

Original Article

Oncogene (2008) 27, 3923–3934; doi:10.1038/onc.2008.38; published online 10 March 2008

Cisplatin treatment increases survival and expansion of a highly tumorigenic side-population fraction by upregulating VEGF/Flt1 autocrine signaling

R Tsuchida^{1,8}, B Das^{1,2,8}, H Yeger^{3,4}, G Koren², M Shibuya⁵, P S Thorner^{3,4}, S Baruchel^{1,2,6} and D Malkin^{1,2,6,7}

¹Division of Hematology/Oncology, Department of Paediatrics, The Hospital for Sick Children, Toronto, Ontario, Canada

²Institute of Medical Science, University of Toronto, Toronto, Ontario, Canada

³Division of Pathology, The Hospital for Sick Children, Toronto, Ontario, Canada

⁴Department of Laboratory Medicine and Pathobiology, University of Toronto, Toronto, Ontario, Canada

⁵Department of Genetics, Institute of Medical Science, University of Tokyo, Division of Genetics, Tokyo, Japan

⁶Department of Paediatrics, University of Toronto, Toronto, Ontario, Canada

⁷Department of Medical Biophysics, University of Toronto, Toronto, Ontario, Canada

Correspondence: Dr D Malkin, Division of Hematology/Oncology, Department of Paediatrics, The Hospital for Sick Children, 555 University Avenue, Toronto, Ontario, Canada M5G 1X8. E-mail: david.malkin@sickkids.ca

⁸These authors contributed equally to this work.

Received 15 August 2007; Revised 28 November 2007; Accepted 21 December 2007; Published online 10 March 2008.

Abstract

The cellular and molecular mechanisms of tumor progression following chemotherapy are largely unknown. Here, we demonstrate that cisplatin (CDDP) treatment upregulates VEGF and Flt1 expression leading to the survival and expansion of a highly tumorigenic fraction of side-population (SP) cells in osteosarcoma (HOS), neuroblastoma (SK-N-BE2) and rhabdomyosarcoma (RH-4) cell lines. In all three lines, we show that CDDP treatment increases levels of VEGF and Flt1 expression, and induces enhanced clonogenic capacity and increased expression of the 'stemness'-associated genes Nanog, Bmi-1 and Oct-4 in the SP fraction. In HOS, these changes are associated with the transformation of a non-tumorigenic osteosarcoma SP fraction to a highly tumorigenic phenotype. Inhibition of Flt1 led to complete reduction of tumorigenicity in the HOS SP fraction, and reduction of clonogenic capacity and expression of stemness genes in the SK-N-BE(2) and RH-4 SP fractions. Treatment with U0126, a specific inhibitor of MAPK/ERK1,2 completely downregulates CDDP-induced VEGF and Flt1 expression and induction/expansion of SP fraction in all three cell lines, indicating that these effects are mediated through MAPK/ERK1,2 signaling. In conclusion, we report a novel mechanism of CDDP-induced tumor progression, whereby the activation of VEGF/Flt1 autocrine signaling leads to the survival and expansion of a highly tumorigenic SP fraction.

Keywords: osteosarcoma, cisplatin, tumorigenic SP, VEGF/Flt1 autocrine