

木耳萃取物抑制蝕骨細胞活性

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摘要

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本試驗以大鼠卵巢切除 (OVX) 模式，探討餵食木耳子實體 (AP) 或木耳萃取物 (APE) 對骨質代謝的影響及其機制。SD 雌鼠分成 3 組，偽手術正常飼料組 (V0)，卵巢切除給予木耳子實體粉末組 (A1)，或卵巢切除給予木耳萃取物組 (A2)。研究結果顯示，餵食木耳子實體粉末可以有效改善 OVX 大鼠的血脂肪值，但對改善骨質代謝效果不顯著。餵食木耳萃取物大鼠的血液生化值及其骨質密度可以改善至接近偽手術組，探討其機制發現餵食木耳萃取物顯著增加卵巢切除大鼠血漿中的蝕骨細胞抑制因子濃度及血鈣濃度。

關鍵詞：木耳子實體、木耳萃取物、卵巢切除、骨質密度、骨源鹼性磷酸酶、蝕骨細胞抑制因子。

正常骨骼的代謝過程稱為骨質再造 (bone remodeling)，人體每年約有 10% 的骨質會進行更換，蝕骨細胞 (osteoclast) 溶解骨質與造骨細胞 (osteoblast) 填補骨質的作用，周而復始進行，維持骨質恆定狀態 (Wade 2001)。骨源鹼性磷酸酶 (bone-specific alkaline phosphatase; BAP) 是造骨細胞活性的指標，BAP 濃度增加代表造骨細胞活性增加 (Bruder *et al.* 1990)。蝕骨細胞抑制因子 (osteoprotegerin; OPG) 為調節骨質吸收的醣蛋白，可抑制骨吸收作用。OPG 會抑制蝕骨細胞活性 (Yasuda *et al.* 1998)，缺乏 OPG 小鼠會有嚴重的骨質疏鬆情形 (Bucay *et al.* 1998)。木耳水萃取物之前期研究，顯示具有改善卵巢切除大鼠血脂及骨質疏鬆之功效。又研究顯示杏鮑菇 (*Pleurotus eryngii*) 萃取物可減少卵巢切除 CD 大鼠的骨骼鈣質流失 (Shimizu *et al.* 2006)，攝食菇類之麥角固醇經 UV 照射轉化產生的維生素 D₂ 可以改善骨骼健康 (Chen *et al.* 2015)。由紅棗、黑大豆、黑木耳所組成的采坤茶萃取

物，動物實驗證明具有明顯的雌激素活性 (Su *et al.* 2003)。

本試驗以 6 wk 齡雌性 SD 大鼠 (Sprague Dawley rat) 為實驗動物，動物育養 1 wk 後施行卵巢切除術 (ovariectomized, OVX)。經 3 wk 復原期，先由尾靜脈採血後進行隨機組分成 3 組 ($n = 25$)，偽手術正常飲食組 (X0) 9 隻及實驗組添加木耳子實體粉末 A1 組 ($1.25 \text{ g Kg}^{-1} \text{ BW AP}$)，添加木耳萃取物 A2 ($0.5 \text{ g Kg}^{-1} \text{ BW APE}$) 各 8 隻。於飼育期 16 mo 結束後犧牲實驗動物，採集血液檢體及下肢股骨進行生化分析及切片判讀。

在本試驗開始的第 0 週 (卵巢切除手術後 3 wk) 的血液生化值分析值顯示，試驗組大鼠的膽固醇、三酸甘油酯及極低密度脂蛋白膽固醇指標高於 X0 組，呈現出類更年期的血液生化值變化。經 16 wk 餵食木耳子實體 (A1) 組或木耳萃取物 (A2) 組，三項血脂肪指標改善至接近 X0 組數值，如表 1 所示。試驗組的總膽固醇濃度由 $86.25\text{--}96.25 \text{ mg dL}^{-1}$ 下降至與 X0

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表 1. 實驗期間各組實驗動物的血液生化值。

Table 1. The blood biochemistry value of Sprague Dawley rats.

