Comparative study of the functional coupling of human chorionic gonadotropin and ca^{2+} activated potassium channels on myometrial contractility in different trimesters of pregnancy

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ABSTRACT

Human chorionic gonadotropin, hCG, is central to the regulation of uterine quiescence during pregnancy. The present study was designed to investigate if the relaxant effect of hCG would vary between the different trimesters of pregnancy, whether this effect is linked functionally to the Ca^{2+} activated potassium channels, BK_{Ca}, and if this link would differ between the different stages of pregnancy. ***Methods:*** *32 female rats were used in the present study, 8 non-pregnant, 8 pregnant at day 4-5, 8 pregnant at day 11-12 and 8 pregnant at day 20-21, representing the 3 trimesters of pregnancy. Frequency and amplitude of isometric contractions were recorded from uterine strips from each group. These recordings were obtained either from spontaneous contractions, subgroup 1; contractions after addition of 20 iu hCG, subgroup 2; contractions after addition of 10 mM BK_{Ca} channels blocker, tetraethyl ammonium TEA, subgroup 3 and contractions after addition of both drugs, subgroup 4.* ***Results:*** *Our results showed that frequency and amplitude of spontaneous uterine contractions were significantly decreased in all trimesters of pregnancy when compared to non-pregnant group. The results also revealed that hCG significantly reduced frequency and amplitude of contraction in all groups and this reduction was more significant in the 2^{nd} and 3^{rd} trimesters of pregnancy. Both parameters of contraction were significantly increased by TEA, while adding both drugs did not alter frequency or amplitude of contraction in both non-pregnant and 1^{st} trimester pregnant strips, but significantly decreased both of them in 2^{nd} and 3^{rd} trimesters pregnant strips.* ***Conclusion****: Our findings outlined that hCG exerted a more significant uterorelaxant effect in the late stages of pregnancy and that activation of BK_{Ca} channels may explain this relaxant effect.*

Key words: *Myometrial contraction – rat – human chorionic gonadotropin – calcium activated potassium channels.*

INTRODUCTION

An increased knowledge of the molecular and physiological mechanisms implicated in the control of myometrial excitability and contractility is essential to the development of effective tocolytic agents that can be used in management of preterm labor which is the largest cause of perinatal

morbidity and mortality, particularly in developing countries.

Myometrial tissue is spontaneously myogenic and can generate slow wave activity as well as simple and complex action potentials. Changes in wave form of myometrial electrical activity and transition from quiescence to excitability at term, suggests extensive fluidity and remodeling of ion channels contributing to the dramatically altered excitability towards time of labor[1].

Human chorionic gonadotropin [hCG], is a hormone produced primarily by the placenta to stimulate progesterone production by the corpus luteum in the early pregnancy[2]. It is a member of a glycoprotein hormone family that consists of leutinising hormone [LH], follicle stimulating hormone [FSH], and thyroid stimulating hormone [TSH]. The members of this family are heterodimers, with similar α-subunits and dissimilar β-subunits[3]. hCG and LH have similar biological actions and bind to a common receptor, which is a member of a G-protein coupled family of receptors[4]. The hCG/LH receptors are present in the endometrial and myometrial layers of the uterus, fallopian tubes, gonads, fetal membranes, deciduas, placenta and umbilical cord[5].

Unlike the previous belief that human myometrium is an indirect target of hCG via ovarian steroid hormones, studies have shown that it is a direct target of hCG regulation as the myometrium contains hCG receptors and exogenous hCG was found to directly increase myometrial hyperplasia and hypertrophy[6]. hCG also inhibited oxytocin stimulated myometrial contraction[7]. The importance for the enhancement of fertility, successful implantation and survival of the conceptus in early gestation is recognized, however, studies conducted worldwide in recent years indicate that hCG may also play a significant role in maintaining pregnancy well after the first trimester raising the question about its potential future role for tocolysis in human preterm labor[8].

K^+ channels have a widespread distribution in virtually all cell types. Their main function is to dampen cellular excitability by maintaining the cell membrane potential close to the equilibrium potential of K^+ ions. By the generation of an outward K^+ current, these channels produce hyperpolarization or repolarization thereby terminating or rendering action potential generation and ultimately contraction less likely[9].

