

Supplementary Figures

Supplementary Figure 1. Detection of cell death in isogenic eIF2 α S/S and eIF2 α A/A MEFs in response to glucose deficiency. eIF2 α S/S and eIF2 α A/A MEFs were maintained in media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM 2-deoxyglucose (2-DG) for 24 or 48 hours. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The graphs represent one out of seven reproducible experiments.

Supplementary Figure 2. Detection of cell death in isogenic PERK^{+/+} and PERK^{-/-} MEFs in response to glucose deficiency. PERK^{+/+} and PERK^{-/-} MEFs were maintained in media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM 2-deoxyglucose (2-DG) for 24 or 48 hours. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The graphs represent one out of six reproducible experiments.

Supplementary Figure 3. Detection of cell death in isogenic PKR^{+/+} and PKR^{-/-} MEFs in response to glucose deficiency. PKR^{+/+} and PKR^{-/-} MEFs were maintained media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM 2-deoxyglucose (2-DG) for 24 or 48 hours. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the

population of cells in sub-G₁. The graphs represent one out of five reproducible experiments.

Supplementary Figure 4. Detection of cell death in isogenic GCN2^{+/+} and GCN2^{-/-} MEFs in response to glucose deficiency. GCN2^{+/+} and GCN2^{-/-} MEFs were maintained in media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM 2-deoxyglucose (2-DG) for 24 or 48 hours. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The graphs represent one out of six reproducible experiments.

Supplementary Figure 5. Detection of eIF2 α phosphorylation in MEFs deficient for PKR, PERK or GCN2 and in their isogenic counterparts.

Isogenic PERK^{+/+} and PERK^{-/-} MEFs (A), PKR^{+/+} and PKR^{-/-} MEFs (B) or GCN2^{+/+} and GCN2^{-/-} MEFs (C) were treated as described in Supplementary Figure 4. Whole cell extract (50 μ g of protein) were subjected to immunoblot analysis for eIF2 α -pSer51 (panel a), eIF2 α (panel b), or actin (panel c). N.S refers to nonspecific band.

Supplementary Figure 6. Detection of cell death in HeLa cells treated with siRNA for XIAP under glucose deficiency. HeLa cells subjected to treatment with siRNA specific for XIAP or with scrambled (SCR) siRNA for 24 hours. Cells were then maintained in media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM of 2-deoxyglucose (2-DG) for an additional 24 hours. Cells

were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The data represent one out of three reproducible experiments.

Supplementary Figure 7. Detection of cell death in XIAP^{+/+} and XIAP^{-/-} primary MEFs in response to glucose deficiency. XIAP^{+/+} and XIAP^{-/-} MEFs were maintained in media containing glucose, or media containing glucose supplemented with 50mM of 2-deoxyglucose (2-DG) for 48 hours. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The data represent one out of four reproducible experiments.

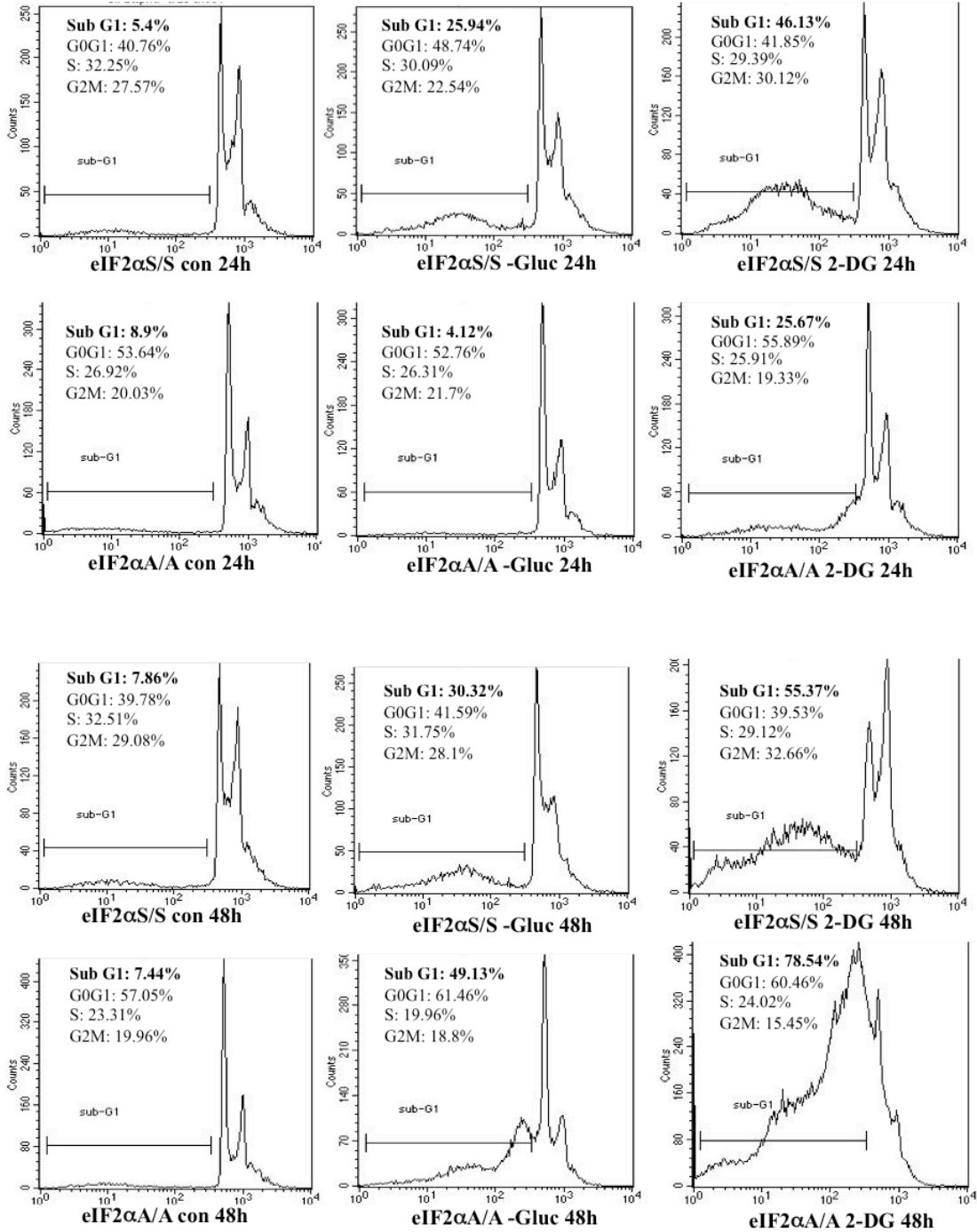
Supplementary Figure 8. Detection of cell death in HCT116 XIAP^{+/+} and HCT116 XIAP^{-/-} cells in response to glucose deficiency. HCT116 XIAP^{+/+} and HCT116 XIAP^{-/-} cells were maintained in media containing glucose, glucose-free media (-Gluc) or media containing glucose supplemented with 50mM of 2-deoxyglucose (2-DG) for 48 or 72 h. Cells were subjected to propidium iodide staining and flow cytometry analysis to detect the population of cells in sub-G₁. The data represent one out of four reproducible experiments.

Supplementary Figure 9. Experimental controls for polysome profile analysis.