Plasma parameter	Rats with different treatments ^z		
	X0	A1	A2
AST (U L ⁻¹)			
wk 0	121.5 ± 11.9 ab ^y	125.0 ± 11.4 a	103.3 ± 5.5 b
wk 16	72.0 ± 2.1 b	92.1 ± 4.3 a	81.4 ± 9.5 ab
ALT (U L ⁻¹)			
wk 0	51.5 ± 2.2 a	50.5 ± 2.2 ab	46.3 ± 2.2 b
wk 16	48.8 ± 2.2 b	54.4 ± 3.1 ab	37.1 ± 2.5 c
Cholesterol (mg dL ⁻¹)			
wk 0	74.3 ± 1.8 c	86.3 ± 0.4 b	96.3 ± 3.4 a
wk 16	68.6 ± 2.4 a	75.3 ± 3.8 a	71.1 ± 2.7 a
Triglyceride (mg dL ⁻¹)			
wk 0	35.8 ± 1.9 d	52.0 ± 2.7 b	60.5 ± 2.1 a
wk 16	33.8 ± 3.2 a	36.0 ± 2.0 a	26.7 ± 2.1 b
HDL-C (mg dL ⁻¹)			
wk 0	23.8 ± 0.5 a	21.8 ± 0.3 b	23.8 ± 0.6 a
wk 16	29.2 ± 1.6 ab	30.1 ± 1.6 a	26.7 ± 0.9 ab
LDL-C (mg dL ⁻¹)			
wk 0	6.3 ± 0.5 a	5.0 ± 0.2 b	6.5 ± 0.3 a
wk 16	4.8 ± 0.4 b	6.1 ± 0.8 ab	5.1 ± 0.3 ab
VLDL (mg dL ⁻¹)			
wk 0	44.3 ± 0.0 c	59.5 ± 0.0 b	66.0 ± 0.1 a
wk 16	34.6 ± 0.0 b	39.0 ± 0.1 a	39.3 ± 0.1 a
Ca (mg dL ⁻¹)			
wk 0	8.1 ± 0.1 b	8.9 ± 0.3 a	6.3 ± 0.3 c
wk 16	9.4 ± 0.2 a	8.9 ± 0.3 ab	18.8 ± 0.2 b

^z X0: sham-operated rat fed on chow diet; A1: OVX rat fed on chow diet supplemented with 1.25 g kg⁻¹ BW AP; and A2: OVX rat fed on chow diet supplemented with 0.5 g kg⁻¹ BW APE.

^y Values are presented as mean ± SEM., Values shared with different superscripted letter(s) are significant differences from each group ($P < 0.05$).

組無顯著差異 ($P > 0.05$)；A2 組的三酸甘油酯濃度 26.71 mg dL⁻¹ 顯著低於 X0、A1 組 ($P < 0.05$)。試驗組的極低密度膽固醇 (VLDL-C) 濃度由 59.5–66.0 mg dL⁻¹ 下降至 39.0–39.3 mg dL⁻¹。結果顯示，餵食木耳子實體粉末或木耳萃取物，均可以有效改善 OVX 大鼠的血液血脂值。肝功能指標評估，試驗 A2 組的 AST 值由 103.25 μL⁻¹ 下降至 81.43 μL⁻¹；ALT 值由 46.25 μL⁻¹ 下降至 37.14 μL⁻¹，顯示餵食木耳萃取物可以有效改善 OVX 大鼠的發炎情形。

利用切片影像分析骨切片，如圖 1 及圖 2 所示，計數骨質密度，X0 組 69.5% 與 A2 組 62.1% 無顯著差異 ($P > 0.05$)，而顯著高於 A1 組 54.3% ($P < 0.05$)。血漿中骨源鹼性磷酸酶 (bone-specific alkaline phosphatase; BAP) 濃度其代表含意為成骨細胞的活性，如圖 3 所示，X0 組 4.0 pmol L⁻¹ 與 A2 組 2.65 pmol L⁻¹ 無統計上差異 ($P > 0.05$) 但顯著高於 A1 組 1.75 pmol L⁻¹ ($P < 0.05$)。蝕骨細胞抑制因子 (osteoprotegerin; OPG) 濃度如圖 4 所示，A2 組血漿中 OPG 濃度 0.27 pmol L⁻¹ 顯著高於

