

**EXPRESSION AND RELEASE OF HEAT SHOCK PROTEIN-60
(HSP60) FROM HUMAN OSTEOBLASTS**

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ABSTRACT

Thermal trauma from orthopedics and oral bone surgery can affect bone healing process. Although many evidences show that heat above 47°C cause irreversibly damage to bone cells and cell death, but how this event contributes to bone healing is largely unknown. The aim of this study was to determine the effects of heat stress on normal human osteoblasts and the synthesis of Heat Shock Protein-60 (HSP60) in normal human osteoblast. To identify the level of lethal heat stress, we exposed normal human osteoblasts to heat stress from 40°C to 46°C and analyzed by MTT assay. The results showed the significant cell death after treatment at 43°C to 46°C for 5 to 30 min in dose- and time-dependent manner. To test whether heat stress could induce HSP60 expression and release, we analyzed the total protein lysate and conditioned media using Western blot analysis at 24 hr after treatment. We found that heat stress from 40°C to 46°C did not induce HSP60 expression and release from normal human osteoblasts. Immunocytochemistry analysis also showed the modest changes in subcellular localization of HSP60 after heat treatment. Finally, we demonstrated the release of HSP60 from necrotic normal human osteoblast after freezing and thawing process. We concluded that hyperthermia at as low as 43°C for 5 min could cause cell death. However, necrotic human osteoblasts, but not heat-treated cells, release HSP60 into the extracellular milieu.