**Supplementary Figure Legends**

**Supplementary Figure 1: Arginase II is the principal arginase isoform in neuroblastoma**

**a)** T cell CD3 expression is downregulated following culture with neuroblastoma cell line conditioned supernatant or arginine free media. **b)** Addition of 1.25g/ml exogenous arginine to cell line condition supernatants moderately restores T cell proliferation. **c)** Arginase II and iNOS expression was determined by RT-PCR in neuroblastoma cell lines. GAPDH was used as the housekeeping gene to ensure equal loading **d)** Arginase II expression was determined by western blotting of whole cell lysates. Actin was used as the housekeeping gene to ensure equal loading **e)** Absence of Arginase I expression was determined by western blotting of whole cell lysates. Liver extract was used as a control. GAPDH was used as the housekeeping gene to ensure equal loading. **f)** Absence of iNOS expression was determined by western blotting of whole cell lysates. RAW cells treated with LPS were used as a control. Actin was used as the housekeeping gene to ensure equal loading.  **g)** siRNA silencing of Arginase II in Kelly cell line. **h)** siRNA silencing of Arginase II in Kelly line restores T cell proliferation.

**Supplementary Figure 2: Arginase II expression by neuroblastoma tumours creates a suppressive microenvironment**

**a)** Staining of sections from neuroblastoma tumours at diagnosiswith DAPI alone (left), anti-human GD2(left middle), anti-arginase II (middle), anti-arginase I (middle right), and iNOS (right). Representative sections from 3 neuroblastoma patients shown. **b)** No significant difference in the arginase activity from plasma of 16 newly diagnosed neuroblastoma patients and 10 healthy donors **c)** Neuroblastoma plasma inhibits T cell proliferation. Data are representative of two independent experiments (error bars, SD). **d)** T cell CD3 expression is downregulated following culture with neuroblastoma patients’ plasma or arginine free media. **e)** Addition of 1.25g/ml exogenous arginine to patient plasma moderately restores T cell proliferation (p=0.0038 plasma vs arginine supplemented).

**Supplementary Figure 3: Neuroblastoma alters myeloid cell frequency in a murine model**

**a)** Arginase I, II and iNOS expression was determined by western blotting of whole cell lysates of sorted murine GD2+ cells. Actin was used as the housekeeping gene to ensure equal loading. **b)** The frequency of F4/80+ (top), Ly6G+ (middle), Ly6C+ (middle), and CD3+ (bottom) cells in the spleens of tumour bearing and control mice were examined by flow cytometry. **c)** The frequency of F4/80+ (top), Ly6G+ (middle), Ly6C+ (middle), and CD3+ (bottom)cells in the blood of tumour bearing and control mice were examined by flow cytometry

**Supplementary Figure 4: Neuroblastoma arginase inactivates CD14+ cells and CAR T cells**

**a)** CD14+ cells cultured with neuroblastoma or arginine deplete media had significant decreases in LPS-driven IL-12 release. All data are representative of three independent experiments (error bars, SD). **b)** No increase in the expression of CD86 on CD14 cells cultured in the presence of neuroblastoma cell lines or arginine deplete medium for 72hours, before stimulation with LPS. All data are representative of two independent experiments (S1 and S2) (error bars, SD). **c)** Myeloid populations in the blood of neuroblastoma patients, gated by FSC/SSC, PI staining for live cells, and CD14 or CD15 staining **d)** Anti-GD2 CAR T cell proliferation is rescued by arginase inhibition. Data are representative of 2 independent experiments.

**Supplementary Figure 5: Arginase 2 and CD14 expression are associated with a worse overall survival.**

**a)** Gene expression analysis of arginase 1 and 2, in neuroblastoma tumours, divided according to INSS stage. Arginase 2 has higher expression that arginase 1 in all stages of neuroblastoma, except Stage 4s.**b)** Kaplan-Meyer curve for neuroblastoma patients, divided according to Arginase 2 expression. High arginase 2 expression is associated a worse overall survival. **c)** Kaplan-Meyer curve for neuroblastoma patients, divided according to CD3 expression. Low CD3 expression is associated a worse overall survival. **d)** Kaplan-Meyer curve for neuroblastoma patients, divided according to CD14 expression. Low CD14 expression is associated a worse overall survival. All data are analysed in accordance with the public Versteeg database ‘R2: microarray analysis and visualization platform’ (http://r2.aml.nl).