

# IL-1 $\beta$ induces apoptosis and autophagy via mitochondria pathway in human degenerative nucleus pulposus cells

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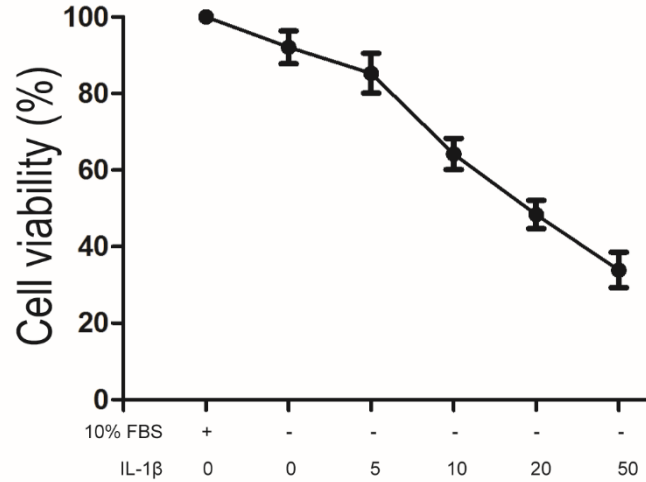
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**Table 1. Supplementary information for human nucleus pulposus sampling**

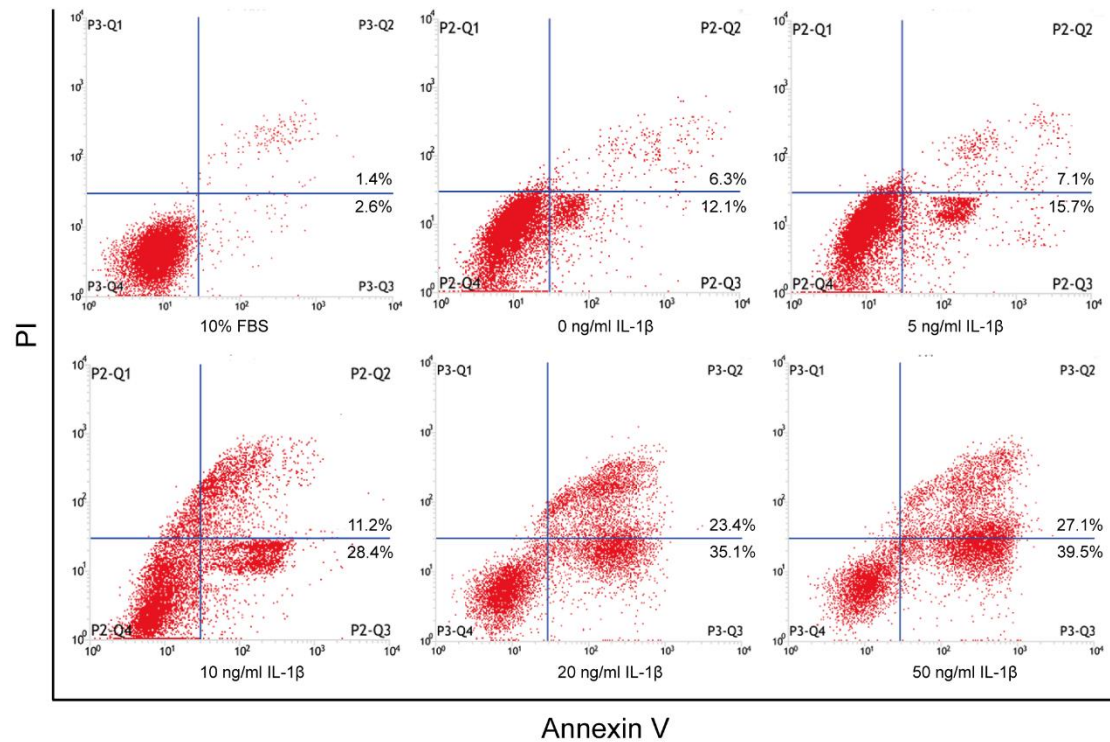
LVF	Age	Gender	Disc level investigate	Degree of degeneration	Application
1	36	F	L2/3、L3/4	Pfirschmann I-II	Tissue research for histological staining and western blot
2	37	M	L1/2	Pfirschmann I	Tissue research for histological staining
3	41	M	L1/2	Pfirschmann II	Tissue research for histological staining
4	33	F	T12/L1、L1/2	Pfirschmann I-II	Tissue research for western blot
5	45	M	L3/4	Pfirschmann II	Tissue research for western blot
LDH	Age	Gender	Disc level investigate	Degree of degeneration	Application
1	42	F	L4/5、L5/S1	Pfirschmann IV	Tissue research for histological staining and western blot
2	44	M	L5/S1	Pfirschmann V	Tissue research for western blot and cell research
3	36	M	L4/5	Pfirschmann IV	In situ histological staining and cell research
4	35	F	L3/4、L5/S1	Pfirschmann IV	Tissue research for histological staining and cell research
5	51	F	L4/5	Pfirschmann V	Cell research
6	42	M	L2/3	Pfirschmann IV	Tissue research for histological staining and cell research
7	38	M	L4/5	Pfirschmann IV	Cell research