Ca^{2+} activated potassium channels are a class of potassium channels that are activated when intracellular Ca^{2+} levels rise. There are three subclasses of calcium-activated potassium channels, the large conductance channels, denoted BK_{Ca} or maxi K, are probably the best characterized and most ubiquitous being expressed in all tissues with the exception of the heart. Two other subclasses are the intermediate, IK_{Ca} and the small, SK_{Ca} conductance calcium-activated K^+ channels but these have not been described in any detail in the human myometrium[10].

The BK_{Ca} or maxi K channel is the predominant K^+ channel type encountered in non-pregnant and pregnant human myometrium[11,12].

Channels belonging to this class are $Ca^{2+}$and voltage-dependent with channel open-state probability increasing with depolarization. BK_{Ca} channels are associated with a variety of cellular functions including control of smooth muscle tone and neuronal firing[13].

BK_{Ca} channels exist in vivo as a complex of α-subunit (a 125 kDa protein) and a 31 kDa glycosylated regulatory protein, the β-subunit[14].The mechanisms by which BK_{Ca} channels sense Ca^{2+} remain an enigma. Meera et al.[15] have reported that the myometrial β-subunit is functionally uncoupled from the α-subunit at resting intracellular Ca^{2+} levels and that as Ca^{2+} rises inside the cell the β-subunit switches to acting as a Ca^{2+} sensor. In addition to the role of the β-subunit, the Ca^{2+} sensing function of the BK_{Ca} channel may also be attributable to the string of negatively charged aspartate residues that constitute the Ca^{2+} bowel located in the α- subunit tail region[13].

Modulation of BK_{Ca} channels by hCG in Leydig cells from mature mouse testis was documented[16], while still conflicting reports have appeared concerning the effects of hCG on these channels in the uterus. Moreover, their role in controlling myometrial contractility during pregnancy is still not clear.

In the present study, we addressed the question of whether the uterorelaxant effect of hCG on myometrial contractility would differ between different stages of pregnancy. Also the involvement of BK_{Ca} in such relaxant effect would be assessed and compared in the three trimesters of pregnancy.

MATERIAL & METHODS

Animals:

Twenty four pregnant and 8 non pregnant rats of the local strain were used in the present study. They were brought from the Eye Research Institute and kept in the animal house of Kasr Al-aini faculty of Medicine, they had food and water ad libitum.

Experimental procedure:

Pregnant and non-pregnant rats were killed by decapitation. The preparation of myometrial strips followed the method of Karsli et al. (2003)[17]. A full-length longitudinal abdominal incision was made to expose both uterine horns which were rapidly excised, carefully cleaned of surrounding connective tissue and opened longitudinally along the mesenteric border. Fetuses of the late-stage pregnant rats were removed and non-uterine tissues were dissected and discarded. We obtained 4 longitudinal full-thickness myometrial muscle strips, measuring 4.5 mm in length and 1.5 mm in width, from each animal.

The strips were mounted vertically, one end of the strip was connected to the lower hook of the bath and the other end of the strip was connected to a force transducer. The strips were incubated in a 20 ml tissue bath containing Krebs-Henseleit physiological solution (composition in mM: KCL 4.6; $MgSO_4$ 1.16; NaH_2PO_4 1.16; $CaCl_2$ 2.5; NaCl 115.5; $NaHCO_3$ 21.9; and glucose 11.1.), which were aerated continuously with 95% oxygen and 5% carbon dioxide. The pH was kept at 7.4 and the temperature was

maintained at 37°C. The solution was prepared daily for each experiment. Myometrial strips of rats were allowed to equilibrate for 20 min. The characteristics of the contraction including frequency and amplitude were recorded by an isometric force transducer of the Intracel computer using analyzing software Phys. 4 (Intracel company; England). When the contractions of the strips became regular, the contraction frequency and amplitude were recorded as baseline activity, then the following drugs, hCG (1 iu/ ml bath solution), TEA (10 mM) as well as hCG in the presence of TEA were added according to the experimental protocol. Each strip was exposed to only one agent. Drug-containing solutions were prepared immediately before the experiment.

