Table 1) Global miRNA profiling of pancreatic islets of pregnant rats at day 14 of gestation
We compared by microarray analysis the expression of 350 miRNAs in pancreatic islets of four pregnant rats (at day 14 of gestation) with four age-matched female control rats. The table shows the miRNAs displaying significant expression changes ($p<0.05$) between the two groups. The data are given in a Log_2 scale and are the mean $\pm$ SD of four individual rats.

Microarray	Systematic Name	Control rats (Log_2 scale)	Pregnant rats (Log_2 scale)	Fold change	Expression change	p-values
Pregnant rats vs control rats	rno-miR-144	4.81±0.89	6.73±0.62	3.77	up	0.0124
	rno-miR-188	6.46±0.93	5.11±0.56	2.56	down	0.0469
	rno-miR-218	4.99±0.28	3.34±0.29	3.14	down	0.00017
	rno-miR-325-5p	1.83±0.88	3.34+0.29	2.86	up	0.0143
	rno-miR-338-3p	7.70±0.24	6.67±0.20	2.04	down	0.00057
	rno-miR-451	8.52±0.64	9.77±0.60	2.38	up	0.0286
	rno-miR-874	3.21±1.31	0.52±0.93	6.46	down	0.0155

Table 2) Impact of miR-338-3p inhibition and of different hormonal treatments on human β-cell proliferation. Dissociated human islet cells were transfected with a scrambled anti-miR or with anti-miR-338-3p. 48h treatments with prolactin (500ng/ml), estradiol (100nM), G1 (100nM), Exendin-4 (100nM) were used as controls for β-cell proliferation. The results are the mean ± SD of 3 independent experiments.

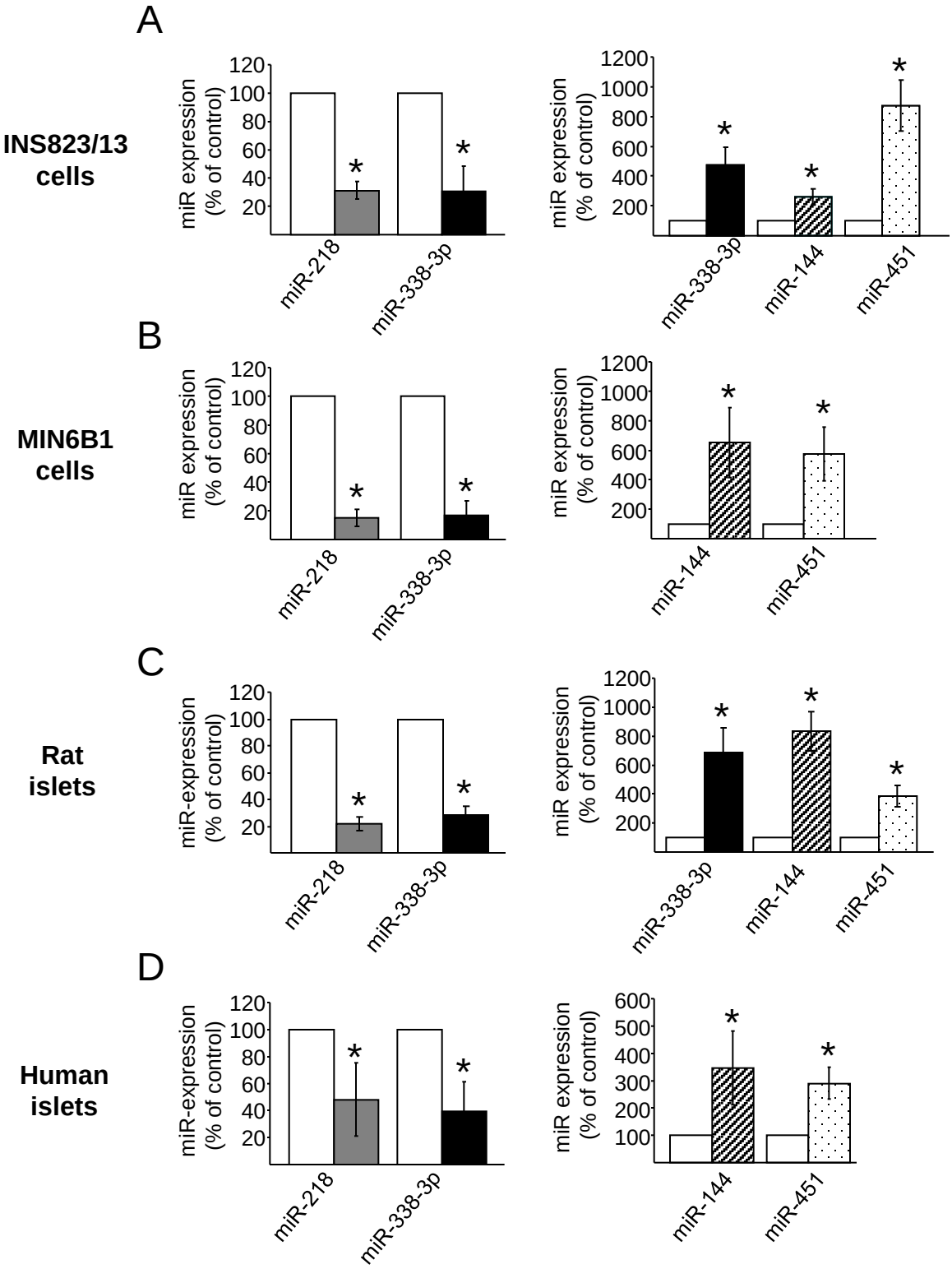
	Anti-control	Anti-338-3p	Prolactin	Estradiol	G1	Exendin-4
$Ki67^+$ cells	0	0	0	0	0	0
Ins^+ cells	715	683	409	500	391	432
Total cells	1241	1103	706	826	678	794
% Ins^+ cells	57.6 ± 6.7	61.9 ± 9.7	57.9 ± 9.8	60.5 ± 4.8	57.7 ± 6.1	54.4 ± 2.9

