## Cardiac automaticity is modulated by $I_{\mbox{\scriptsize KACh}}$ in sinoatrial node during pregnancy.

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## SUPPLEMENTAL DATA

Table S1. Sequences of primers.

Primers	Sequences
Kcnj3	Forward: 5' CAG TTC GAG GTT GTC GTC ATC C 3'
	Reverse: 5' CCC AAA GCA CTT CGT CCT CTG T 3'
Kcnj5	Forward: 5' GGA GAT GTC TCG TGC TCA ACT G 3'
	Reverse: 5' CCA GAG CAC CTC TGT ATC CAT G 3'
Adora1	Forward: 5' ATC CCT CTC CGG TAC AAG ACA GT 3'
	Reverse: 5' ACT CAG GTT GTT CCA GCC AAA C 3'
Chrm2	Forward: 5' GTC AGC AAT GCC TCC GTT AT 3'
	Reverse: 5' CCT ACG ATG AAC TGC CAG AA 3'
Rgs4	Forward: 5' AGT CAG ATT CCT GCG AAC AC 3'
	Reverse: 5' TCC TCG CTG TAT TCC GAC TT 3'
Rgs6	Forward: 5' AAT CTC AGG TTC TGG CTG TCT GTC 3'
	Reverse: 5' TTG CCA GAT TTC CTC CAC CCT CTT 3'
Hprt	Forward: 5' TGA ATC ACG TTT GTG TCA TTA GTG A 3'
	Reverse: 5' TTC AAC TTG CGC TCA TCT TAG G 3'
Sdha	Forward: 5' GAG GAA GCA CAC CCT CTC ATA 3'
	Reverse: 5' GCA CAG TCA GCC TCA TTC AA 3'
Cyclo	Forward: 5' CCG ATG ACG AGC CCT TGG 3'
	Reverse: 5' GCC GCC AGT GCC ATT ATG 3'

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Figure S1. Activation of the G-protein-gated K<sup>+</sup> current I<sub>KACh</sub> by carbachol (CCh). IV curves

under baseline condition (black), in presence of CCh at 1  $\mu$ M (blue) and 10  $\mu$ M (red).

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Figure S2. Comparison of a ramp and voltage-step protocol for  $I_{KACh}$  recording. (A) Comparison of  $I_{KACh}$  recorded from a ramp (500 ms ramp from +50 mV to -120 mV with a holding potential of -40 mV) and a step (60-ms steps ranging between +50 mV to -120 mV in 10-mV increments from a holding potential of -40 mV) protocol on NP SAN cells. Insets show ramp and steps protocols. (B) Current-voltage (IV) curves of  $I_{KACh}$  recorded with both voltage-clamp protocols were comparable. (C) IV curves of baseline, acetylcholineactivated (+CCh; carbachol, 10  $\mu$ M) and washout (WO) current. Complete return to baseline values is observed in WO conditions.

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**Figure S3. Stain-free blots used to normalize Kir3.1, Kir3.4 and M2R expression.** (*left*) Protein (20 μg) isolated from NP and P mice (N=4 per group) were visualized using stainfree technology. All visible bands were used for normalization of Kir3.1 (*upper*), Kir3.4 (*middle*) and M2R (*lower*). (*right*) Ponceau S of the nitrocellulose membrane right after the transfer and the stain-free image obtained with the ChemiDoc system.