Role of the antidiabetic drugs: Glibenclamide and metformin on the contractility of isolated rat uteri

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Abstract: The current investigation has designed to study the role of two antidiabetics, glibenclamide and metformin on the spontaneous uterine contractions in the non-diabetic non-pregnant female rats. The rat uteri were isolated and allocated to two groups: 1) the glibenclamide group: After recording the normal spontaneous uterine contractions, the vehicle (ethanol) and glibenclamide molar concentrations (10^{-7}, 10^{-6} and 10^{-5} M) were analyzed on uterine contractions by recording on smoked paper on a rotating kymograph drum, and 2) the metformin group: After recording the normal spontaneous uterine contractions, the metformin concentrations (10^{-7}, 10^{-6} and 10^{-5} M) were analyzed on uterine contractions. Responses to the two drugs and vehicle control (ethanol) were recorded for 30 min. Glibenclamide has not significantly effected on the amplitude and frequency of spontaneous contractions of the isolated rat uteri. Metformin also has no significant effect on the amplitude and frequency of spontaneous contractions of the isolated rat uteri. In conclusion, the two oral antidiabetics glibenclamide and metformin have not changed both the amplitude and frequency of spontaneous uterine contractions in the non-pregnant non-diabetic female rats.

Keywords: Glibenclamide, metformin, isolated uterus, myometrium, spontaneous contractions, non-pregnant non-diabetic female rats.

INTRODUCTION

Pregnancy complicated by diabetes is a medical challenge, which is associated with maternal, fetal and neonatal ill health (NICE, 2008; Bell *et al*., 2008). Pregnancy complications due to diabetes have been reported such as macrosomia (a birth weight >4000g), fetal distress, neonatal hypoglycemia, congenital malformation, Cesarean section (CS) and preeclampsia (El Mallah *et al*., 1997; Jensen *et al*., 2004). Increased fetal insulin (an anabolic hormone) levels in response to the maternal hyperglycemia result in development of macrosomic infant and predispose to fetal hypoglycemia during neonatal period (catalano *et al*., 1995).

According to Evers *et al* (2004) 51% of emergency CS have accounted with the failure of induction labour and prolonged labour, which are associated with diabetes. Postpartum hemorrhage due to poor myometrial contractility is reported to be more common in diabetic women (Dunne *et al*., 2003). In streptozotocin-induced diabetic animal models, these studies found either no effect (Franchi *et al*., 1988) or inhibition of uterine contractility (McMurtrie *et al*., 1985; Jawerbaum *et al*., 1996). In addition, the force of myometrial contractions was reduced faster in diabetics than non-diabetic controls (Kaya *et al*., 1999). The myometrium of diabetics is exposed during pregnancy and labour to high levels of insulin given to achieve glycaemic control (Balsells *et al*., 2000; Stenninger *et al*., 2008). It has been reported that insulin interferes with uterine contractions in rat and human. The ability of insulin to induce myometrial relaxation in diabetic pregnant women is less compared to non-diabetic controls. This may be explained by the insulin resistance, which usually occurs in diabetes (Kuznetsova *et al*., 2006; Kuznetsova *et al*., 2005).

Augmentation of labour with oxytocin fails in many diabetics (Evers *et al*., 2004). Also, oxytocin-induced myometrial contraction in diabetic rats was less as compared to non-diabetic animals (McMurtrie *et al*., 1985). Diabetes has been shown to have an inhibitory effect on myometrial contractility during late pregnancy (Jawerbaum *et al*., 1996) and in non-pregnant (McMurtrie *et al*., 1985) rats. However, diabetes is reported to have no effect on myometrial contractility in non-pregnant rats (Franchi *et al*., 1988). Also, as noted above, the rate of CS is high in women sufferers with diabetes when compared to non-diabetics indicating reduced contractility (Dunne *et al*., 2003; Jensen *et al*., 2004; Crowther *et al*., 2005).

