

# Expression of Adrenomedullin signalling factors in bovine corpus luteum during oestrous cycle, pregnancy and induced luteolysis

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## INTRODUCTION

Investigations on "nonclassic" regulators of angiogenesis could open new perspectives in understanding angiogenesis under physiological and pathological conditions. A pro-angiogenic effect has been demonstrated for adrenomedullin (AM), its co-factor activity-modifying protein 2 (RAMP-2) and the Calcitonin receptor-like Receptor (CALCLR). This signalling pathway is besides associated with lymphangiogenesis, that (in a physiologically way) is still only poorly understood in ovary and its structures (corpus luteum and follicles).

## MATERIAL AND METHODS

The aim of this study was therefore to evaluate mRNA expression of AM, RAMP-2 and CALCLR in bovine corpora lutea (CL) during different physiological stages. Experiment 1: CL were assigned days 1-2, 3-4, 5-7, 8-12, 13-16, >18 (after regression) of oestrous cycle and of gravidity (month <3, 3-5, 6-7 and >8). Experiment 2: Induced luteolysis. Cows on days 8-12 were injected with a PGF2 alpha analogue and CL were collected by transvaginal ovariectomy before and 0.5, 2, 4, 12, 24, 48 and 64 h after PGF2 alpha injection. Tissue levels of mRNA were characterized by real-time RT-PCR.