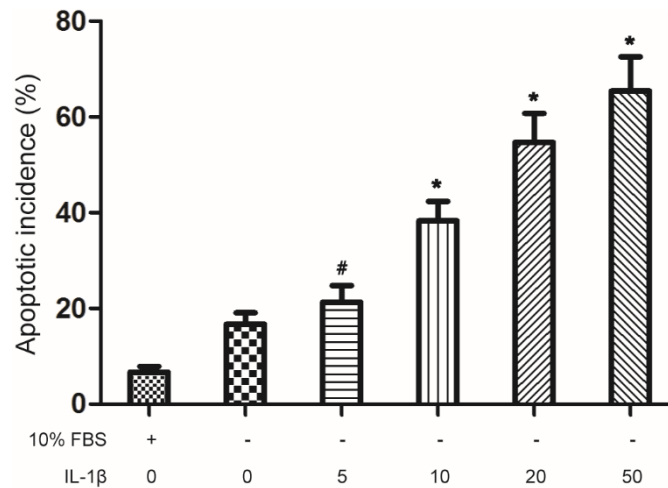
**Figure 1 Concentration gradient experiment for IL-1 $\beta$**

**Figure 1A.** CCK-8 assay for the cell viability. NP cells were treated with different concentrations of IL-1 $\beta$  for 24 hours.



**Figure 1B.** Flow cytometric analysis for apoptosis incidence. The apoptosis incidence was calculated by the percentage of early apoptotic (Annexin V+/PI-) cells plus the percentage of late apoptotic (Annexin V+/PI+) cells.





#P>0.05 Vs. 0 ng/ml IL-1 $\beta$ , and \*P<0.05 Vs. 5 ng/ml IL-1 $\beta$

### Figure Legends

Supplementary Figure 1A showed CCK-8 assay for the cell viability. Results showed that the viable cells reduced to 92.1%, 85.3%, 64.2% , 48.4% and 33.9% respectively, exposing with different concentrations of 0, 5, 10, 20, 50 ng/ml IL-1 $\beta$ . Supplementary Figure 1B showed flow cytometric analysis for apoptotic incidence. Results indicated that 5 ng/ml IL-1 $\beta$  treatment slightly increase the apoptotic rate compared with that in 0 ng/ml group (from 16.7% to 21.3%), but 10 ng/ml IL-1 $\beta$  treatment significantly increased the apoptotic rate upto 38.3%. With the further increased concentration, apoptotic rate increased as high as 54.7% and 65.4% under 20 and 50 ng/ml treatment respectively. All these results suggested that 10 ng/ml IL-1 $\beta$  with serum-free had a definite cytotoxicity, and higher concentrations resulted in insufficient cell viability and too much apoptosis , thus we used the concentration of 10 ng/ml in this study.