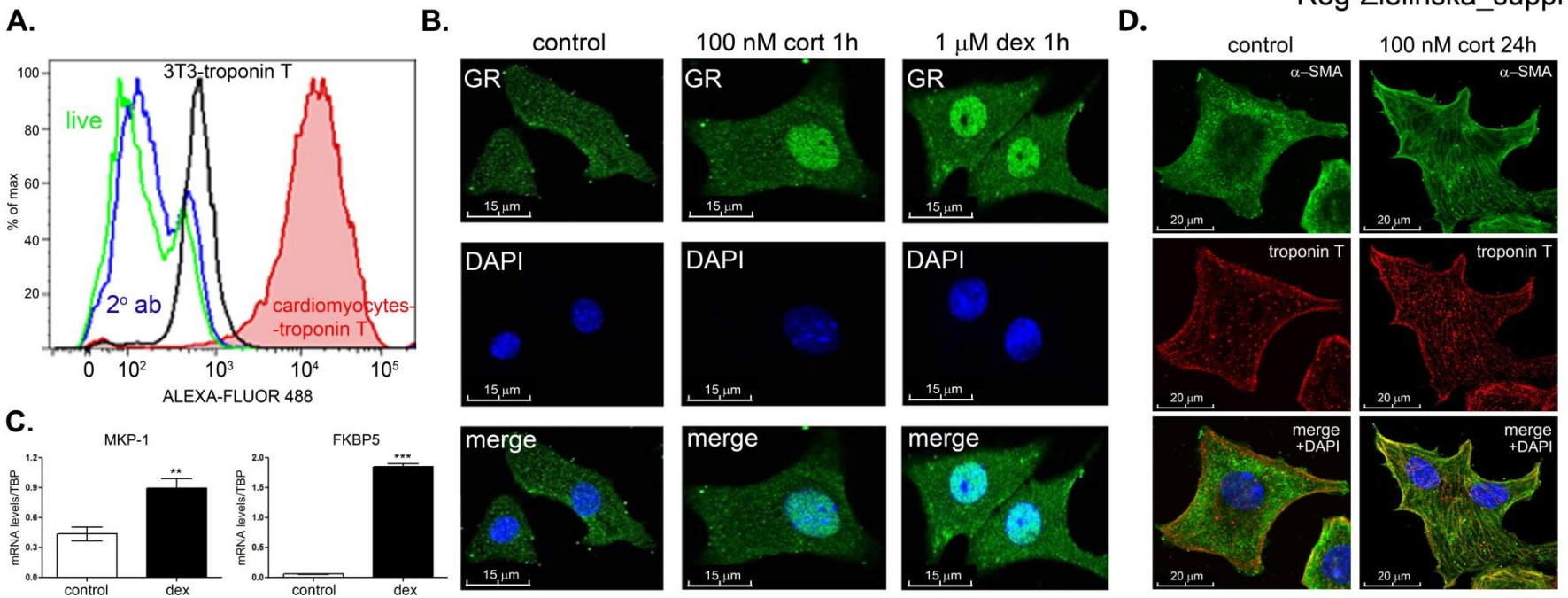

Supplementary figure 1. **(A)** Purity of primary foetal C57BL/6 mouse cardiomyocytes was assessed using a BD LSR Fortessa flow cytometer following labelling with anti-troponin T antibody (Abcam; Cambridge, UK) and fluorophore-conjugated secondary antibody (Alexa488, Invitrogen; Paisley, UK); undifferentiated 3T3-L1 (preadipocyte) cells were used as a negative control. Troponin T-positive cardiomyocytes are shown in red; 3T3-L1 cells stained with troponin-T and used as a negative control are shown in black. Live cells are shown in blue and cells stained with secondary antibody only, in green. Troponin T-positive cardiomyocytes represented 98% of total cells recovered from foetal hearts. **(B)** GR translocates to the nucleus of primary murine cardiomyocytes following 1 h treatment with 100 nM corticosterone or 1 μM dexamethasone. Cells were stained with anti-GR antibody (green) and DAPI (nuclear counterstain, blue); images were captured using a confocal microscope and are representative of n=5 (replicates). (**C**) Levels of mRNA encoding MKP-1 (*Dusp-1*) and FKBP5, known GR targets, were increased in primary cardiomyocytes following treatment with 1 μM dexamethasone for 24 h. Data were analysed by Student's *t*-test; **p<0.01, ***p<0.001, n=3 replicates. (**D**) Corticosterone induced the formation of troponin T-associated α-smooth muscle actin fibers in primary foetal cardiomyocytes. Cells were treated with 100 nM corticosterone for 24 h, fixed and stained for a-smooth muscle actin (a-SMA, green) and troponin T (red). DAPI (blue) was used as a nuclear counterstain. Bottom panel shows merged images. Images representative of n=12 replicates over two independent experiments.

