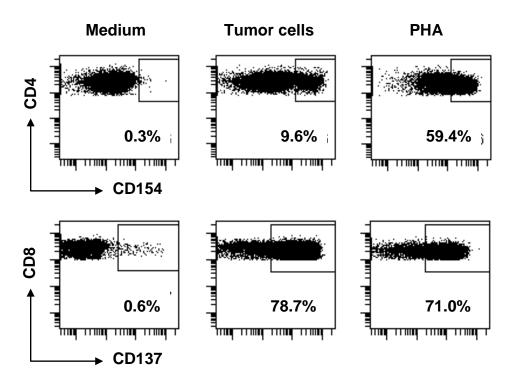
## Successful treatment of metastatic melanoma by adoptive transfer of blood-derived polyclonal tumor-specific CD4+ and CD8+ T cells in combination with low dose Interferon- $\alpha$ .

## Els M.E. Verdegaal et al <u>supplementary material</u>

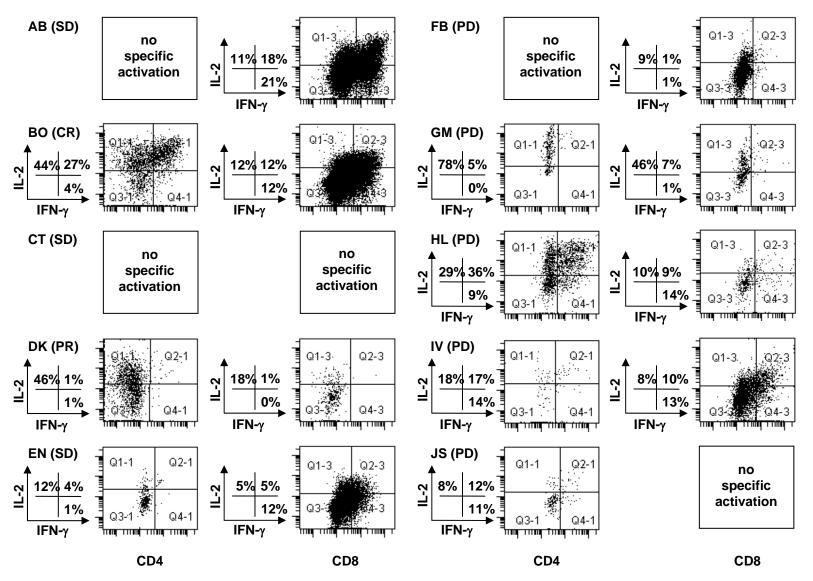
## Supplementary Table 1 Serious adverse events

| Patient | Serious adverse event <sup>a</sup>  | Relation to disease and treatment   |
|---------|---|---|
| AB      | None  | n.a.  |
| ВО      | None  | n.a.  |
| СТ      | Necrotic finger tip lesions, folliculitis-like inflammation of the skin   | Possibly related to treatment   |
| DK      | None  | n.a.  |
| EN      | Bleeding of brain metastasis  | Related to disease but not to treatment   |
| FB      | Addisonse crisis  | Related to disease but not to treatment   |
| GM      | Atrial fibrillation and dyspnoea  | Related to disease but not likely related to treatment  |
| HL      | None  | n.a.  |
| IV      | Abdominal pain due to constipation, cerebellar edema Edema of arm due to s.c injection of part of the T cell infusion | Related to disease but not to treatment Treatment related, patient was hospitalized one night for observation |
| JS      | None  | n.a.  |

<sup>&</sup>lt;sup>a</sup> All serious adverse events, both disease related and (possibly) treatment related that required hospitalization are described.



**Supplementary Figure. 1** Analysis of tumor-reactivity of infused T cells. T cell batches were stimulated with autologous tumor cells and stained for phenotypic and activation markers as described in the Methods section. Representative dot plots (gated on CD3+ cells) for CD154+CD4+ (upper panels) and CD137+CD8+ (lower panels) T cells are shown. Medium controls (left panels) were included as reference and used to adjust the gate settings. Responses after stimulation with autologous tumor cells (middle panels) were considered positive when the percentage of tumor-stimulated cells was at least three times the medium control. T cells stimulated with PHA (5 mg/ml, right panels) were included as positive controls to check for T cell responsiveness



**Supplementary Figure. 2** Cytokine production of tumor-reactive T cells. T cell batches were stimulated with autologous tumor cells and stained for phenotypic and activation markers and for IFN-gamma and IL-2, as described in the Methods section. Tumor-reactive cytokine producing T cells gated on CD3+CD4+CD154+ and CD3+CD8+CD137+ T cells are plotted and the percentage cytokine positive cells (i.e. 3 times above background in medium alone) are given in the squares on the left of each dot plot. Left from the dot plots the corresponding patient ID and best overall clinical response between brackets is given