寧夏醫科大學
Ningxia Medical University

结合临床 深挖数据
髋关节软骨的全基因组 DNA 甲基化分析与股骨头坏死的相关性

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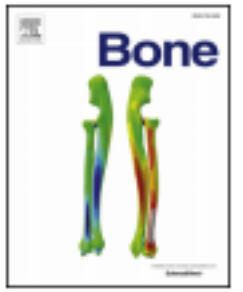

Full Length Article

Genome-wide DNA methylation profiling of hip articular cartilage identifies differentially methylated loci associated with osteonecrosis of the femoral head

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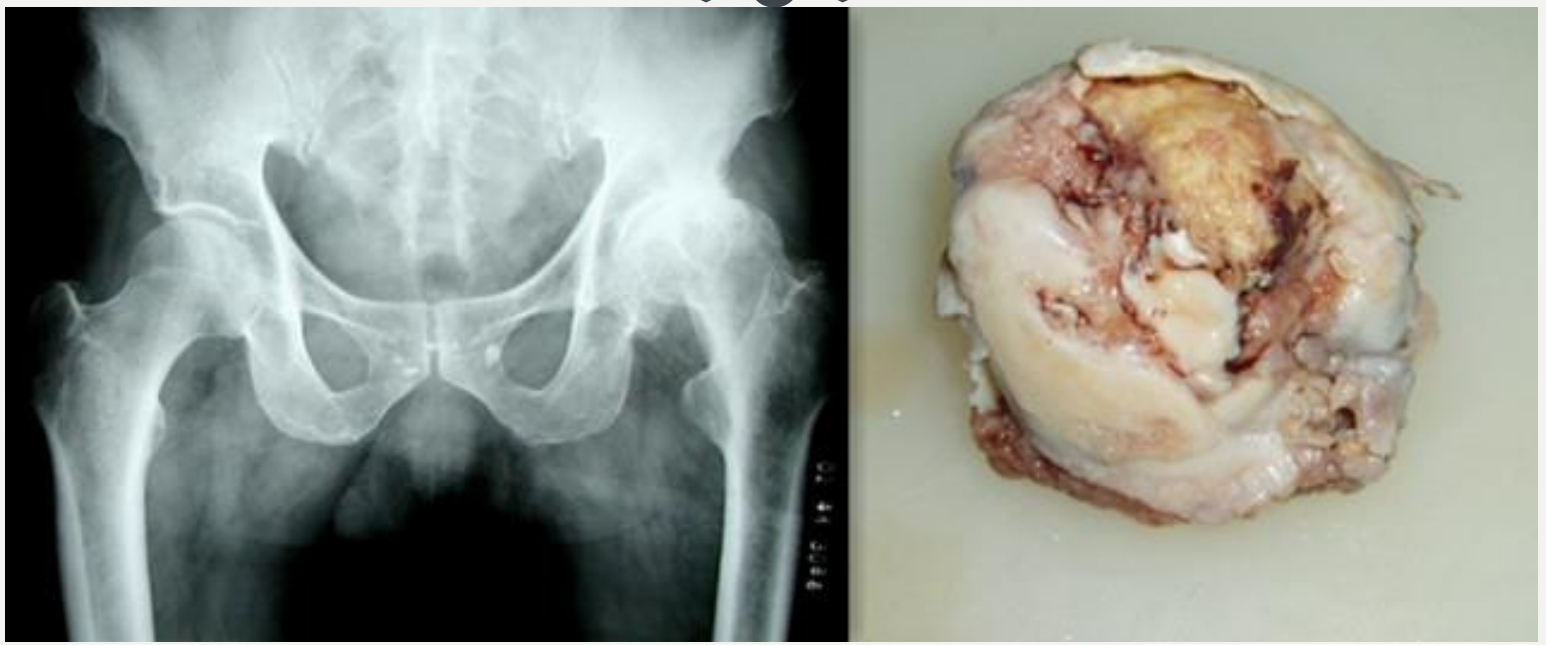
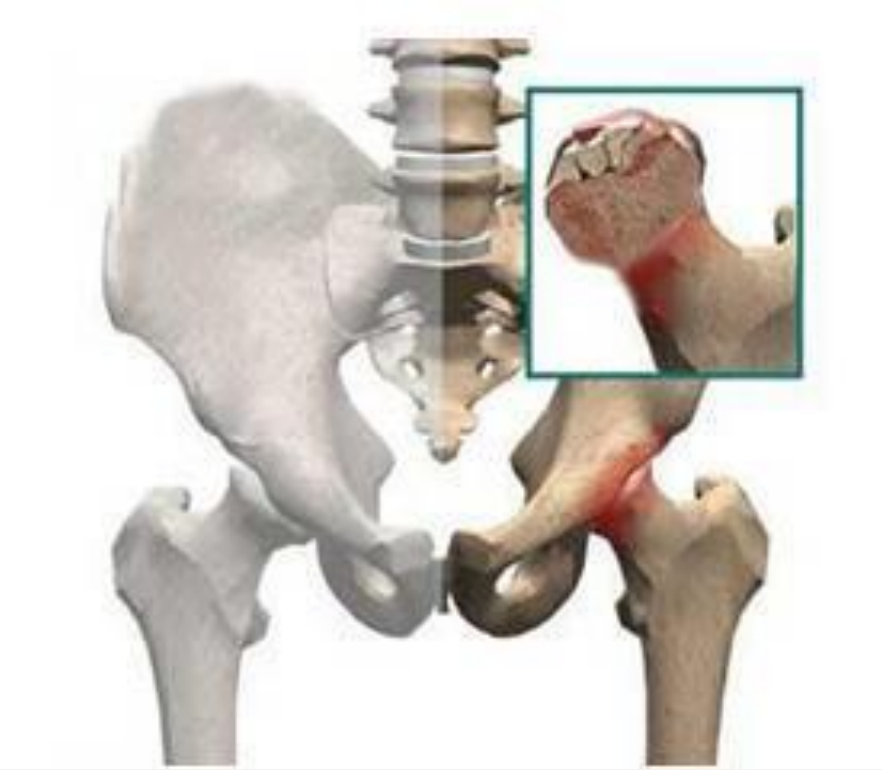
Introduction