Experimental protocol:

A total of 128 myometrial strips were obtained from pregnant and non pregnant rats, and were randomly assigned to 1 of 4 main groups (n=32 strips, each group):

Group I: non-pregnant group

Group II: pregnant 1st trimester group (at day 3 - 4 of pregnancy).

Group III: pregnant 2nd trimester group (at day 11-12 of pregnancy).

Group IV: pregnant 3rd trimester group (at day 20- 21 of pregnancy)[18].

After spontaneous myometrial contractions were induced in the Krebs-Henseleit physiological solution, each group was further subclassified into 4 subgroups (n=8 strips, each subgroup):

Subgroup 1, control group: frequency and amplitude of contraction were recorded 20 minutes after equilibration without addition of any drugs to the bathing solution.

Subgroup 2, hCG group: frequency and amplitude of contraction were recorded 20 minutes after addition of 1 iu/ ml hCG to the bathing solution[19].

Subgroup 3, TEA group: frequency and amplitude of contraction were recorded 20 minutes after addition of 10 mM tetraethyl ammonium [TEA] to the bathing solution[20].

Subgroup 4, hCG and TEA group: TEA was added to the bathing solution 20 minutes before adding hCG and after another 20 minutes, recordings of frequency and amplitude of contraction from uterine strips were carried out.

Drugs: hCG was purchased from Organon Co- Switzerland.
TEA was purchased from Sigma Chemical Co., St Louis, Mo, USA
hCG and TEA were dissolved in distilled water.

Statistical Analysis:

The results were expressed as mean values ± SD. Comparisons between subgroups in each main group or between subgroups in different main groups were carried out by one-way analysis of variance (ANOVA) for multiple comparisons. A P value <0.05 was regarded as statistically significant.

RESULTS

Results are shown in tables (1- 3) and figures (1- 10).

Spontaneous myometrial contractions were less frequent and having less amplitude in pregnant uterine non treated strips from any of the three trimesters when compared with non-pregnant, non treated strips. $P<0.001$ (pregnant 1^{st}, 2^{nd}, and 3^{rd} trimesters versus non-pregnant) as regard the frequency of contraction, $P<0.005$ (pregnant 1^{st} and 2^{nd} trimesters versus non-pregnant) and $P<0.001$ (pregnant 3^{rd} trimester versus non-pregnant) as regard the amplitude of contraction.

Effect of hCG on the contractility of isolated uterine muscle strips from pregnant and non-pregnant female rats:

Comparing the frequency of contraction between subgroups in each main group revealed that addition of hCG to the bathing solution significantly decreased frequency of contraction in uterine strips from all groups when compared with non treated control strips in each corresponding group ($P<0.05$ in non-pregnant strips and $P<0.005$ in all pregnant strips). This decrease was more significant in pregnant than non pregnant strips.

As regards the effect of hCG on amplitude of contraction, it reduced amplitude of contraction in non-pregnant strips but this reduction did not reach statistical significance. hCG significantly decreased amplitude of contraction in 1^{st}, 2^{nd} and 3^{rd} trimester pregnant strips, $P< 0.005$, $P<0.05$ and $P< 0.001$ respectively when compared to non treated strips in each group.

Effect of TEA on the contractility of isolated uterine muscle strips from pregnant and non-pregnant female rats:

The large conductance Ca^{2+} sensitive K^+ channels blocker, tetraethyl ammonium, TEA, was used to study the role of these channels in regulating basal contractility in myometrial smooth muscle. We found that TEA significantly increased frequency of contraction in non pregnant, 1^{st}, 2^{nd} and 3^{rd} trimester pregnant strip $P< 0.001$, $P<0.001$, $P< 0.001$ and $P< 0.005$ respectively.

The same finding was obtained on observing the effect of TEA on amplitude of contraction, it significantly increased amplitude of contraction in all uterine strips $P< 0.01$, $P<0.005$, $P< 0.5$ and $P<0.05$ respectively when compared with control non-treated strips in each group.