Table 3) List of gene ontology functions significantly enriched upon miR-338-3p down-regulation

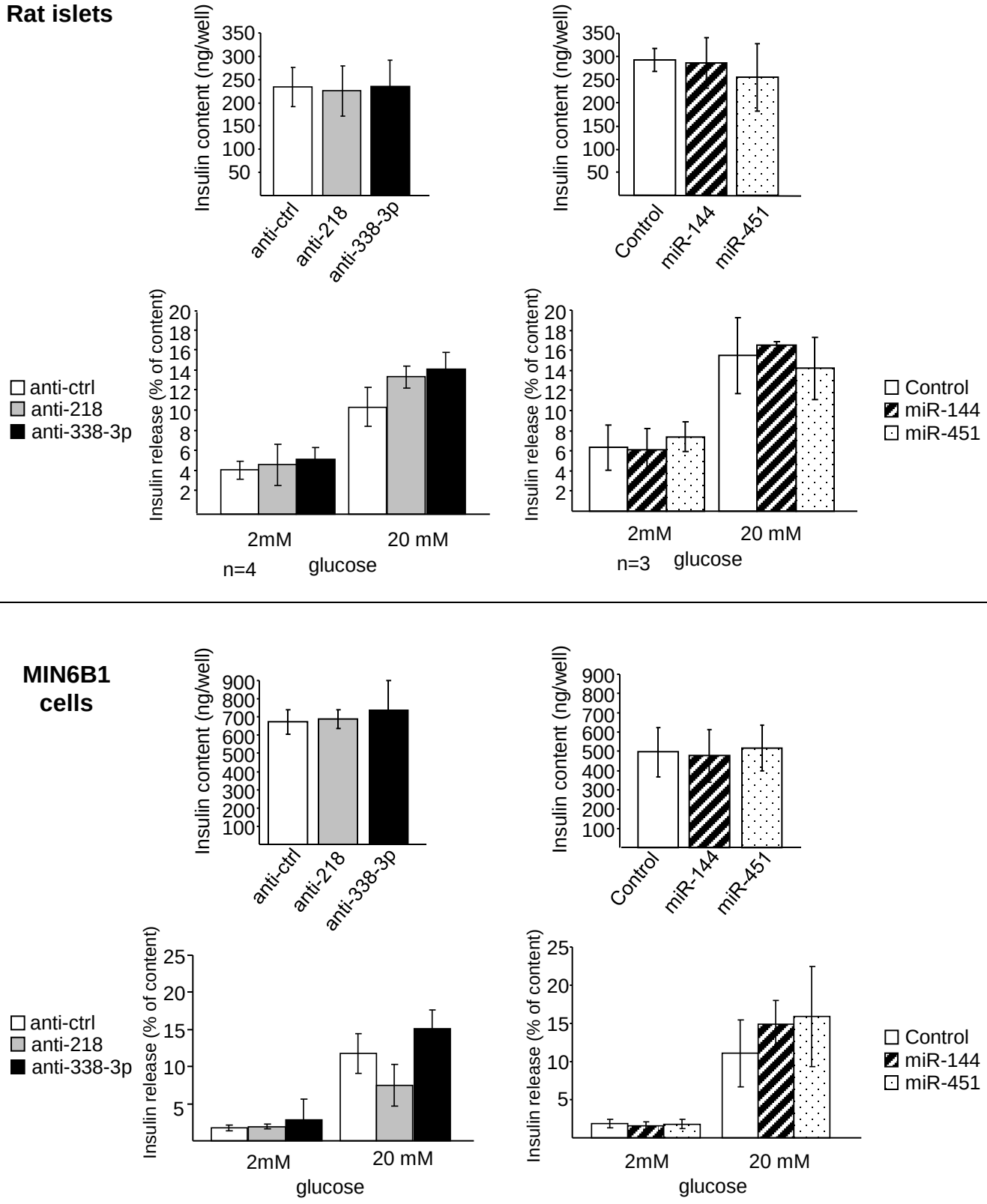
The genes involved in cell cycle and cell growth are highlighted in green.

	No. of genes	Benjamini *P* value
cellular process	284	1.49E-04
plasma membrane	105	0.03420889
multicellular organismal development	102	0.02322373
system development	90	0.03954513
nervous system development	51	0.02848449
cytoskeletal part	39	0.03434116
cytoskeleton	53	0.00740984
response to external stimulus	44	0.03155333
response to organic substance	51	0.01025913
cell projection	42	0.01987947
endomembrane system	37	0.03217145
endoplasmic reticulum	46	0.00897908
cellular ketone metabolic process	32	0.04672126
organic acid metabolic process	32	0.03866164
carboxylic acid metabolic process	32	0.03794723
oxoacid metabolic process	32	0.03794723
neuron projection	31	0.01076592
cellular carbohydrate metabolic process	24	0.03422616
regulation of growth	22	0.03075852
microtubule cytoskeleton	29	0.01197663
response to endogenous stimulus	43	1.19E-04
nucleotide metabolic process	22	0.02641974
nucleoside phosphate metabolic process	22	0.02641974
organelle fission	15	9.70E-04
mitosis	15	5.22E-04
nuclear division	15	5.22E-04
M phase of mitotic cell cycle	16	2.24E-04
cholesterol metabolic process	11	0.00896151
sterol metabolic process	12	0.00423696
organ regeneration	9	0.03075862