Osteonecrosis of the femoral head (ONFH) is a seriously disabling disease, usually affecting young adults aged between 35 and 55, There are about 8.12 million patients with non-traumatic ONFH in China now。

Introduction



1、ONFH is pathologically characterized by the death of osteocytes and bone marrow cells due to inadequate blood supply of subchondral bone。

2、recent studies demonstrated a critical role of hip articular cartilage in the development of ONFH and articular cartilage degeneration occurs at the early stage of ONFH。

Introduction

age

fat

hormone

heredity

cartilage change

subchondral bone change

trauma

Results

十大高甲基化基因座列表。

糖蛋白	P值	贝塔差	基因名称
cg10161198cg10161198		1.71×10^{-6}	0.37
	fam178bfam178b		
cg03053125cg03053125	4.61×10^{-6}	0.23	
CG0804300	5.31×10^{-6}	0.21	fam178bf
			am178b
cg13990585cg13990585		1.09×10^{-5}	0.23 格玛
cg15140902cg15	3.20×10^{-5}	0.21	f1j22536f1j2
cg18699025cg18	5.00×10^{-5}	0.24	fgfrl1fgfrl1
CG2698799	5.24×10^{-5}	0.20	
CG0456511	5.38×10^{-5}	0.23	
			ptpn6p
		tpn6	
cg05255811cg05255811		5.44×10^{-5}	0.20
	kcnk5kcnk5		
CG26470219	6.33×10^{-5}	0.21	stk24stk24

Results

十大次甲基化基因座列表。

糖蛋白	P值	贝塔差	基因名称
CG0911488	2.28×10^{-6}	−0.20	汽车
CG26440334	5.07×10^{-6}	−0.22	
cg21724915cg21724915		−0.32	fto
	5.15×10^{-6}	邀请	
cg11849638cg11849638		1.36×10^{-5}	−0.26
	汽车		
cg03180359cg03180359		1.50×10^{-5}	−0.26
CG0910438	1.73×10^{-5}	−0.35	
	arhgap26arhgap26		
cg01886323cg01886323		2.68×10^{-5}	−0.42
cg10925915cg10925915	3.65×10^{-5}	−0.22	
CG7052537	3.85×10^{-5}	−0.21	
CG0220786	3.92×10^{-5}	−0.22	

Results

质谱仪在蛋白水平验证

The results of MS validation were consistent with that of genome-wide DNA methylation profiling, confirming the accuracy of DNA methylation profiles data.

Results

质谱

The results of MS validation were consistent with that of genome-wide DNA methylation profiling, confirming the accuracy of DNA methylation profiles data.