Effect of TEA on hCG induced relaxations in isolated uterine muscle strips from pregnant and non-pregnant female rats:

In an attempt to examine whether the blockade of the large conductance Ca^{2+} sensitive K^+ channels interferes with hCG-induced relaxation of myometrial smooth muscles, incubation of uterine strips with TEA before addition of hCG to the bathing solution was performed. We obtained different responses on frequency and amplitude of contraction in different trimester strips. Both drugs when added together did not alter any of the two parameters of contraction in non-pregnant and 1^{st} trimester pregnant uterine strips.

On the other hand in 2nd and 3rd trimester pregnant uterine strips, adding hCG and TEA together decreased frequency of contraction which was significant in 3rd trimester uterine strips P<0.05, when compared to control non treated uterine 3rd trimester strips. Both drugs together reduced the amplitude of contraction in 2nd and 3rd trimester pregnant strips and this reduction was significant in both trimesters, P< 0.05 and P< 0.001.

Table 1: frequency (cycles/ min.) and amplitude (screen units) of contraction of uterine strips from control non-pregnant, pregnant 1st, 2nd and 3rd trimesters.

	Control non-pregnant	**Pregnant 1st trimester**	**Pregnant 2nd trimester**	**Pregnant 3rd trimester**
Frequency	1.19 ± 0.12	0.89 ± 0.17 P < 0.001 *	0.71 ± 0.12 P < 0.001 *	0.86 ± 0.17 P < 0.001 *
Amplitude	19.25 ±2.12	15.63 ±1.92 P < 0.005 *	15.5 ±2.14 P < 0.005 *	15 ± 1.85 P < 0.001 *

** Significant when compared with non-pregnant control group.*

Table 2: frequency (cycles/ min.) and amplitude (screen units) of contraction of uterine strips from non- pregnant and pregnant 1st trimester

	Non-pregnant				**Pregnant 1st trimester**			
	Control	**hCG**	**TEA**	**hCG + TEA**	**Control**	**hCG**	**TEA**	**hCG + TEA**
Frequency	1.19 ± 0.12	1.03 ± 0.13 P < 0.05*	1.64 ± 0.14 P < 0.001*	1.21 ± 0.12	0.89 ± 0.17	0.58 ± 0.18 P < 0.005 *	1.29 ± 0.18 P < 0.001*	0.94 ± 0.17
Amplitude	19.25 ± 2.12	16.75 ± 1.91	22.88 ±3.27 P < 0.01*	19.38 ±2.39	15.63 ± 1.92	12.5 ± 1.77 P < 0.005 *	19.13 ± 2.23 P < 0.005 *	14.25 ± 1.67

***** Significant when compared with the corresponding control.
Control: no drugs added to the bathing solution.
hCG: hCG 20 iu were added to the bathing solution.
TEA: TEA 10 mM were added to the bathing solution.
hCG + TEA: both drugs were added to the bathing solution.

Table 3: frequency (cycles/ min.) and amplitude (screen units) of contraction of uterine strips from pregnant 2nd and 3rd trimesters

	Pregnant 2nd trimester				Pregnant 3rd trimester			
	Control	**hCG**	**TEA**	**hCG + TEA**	**Control**	**hCG**	**TEA**	**hCG + TEA**
Frequency	0.71 ± 0.12	0.46 ± 0.13 P < 0.005*	1.11 ± 0.2 P < 0.001*	0.6 ± 0.13	0.86 ± 0.17	0.59 ± 0.16 P < 0.005*	1.2 ± 0.2 P < 0.005*	0.66 ± 0.16 P<0.05*
Amplitude	15.5 ± 2.14	12.63 ±1.92 P < 0.05*	18.13 ± 2.79 P < 0.05*	12.63 ± 2.26 P < 0.05*	15 ± 1.85	11.13± 1.25 P < 0.001*	16.88 ± 2.1 P < 0.05*	11.38 ± 1.41 P<0.001*

***** Significant when compared with the corresponding control.
Control: no drugs added to the bathing solution.
hCG: hCG 20 iu were added to the bathing solution.
TEA: TEA 10 mM were added to the bathing solution.
hCG + TEA: both drugs were added to the bathing solution.

