

MODULATION OF INSULIN RESPONSIVENESS IN L₆ MYOBLASTS

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Previously it has been reported that insulin induces insulin resistance in cultured adipocytes (Garvey et al. 1986). More recently, Garvey et al. (1987) reported that glucose could modulate insulin's ability to regulate glucose utilisation in primary adipocytes pretreated for 24 hours with various concentrations of glucose and insulin. The aim of the present study was to investigate whether this effect could also be demonstrated in the L₆ muscle cell line. L₆ myoblasts are originally derived from rat skeletal muscle and share many characteristics of mature tissue. Cells are maintained in Dulbecco MEM containing 10% fetal calf serum (FCS) and 25 mM glucose. For measurement of glucose utilisation cells were transferred to 2.5 cm² Falcon wells and incubated in the same medium at 37°C. After reaching confluence the cells were incubated for 24 hours in Dulbecco MEM, 2% FCS with either 25 mM glucose or 5 mM glucose both containing 0.25 μM insulin. Cells were then removed into Krebs Ringer Bicarbonate Hepes buffer, 10% BSA with washing, 3 hours prior to stimulation with insulin. The incubation without serum was necessary to lower basal rates of glucose transport to optimise insulin responsiveness of glucose utilisation. Basal and maximally insulin-stimulated glucose utilisation were measured at 37°C. Cells were preincubated for 10 mins in appropriate insulin concentrations and then glucose utilisation (3H-glucose) was measured after 30 mins. Insulin significantly stimulated glucose utilisation in cells grown in 5 mM glucose and 0.25 μM insulin overnight. However in cells grown in 25 mM glucose and 0.25 μM insulin, insulin responsiveness of glucose utilisation was reduced. This demonstrates that glucose can regulate insulin's ability to stimulate glucose metabolism in L₆ myoblasts in a manner analogous to that demonstrated in primary adipocytes.

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