48. CLONING AND CHARACTERISATION OF MOUSE GLUT12 IN PREIMPLANTATION EMBRYOS

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Glucose transport in preimplantation mouse embryos is mediated by a family of facilitative glucose transporters known as GLUT. Whilst several isoforms are expressed and critical during early embryonic development (1), the recent identification of novel mammalian GLUTs and their classification into three sub-classes necessitates re-evaluation of embryonic transporter expression. Here we report the cloning and characterisation of the murine homologue of GLUT12 from preimplantation embryos. Using an antiserum against the human C-terminal GLUT12 dodecapeptide (2), positive immunoreactivity was observed in mouse 2-cell embryos by western immunoblotting. To confirm this observation and identify GLUT12 mRNA transcripts, the mouse genome and the expressed sequence tag (EST) databases were searched and an EST clone (ID 6542091, Genbank Acc: BF139811) was identified that corresponded to the antigenic sequence. RNA from 2-cell embryos was subjected to 5' RACE RT-PCR using primers designed against this EST clone. The results indicate that the mGLUT12 gene contains an open reading frame of 1869 base pairs, potentially encoding a polypeptide of 622 amino acids, which shows 83% sequence homology to hGLUT12. Like its human homologue, GLUT12 mRNA is found predominantly in skeletal and cardiac muscle and fat. However, it is also found in the uterus and embryos. GLUT12 expression is apparent during early development to the 2-cell stage and declines thereafter until E11. GLUT12 expression in classically insulin-responsive tissues such as muscle and fat has led to the suggestion that this may be an insulin responsive transporter (2). However the significance of GLUT12 expression in oocytes and 2-cell embryos in the absence of insulin receptor expression and the decline of its expression following genome activation and insulin receptor expression is unclear.

(1) Pantaleon & Kaye (1998) *Rev Reprod* **3**: 77–81. (2) Rogers *et al.* (2002) *Am. J. Physiol. Endocrinol. Metab.* **283**: E733–E738.

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