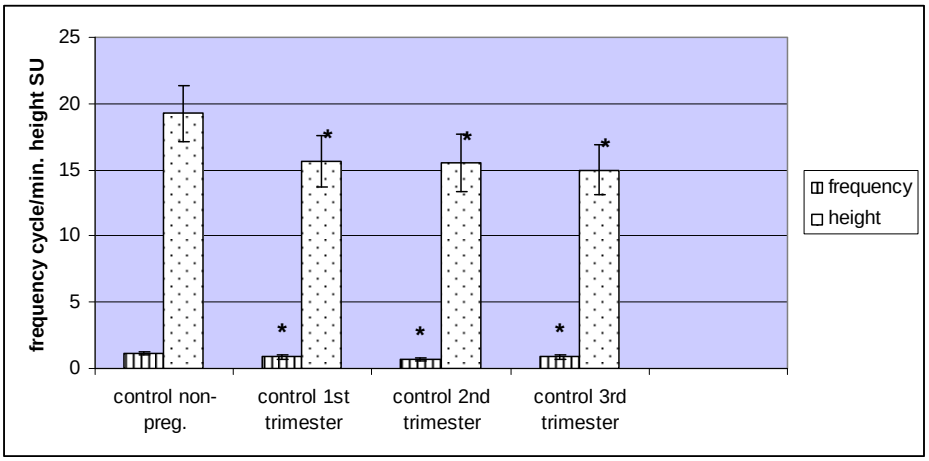

Figure 1: frequency (cycles/ min.) and amplitude (screen units) of contraction of uterine strips from control non-pregnant, pregnant 1st, 2nd and 3rd trimesters
** Significant when compared with non-pregnant control group.*

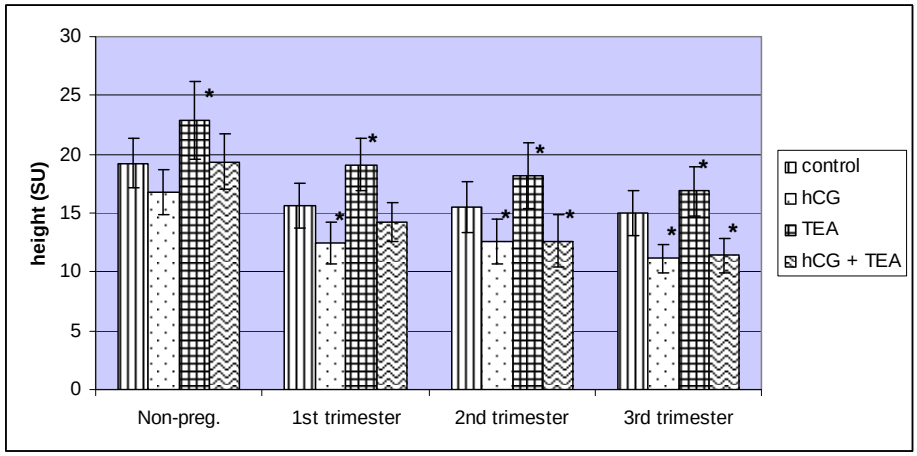

Figure 2: amplitude of contraction (screen units) of uterine strips from non-pregnant, pregnant 1st, 2nd and 3rd trimesters
Control: no drugs added to the bathing solution.
hCG: hCG 20 iu were added to the bathing solution.
TEA: TEA 10 mM were added to the bathing solution.
hCG + TEA: both drugs were added to the bathing solution.
****** Significant when compared with the corresponding control.*