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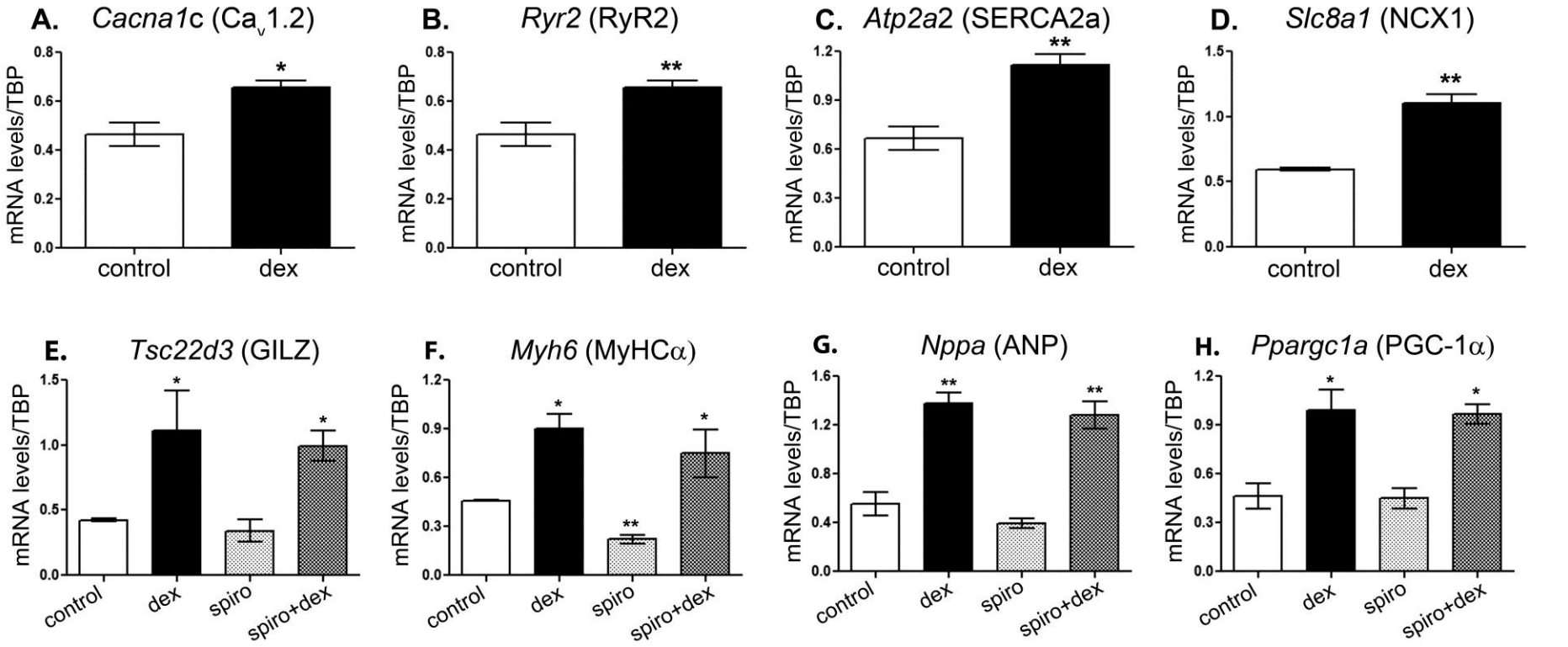
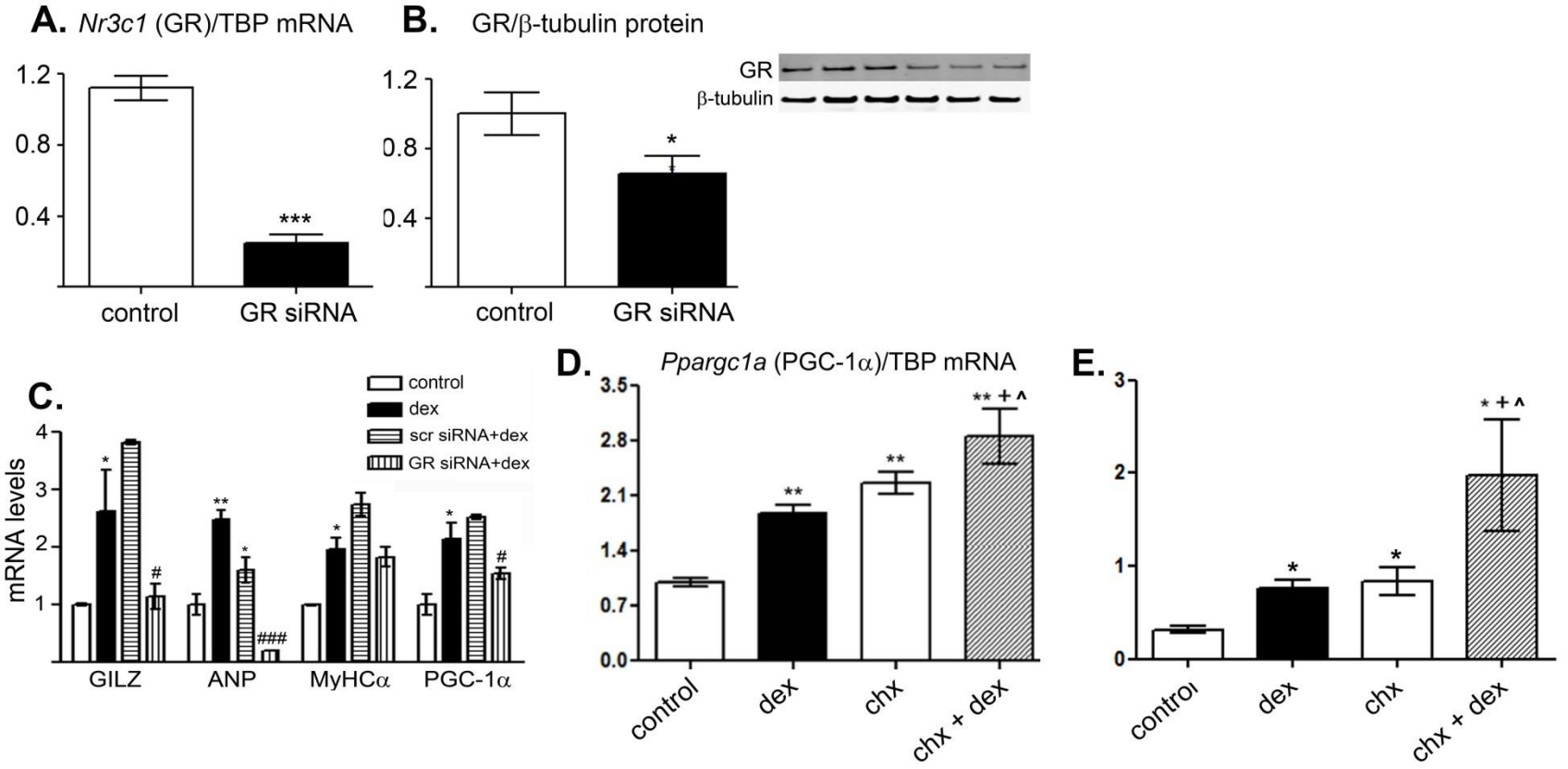
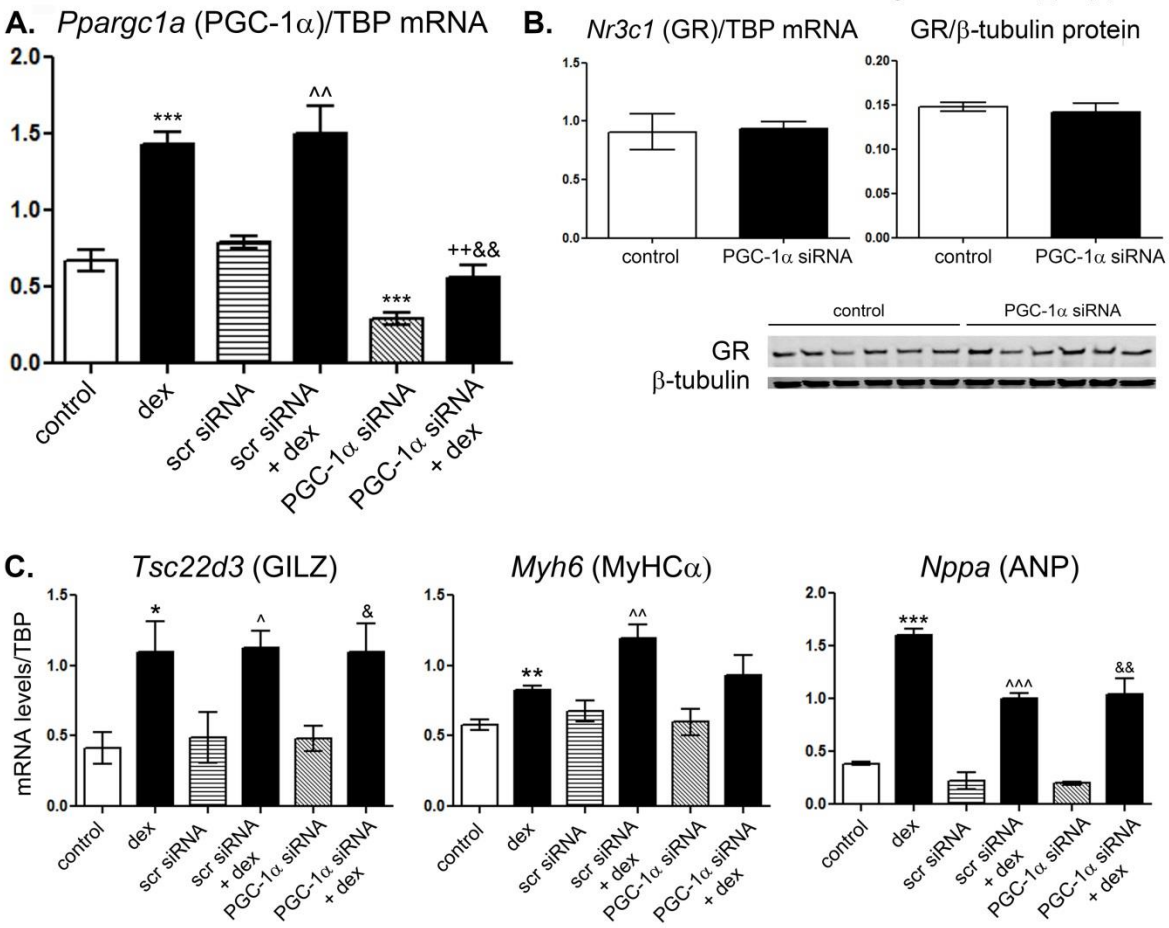


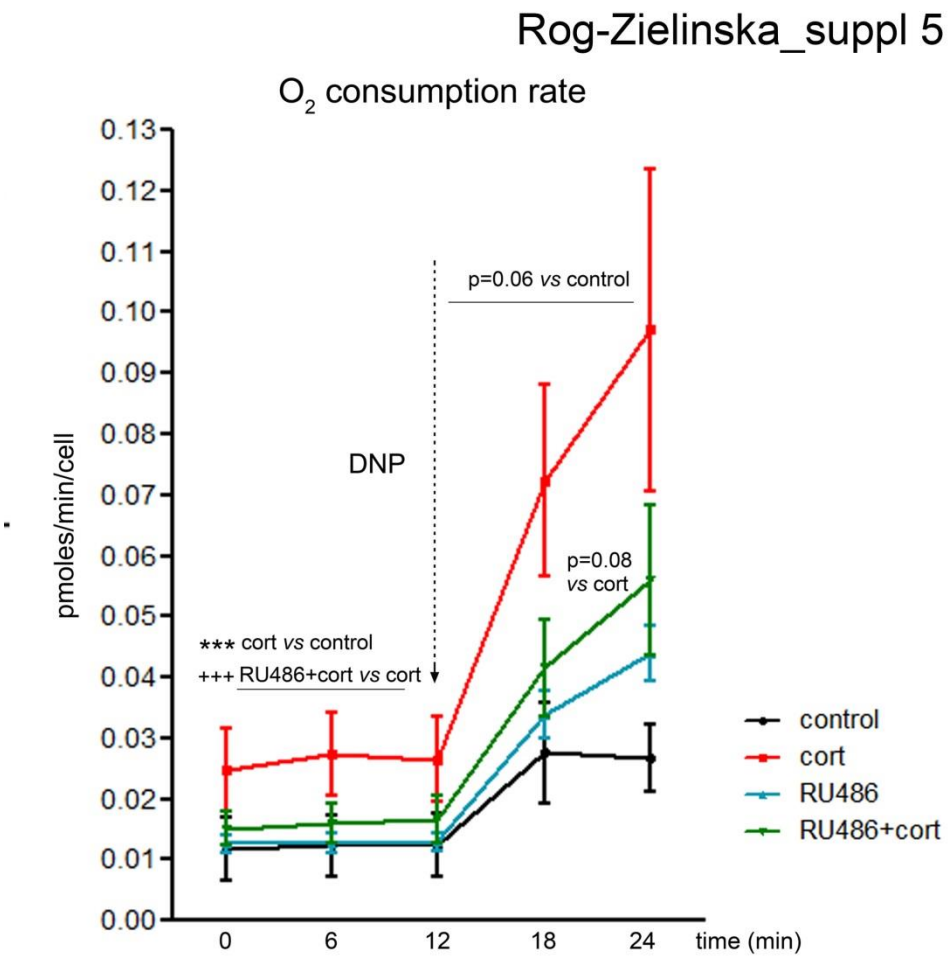
Supplementary figure 2. **(A-D)** Dexamethasone treatment (1 µM, 24 h) of primary cardiomyocytes induced mRNAs encoding proteins involved in calcium handling. Data were analysed by Student's *t*-test; *p<0.05, **p<0.01; n=3 replicates. **(E-H)** Pre-treatment of primary foetal cardiomyocytes with the mineralocorticoid receptor antagonist, spironolactone (spiro, 30 min, 10 µM) had no effect on dexamethasone (1 µM, 16 h) induction of mRNAs encoding GILZ, MyHCα, ANP and PGC-1α. Data were analysed by one-way ANOVA with Bonferroni's *post-hoc* test; *p<0.05, **p<0.01 *vs* control; n=4 replicates.