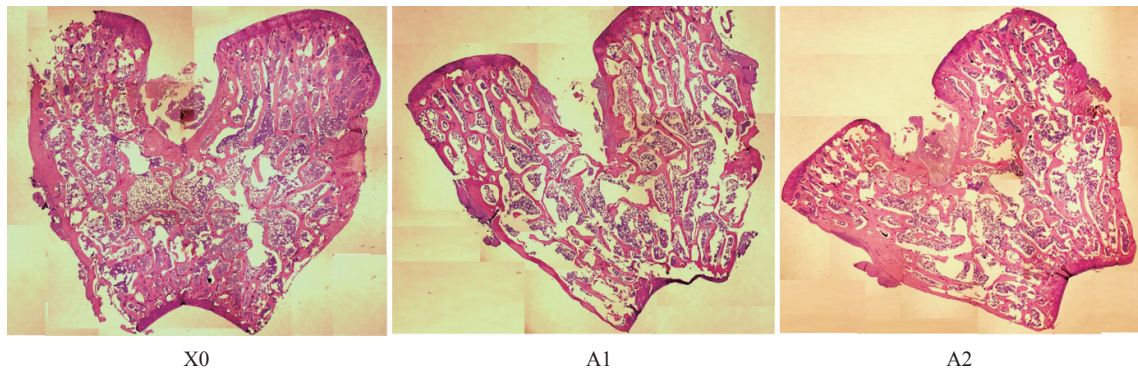


圖 1. 不同處理之 SD 大鼠骨組織切片。

Fig. 1. Bone histology sections under H&E stain with different treatments. X0: sham rat + chow diet; A1: ovx rat + 1.25 g kg⁻¹ BW AP; A2: ovx rat + 0.5 g kg⁻¹ BW APE.

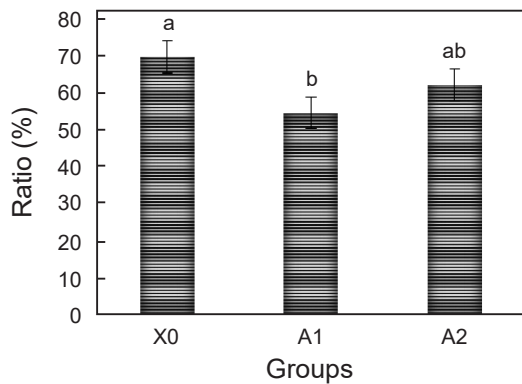


圖 2. 不同處理之 SD 大鼠的骨質密度。

Fig. 2. Bone density of rats with different treatments. Values are means with SEM represented by vertical bar. Values shared with different superscripted letters are significant differences from each group ($P < 0.05$). X0: sham-operated rat fed on chow diet; A1: OVX rat fed on chow diet supplemented with 1.25 g kg⁻¹ BW AP; A2: OVX rat fed on chow diet supplemented with 0.5 g kg⁻¹ BW APE.

X0 組 0.13 pmol L⁻¹、A1 組 0.07 pmol L⁻¹ ($P < 0.05$)。Yasuda *et al.* (1998) 研究顯示，OPG 可經由 1,25 (OH)₂D₃、PTH 或 IL-11 刺激 signaling pathways 抑制類蝕骨細胞的形成，Bucay *et al.* (1998) 研究也顯示，小鼠缺乏 OPG 時，蝕骨細胞活性增加造成骨吸收，而出現嚴重的骨質疏鬆情形。試驗初期至犧牲點血漿中鈣離子濃度，各組就統計而言無顯著差異，唯比較試驗期間變化，A2 組之血漿鈣離子濃度

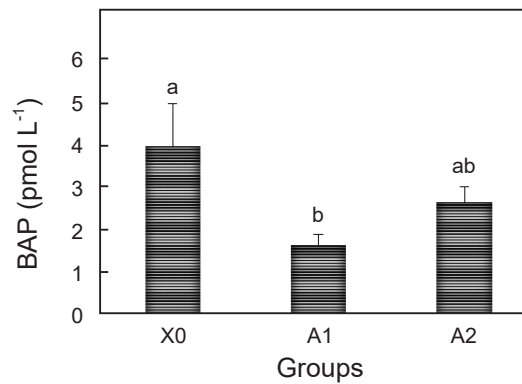


圖 3. 不同處理之 SD 大鼠血漿中骨源鹼性磷酸酶濃度。

Fig. 3. Plasma bone-specific alkaline phosphatase concentration of rats with different treatments. SD: Sprague-Dawley rat. Values are means with SEM represented by vertical bar. Values shared with different superscripted letters are significant differences from each group ($P < 0.05$). ALP: Alkaline Phosphatase; X0: sham-operated rat fed on chow diet; A1: OVX rat fed on chow diet supplemented with 1.25 g kg⁻¹ BW AP; and A2: OVX rat fed on chow diet supplemented with 0.5 g kg⁻¹ BW APE.