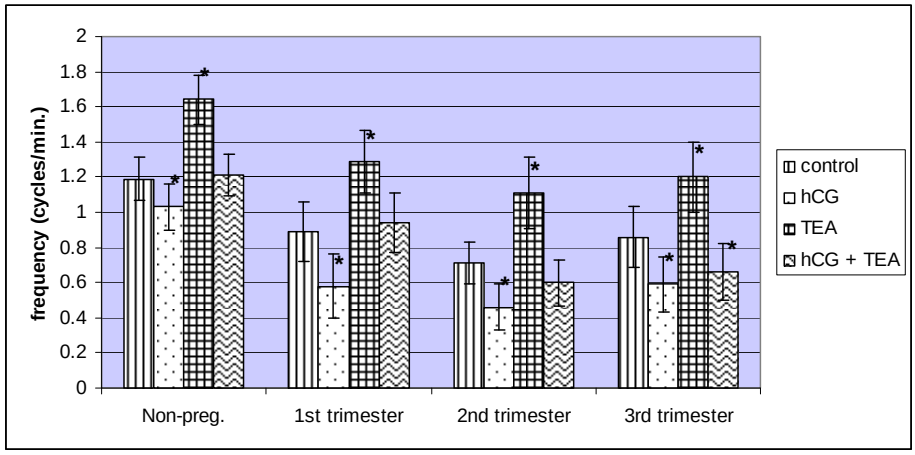

Figure 3: frequency of contraction (cycles/ min.) of uterine strips from non-pregnant, pregnant 1st, 2nd and 3rd trimesters
Control: no drugs added to the bathing solution.
hCG: hCG 20 iu were added to the bathing solution.
TEA: TEA 10 mM were added to the bathing solution.
hCG + TEA: both drugs were added to the bathing solution.
****** Significant when compared with the corresponding control.*

Figure 4: Control non pregnant rat uterus

Figure 5: Effect of hCG on non pregnant uterus

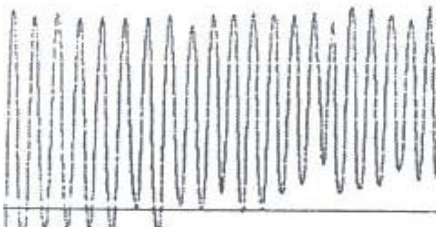

Figure 6: Effect of TEA on non pregnant uterus

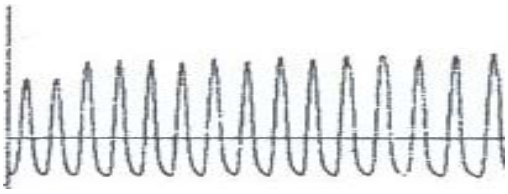

Figure 7: Effect of hCG + TEA on non pregnant uterus

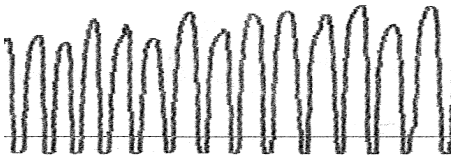

Figure 8: Effect of hCG + TEA on 1st trimester pregnant uterus

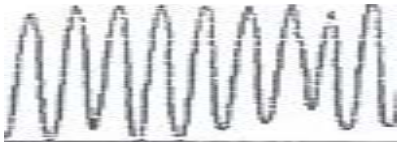

Figure 9: Effect of hCG + TEA on 2nd trimester pregnant uterus

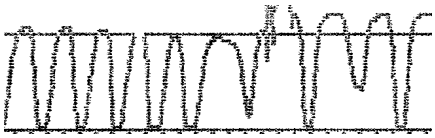

Figure 10: Effect of hCG + TEA on 3rd trimester pregnant uterus

DISCUSSION

The uterorelaxant effect of human chorionic gonadotropin is regarded as an important mediator in maintenance of uterine quiescence during pregnancy, while the high conductance, calcium activated potassium channels were shown to play a pivotal role in modulating myometrial excitability.

We aimed to compare the involvement of BK_{Ca} channel function in the response of uterine contractility to hCG between different trimesters of pregnancy.

The results of the present study revealed that frequency and amplitude of contraction decreased significantly in pregnant strips when compared to non-pregnant ones.

