

VITAMIN D TREATMENT ENHANCES ADIPOGENIC MRNA EXPRESSION IN STROMAL VASCULAR CELLS ISOLATED FROM BEEF CATTLE

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I. INTRODUCTION

Intramuscular fat (IMF), also known as marbling, is an important quality trait of meat, contributing to its flavour, juiciness, and tenderness. Adipocytes are specialized fat cells, which in turn affect the extent and distribution of IMF within the muscle tissue. Adipogenic differentiation, the process by which stem cells differentiate into adipocytes, plays a crucial role in the development of IMF. Vitamin D has been suggested to promote adipogenic differentiation and may be a possible strategy to optimize the production of IMF. In this study, we investigated the effect of vitamin D treatment on mRNA expression of adipogenic markers and lipid accumulation in bovine preadipocytes and mature adipocytes. Our findings will provide valuable insights into the development of more efficient and sustainable methods for the production of high-quality meat with desirable organoleptic properties.

II. MATERIALS AND METHODS

Stromal vascular (SV) cells were isolated from muscle tissue of Korean native cattle by a collagenase digestion method. These cells, including an untreated control group, were treated with 1,25-dihydroxyvitamin D₃ (1,25(OH)₂D₃) at three varying doses (1 nM, 10 nM, 100 nM) for either 2 days during the growth phase or 14 days during the differentiation phase. Each treatment condition was replicated four times independently. The mRNA expression of key preadipocyte markers such as zinc finger protein 423 (Zfp423), preadipocyte factor 1 (Pref-1), and KLF2 (Kruppel-like transcription factor 2), as well as mature adipocyte markers such as peroxisome proliferator-activated receptor gamma (PPAR γ) and fatty acid-binding protein 4 (FABP4), were measured using real-time quantitative PCR after 14 days of differentiation. Lipid accumulation was visualized using Oil Red O staining. Data were statistically analysed using the GLM procedure in SAS, Tukey's test, and one-way ANOVA.

III. RESULTS AND DISCUSSION

The results of our study demonstrate that treatment with 1,25-dihydroxyvitamin D₃ during the differentiation phase can significantly enhance the production of mature adipocytes in cultured meat. Specifically, treatment with 10 nM vitamin D resulted in increased mRNA expression of key adipogenic markers such as PPAR γ and FABP4 ($P < 0.05$), which are involved in adipocyte differentiation and lipid metabolism. However, we also observed a decrease in the expression of Zfp423 with vitamin D treatment, which could suggest that vitamin D promotes adipogenesis by suppressing its inhibitory effects. The exact mechanism of this effect, however, is still unclear and warrants further investigation. In contrast, during the growth phase, vitamin D treatment only had a significant effect on the mRNA expression of FABP4 at both 10 nM and 100 nM concentrations ($P < 0.01$), with no significant changes observed in the expression of PPAR γ , Zfp423, and Pref-1. This suggests that vitamin D may have a different mode of action during the growth phase compared to the differentiation phase. Additionally, our study found that treatments with vitamin D during differentiation phases can increase lipid accumulation in SV cells, as visualized by Oil Red O staining ($P < 0.05$). This is an important finding,

as lipid accumulation is essential for the desirable organoleptic properties of meat, such as tenderness and juiciness.

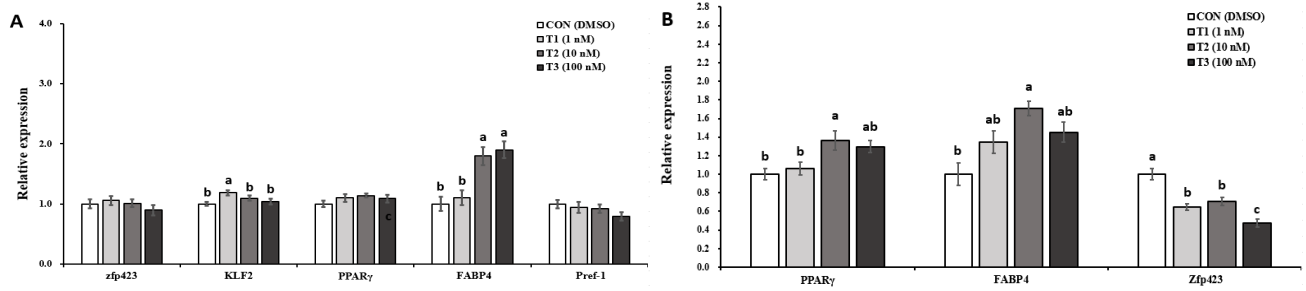


Figure 1. mRNA expression of key preadipocyte and adipocyte markers in Bovine adipocytes treated with varying doses of vitamin D during the growth or differentiation phase. Data are presented as mean \pm SEM (n=3). Different letters (a, b, c) indicate significant differences (P < 0.05) between treatment groups. A) Growth phase: cells were treated with varying doses of 1,25-dihydroxyvitamin D3 for 2 days. B) Differentiation phase: cells were treated with varying doses of 1,25-dihydroxyvitamin D3 for 14 days.

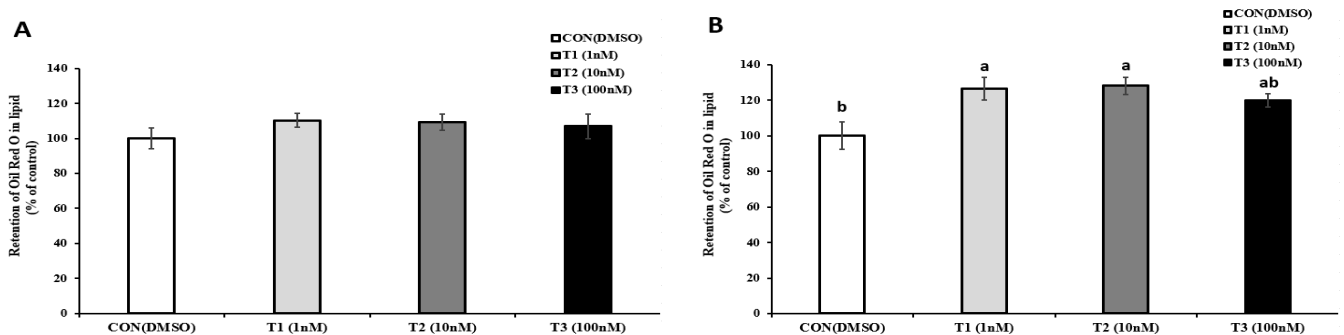


Figure 2. Lipid accumulation in bovine adipocytes treated with 1,25-dihydroxyvitamin D3 at varying doses (1 nM, 10 nM, 100 nM) during the growth phase (2A) or differentiation phase (2B). Lipid accumulation was visualized using Oil Red O staining and quantified. Data are presented as mean \pm SEM (n=3). Different letters (a, b, c) indicate significant differences (P < 0.05) between treatment groups.

IV. CONCLUSION

Our study demonstrates the potential of vitamin D treatment in enhancing the production of high-quality meat through the increase in adipogenic differentiation. Our findings suggest that vitamin D treatment may play a role in promoting adipogenesis by increasing the expression of key adipogenic markers and promoting lipid accumulation in bovine SV cell, particularly during the differentiation phase.

ACKNOWLEDGEMENTS

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