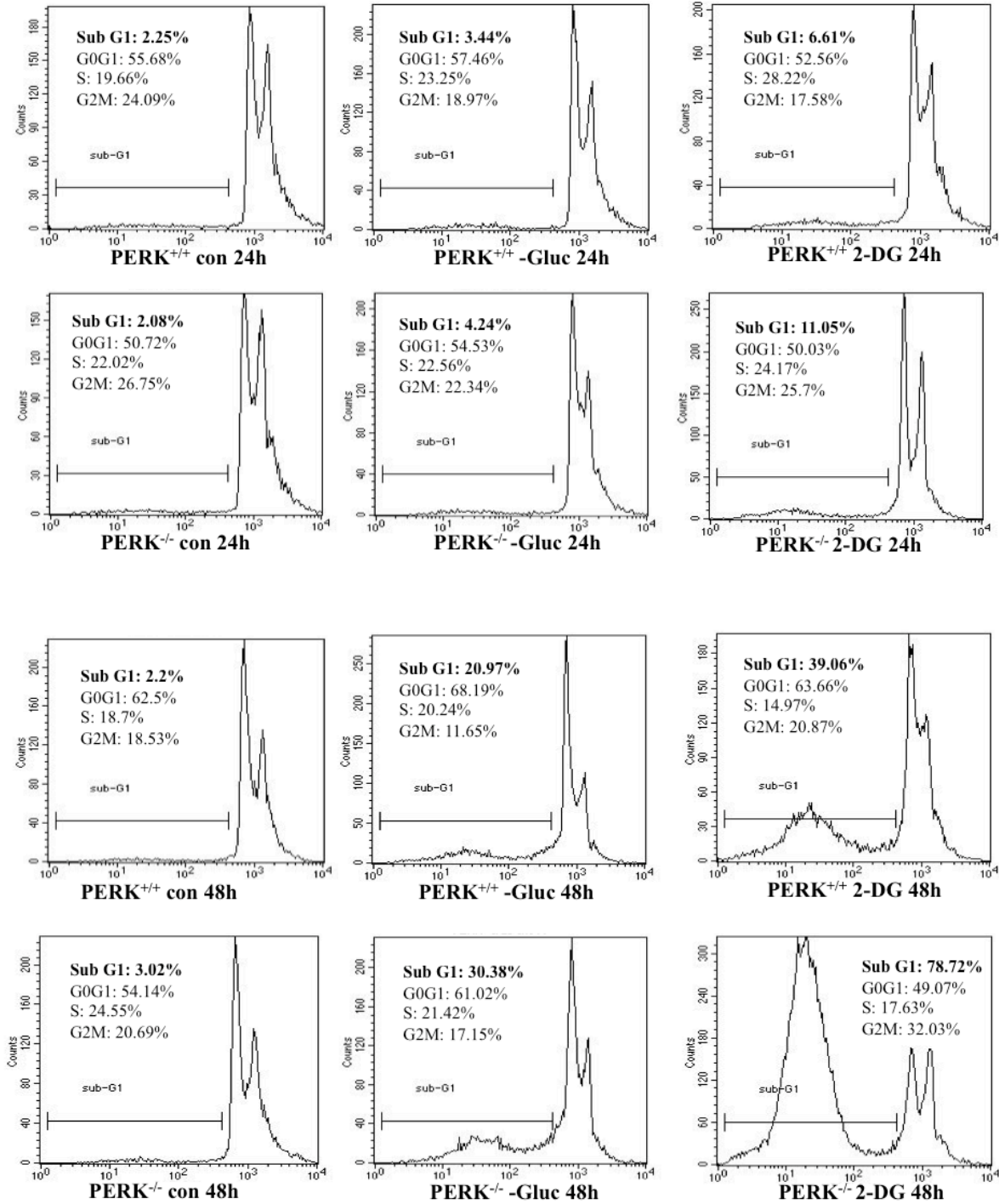
eIF2 α S/S (A) and eIF2 α A/A MEFs (B) were kept in media containing glucose (Con) or in glucose-free media (-Gluc) for 30 h. Lysates were subjected to polysome profile analysis as described in Methods. Gradients were fractionated and absorbance at 254 nm was recorded. As experimental controls, the distribution of rpL27 (upper panel) and

ATF4 (lower panel) mRNAs in the fractions of the sucrose gradients was quantified by qRT-PCR. The underlined fractions in the graphs signify the polysomes. The graphs represent the mean \pm SEM from two independent experiments. The primers that were used to visualize rpL27 mRNA are as follows: rpL27 forward primer 5'-GCAAGAAGAAGATCGCCAAG-3', rpL27 reverse primer 5'-CGCTCCTCAAACCTTGACCTT-3'. The primers that were used to visualize ATF4 mRNA are as follows: ATF4 forward primer 5'-GTTTGACTTCGATGCTCTGTTTC-3', ATF4 reverse primer 5'-GGGCTCCTTATTAGTCTCTTGG-3'