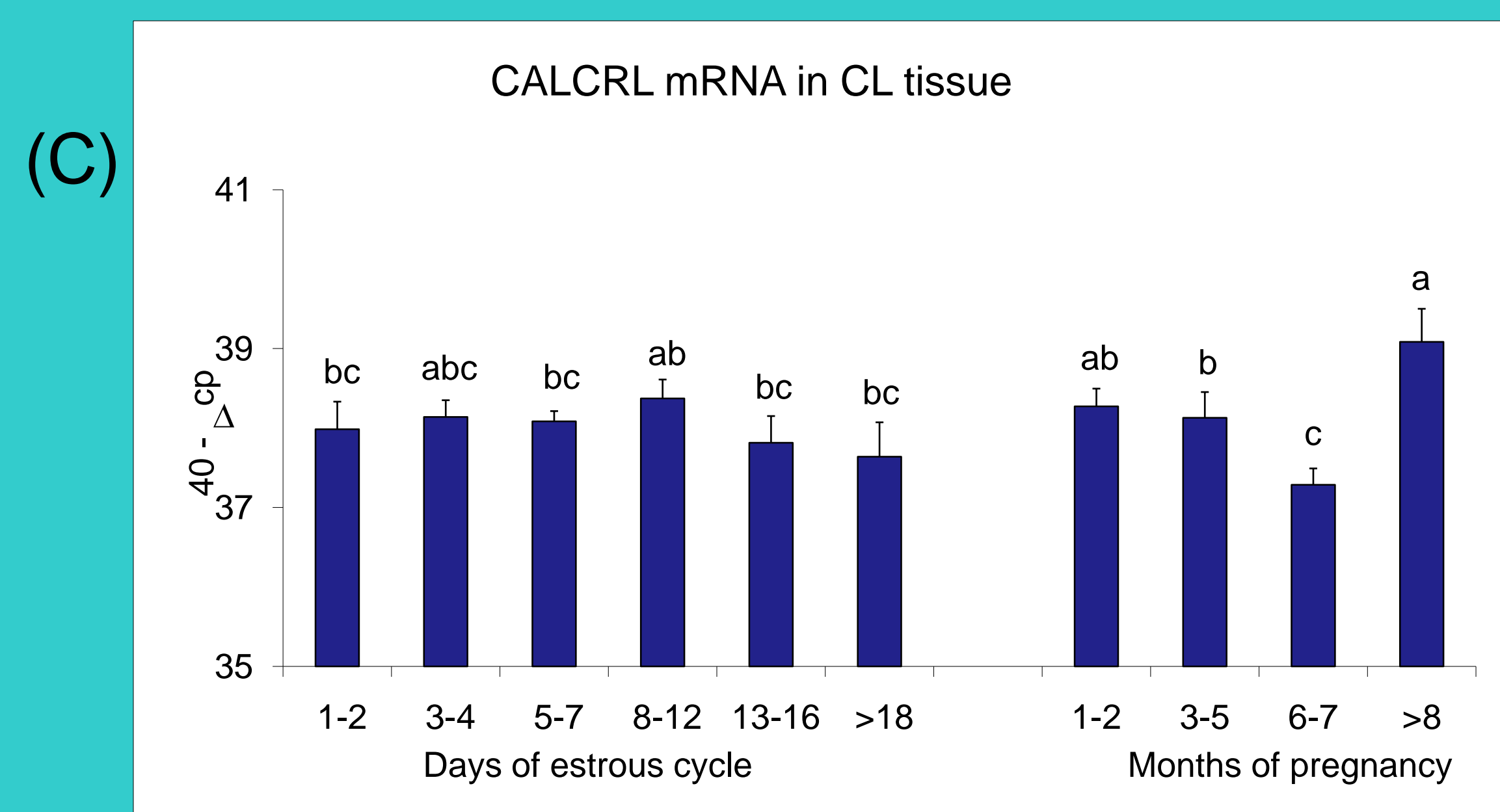
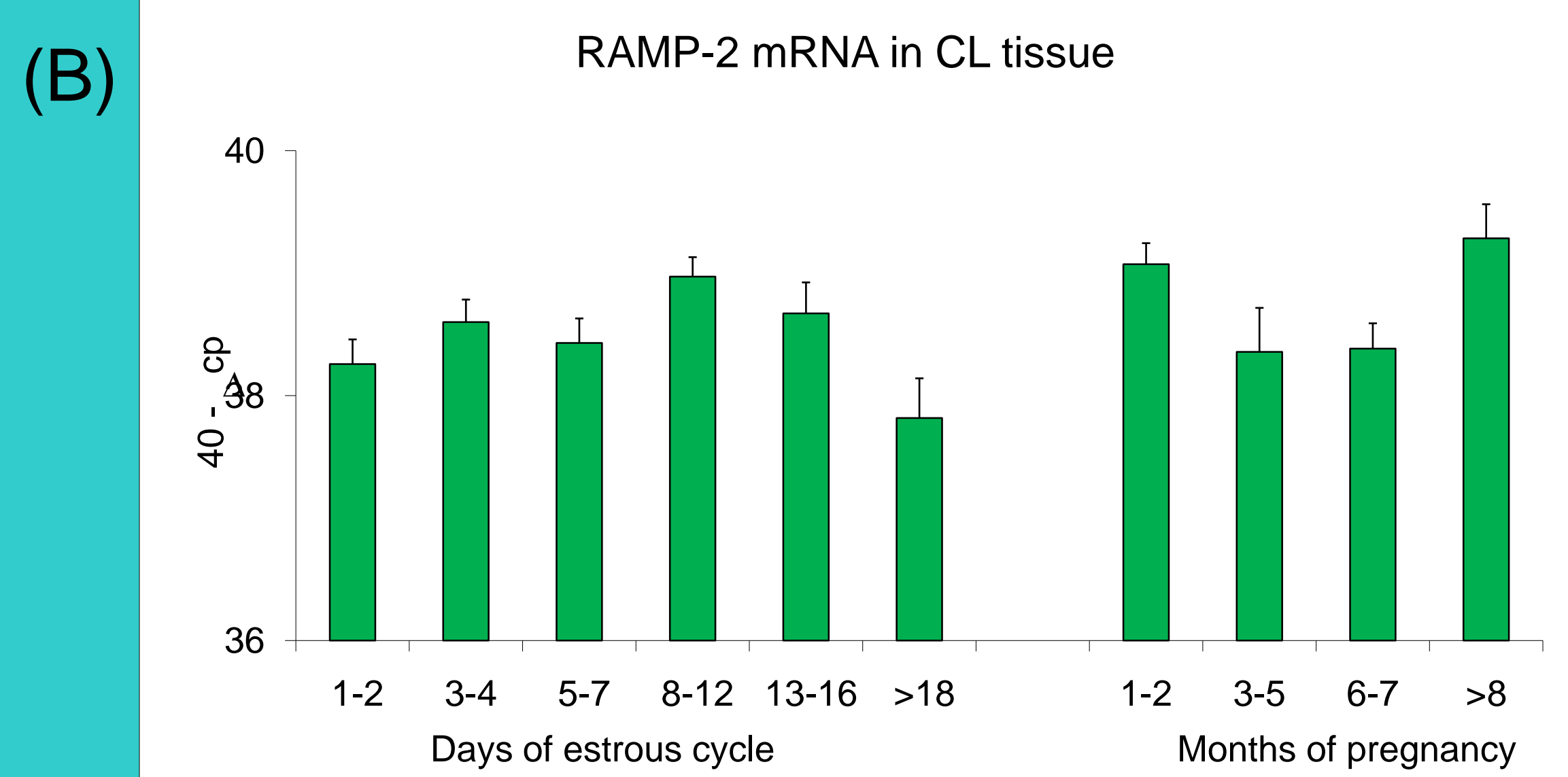