The only treatment for type-1 diabetes during pregnancy is insulin. To maintain the normal levels of blood glucose the gestational diabetes mellitus or type 2 diabetes women may be treated with oral antidiabetics and diet therapy. Using an oral hypoglycemic agent instead of insulin is now widely acceptable as a treatment of diabetes during pregnancy; both type-2 and GDM, despite initial concerns about the safety of these medications during pregnancy, especially if crossing the placenta (Anonymous, 2003;

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Langer *et al*., 2000; Koren, 2001). Glibenclamide (also known as glyburide), a second-generation oral hypoglycemic sulfonylurea, is considered a safe drug to be used to achieve euglycemia during pregnancy in women with type-2 or GDM (Langer *et al*., 2000; Koren, 2001). Glibenclamide selectively blocks the ATP-dependent K^+- channels (Escande *et al*., 1989; Piper *et al*., 1990). Metformin is a biguanide oral euglycemic drug (Rowan *et al*., 2008). The effects of antidiabetics on the myometrium of either animals or women have not been much studied. Insulin attenuates KCl-induced myometrial contractions in human and rats (Kuznetsova *et al*., 2006), indicating that insulin may affect force-producing pathways in myometrium beyond the surface membrane depolarization (Wray, 2007). However, Goldraij *et al*. showed that insulin enhances the contractility of the myometrium in rats. Further, McMurtrie *et al*. indicated that insulin enhances the oxytocin-induced myometrial contraction in rat.

There are no studies related to the action of glibenclamide and metformin on the contraction of rat uterine in the literature to the best of our knowledge, so for the first time we aimed our research to determine whether the two oral antidiabetics are advantageous over insulin, concerning their effects on myometrial contractility during labour. Therefore, we studied the effects of glibenclamide and metformin on the spontaneous uterine contractions in the female non-diabetic non-pregnant rats.

MATERIALS AND METHODS

The drugs and chemicals

Glibenclamide (glyburide) powder was dissolved in 95% ethanol (Piper *et al*., 1990) (vehicle). Metformin powder was dissolved in distilled water (Martindale, 2010). The drug molar concentrations (M) were calculated and expressed in terms of the final bath concentrations. All solutions were freshly prepared on a daily basis and were maintained at 35°C (Ghosh, 1971).

The animals

The present study was carried out using the adult non-pregnant female albino rats weighing 200-250g. The female rats were housed in an air conditional animal room at 23±2ºC with 12/12h light/dark photoperiod, and free access to water and normal laboratory rat chow. The Ethics Committee of King Saud University approved all experiments. Before killing the rat, all equipment should be checked for their perfect working condition. The organ bath should be scrupulously cleaned.

Tissue isolation and recording of uterine contractions

The physiological salt (De Jalon) solution (PSS) was in the following composition (g/L): NaCl 9.00, KCl 0.42, $CaCl_2$ 0.06, $NaHCO_3$ 0.50 and glucose 0.50 and bubbled with the gas mixture of 95% O_2 and 5% CO_2 and maintained at 31ºC (Ghosh, 1971).

The female virgin (non-pregnant) rats were sacrificed by a head blow and cervical dislocation. The uteri were removed, cleaned of fat and immediately placed into the PSS in a shallow Petri dish. Then, the two horns were divided. Each uterine horn was opened longitudinally and cut into longitudinal strips of approximately 1 cm length. The strip was mounted vertically in a 10 ml-organ bath containing the PSS; one end of the tissue strip was tied by a thin cotton thread to a fixed support and the other to the recording lever and the other horn kept in the salt solution at fridge for later use, if needed. The regular spontaneous contractile activity of healthy myometrium developed within a 30-60min. The writing lever of the kymograph was adjusted for the desired degree of magnification and tension (Ghosh, 1971). The organ bath is emptied every 5 min and the solution replaced to avoid alteration of pH or other metabolites (Aaronson *et al*., 2006; Downing and Hollingsworth, 1991).

Either drug or vehicle [ethanol 95% (Piper *et al*., 1990) dose (0.1ml) was added into the bath fluid via a 1-ml graduated tuberculin syringe (at uniform speed). Drug or vehicle was quickly mixed throughout due to bubbling of the gas. Simultaneously with the addition of the vehicle or drug, the uterine contractility was recorded.

The vehicle or drug was allowed to act until the uterine contractility reaches a steady level for 30 min after which the bath fluid containing the vehicle or drug was washed out and the fresh PSS was allowed to refill the bath. Thus, PSS was added and replaced two or three times, to allow the uterine contractility to return to control level. At the time of washing, the recording is stopped so that any contraction due to the momentary exposure of the tissue to air is recorded as vertical kick and readily discernible from true contractions. The uterine contractions were recorded on the smoked paper rounded on the rotating kymograph drum. After the end of experiment, fixing of the tracings on smoked surface was done (Ghosh, 1971; Aaronson *et al*., 2006).

