

# Proteomic Analysis of Exosomes and Exosome-Free Conditioned Media From Human Osteosarcoma Cell Lines Reveals Secretion of Proteins Related to Tumor Progression

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© 2016 Wiley Periodicals, Inc. Osteosarcomas are the most prevalent bone tumors in pediatric patients, but can also occur later in life. Bone tumors have the potential to metastasize to lung and occasionally other vital organs. To understand how osteosarcoma cells interact with their micro-environment to support bone tumor progression and metastasis, we analyzed secreted proteins and exosomes from three human osteosarcoma cell lines. Exosome isolation was validated by transmission electron microscopy (TEM) and immuno-blotting for characteristic biomarkers (CD63, CD9, and CD81). Exosomal and soluble proteins (less than 100 kDa) were identified by mass spectrometry analysis using nanoLC-MS/MS and classified by functional gene ontology clustering. We identified a secretome set of >3,000 proteins for both fractions, and detected proteins that are either common or unique among the three osteosarcoma cell lines. Protein ontology comparison of proteomes from exosomes and exosome-free fractions