免疫组织化学分析

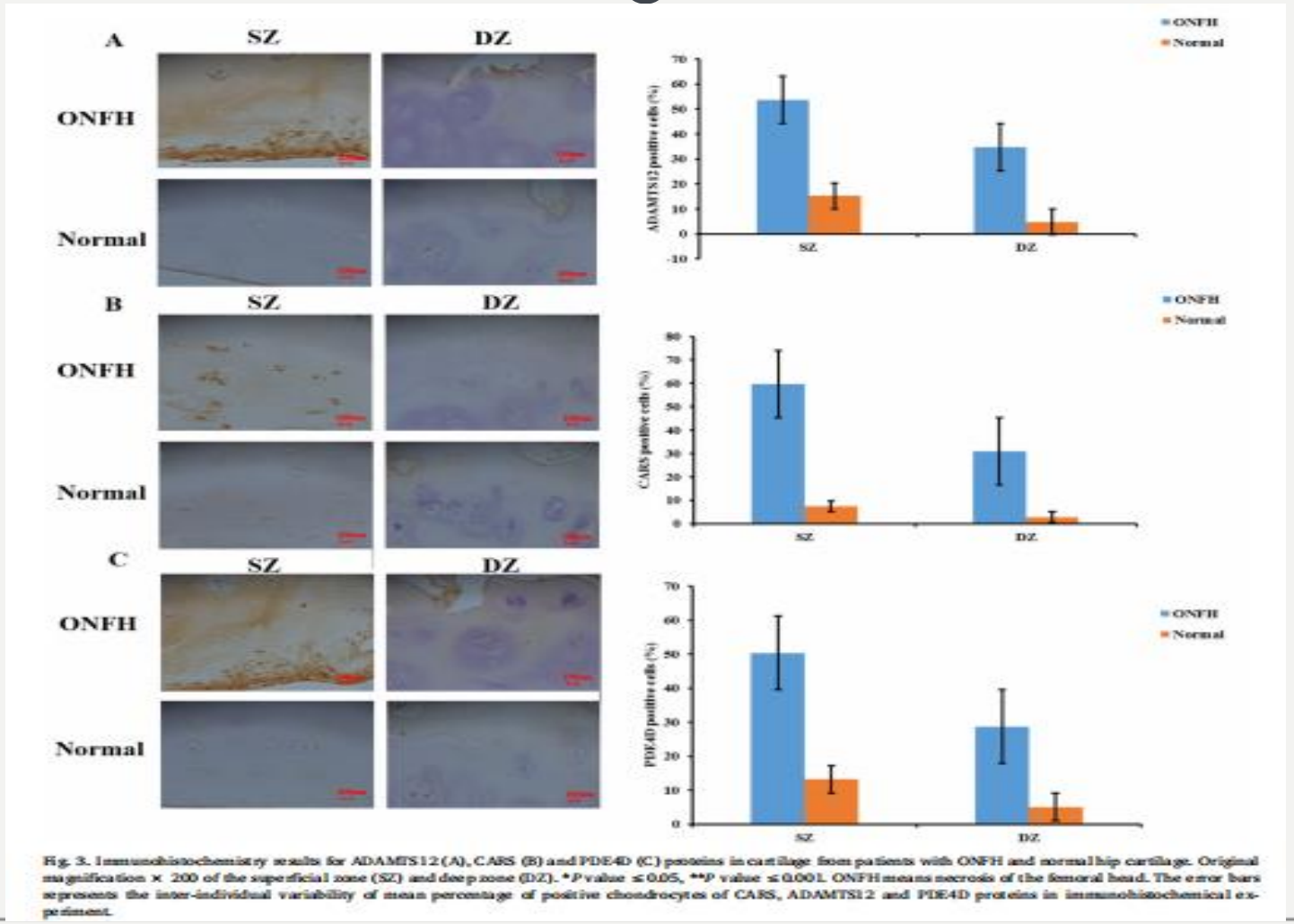


Fig. 3. Immunohistochemistry results for ADAMTS12 (A), CARS (B) and PDE4D (C) proteins in cartilage from patients with ONFH and normal hip cartilage. Original magnification × 200 of the superficial zone (SZ) and deep zone (DZ). *P value ≤ 0.05, **P value ≤ 0.001. ONFH means necrosis of the femoral head. The error bars represents the inter-individual variability of mean percentage of positive chondrocytes of CARS, ADAMTS12 and PDE4D proteins in immunohistochemical experiment.