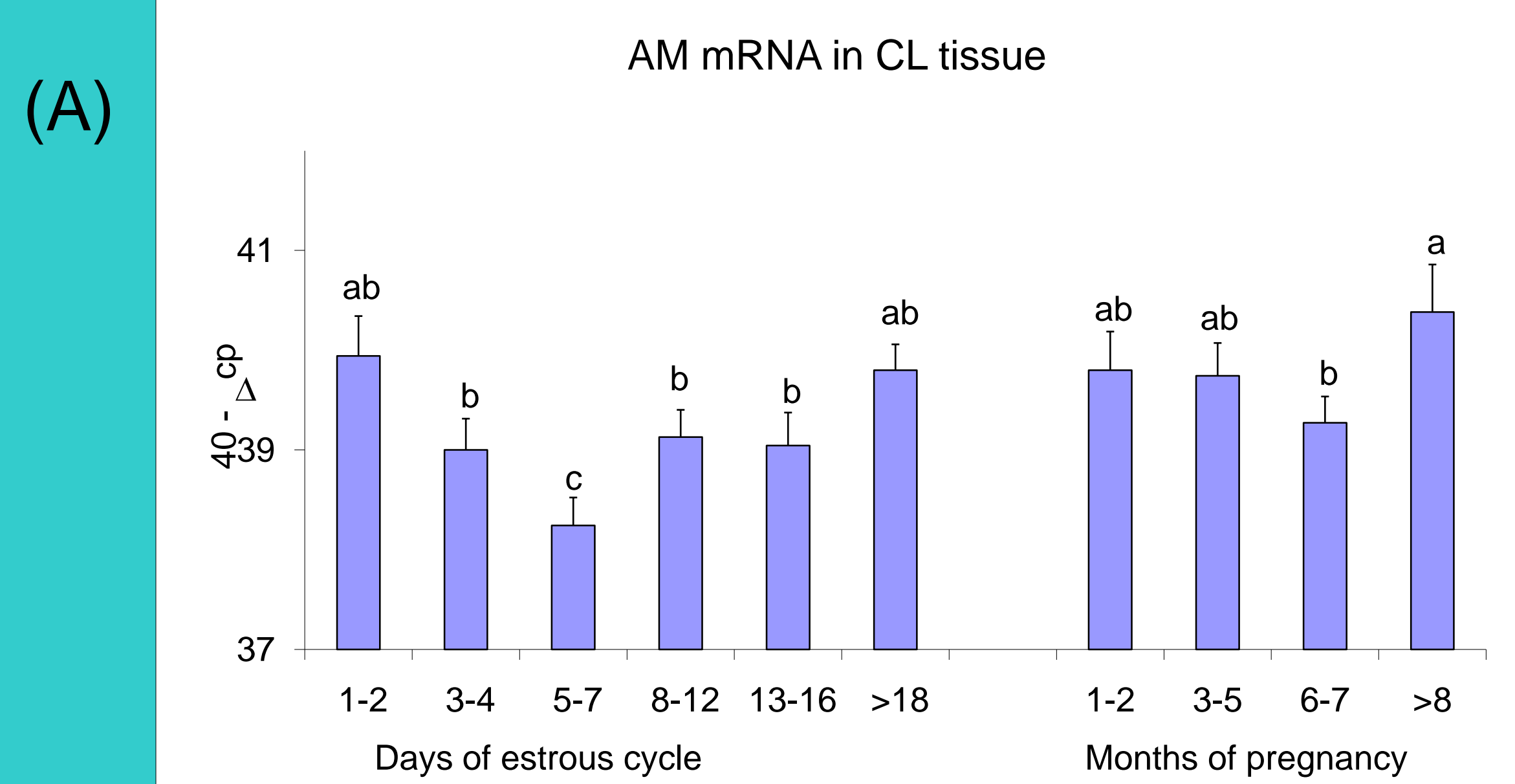