Supplementary figure 3. Transfection with GR siRNA reduced GR mRNA (**A**) and protein levels (**B**) in primary foetal cardiomyocytes. Data were analysed by *t*-test; *p<0.05, ***p<0.001; n=7 replicates over 2 independent experiments. (**C**) GR siRNA attenuated the dexamethasone induction of mRNAs encoding GILZ, ANP, MyHCα and PGC-1α but transfection with scrambled siRNA (scr siRNA) for 40 h prior to glucocorticoid treatment did not impair dexamethasone induction of these mRNAs. Data were analysed by one-way ANOVA with Bonferroni's *post-hoc* test; *p<0.05, **p<0.01; n=6-8 replicates of two independent experiments. **(D-E)** Dexamethasone (100 nM) increases *Ppargc1a* mRNA levels after 2h (D) and 8h (E) in primary foetal cardiomyocytes, effect that in not abolished by pre-treatment with cycloheximide. Data were analysed by one-way ANOVA with Bonferroni's post-hoc test; *p<0.05, **p<0.01 *vs* control, $^{+}$p<0.05 *vs* dex, ^p<0.05 *vs* chx; n=3 replicates.



Supplementary figure 4. (**A**) Transfection with PGC-1α siRNA reduced PGC-1α mRNA levels in primary foetal cardiomyocytes whereas scrambled (scr) siRNA had no effect. Data were analysed by one-way ANOVA with Bonferroni's *post-hoc* test; **/++/^^/&&p<0.01, ***p<0.001; **vs* untreated control, *+vs* dex, *^vs* scrambled siRNA, *&vs* PGC-1α siRNA; n=6-8 replicates of two independent experiments. **(B)** Transfection with PGC-1α siRNA did not affect mRNA or protein levels of GR. **(C)** siRNA-mediated knock down of PGC-1α does not impair transcriptional responses to dexamethasone in primary foetal cardiomyocytes. Cells were treated with 1 μM dexamethasone for 16 h. To block PGC-1α -mediated effects, cells were transfected with PGC-1α siRNA 40 h prior to glucocorticoid treatment**.** Data were analysed by one-way ANOVA with Bonferroni's *post-hoc* test; */^/$^{\&}$p<0.05, **/^^/$^{\&}$p<0.01, ***/^^^p<0.001; **vs* untreated control, *^vs* scrambled siRNA, *$^{\&}$vs* PGC-1α siRNA; n=3 replicates.



Supplementary figure 5. Glucocorticoids increase mitochondrial respiration in primary foetal cardiomyocytes in a GR-dependent manner. Real-time oxygen consumption rate (OCR) was measured in untreated (control) cardiomyocytes or following treatment with 100 nM corticosterone for 24 h (cort). To test mitochondrial reserve, 2,4-DNP was added to acutely uncouple ATP synthesis from respiration. Cells were pre-treated with RU486 (1 µM, 30 min). Data were analysed by one-way ANOVA with Bonferroni's *post-hoc* test (first 3 time points), and the area under the curve (AUC) was measured for subsequent readouts; ***p<0.001 *vs* control, $^{+++}$p<0.001 *vs* cort; n=5 replicates.