免疫组织化学分析

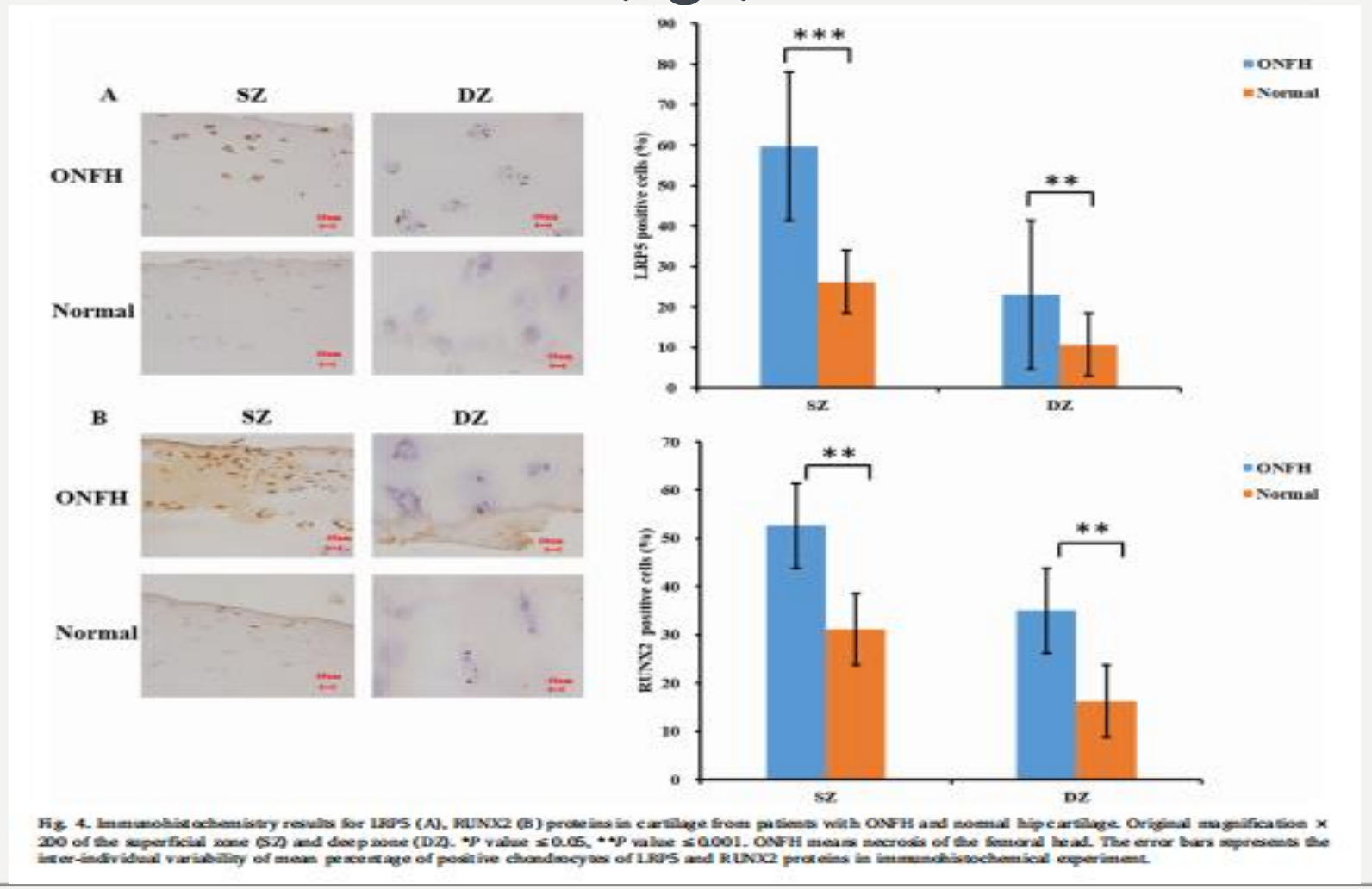


Fig. 4. Immunohistochemistry results for LRP5 (A), RUNX2 (B) proteins in cartilage from patients with ONFH and normal hip cartilage. Original magnification × 200 of the superficial zone (SZ) and deep zone (DZ). *P value ≤ 0.05, **P value ≤ 0.001. ONFH means necrosis of the femoral head. The error bars represents the inter-individual variability of mean percentage of positive chondrocytes of LRP5 and RUNX2 proteins in immunohistochemical experiment.

Materials and methods

First A genome-wide DNA methylation profiling .
Second The top two of differentially methylated genes were selected for further MS validation.
Finally IHC was conducted to compare the proteins expression levels of identified candidate genes between ONFH cartilage and control cartilage specimens.

Materials and methods

15 例 ONFH 患者
11 例男性和 4 例女性

15 例对照组
11 例男性和 4 例女性

5 个 ONFH 和 5 个对照软骨的全基因组 DNA 甲基化谱

对 10 个 ONFH 软骨和 10 个正常软骨进行了质谱（MS）分析，以验证全基因组 DNA 甲基化谱分析的结果。

4 个 ONFH 软骨和 4 个对照软骨的免疫组织化学。

共鉴定了 2872 个差异甲基化的 CpG 位点，分别注释了 480 个 ONFH 的超甲基化基因和 1335 个低甲基化基因。

Discussion

Recent studies have demonstrated the important roles of hip ar_x0002_ticular cartilage damage in the development of ONFH . Clarifying the molecular mechanism underlying the destruction of ONFH articular cartilage may provide insight into the pathogenetic and therapic studies of ONFH.

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