	No. of genes	Benjamini *P* value
nucleobase, nucleoside and nucleotide metabolic process	25	0.00985475
response to hormone stimulus	40	1.52E-04
cell cycle	37	1.31E-04
response to peptide hormone stimulus	18	0.03782789
synaptic transmission	18	0.03344988
response to steroid hormone stimulus	24	0.0057367
alcohol metabolic process	33	2.15E-04
response to organic cyclic substance	18	0.02697827
cell cycle process	31	2.14E-04
Golgi membrane	14	0.03482567
regulation of cell growth	15	0.04781072
axon	19	0.01351035
mitotic cell cycle	23	4.12E-04
cellular response to hormone stimulus	15	0.01341517
response to corticosteroid stimulus	14	0.02145489
cell cycle phase	25	1.29E-04
cell division	16	0.00722932
regeneration	13	0.02535536
response to glucocorticoid stimulus	14	0.0128922
spindle	11	0.03353471
M phase	20	4.41E-04
axon part	10	0.03259455
condensed chromosome	12	0.01229556
liver development	11	0.00738015
steroid biosynthetic process	12	0.00271587
condensed chromosome, centromeric region	7	0.03261501
pyruvate metabolic process	8	0.03004931
sterol biosynthetic process	11	3.23E-05
cholesterol biosynthetic process	10	4.06E-05
Steroid biosynthesis	8	7.79E-05

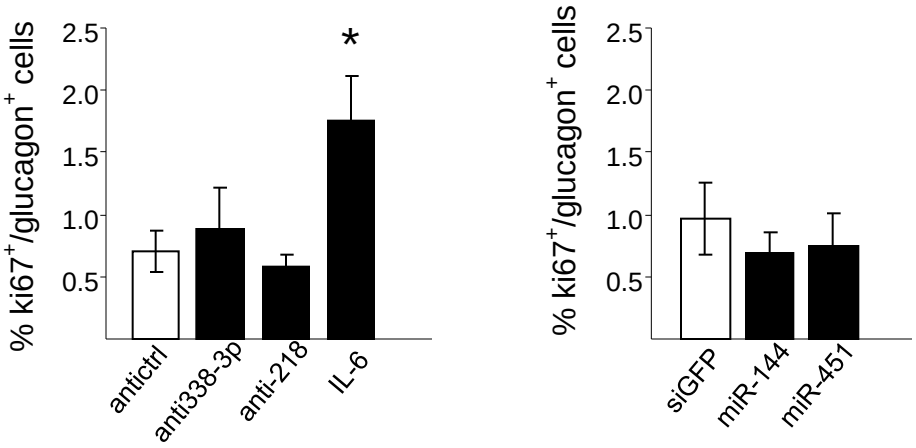


Supplementary Fig.1) Down- and up-regulation of miRNAs in cell lines and dispersed rat and human islet cells. INS832/13 cells (A), MIN6B1 cells (B), dissociated rat (C) and human islets (D) were transfected with either single stranded antisense oligonucleotides to reduce (left panels) or with oligonucleotide mimic to increase (right panels) the expression of the indicated miRNAs. A scrambled anti-miR sequence and a siRNA duplex against GFP were used as controls for the experiments on the left and on the right, respectively. The miRNA levels were measured 48h later by qRT-PCR. The results are the mean ± SD of 4 (INS832/13 and MIN6B1 cells) or 3 (rat and human islets) independent experiments. Statistical differences were determined by ANOVA (* $p<0.05$).

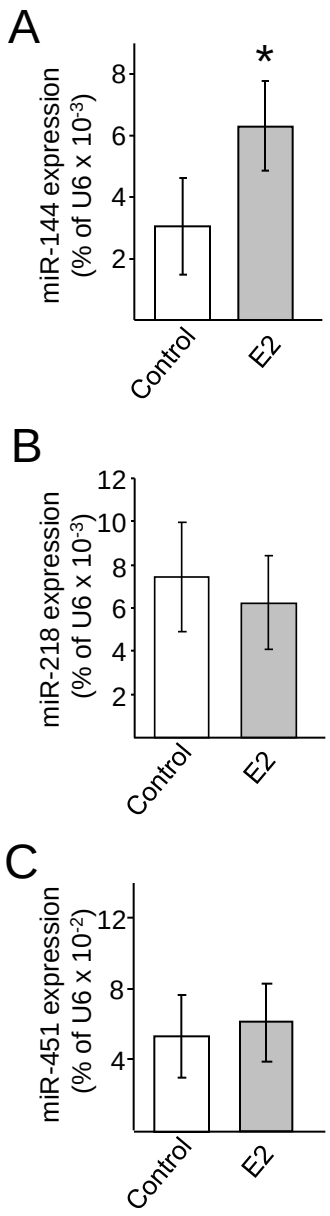


Supplementary Fig.2) Changes in miRNA expression mimicking those occurring during pregnancy do not impair glucose-induced insulin secretion.