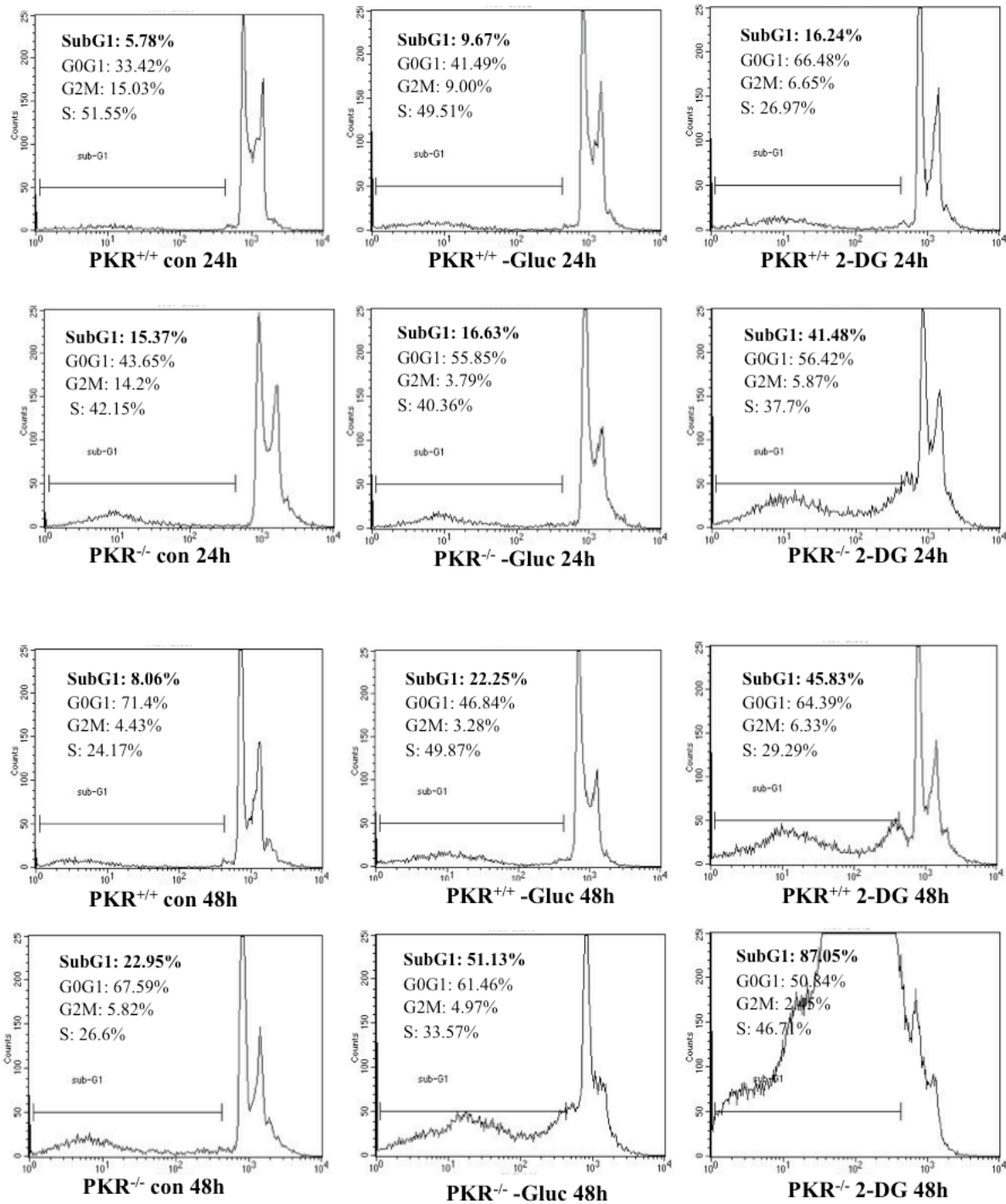
Supplementary Figure 1



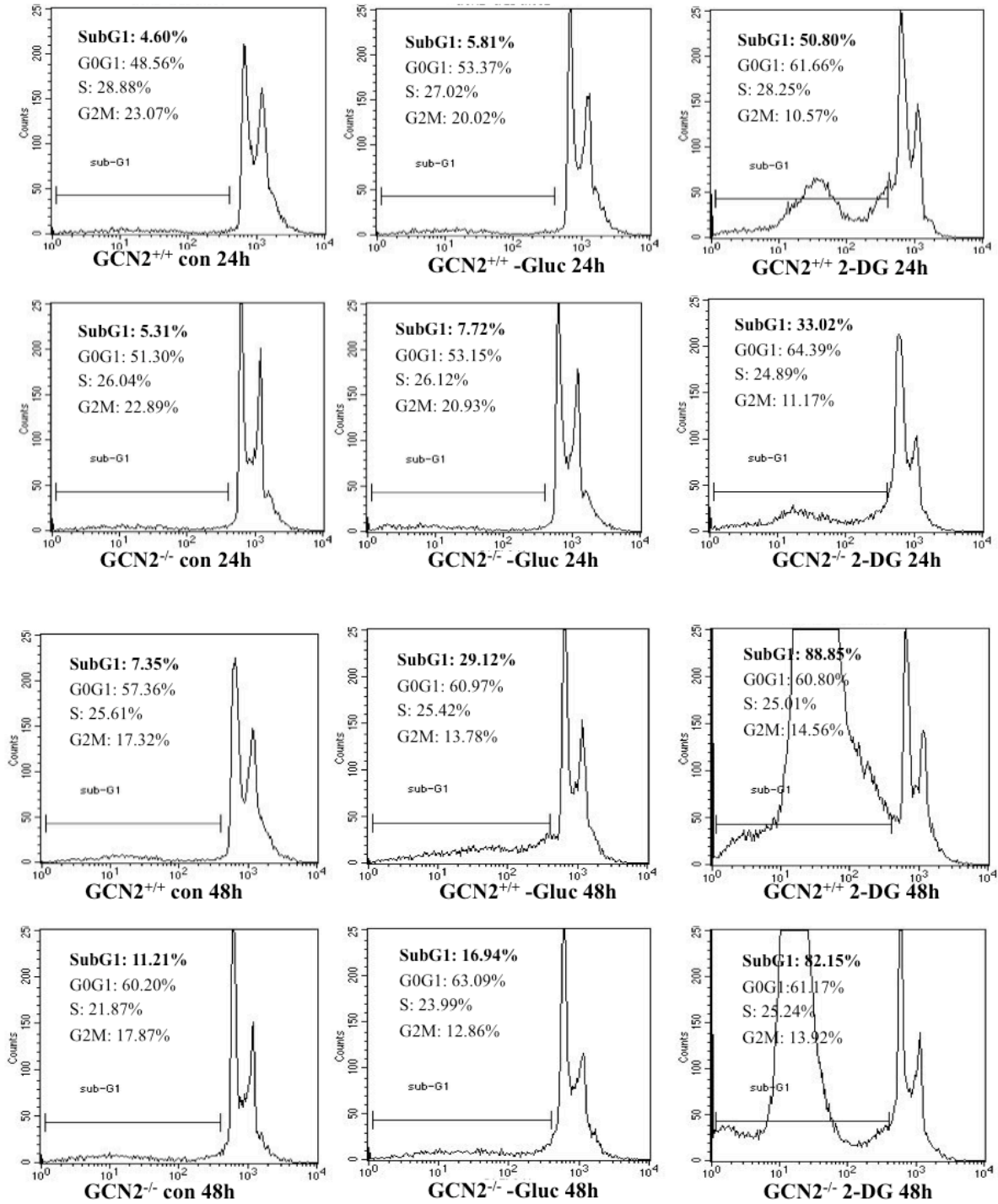
Supplementary Figure 2



Supplementary Figure 3

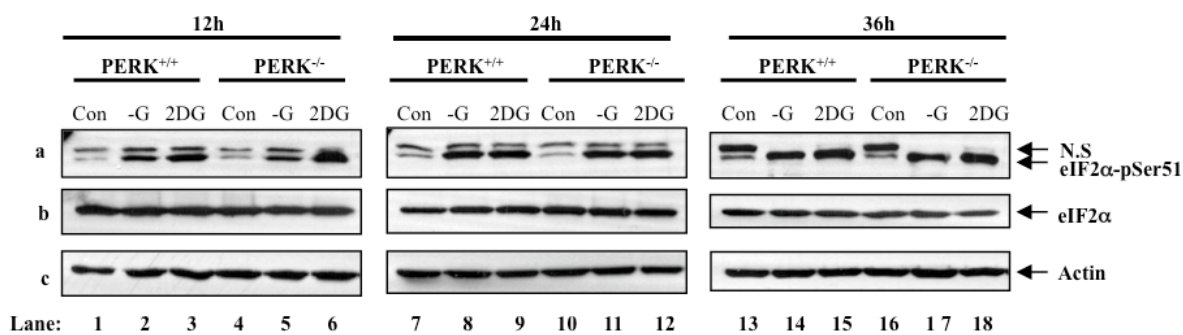


Supplementary Figure 4

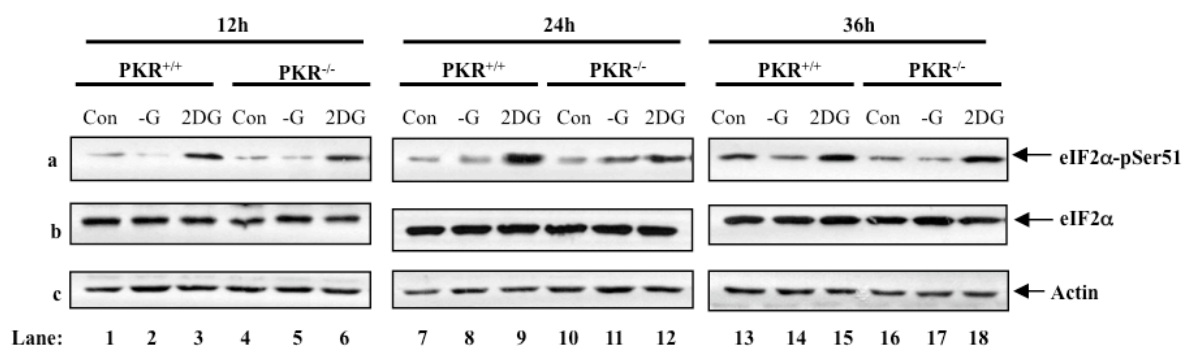


Supplementary Figure 5

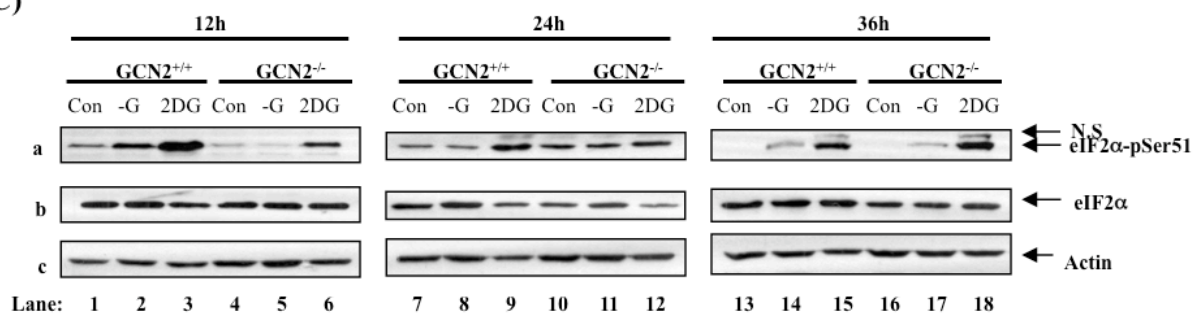
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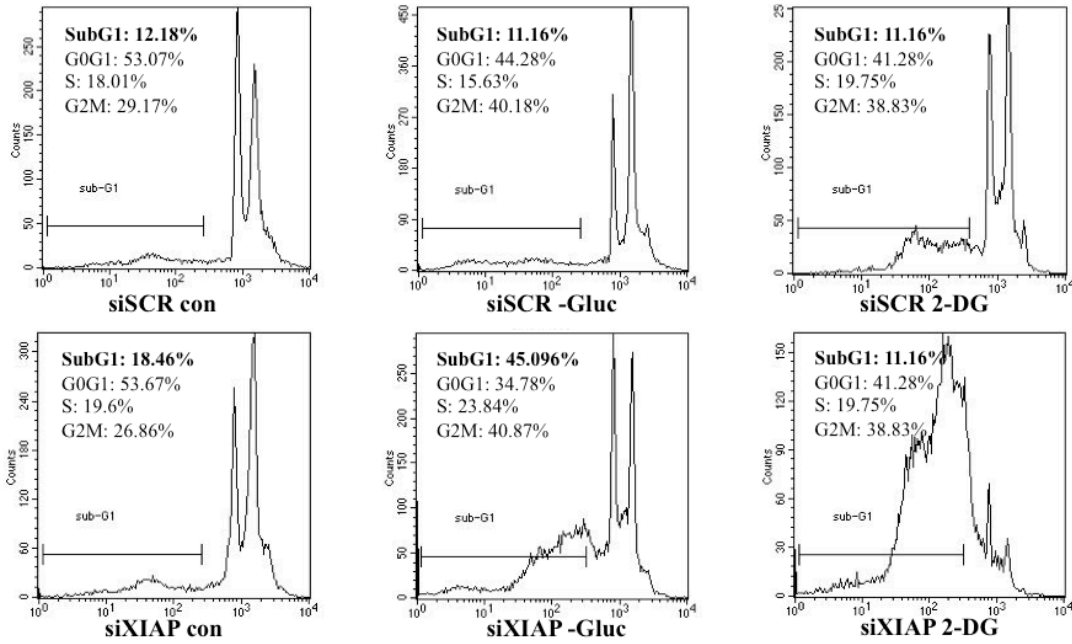