由 6.3 mg dL⁻¹ 增加至 8.79 mg dL⁻¹ 最為顯著，如圖 5 所示。

以上結果顯示，餵食木耳或萃取物均可有效改善 OVX 大鼠的血液血脂肪值。攝食木耳萃取物可以有效將 OVX 大鼠的血液生化值及骨質代謝問題，並改善至接近正常組狀態。此結果可能與萃取物增加蝕骨細胞抑制因子濃

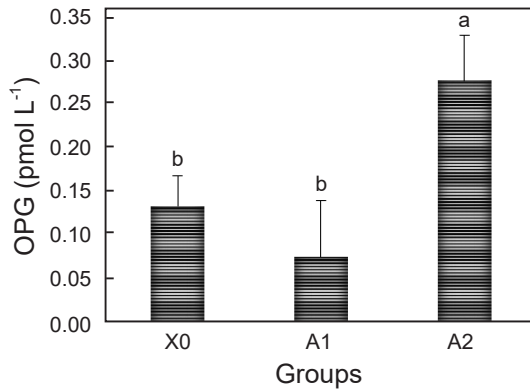


圖 4. 大鼠血漿中蝕骨細胞抑制因子 (osteoprotegerin) 濃度。

Fig. 4. Plasma osteoprotegerin concentration of different groups. Values are means with SEM represented by vertical bar. Values shared with different superscripted letters are significant differences from each group ($P < 0.05$). X0: sham-operated rat fed on chow diet; A1: OVX rat fed on chow diet supplemented with 1.25 g kg⁻¹ BW AP; and A2: OVX rat fed on chow diet supplemented with 0.5 g kg⁻¹ BW APE.

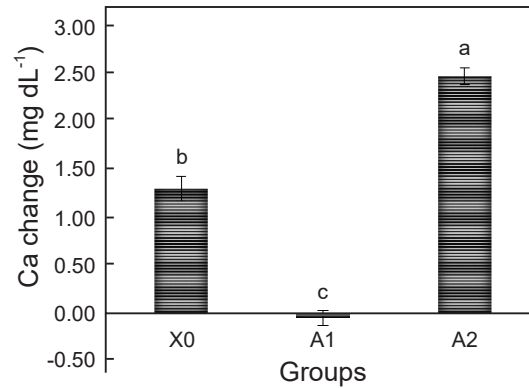


圖 5. 餵食 16 週大鼠血漿中血鈣變化。

Fig. 5. Plasma Ca concentration change at 16 wk of different groups. Values are means with SEM represented by vertical bar. Values shared with different superscripted letters are significant differences from each group ($P < 0.05$). X0: sham-operated rat fed on chow diet; A1: OVX rat fed on chow diet supplemented with 1.25 g kg⁻¹ BW AP; and A2: OVX rat fed on chow diet supplemented with 0.5 g kg⁻¹ BW APE. Value change = Ca (W16) – Ca (W0).

度，抑制蝕骨細胞活性，而減少骨質流失有關。木耳 (*Auricularia polytricha*) 萃取物含有水溶性多醣、多酚類、麥角固醇等物質，至於何者為改善骨質疏鬆之功效指標成分有待進一步研究。

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Inhibition on Osteoclasts Activity of *Auricularia polytricha* Extracts

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Abstract

Yang, S. H., H. T. Yang, and H. C. Liu. 2017. Inhibition on osteoclasts activity of *Auricularia polytricha* extracts. *J. Taiwan Agric. Res.* 66(3):261–265.

This study investigated the effect of bone metabolism and mechanism of feeding *Auricularia polytricha* fruiting body powder (AP) or extract (APE) on ovariectomized (OVX) rats. Female Sprague Dawley rats were divided into 3 groups, sham operated group fed on chow diet (V0), OVX fed on AP (A1) group or OVX fed on APE (A2) group. AP treatment effectively improved blood lipid values, but not significantly affected the bone metabolism. The blood biochemistry values and bone density of APE treatment can be improved to near sham operated group. APE treatment significantly increased osteoprotegerin concentration and calcium concentration of plasma on ovariectomized rats.

Key words: *Auricularia polytricha* powder, *Auricularia polytricha* extracts, Ovariectomy, Bone density, Bone-specific alkaline phosphatase, Osteoprotegerin.

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