Fig. 1. Expression of mRNA of (A) Adrenomedullin, (B) RAMP-1 and (C) CALCLR in the CL of oestrous cycle and gravidity. Results are presented by 40 minus mean of normalized crossing points ( $\Delta cp$ ) SEM (n=6-7). Different superscripts denote statistically different values (P<0.05).

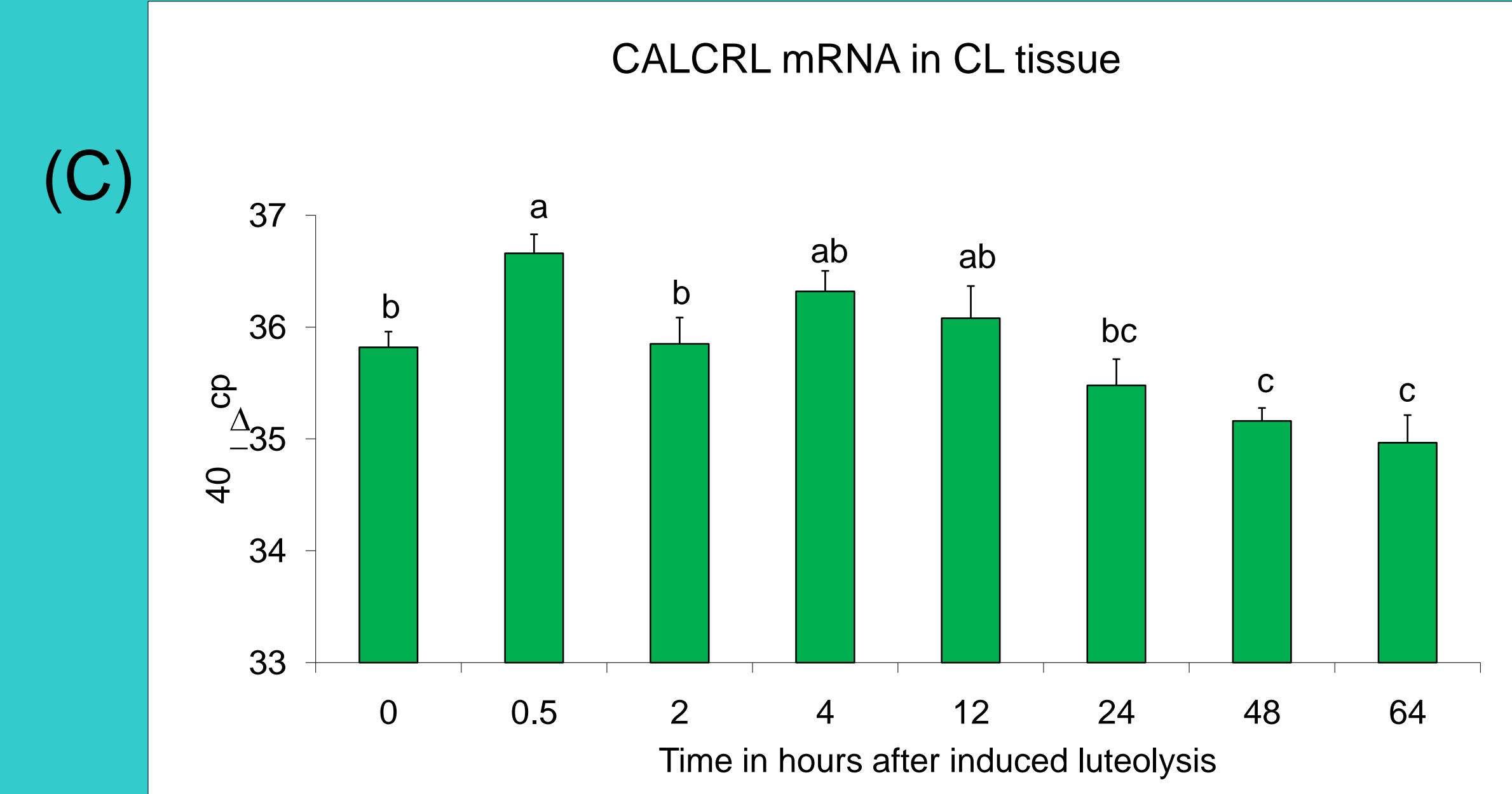
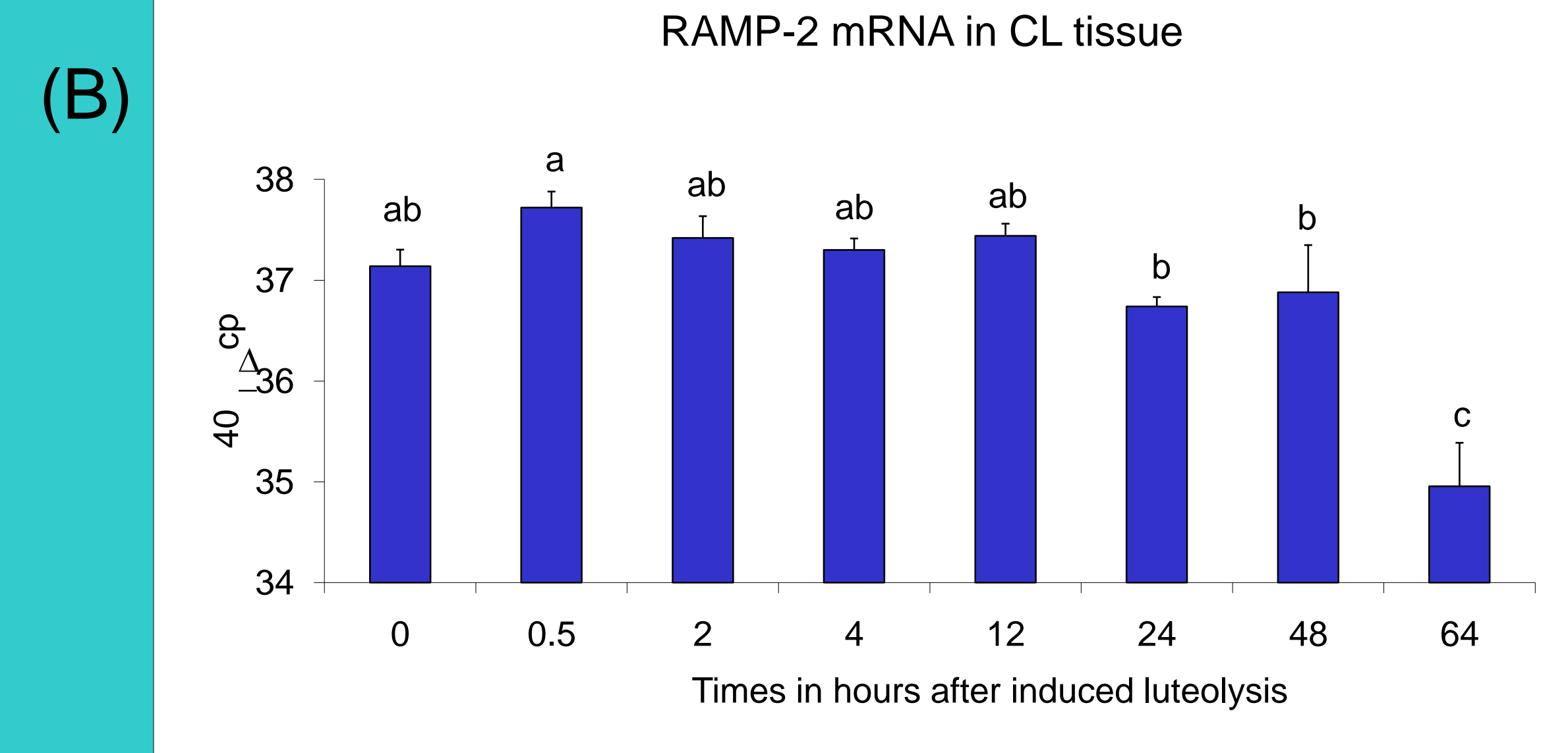
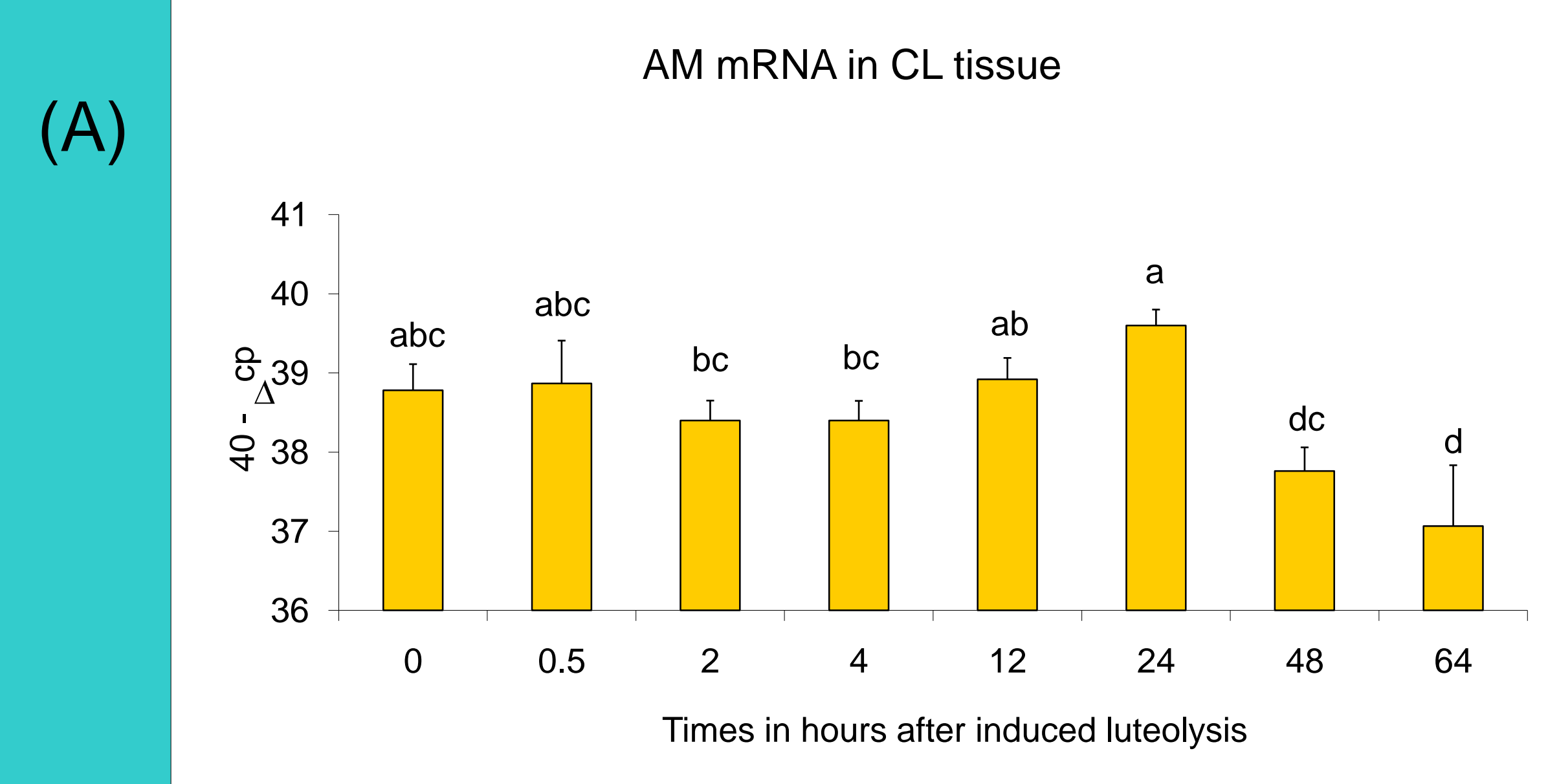


Fig. 2. Expression of mRNA of (A) Adrenomedullin, (B) RAMP-2 and (C) CALCLR in the CL during induced luteolysis. Results are presented by 40 minus mean of normalized crossing points ( $\Delta cp$ ) SEM (n=6-7). Different superscripts denote statistically different values (P<0.05).

## RESULTS AND DISCUSSION

All three examined factors (AM, RAMP-2, CALCLR) were clearly expressed. AM and CALCLR showed significant changes in both experiments, RAMP-2 during induced luteolysis. For AM highest levels could be observed at the beginning and the end of the luteal phase, CALCLR remained at comparable levels. Both factors showed their highest expression at the end of pregnancy. After induced luteolysis the expression of all factors started to decline (significantly during structural luteolysis). In conclusion, our results could lead to the assumption that factors investigated may be involved in mechanisms regulating CL formation, function and regression – with a special emphasis during the final months of pregnancy.