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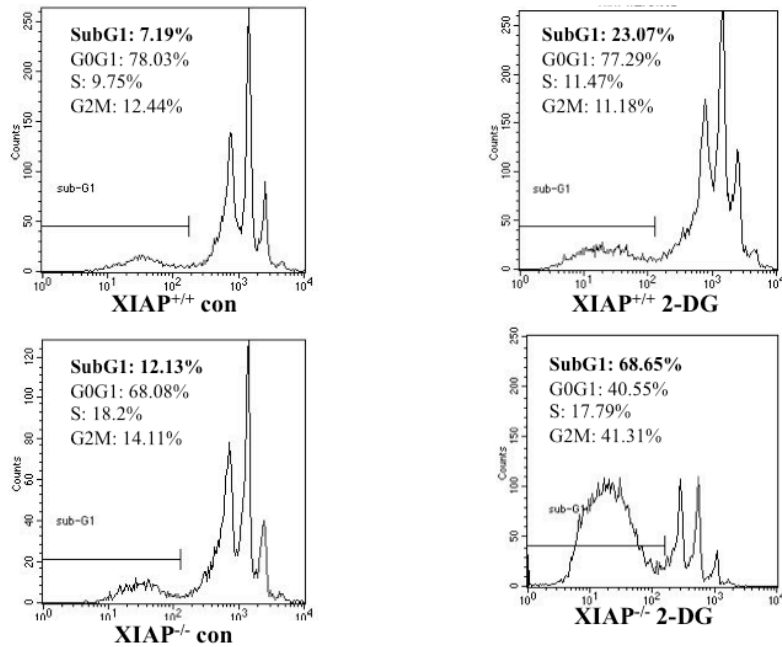
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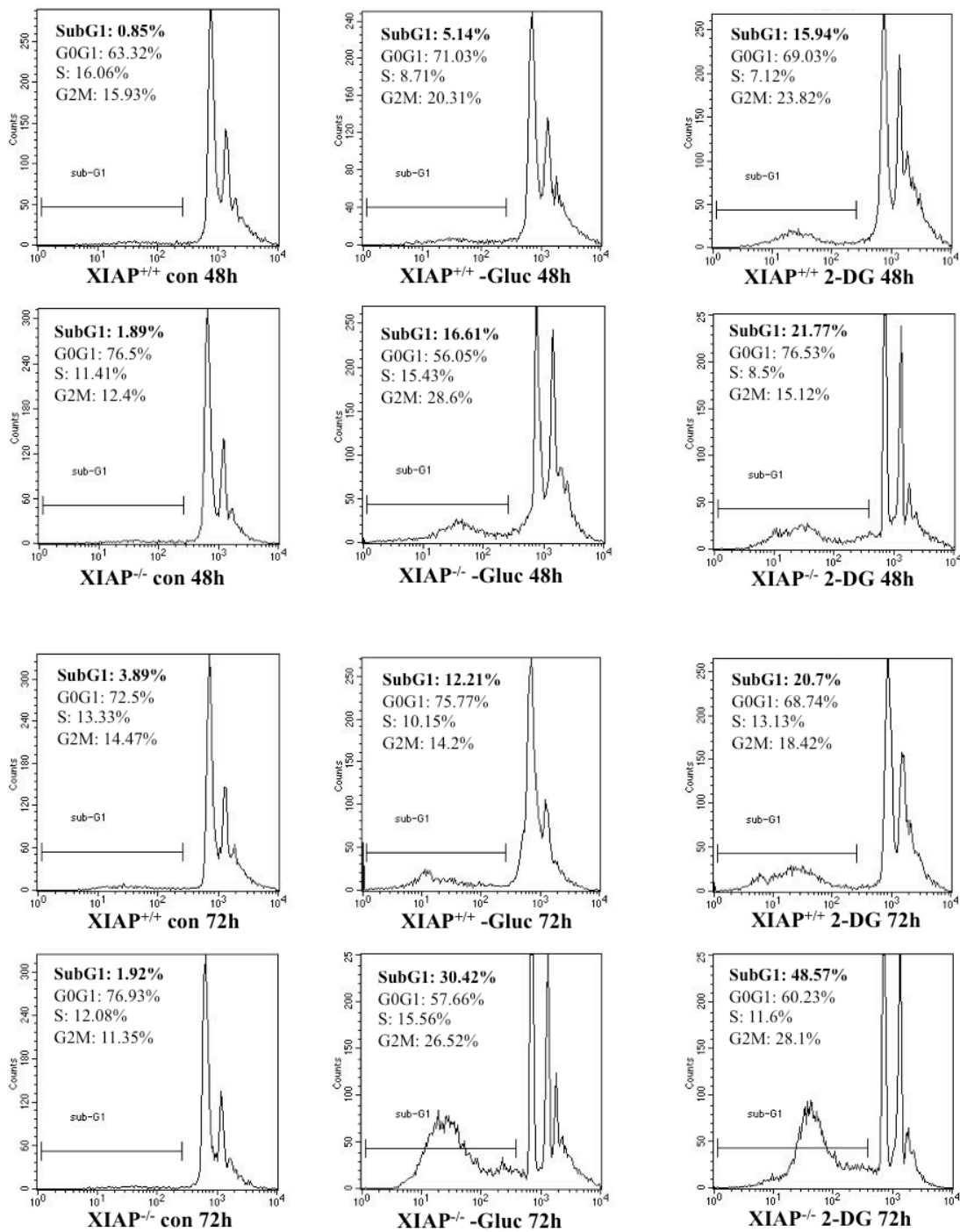
Supplementary Figure 6



Supplementary Figure 7



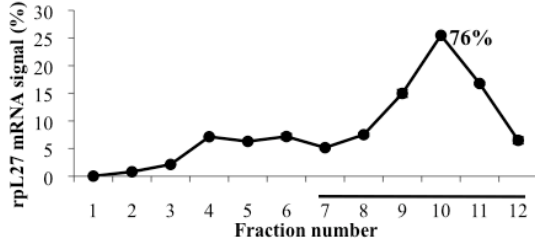
Supplementary Figure 8



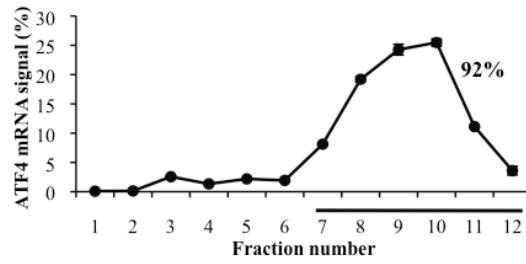
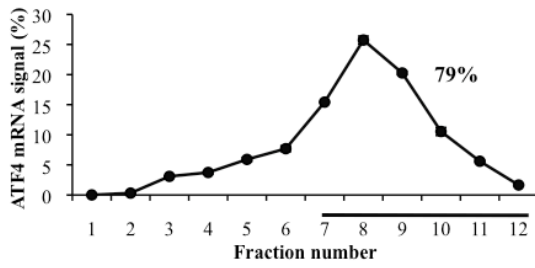
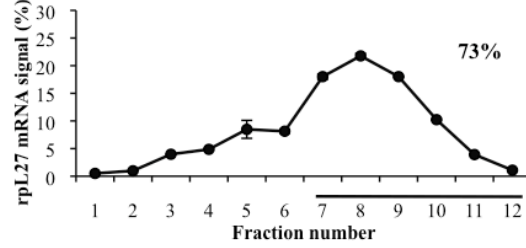
Supplementary Figure 9

