**Nutlin-3 Treatment Attenuates Cisplatin-induced Inhibition Of Bone Healing: Implications For Osteosarcoma Surgery**

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**Disclosures:**


**Introduction:** Osteosarcoma (OS) is the most common primary bone cancer that affects children and adolescents. The majority of patients are treated by a closely timed combination of chemotherapy and surgery, yet these patients continue to be at risk for postoperative complications especially associated with decreased bone repair. Cisplatin (CDP) is a commonly used OS chemotherapy, yet the effects of CDP on bone repair are not well understood. CDP exposure can occur preoperatively, postoperatively, and even simultaneously with bone repair in OS surgery. We plan to demonstrate that the inhibition of bone repair (during distraction osteogenesis; DO) following CDP treatment can be protected by the administration of a small molecule (nutlin-3) but also demonstrate that Nutlin-3 potentiates the effects of CDP. Nutlin-3 is a p53 inducer that has been found to 1) reversibly induce cell arrest in non-cancerous bone progenitor cells, and 2) be toxic to a wide range of human osteosarcoma cells.

**Methods:** In Study 1, 9-week-old C57BL/6 male mice received either CDP + vehicle (n=10) or CDP + nutlin-3 (n=10). All mice received an IP injection of either vehicle (50% DMSO) or nutlin-3 (20mg/kg) on day 1, 3, and 4 prior to surgery. The day after the first nutlin injection, all mice received an IP injection of CDP (2mg/kg/day) for two days (day 2 and 3 prior to surgery). On the fourth day after the second injection of CDP, the mice underwent placement of an external fixator and osteotomy to the left tibia. DO began three days after surgery at a rate of 0.075 mm b.i.d for 11 days. At sacrifice both the distracted and contra lateral tibiae were harvested. Animal weights at sacrifice were not significantly different.

In Study 2, 9-week-old C57BL/6 male mice received either CDP + vehicle (n=10) or CDP + nutlin-3 (n=10). All mice received an IP injection of either vehicle (50% DMSO) or nutlin-3 (20mg/kg) on day 1, 3, and 4 prior to surgery. The day after the first nutlin injection, all mice received an IP injection of CDP (2mg/kg/day) for two days (day 2 and 3 prior to surgery). On the fourth day after the second injection of CDP, the mice underwent placement of an external fixator and osteotomy to the left tibia. DO began three days after surgery at a rate of 0.075 mm b.i.d for 11 days. At sacrifice both the distracted and contra lateral tibiae were harvested. Animal weights at sacrifice were not significantly different.

**Results:** In study 1, comparison of the distracted tibia (radiographs and microCT) demonstrated a significant increase in the mineralized area of distraction gaps of CDP/nutlin-3 treated (52.8% ± 3.7) versus CDP treated mice (30.9% ± 6.5) mice (P<0.010). Histological analysis of the DO gaps confirmed the significant increase in bone formation in CDP/nutlin-3 (79.8% ± 2.1) versus CDP (51.3% ± 4.6) (P<0.001). In study 2, both CDP and CDP + nutlin-3 treatment resulted in a significant 67% (P<0.002) and 78% (P<0.001) respectively inhibition of tumor growth relative to untreated vehicle controls.

**Discussion:** Collectively, these results demonstrate that an intervention involving the induction of p53, prior to surgery protects mesenchymal osteoprogenitors from the toxic effects of CDP, thereby facilitating bone healing, while maintaining the anti-tumor effect of CDP treatment. This scenario bodes well for the treatment of OS while allowing full potential for bone healing post-surgery.

**Significance:** This study provide relevant and appropriate preclinical data for the prevention of the negative effects of chemotherapy on bone healing and repair.

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**References:**

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