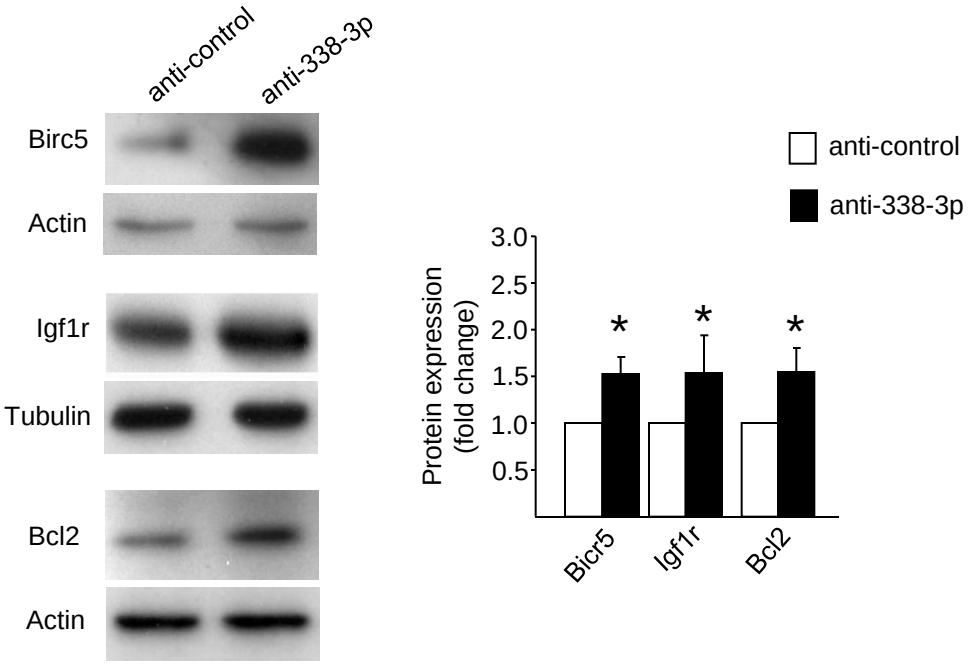
Dissociated rat islet cells (upper panels) or MIN6B1 cells (lower panels) were transfected with the indicated anti-miRs (left panels) or with miRNA mimics (right panels). Two days later the cells were incubated at 2 or 20 mM glucose for 45 minutes. Insulin content and insulin secretion were measured by ELISA. The results are the mean ± SD of 4 independent experiments.* Significantly different from control condition ($p < 0.05$ by ANOVA analysis).



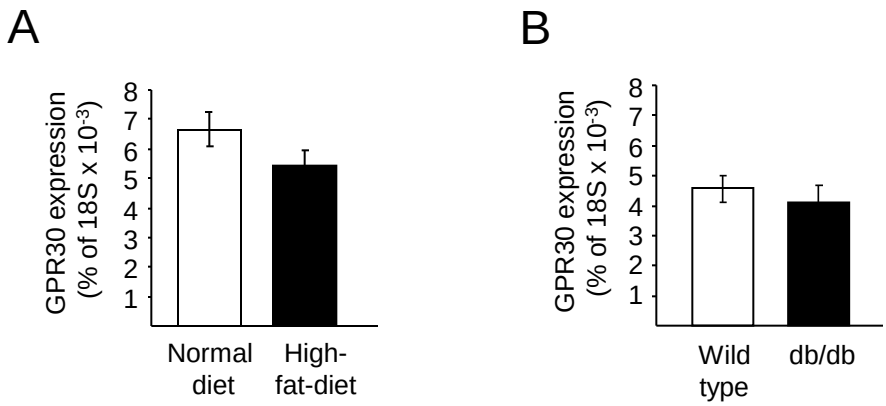
Supplementary Fig.3) Impact of modifications in miRNA levels on α-cell proliferation. Dissociated rat islet cells were transfected with the indicated anti-miR (left panels) or with oligonucleotide mimics (right panels) to modulate the expression of the indicated miRNAs. IL-6 treatment (100ng/ml for 48h) was used as positive control for α-cell proliferation. The fraction of proliferating glucagon-positive cells was assessed using a Ki67 antibody. The data are the mean ± SD of 5 independent experiments. * Significantly different from control condition ($p < 0.05$ by ANOVA analysis).



