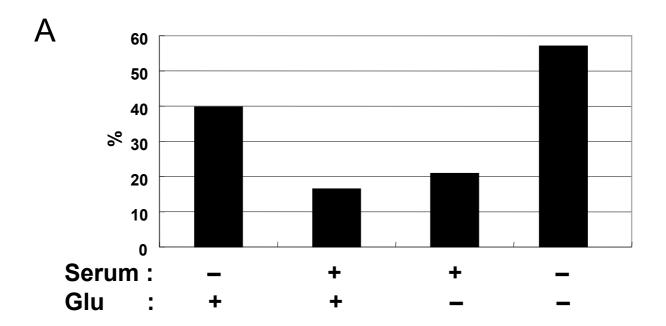
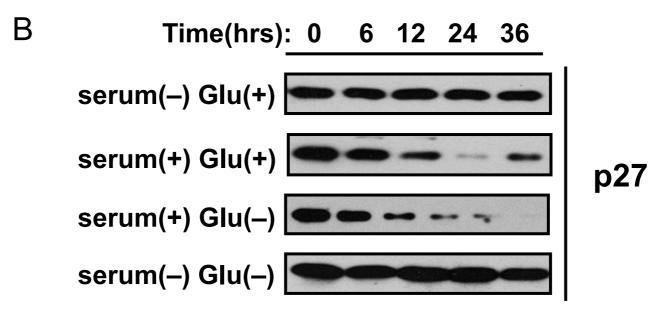
Supplementary FIG. 1. Nagano et al.

## **Materials and Methods for Supplementary Figures**

## SA-β-Gal assay

Serum-starved MEFs were released into serum and/or glucose free media. After 24 hours, the cells were washed with PBS, and fixed with 0.5% glutaraldehyde for 10 min at room temperature. After two ten-minute washings with 1 mM  $MgCl_2$  the cells were stained with X-Gal.





Supplementary Figure 1. Glucose limitation-induced p27 degradation is cell cycle- and senecence-independent. (A) Serum-starve MEFs were released into serum and/or glucose free media. After 24 hours, cells were stained for SA- $\beta$ -Gal. The percentage of SA- $\beta$ -Gal stained cells was indicated in graph. (B) The cell cycle of wild-type MEFs was synchronized by serum starvation for 48 hours and the cells cultured in media containing 0.5% FBS (-serum), 10% FBS (+serum), 0.1 mM glucose (-Glu), or normal glucose (+Glu) in various combinations for the indicated times. Cell lysates were prepared and subjected to immunoblotting using anti-p27 antibody.