

Supplementary material

Renal carcinoma CD105-/CD44- cells display stem-like properties *in vitro* and form aggressive tumors *in vivo*

Fiedorowicz M.^{1,*,#}, Khan M.I.^{2,3,#}, Strzemecki D.¹, Orzeł J.^{1,4}, Wełniak-Kamińska M.¹, Sobiborowicz A.^{5,6}, Wieteska M.^{1,4}, Rogulski Z.⁷, Cheda L.⁷, Wargocka-Matuszewska W.⁷, Kilian K.⁸, Szczylik C.^{2,9,10}, Czarnecka A.M.^{1,2,11}

¹ Mossakowski Medical Research Centre, Polish Academy of Sciences, Warsaw, Poland

² Department of Oncology with Laboratory of Molecular Oncology, Military Institute of Medicine, Warsaw, Poland

³ Current address: Department of Otolaryngology - Head & Neck Surgery, Western University, London, ON, N6A 3K7, Canada.

⁴ Faculty of Electronics and Information Technology, Warsaw University of Technology, Warsaw, Poland

⁵ Faculty of Medicine, Medical University of Warsaw, Warsaw, Poland

⁶ Department of Soft Tissue/Bone Sarcoma and Melanoma, Maria Skłodowska-Curie Memorial Cancer Center and Institute of Oncology, Warsaw, Poland

⁷ Faculty of Chemistry, Biological and Chemical Research Centre, University of Warsaw, Warsaw, Poland

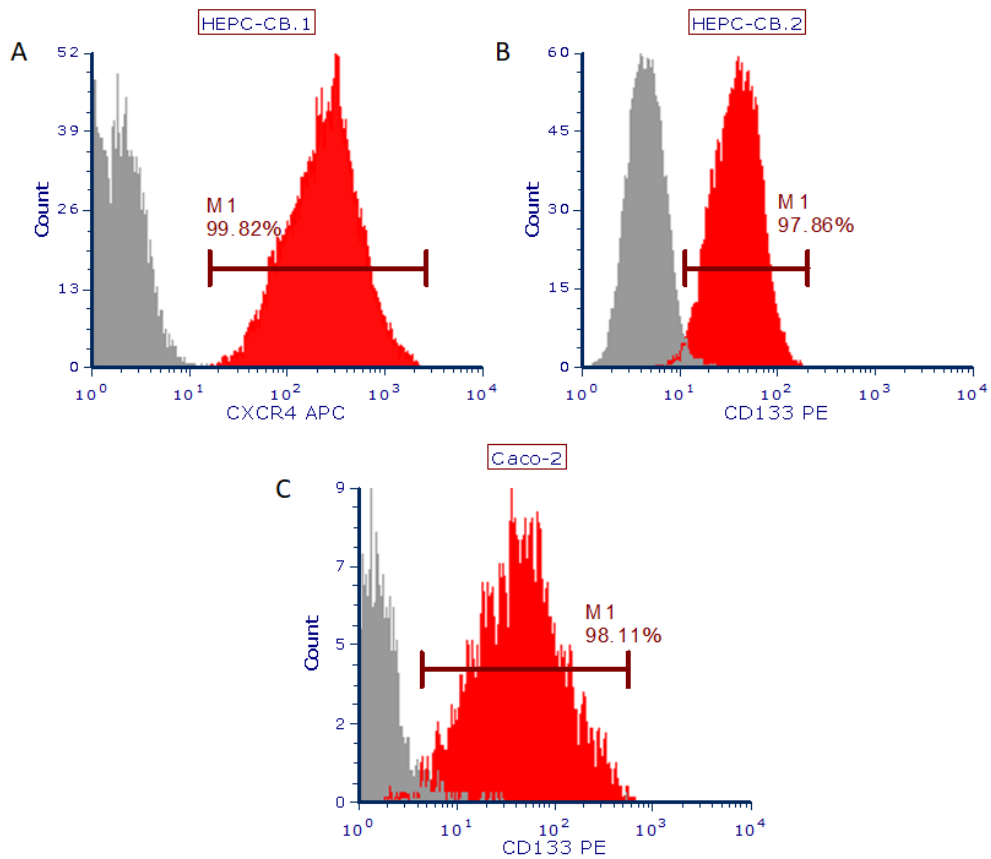
⁸ Heavy Ion Laboratory, Faculty of Physics, University of Warsaw, Warsaw, Poland

⁹ Current address: Department of Oncology, European Health Centre, Otwock, Poland.

