

# APPENDICES

## Appendix 1: *Irs2* silencing and insulin sensitivity

In this case, a different pattern from the analysed in the current study with *Irs2* overexpression was observed. Generally, it was observed that *Irs2* silencing decreased phosphorylation of most of the signalling proteins when these were stimulated with IGF-1. Whilst, in general, no significant differences were observed between control and shIRS2 cells with insulin stimulation.

In conclusion, these results suggested that *Irs2* silencing altered downstream signalling pathways in a different way than with *Irs2* overexpression.

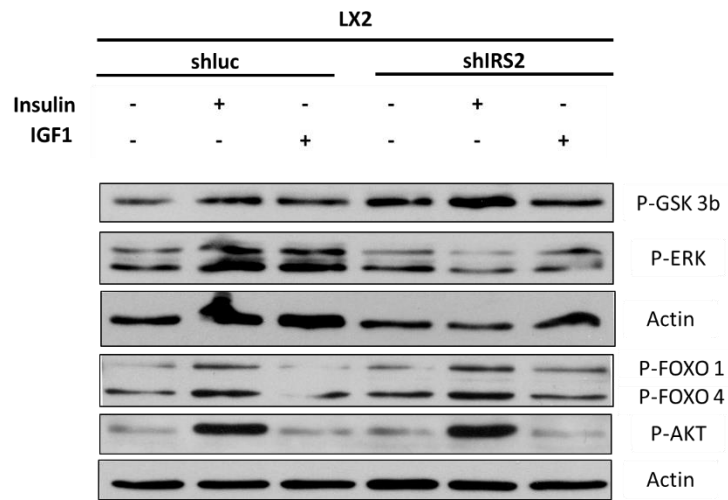


Figure 1. Western blot of insulin/IGF-1 pathways phosphorylated proteins in GFP and shIRS2 cells. A different pattern of phosphorylation was observed from the *Irs2* overexpression cells.

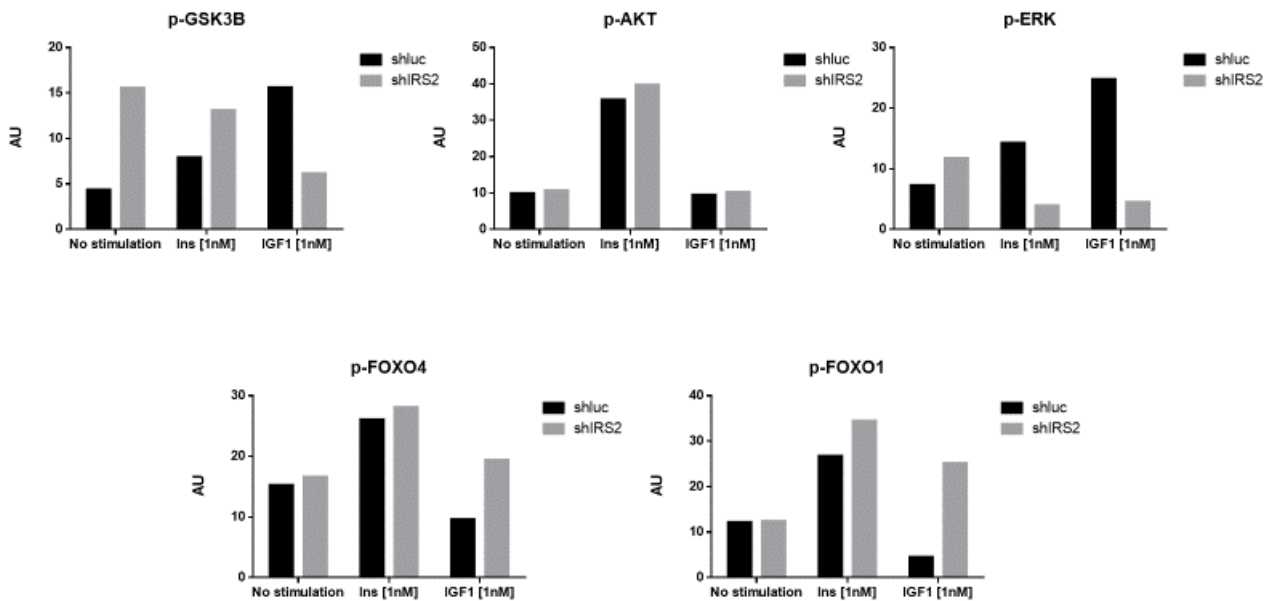


Figure 2. Results of signalling experiment with shIRS2 cells. n=1 experiment. Cells were stimulated with [1nM] of insulin and IGF1.