A)

eIF2 α S/S con 30h

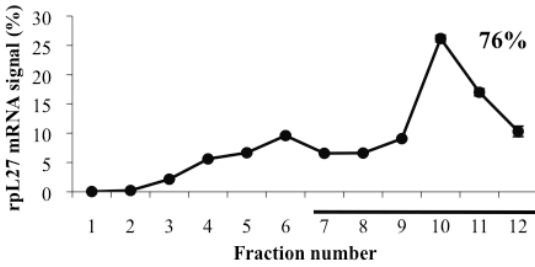


eIF2 α S/S -Gluc 30h

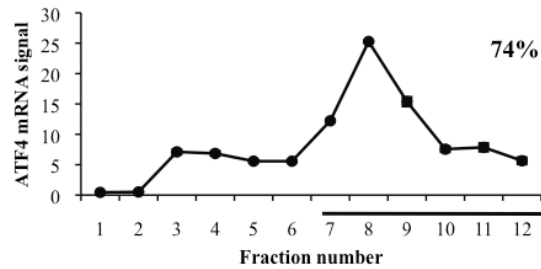
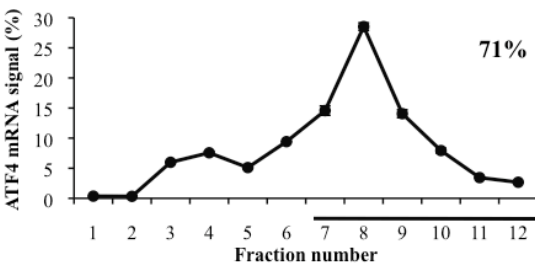
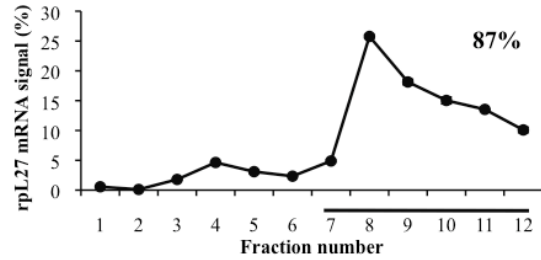


B)

