MITOGENIC EFFECTS OF INSULIN-LIKE GROWTH FACTOR-1 ON VELVET ANTLER CELLS *IN VITRO*

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It has been shown that plasma levels of insulin-like growth factor-1 (IGF1) correlate strongly and positively with rate of velvet antler growth. Specific Type 1 IGF receptors are found in the antler tip. The effect of human recombinant IGF1 on DNA synthesis was studied in primary culture of undifferentiated fibroblast-like cells and differentiated osteoblast-like cells. Primary cultures of fibroblast and osteoblast-like cells were successfully prepared from adult red deer (*Cervus elaphus*) stags whose antlers were harvested after 75 days of growth. Tissues were dissected in zones and dispersed with collagenase, and then the cells were grown in 45% Fitton-Jackson modification media (BGJ_b), 45% F12 nutrient, 10% fetal bovine serum (FBS), penicillin (100 U.ml), and streptomycin (100 µg/ml). Two x 10^4 cells/cm^2 were seeded in 24 well plates and incubated in a humidified 5% CO_2 atmosphere at 37°C. After 48 hr the media was changed to either 10% FBS or serum-free media (SFM) and incubated for a further 24 hr, followed by a 24 hr incubation in either 10% FBS, SFM or 10 nM IGF1. After 23 hr 2.5 µCi ^{3}H thymidine were added to each well for one hour. Reactions were terminated with 10% TCA. ^{3}H thymidine was counted in universal scintillant. Results are the mean of triplicate experiments and were analysed by ANOVA. The table shows the incorporation of ^{3}H thymidine (DNA synthesis) into fibroblast and osteoblast-like cells. Means with different superscripts are significantly different, $P<0.05$.

1st 24 hr incubation	2nd 24 hr incubation	Fibroblast d.p.m./wells	Osteoblast d.p.m./wells
10% FBS	10% FBS	20799[a]	20953[a]
SFM	SFM	34546[b]	27238[b]
SFM	10 nM IGF1	66870[c]	68783[c]
SFM	10% FBS	24042[a]	27795[b]
	sed	2661	2664

IGF1 significantly increased ^{3}H thymidine uptake in both cell types compared to SFM, FBS or SFM followed by FBS. SFM increased ^{3}H thymidine uptake compared with FBS in both cell types indicating either an inhibited effect in FBS or a potential for endogenous growth factor production. Our results provide direct evidence that IGF1 can stimulate proliferation of antler cells.