

Morphological Observation of the Cashmere goat fetal fibroblast after mTOR kinase inhibition with combination of fluorescent dyes and confocal cell imaging

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ABSTRACT The mammalian target of rapamycin (mTOR) is a kind of Ser/Thr kinase in mammalian cells. It can recruit and integrate input signals from nutrients, growth factors, energy and environmental stress to regulate cell growth and proliferation via different cellular processes. The mTOR signaling pathway has been investigated extensively in human and some animals, whereas it is not yet studied in goat. Thus this study uses the fetal fibroblast of

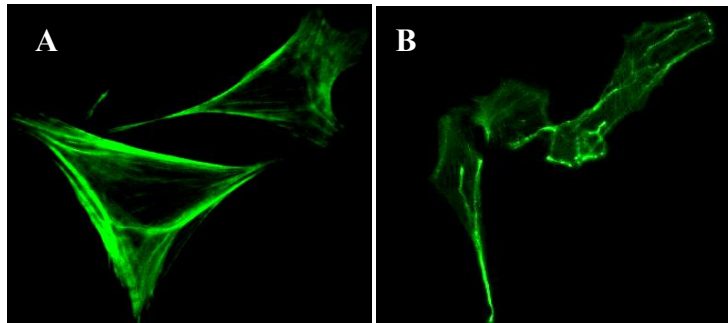


Figure 1: Confocal images of F-actin in GFb cells treated with 20 $\mu\text{mol/L}$ CCI-779 for 48 h.

A control (182.4 μm ×182.4 μm)

B treatment with 20 $\mu\text{mol/L}$ CCI-779 for 48h (216.5 μm ×216.5 μm)

Inner Mongolia Cashmere goat (*Capra hircas*) to prove that the mTOR plays a critical role in the cell growth. The mTOR kinase activity was inhibited in Inner Mongolia Cashmere goat fetal fibroblasts (GFb) after treatment with 20 $\mu\text{mol/L}$ CCI-779, an mTOR specific inhibitor, for 48 h. The results showed that GFb cells were sensitive to CCI-779. CCI-779 inhibits the activity of mTOR signal pathway and cell proliferation, blocks cell cycle.

GFb cells morphology and its cytoskeleton structure changed under confocal laser scanning microscopy stained with the Fluorescent phalloidin (50 $\mu\text{g/ml}$, Phalloidin-FITC5282, Sigma) which combines with F-actin (Figure 1). In summary, mTOR signaling pathway was proved to be functional in GFb cells and acts as a key regulator to regulate cell growth. The Morphological results indicated that perhaps the synthesis of microfilament or organization of cytoskeleton was disrupted in GFb cells when mTOR was inhibited.

KEY WORDS: Confocal, goat fetal fibroblast, mTOR.

ACKNOWLEDGMENT

This work supported by grant from Natural Sciences Foundation of China (No. 30860191) and Major Projects for New Varieties of Genetically Modified Organisms (No.2008ZX08008-002)

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