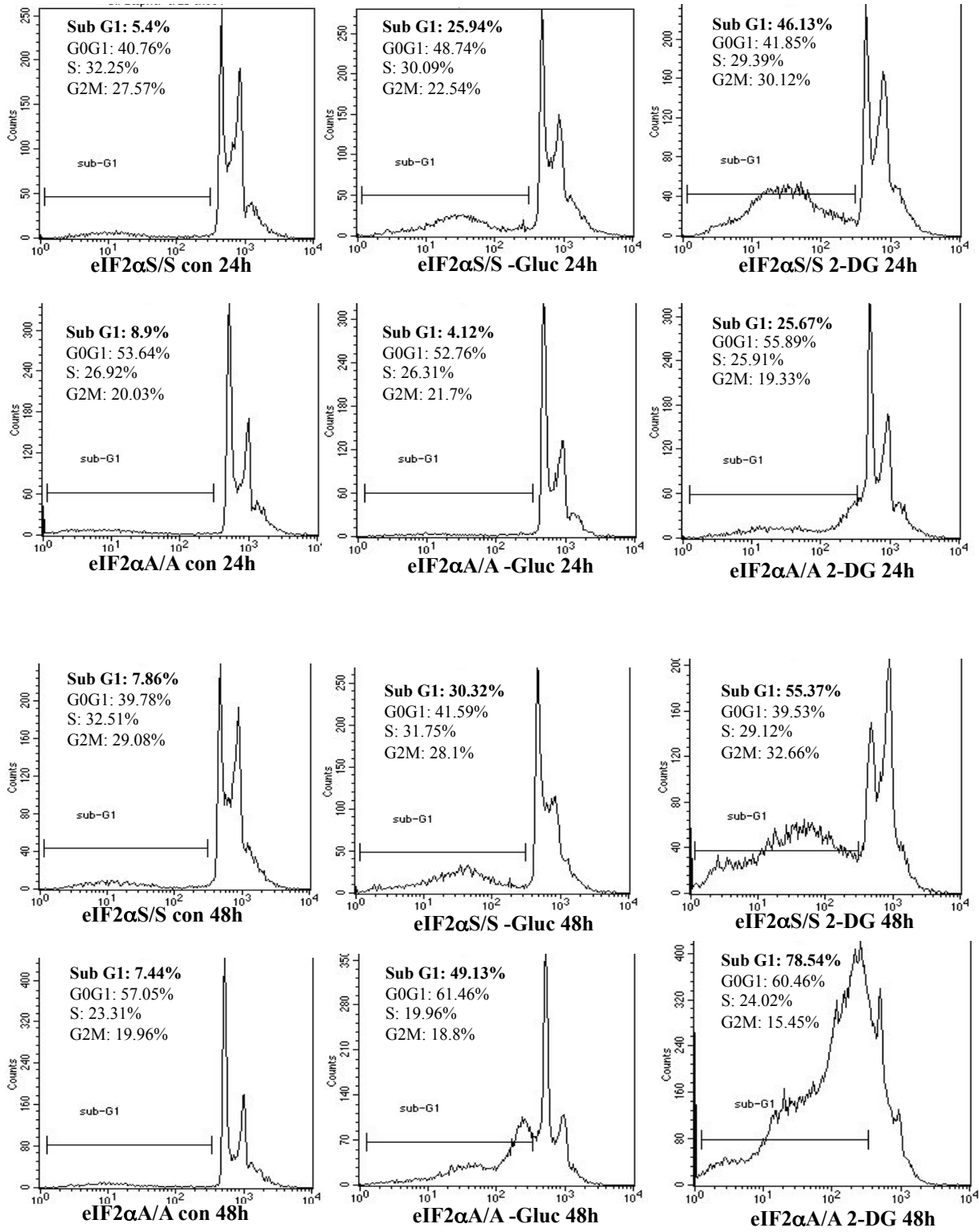
eIF2 α A/A con 30h



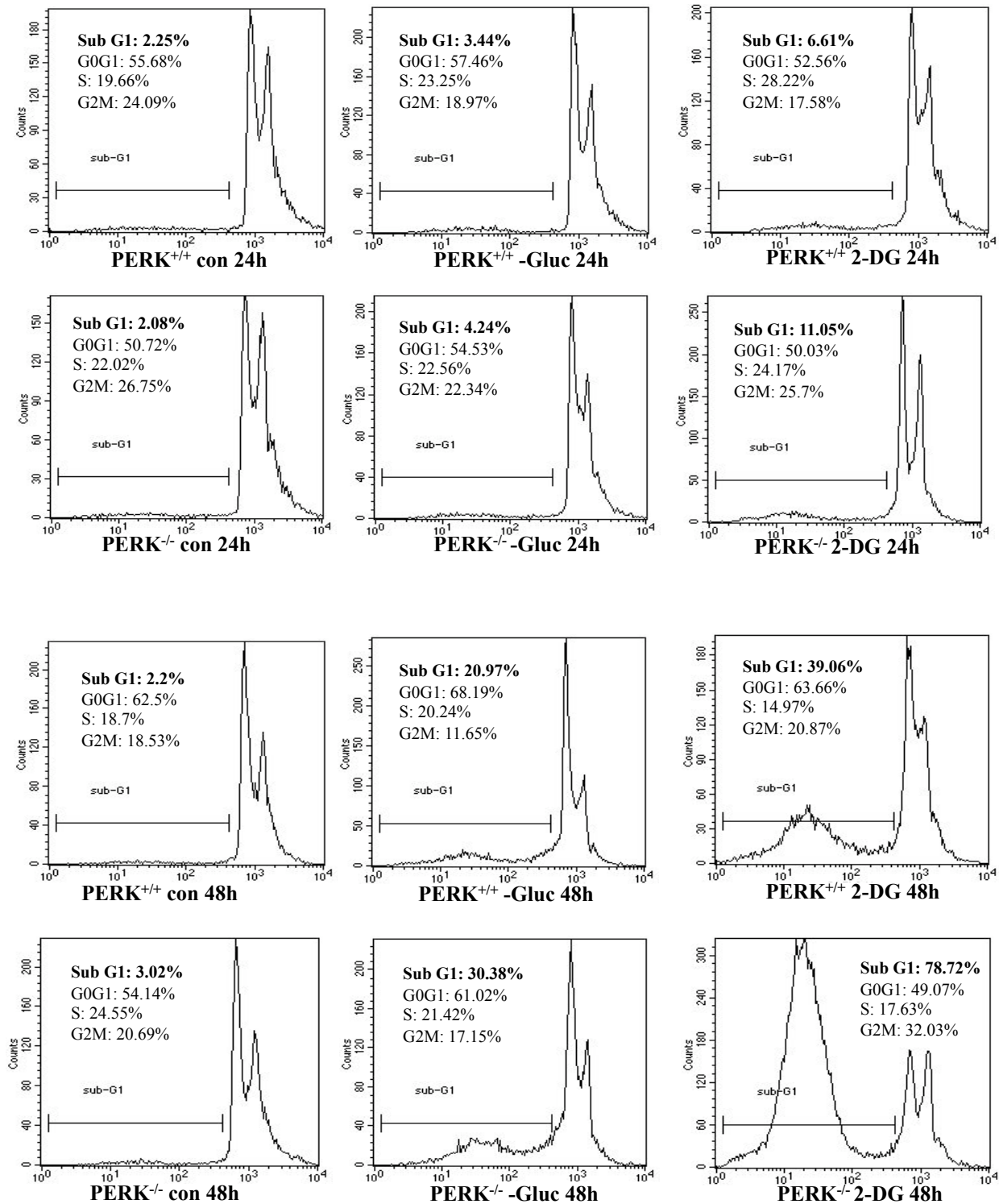
eIF2 α A/A -Gluc 30h



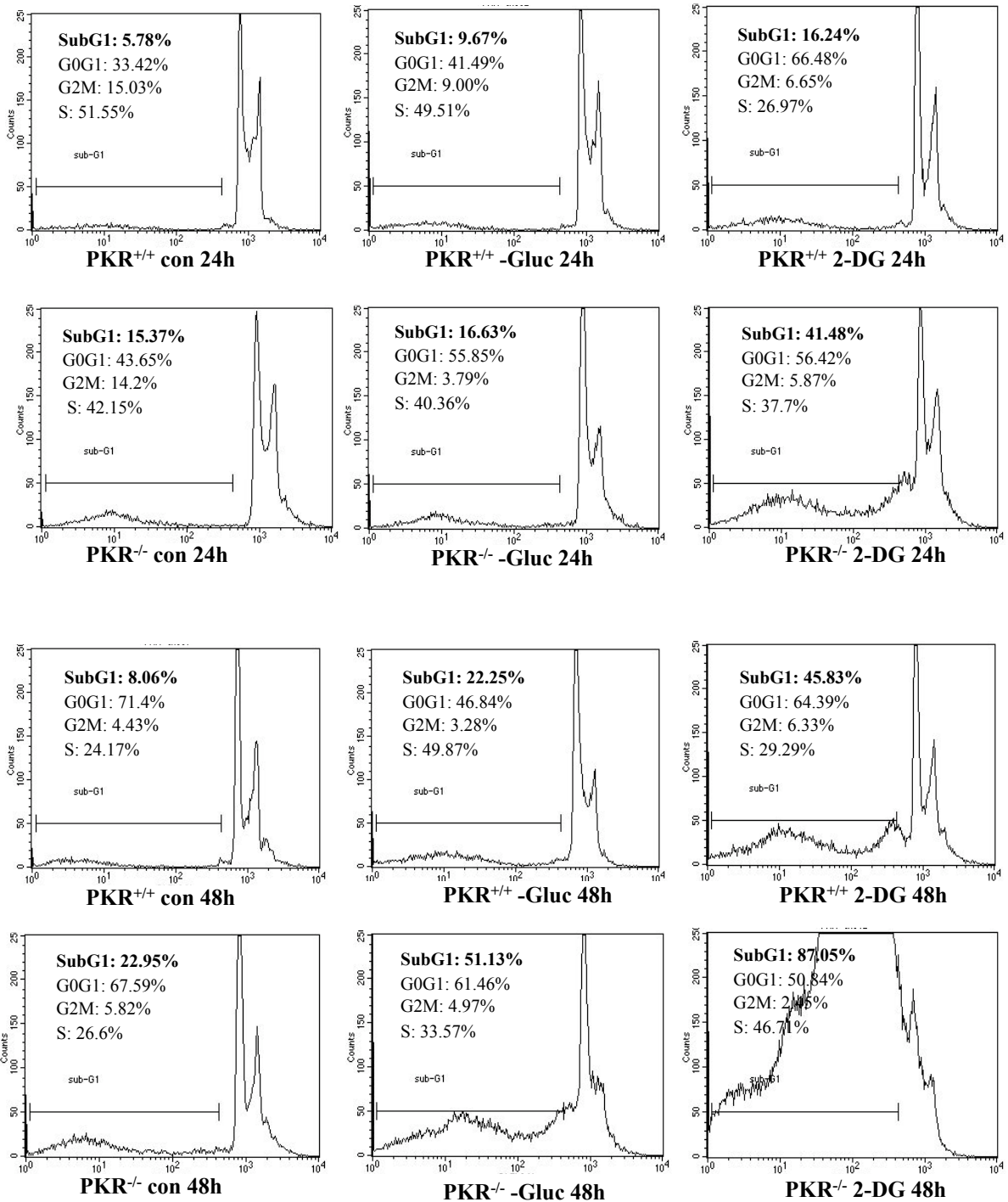
Supplementary Figure 1



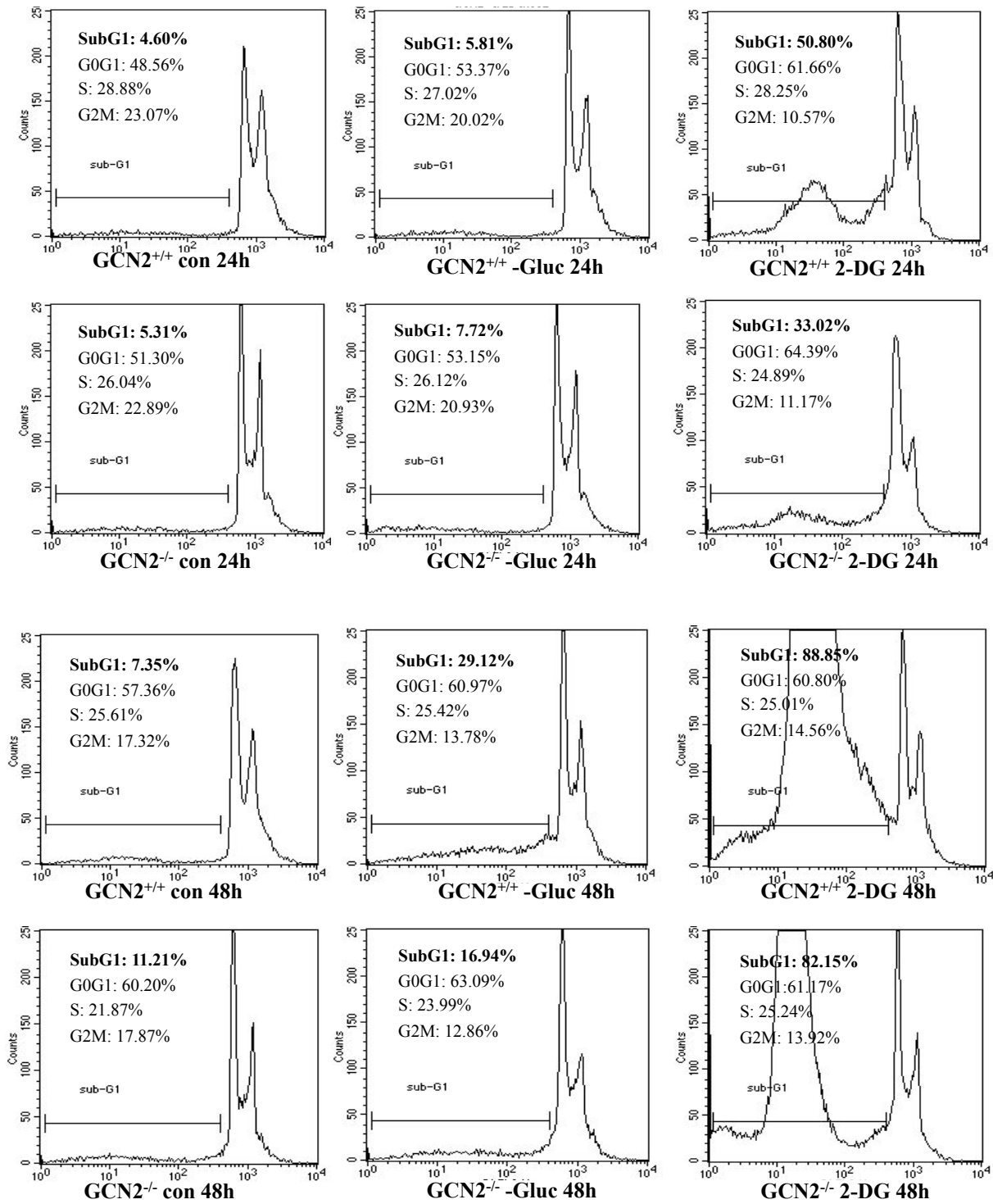