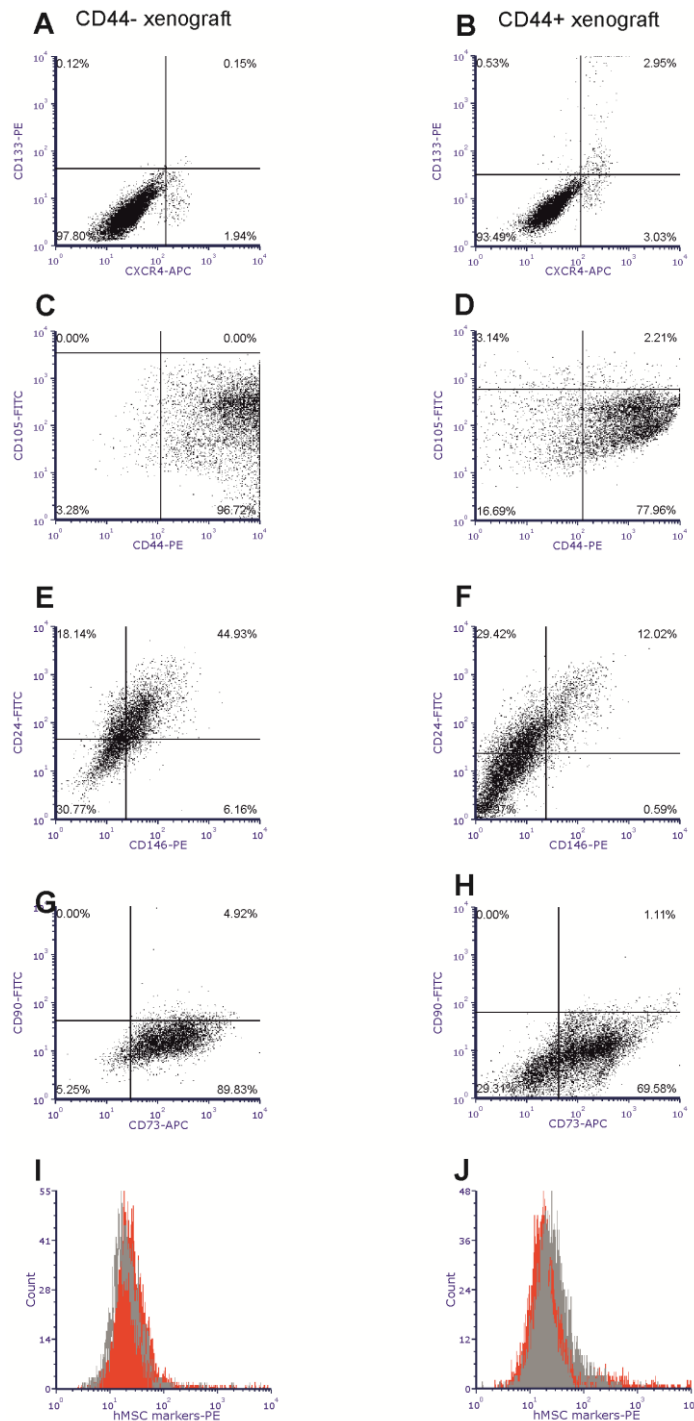
¹⁰ Current address: Medical Center for Postgraduate Education, Warsaw, Poland.

¹¹ Current address: Department of Soft Tissue/Bone Sarcoma and Melanoma, Maria Skłodowska-Curie Memorial Cancer Center and Institute of Oncology, Warsaw, Poland