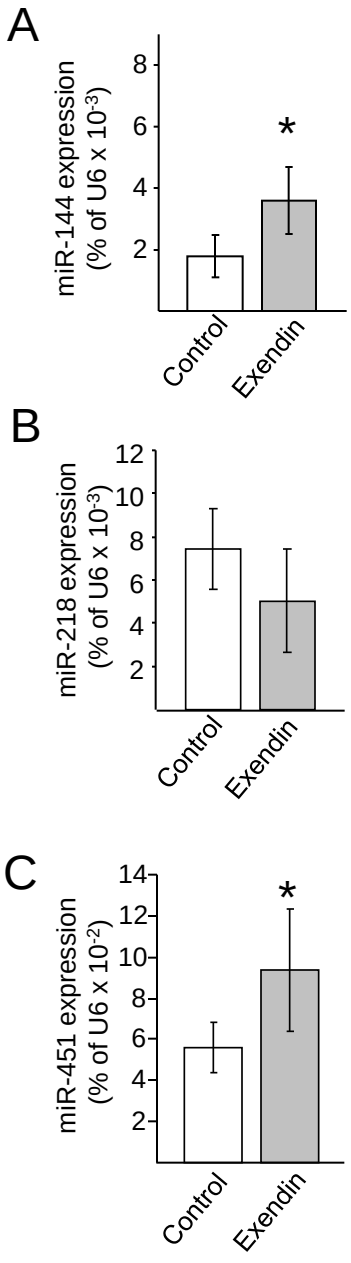
Supplementary Fig.4) miRNAs expression in rat islets after exposure to estradiol.
Rat pancreatic islets were incubated in the absence (Control) or in the presence of 100 nM estradiol (E2) for 48h. The expressions of miR-144 (A), miR-218 (B) and miR-451 (C) were measured by qRT-PCR and are expressed as % of U6. The values correspond to the mean ± SD of 4 independent experiments. * Significantly different from control condition ($p < 0.05$ by ANOVA analysis).



Supplementary Fig.5) Changes in protein expression induced by miR-338-3p down-regulation. INS832/13 cells were transfected with a scrambled anti-miR or with anti-miR-338-3p. Two days later the cells were collected and the expression of Birc5, Igf1r and Bcl2 proteins analyzed by Western Blotting. Examples of blots are shown on the left. Quantification of the bands is given on the right. The results are expressed as fold changes and correspond to the mean ± SD of 4, 5 and 6 independent experiments for Birc5, Ifg1r and Bcl2, respectively. * Significantly different from control condition ($p < 0.05$ by ANOVA analysis).



Supplementary Fig.6) GPR30 expression is not changed in islets from mice fed a high fat diet and from *db/db* mice. GPR30 levels were measured by qRT-PCR in islets from mice fed a high-fat-diet for 8 weeks (A) and in 6 week-old normoglycemic *db/db* mice (B). The data are expressed as % of 18S. They are the means ± SD of 3 different mice per group.



Supplementary Fig.7) Impact of Exendin-4 on miRNAs expression in rat islets.
Rat islets were treated with 100 nM Exendin-4 for 48h. miR-144 (A), miR-218 (B) and miR-451 (C) expression was measured by qRT-PCR.. The data are expressed as % of U6 and correspond to the mean ± SD of 4 independent experiments for miR-144 and miR-218 and of 6 independent experiments for miR-451. * Significantly different from control condition ($p < 0.05$ by ANOVA analysis).