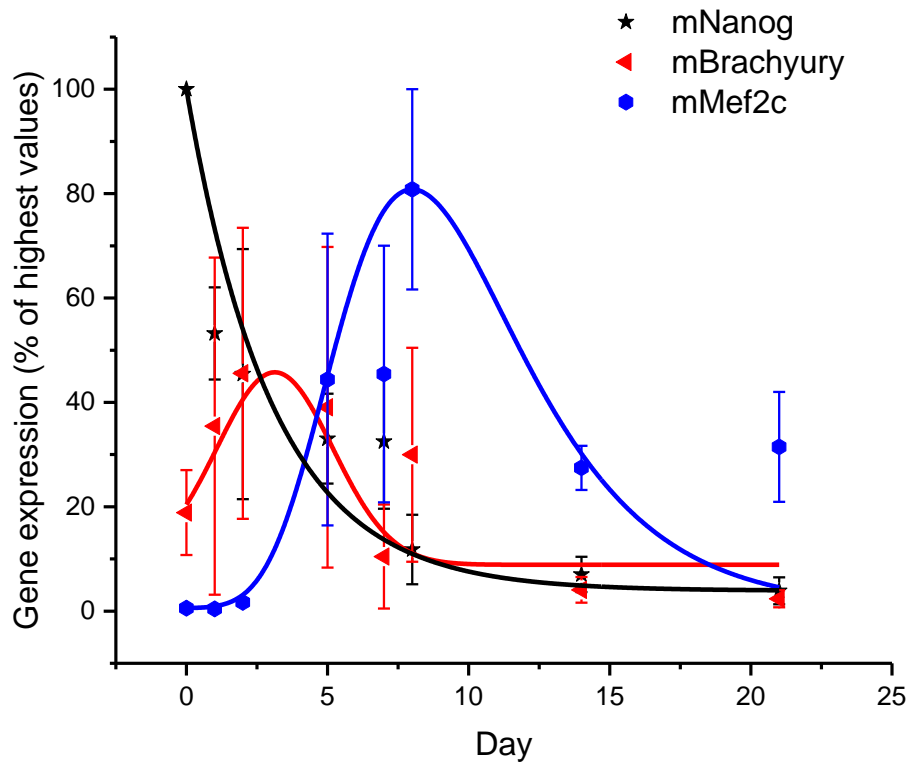
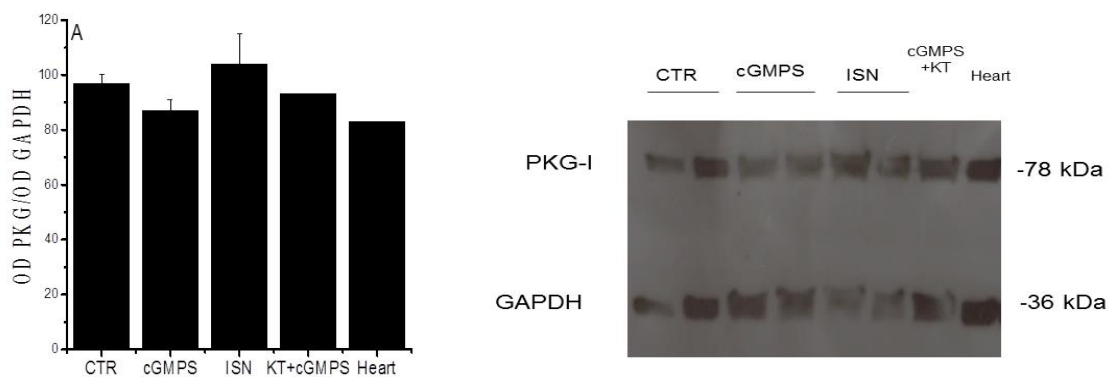