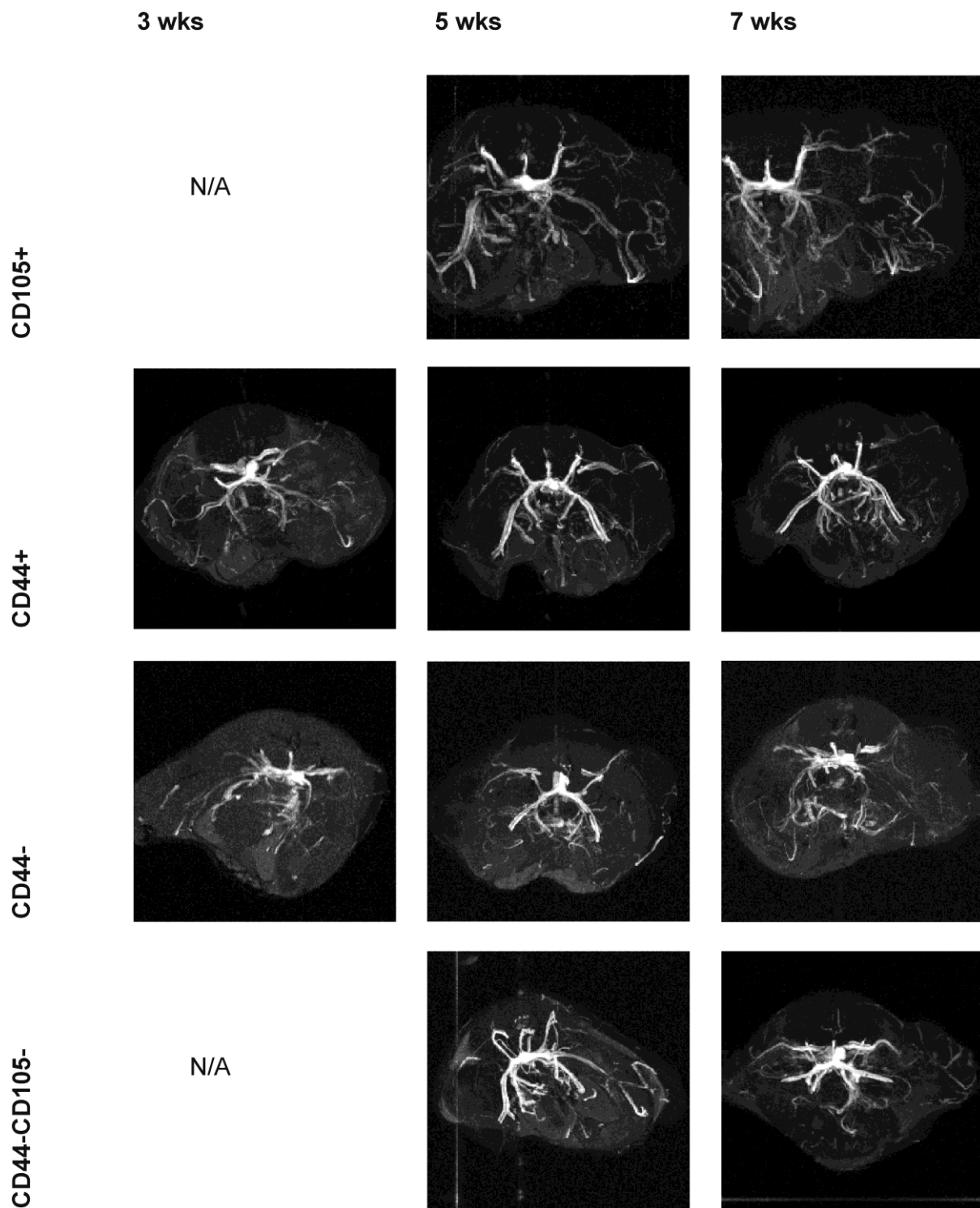
*Correspondence: mfiedorowicz@imdik.pan.pl



Supplementary Figure 1 – Control cell lines used for expression of CD133 and CXCR4 surface markers. Red histograms showing expression of **(A)** CXCR4 in HEPC-CB.1 cells, **(B)** CD133 in HEP-CB.2 cells, and **(C)** CD133 in Caco-2 cells. Grey histograms represent unstained cells.



Supplementary Figure 2 – Dot plots showing re-analysis of cancer stem cells (CSC) and human mesenchymal stem cells (hMSC) markers in CD44^{-/-} and CD44^{+/-} xenografts. Figure **(A)** and **(B)** showing dual expression analysis of CSCs markers (CD133 and CXCR4) in CD44^{-/-} and CD44^{+/-} xenografts, respectively. Figure **(C)** and **(D)** showing dual expression analysis of CSCs markers (CD105 and CD44) in CD44^{-/-} and CD44^{+/-} xenografts, respectively. Figure **(E)** and **(F)** showing dual expression analysis of hMSCs markers (CD24 and CD146) in CD44^{-/-} and CD44^{+/-} xenografts, respectively. Figure **(G)** and **(H)** showing dual expression analysis of hMSCs markers (CD90 and CD73) in CD44^{-/-} and CD44^{+/-} xenografts, respectively. Red histograms **(I)** and **(J)** showing analysis of hMSCs markers (CD34, CD45, CD11b or CD14, CD19 or CD79 α) in CD44^{-/-} and CD44^{+/-} xenografts, respectively. Grey histograms represent unstained cells.



Supplementary Figure 3 - MR *time of flight* angiography in tumors at different timepoints after implantations of various subpopulations of Caki1F cells (3, 5 and 7 weeks after implantation). N/A – tumors not detected in most of the animals, no representative MR angiography.