Doxycycline-regulated 3T3-L1 preadipocyte cell line with inducible, stable expression of adenoviral E4orf1 gene: a cell model to study insulin-independent glucose disposal.

Impaired glycemic control and excessive adiposity are major risk factors for Type 2 Diabetes mellitus. In rodent models, Ad36, a human adenovirus, improves glycemic control, independent of dietary fat intake or adiposity. It is impractical to use Ad36 for therapeutic action. Instead, we identified that E4orf1 protein of Ad36, mediates its anti-hyperglycemic action independent of insulin signaling. To further evaluate the therapeutic potential of E4orf1 to improve glycemic control, we established a stable 3T3-L1 cell system in which E4orf1 expression can be regulated. The development and characterization of this cell line is described here. Full-length adenoviral-36 E4orf1 cDNA obtained by PCR was cloned into a tetracycline responsive element containing vector (pTRE-Tight-E4orf1). Upon screening dozens of pTRE-Tight-E4orf1 clones, we identified the one with the highest expression of E4orf1 in response to doxycycline treatment. Furthermore, using this inducible system we characterized the ability of E4orf1 to improve glucose disposal in a time dependent manner. This stable cell line offers a valuable resource to carefully study the novel signaling pathways E4orf1 uses to enhance cellular glucose disposal independent of insulin.

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