Table 1S – Amplicon context sequences of primers used for Q-PCR

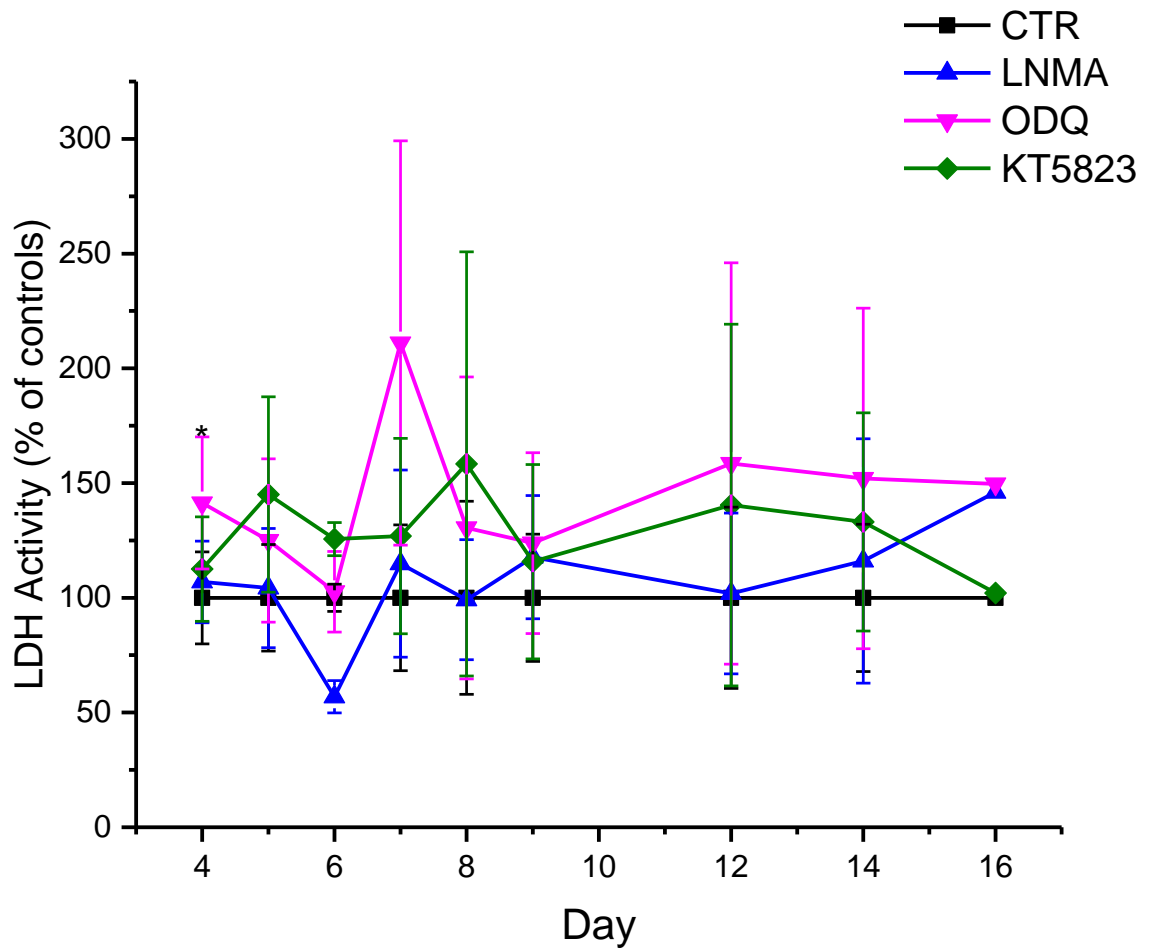
Mouse Glyceraldehyde-3-Phosphate Dehydrogenase (mGapdh)	TGGGAGTTGCTGTTGAAGTCGCAGGAGACAACCTGGTCTCAGTGTAGCCCAAGATGCCCTTCAGTGGGCCCTCAGATGCCTGCTTACCACCTTCTTGATGTCA
Mouse Octamer-binding Transcription factor 4 (mOct4)	ATGCAAATCGGAGACCCTGGTGCAGGCCCGGAAGAGAAA GCGAACTAGCATTGAGAACCCTGTGAGGTGGAGTCTGGAG ACCATGTTTCTGAA
Mouse Brachyury (mBrachyury)	CCACCGGTCATCGCCCTACCCCAGCCCCTATGCTCATCGGA ACAGCTCTCCAACCTATGCGGACAATTCATCTGCTTGTCTG TCCATGCTGCAGTCCCATGATAACTGGTCTAGCCTCGGAGT
Mouse Myocyte Enhancer factor 2C (mMef2c)	TTCAGATTACGAGGATAATGGATGAGCGTAACAGACAGGT GACTTTTACGAAGAGGAAATTTGGATTGATGAAGAAGGT TATGAGCTGAGCGTGTGCGACTGTG AGATTGCACTG
Mouse GATA binding protein 4 (mGata4)	CTGGGACATGGAGCTGCTGTGCCCATAGTGAGATGACAGC CCGGGCTCTGTCTTGATGGGGCGCATCTTCTACTGCTGCT GCTGCTGCTAGTGGCATTGCTGGAGTTACCGCTGGAGGCA CCACTGGAGGGAGGGAGGGTCTCACCAGCAGGACCTGCT GGCGTCTTAGATTTATTCAGGTTCTTGGGCTTCCGTTTTCTG GT
Mouse NK2 transcription factor related, locus 5 (Drosophila), (mNkx2.5)	GTGGTCTCTCGGGCCATCCGTCTCGGCTTTGTCCAGCTCC ACTGCCTTCTGCAGCGCGCACAGCTCTTTTTTATCCGCCCG AGGGTCTTTGGCTGGGTGAGGGTACCCTAGGCTCCCGGG TAAAATGTGGGGGCGGCTGGGAAAGCAGGAGAGCACTTG GGGGCGAAGGCGCG
Mouse Nanog homeobox (mNanog)	TCCAGCAGATGCAAGAACTCTCCTCCATTCTGAACCTGAGC TATAAGCAGGTTAAGACCTGGTTTCAAACCAAAGGATGA AGTGCAAGCGGTGGCAGAAAAACCAGTGGTTGAAGACTA GCAATGGTCTGATTCAGAAGGGCT
Mouse Nitric Oxide Synthase 1 (neuronal) (mNos1)	GGCAACCATGAGGACCTCGTGAATGCACTCATTGAACGGC TGGAGGACGCGCC GCCTGCCAACACGTGGTGAAGGTGGAAATGCTGGAGGAA AGGAACACGGCTC TGGGTGTCATCAGTAATTGGAAGGATGAATCTCGCCTCC
Mouse Nitric Oxide Synthase 2 (inducible) mNos2	ACCACACCAAACCTGTGTGCTGGAGGTTCTGGATGAGAGC GGCAGCTACTGGG TCAAAGACAAGAGGCTGCCCCCTGCTCACTCAGCCAAGC CCTCACCTACTTCC TGGACATTACGACCCCTCCACCC