Supplementary Figure 2



Supplementary Figure 3

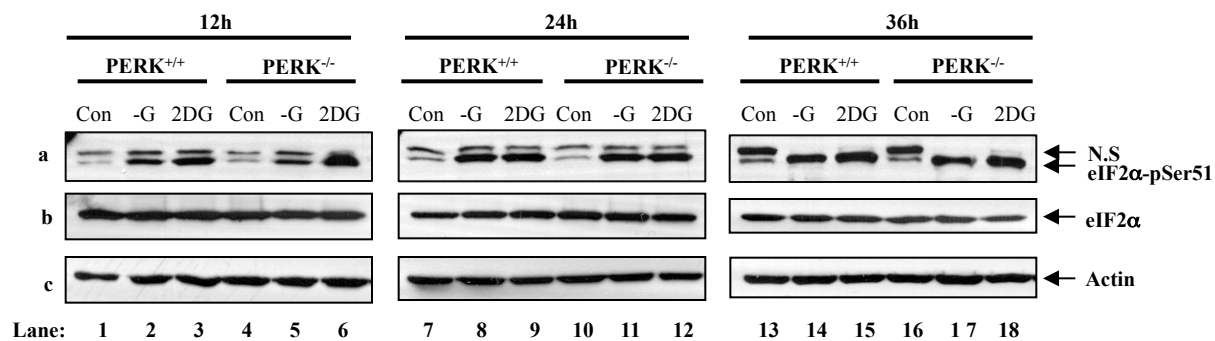


Supplementary Figure 4

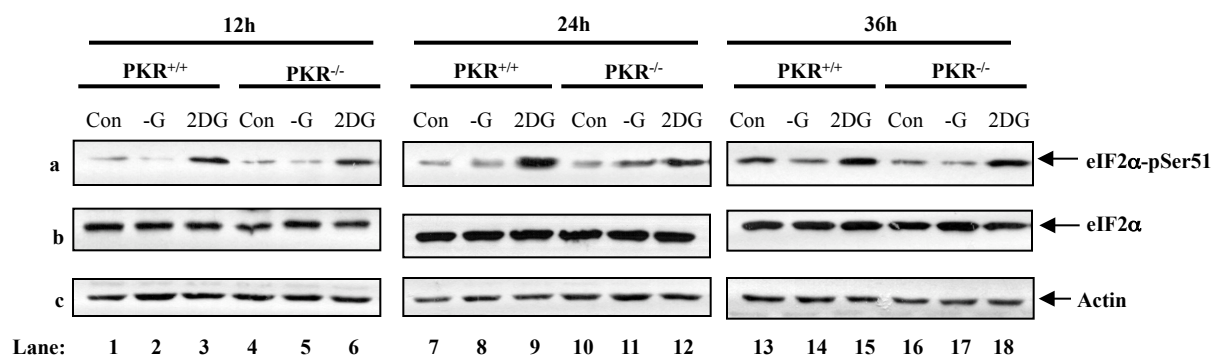


Supplementary Figure 5

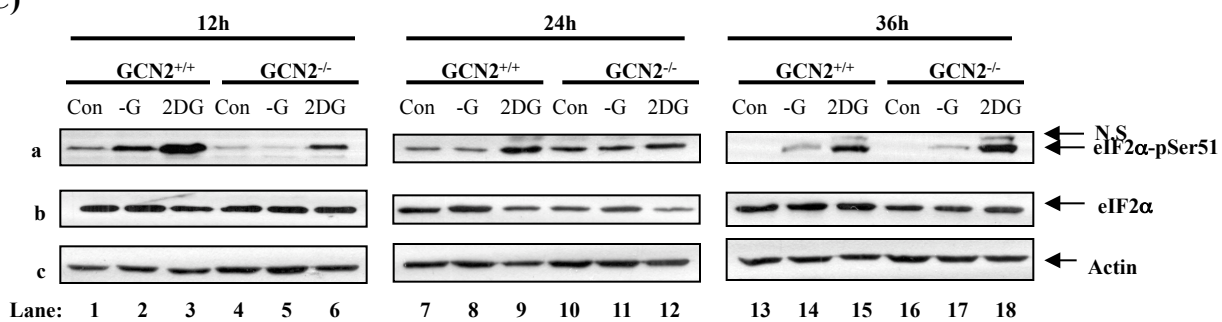
A)