Our results agree with many previous observations which stated that during pregnancy uterine contractility is normally held in abeyance till parturition, when it gets gradually activated giving rise to powerful rhythmic contractions

culminating in delivery of the fetus. The progesterone withdrawal theory was the earliest theory that explained shift in myometrial activity during parturition, it ascribed the relative quiescence of the pregnant uterus to the presence of progesterone and that the decline in its level at term releases myometrium from inhibition[21]. Researches have proved that hCG[7] and Bk_{Ca} channels[1] may be involved in the mechanisms underlying the inhibition of uterine activity during pregnancy. The conversion of the silent pregnant uterus to highly excitable at term had been explained by many observations like the significant decrease in hCG receptors at labor myometrium tissue[22], also the BK_{Ca} channels were found to be either absent or considerably altered in their physiological and pharmacological properties at labor, they have their identical conductance and K^+ selectivity but exhibit no Ca^{++} or voltage sensitivity, allowing cytoplasmic Ca^{++} levels to rise without activating a counteracting K^+ outward current[23].

Changes in electrical activity of the uterus may be considered also in the dramatic change in myometrial contractility at term, Sims et al. (1982)[24] observed a decrease in myometrial cell resting potential in rats at term. Conversion of complex action potentials to simple spikes at labor was also documented[25]. Many compounds have been implicated also such as prostaglandins, oxytocin and heteromeric G protein family[26].

The present study showed that hCG decreased frequency and amplitude of contraction in non pregnant and pregnant uterine strips and that inhibitory effect was more significant in the 3rd trimester pregnant strips. This suggests that the quiescent effect of hCG on the myometrium gets more prominent in pregnant uteri in the 3rd trimester.

In accordance with our suggestion is that of Slattery et al. (2001)[18] who highlighted a potential role of hCG in maintaining uterine quiescence in the 3rd trimester.

In addition, in the study of Rzucidlo et al. (1998)[27], the authors measured the unoccupied binding sites of hCG receptors in the porcine myometrial tissue at different stages of pregnancy, it was significantly higher in the 1st trimester than 2nd and 3rd trimester pregnant uteri, this means that more receptors were occupied in the 2nd and 3rd trimesters, which supports our observation in the present study that hCG has a more prominent effect in the 2nd and 3rd trimester pregnant uterine strips.

Blocking the BK_{Ca} channels in our experiment enhanced contractility of non-pregnant and pregnant uterine strips, as it allows intracellular accumulation of Ca^{2+} without K^+ outflow, i.e. depolarizing the cells and hence increasing contractile activity. This finding concurs with the data from the report of Sanborn, (2000)[28], who stated that BK_{Ca} channels blocking increased contractile activity in rat and human myometrium. Also the study of Kafali et al. (2002)[29] who observed that TEA increased spontaneous myometrial contraction in pregnant rats.

The other important finding in the present work, is that addition of hCG with the Ca^{2+} sensitive K^+ channel blocker, TEA, did not alter frequency

and amplitude of contraction in non-pregnant and 1st term pregnant uteri, but significantly decreased the same parameters in 2nd and 3rd trimester pregnant myometrial strips when compared to control non treated strips in each group.

According to the report of Doheny et al. (2003)[30], who observed that hCG increased open state probability of the BK_{Ca} channels and blocking of these channels significantly attenuated the relaxant effect exerted by hCG in human myometrium of pregnant women undergoing cesarean section. It was expected in our study that incubation of uterine strips with TEA would block the relaxant effect of hCG, this was observed in non-pregnant and 1st trimester pregnant strips, while in the 2nd and 3rd trimester pregnant strips addition of TEA did not block the effect of hCG which still significantly inhibited uterine contractility, this suggests that hCG exerts stronger effect in 2nd and 3rd trimesters which could be mediated by other mechanisms than BK_{Ca} channels.

The discrepancy between the observation of Doheny et al, that BK_{Ca} channel blocking could block the inhibitory effect of hCG on myometrial contraction at those females undergoing cesarean section, and our observation that TEA did not exert the same action in the 2nd and 3rd trimesters may be attributed to the fact that the myometrial strips in their study were taken from uteri at term which are expected to have fewer hCG receptors with unremarkable effect for hCG[22].

