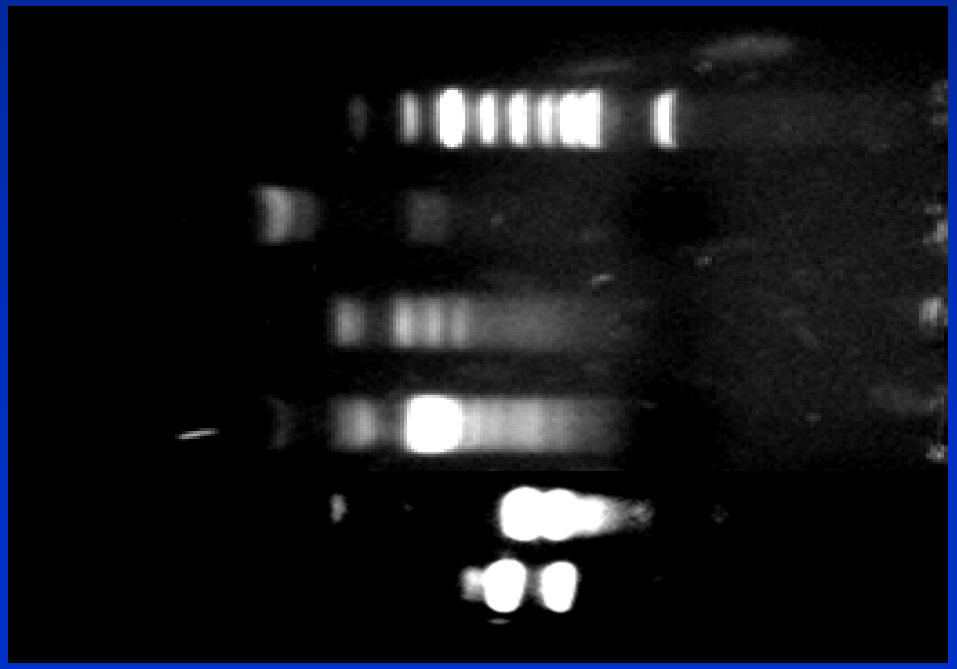

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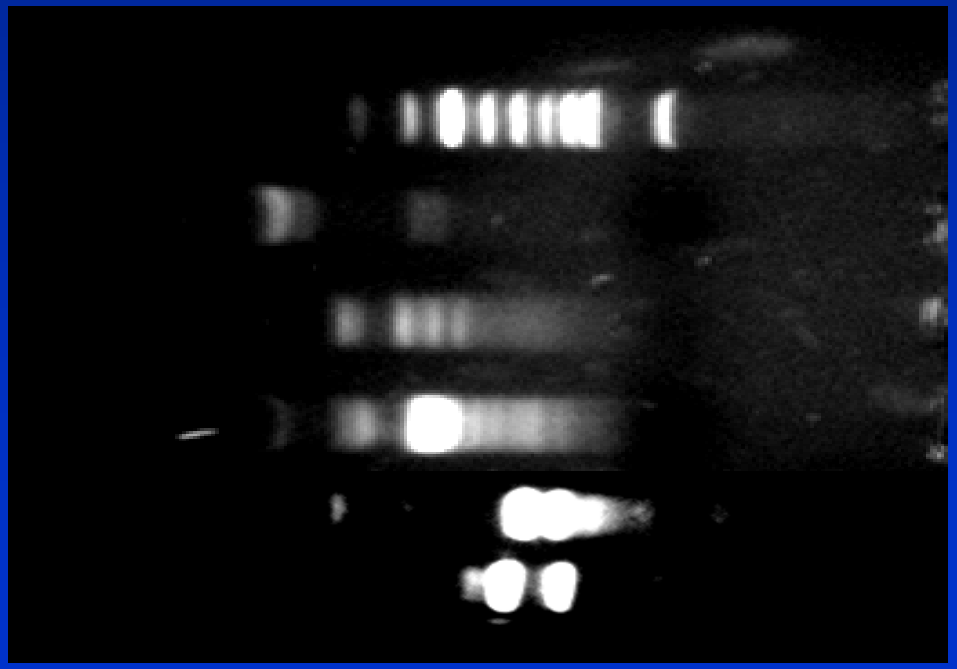

Human DNA involved in the virus integration site

Sequence analyses show non-coding or unknown sequences

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CGACAAATTCTATTAATAGCTTAAATGGGATT

Inv 5 chromosome 15 RP11-573G7 109830-109906

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Unknown sequence from 1 band

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AAATGCAAGAACAGTGTTCCCTTCAAGCCAGACTACATCCTGACTCTCGGCTTTATAAAAGAATGTTGAAGGGCTCTGT
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- Longstanding safety and efficacy data with EBV CTL in humans
- Clinical experience with 14G2a MAb
- Preclinical safety data with GD2 chimeric EBV CTLs
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