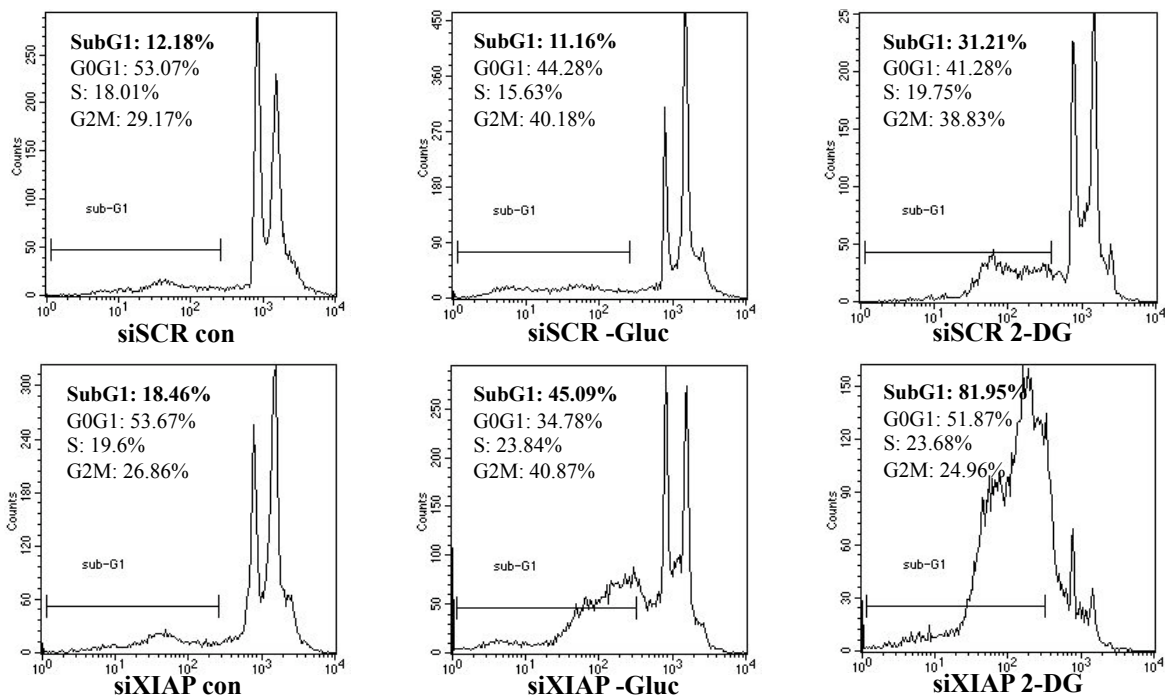
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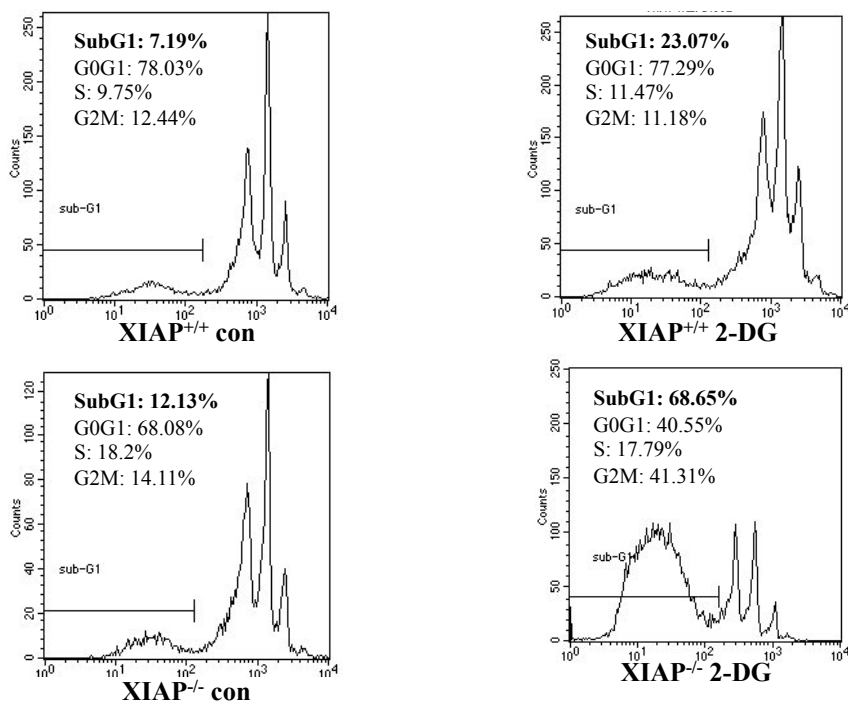
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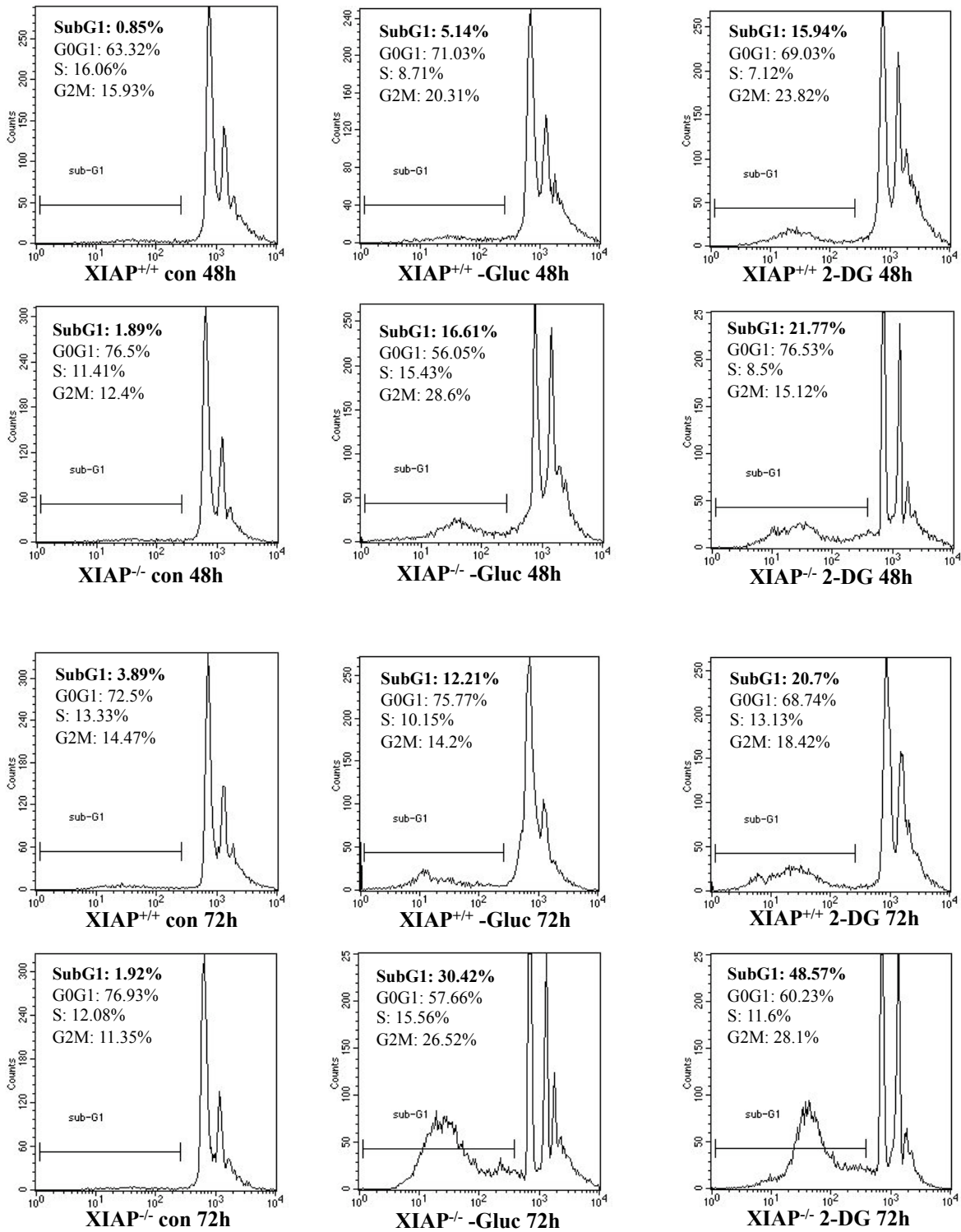
Supplementary Figure 6



Supplementary Figure 7



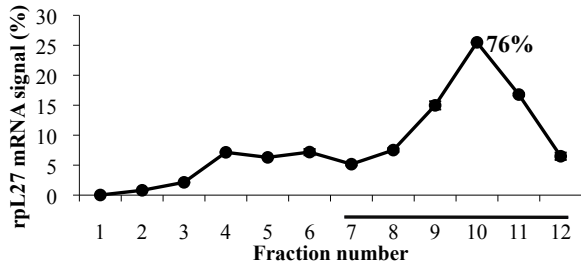
Supplementary Figure 8



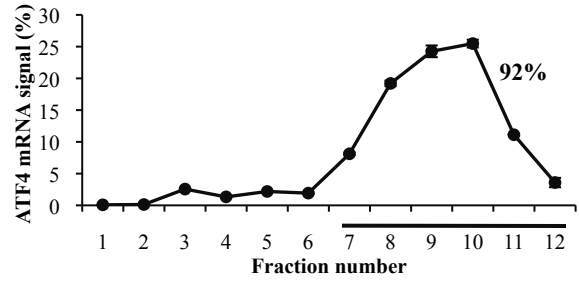
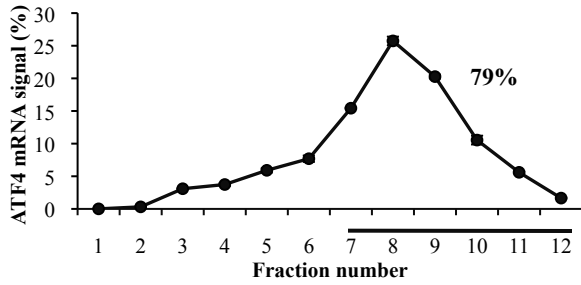
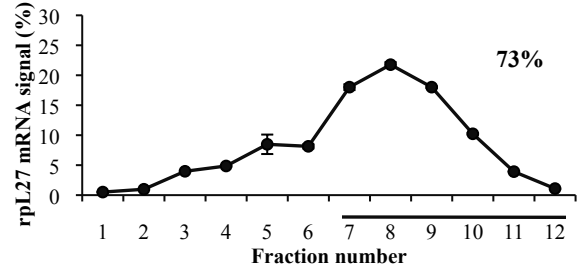
Supplementary Figure 9

A)

eIF2 α /S con 30h

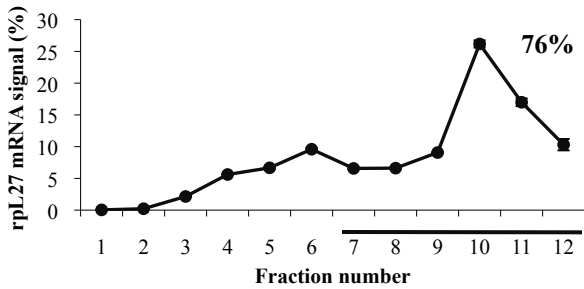


eIF2 α /S -Gluc 30h



B)

eIF2 α /A con 30h



eIF2 α /A -Gluc 30h