The suggested mechanisms for the uterorelaxant effect of hCG included alterations in myometrial gap junctions and hence the electrical coupling between muscle cells, hCG was found to decrease connexin 43, an integral protein in myometrial gap junction, in addition it decreased the assembly of gap junctions and also their permeability in cultured myometrial cells[31]. hCG was found to inhibit uterotropic eicosanoids[22] and reduce intracellular calcium availability[32]. Data also showed that hCG may mediate its action through increasing myometrial biosynthesis of progesterone in rats[33]. By these suggested mechanisms hCG can exert its uterorelaxant effect despite the block of BK_{Ca} channels.

In some groups in our study, the effects of the drugs used, on either frequency or amplitude of contraction, were not exactly the same. It is known that whether excitatory or inhibitory compounds can alter the frequency or amplitude of contraction or even the baseline of the recording. Quite often low concentrations produce changes in frequency, while intermediate concentrations can affect amplitude of contraction and higher concentrations may change the baseline[34]. As we implemented one concentration for each drug in our study, this could explain that frequency and amplitude of contraction did not always show the same significance in change by each drug used.

Conclusion: the present study pointed to the highly significant uterorelaxant effect of hCG in the 3rd trimester. It also confirmed that activation of BK_{Ca} channels could be one of the mechanisms underlying this uterorelaxant effect.

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تأثير العلاقة بين هرمون الحمل و قنوات البوتاسيوم التى تتأثر بالكالسيوم على انقباض الرحم فى المراحل المختلفة للحمل

ان هرمون الحمل (hCG) يلعب دورا هاما فى انبساط عضلة الرحم اثناء الحمل، و قد صممت هذه الدراسة لبحث ما اذا كان تاثير الhCG على عضلة الرحم يختلف اثناء مراحل الحمل، كذلك اذا كانت هناك علاقة بين هذا التأثير و بين قنوات البوتاسيوم التى تتأثر بالكالسيوم و ما اذا كانت هذه العلاقة تختلف اثناء المراحل المختلفة للحمل.اجريت هذه الدراسة على ٣٢ فأرة تم تقسيمها الى ٤ مجموعات، المجموعة الاولى ٨ غير حوامل، المجموعة الثانية ٨ حوامل فى اليوم ٤-٥ ، المجموعة الثالثة ٨ حوامل فى اليوم ١١-١٢ و المجموعة الرابعة ٨ حوامل فى اليوم ٢٠-٢١. فى كل مجموعة تم قياس معدل و ارتفاع الانقباضات فى عضلة الرحم، اما الانقباضات التلقائية، او بعد اضافة ٢٠ وحدة دولية من الhCG ، او بعد اضافة ١٠ ميللى مول من الTEA الذى يلغى عمل قنوات البوتاسيوم او بعد اضافة كلا من الhCG و الTEA.

و قد أظهرت النتائج ان معدل و ارتفاع الانقباضات التلقائية فى رحم المجموعات الحوامل كان اقل بدلالة احصائية من قيمتهما فى رحم المجموعة غير الحوامل. كذلك اظهرت النتائج ان الhCG ادى الى انخفاض هذين المؤشرين فى كل المجموعات و لكن الانخفاض كان أكثر دلالة احصائية فى مجموعة اخر الحمل، بينما ادى ال TEA الى ارتفاع هذين المؤشرين ارتفاعا ذا دلالة احصائية فى كل المجموعات. اظهرت النتائج ايضا ان اضافة كلا من الhCG و الTEA ادى الى عدم تغير اى من معدل او ارتفاع انقباض عضلة الرحم فى المجموعتين الاولى والثانية بينما أدى الى انخفاض ذى دلالة احصائية فى هذين المؤشرين فى مجموعتى الفئران الحوامل فى وسط و اخر الحمل.

تشير نتائج هذه الدراسة الى ان الhCG له تأثير مثبط على عضلة الرحم و خاصة فى اخر الحمل. كذلك تشير الى ان قنوات البوتاسيوم التى تتأثر بالكالسيوم قد تلعب دورا فى طريقة عمل ال hCG.