Mouse Nitric Oxide Synthase 3 (endothelial) (mNos3)	GTCCTGTGCATGGATGAGTATGATGTGGTGTCCCTAGAGC ACGAGGCACTGGTGTGGTGGTGACCAGCACATTTGGCAA TGGGGATCCTCCGGAGAATGGAGAGAGCTTTGCAGCAGCG CTCATGGAAATGTCAGGCCCGTACAACAGCTCCCCTAGGC CTGAGCAGCACAAGAGCTACAAAATCCGATTCAACAGTGT CTCCTGCTCAGACCC ACT
Mouse Protein Kinase, cGMP-dependent, type I (mPrkg1)	TATCATGTCAATCCCCCGCAGTATGATATTGTAGGTTTTCA TTGGATCTGGGCT GAGAAAGGTGGGCTGCCAGTCAGAAGCTCATACATCAGAA TTCCTAGTGACCA
Mouse Guanylate cyclase 1, soluble, beta (mGuCy1b)	AGGCCAGCCAGTATGTAGAGAGCATGTTCCGAGCCTTCGA CAAGAATGGGGACA ACACCATTGACTTCTGGAGTATGTGGCAGCCCTGAACCTC GTGCTG



**Figure 1S. Time-course of gene expression in differentiating mEBs** - Relative expression of mNanog (pluripotency marker), mBrachyury (early mesodermic) and mMef2c (cardiac specific) genes. Data are the mean  $\pm$  s.e. of 3 differentiations; measures were performed in triplicate.



**Figure 2S - PKG-I protein expression in a single experimental preparation used to quantify protein phosphorylation** (see figure 4). Different drug treatments were performed as described in the method (scheme 2).



**Figure 3S - LDH release during mEBs cardiac differentiation** - LDH activity in conditioned medium of mEBs was measured in control conditions or in the presence of either 50  $\mu$ M ISN (NO-donor) or NOS/sGC/PKG-I pathway inhibitors (respectively, 100  $\mu$ M L-NMA, 1  $\mu$ M ODQ or 1  $\mu$ M KT5823) from day 0 to 16. Data are expressed as percentage of control values. \* $P < 0.05$  vs control, one way ANOVA followed by post-hoc Bonferroni's test. Data are the mean  $\pm$  s.e. of at least 3 different differentiations; measures were performed in duplicate.