Both metformin and glyburide are the two oral hypoglycemic agents in the WHO Model List of Essential Medicines (WHO, 2007).

The effects of glibenclamide or metformin on the amplitude and frequency of spontaneous uterine contractions:

After the 30-90 min equilibration period, the control frequency and amplitude of spontaneous uterine contractions were recorded for 30min (control period).

The vehicle which gets mixed up quickly due to bubbling of the gas was allowed to act till the response reaches a steady level for 30min after which the bath fluid containing the vehicle was washed out and the fresh PSS was allowed to refill the bath.

Table 1: The effects of the vehicle (ethanol) and molar concentrations (10^{-7}, 10^{-6} and10^{-5} M) of glibenclamide on the amplitude and frequency of spontaneous contractions of the isolated uterus of female non-pregnant rats (No. in the group (*n)* = 5-8).

Parameter	Vehicle (Ethanol)	Glibenclamide 10^{-7} M	Glibenclamide 10^{-6} M	Glibenclamide 10^{-5} M
The amplitude (Mean% ±S.E)	-4.42%±4.74^{A}	-18.05%±9.45^{A}	-24.24%±14.77^{A}	- 29.34 % ± 9.87^{A}
The frequency (Mean % ± S.E)	3.43%±2.15^{A}	-1.33 %±1.33^{A}	3.18%±3.39^{A}	0.03 % ± 3.29^{A}

Table 2: The effects of the molar concentrations (10^{-7}, 10^{-6} and10^{-5} M) of metformin on the amplitude and frequency of spontaneous contractions of the isolated uterus of female non-pregnant rats (No. in the group (*n*=5).

Parameter	Metformin 10^{-7} M	Metformin 10^{-6} M	Metformin 10^{-5} M
The amplitude (Mean % ± S.E)	0.79 %±5.30^{A}	- 0.09%±8.07^{A}	- 3.37%±3.93^{A}
The frequency (Mean % ± S.E)	- 0.64%±4.03^{A}	- 3.33%±2.22^{A}	- 1.05%±1.05^{A}

No significant differences between groups. Within the same row, values with common superscript capital letters are non-significantly (P>0.05) different (one-way ANOVA).

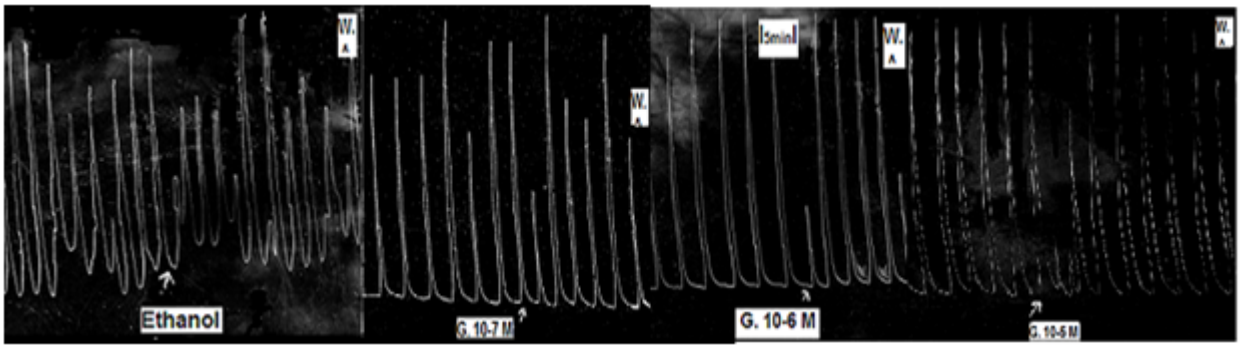

Fig. 1: The tracing showing the effects of the vehicle (ethanol) and molar concentrations (10^{-7}, 10^{-6} and10^{-5} M) of glibenclamide (G.) on the spontaneous contractions of the isolated uterus of female non-pregnant rats (W. = washing the isolated uterus).

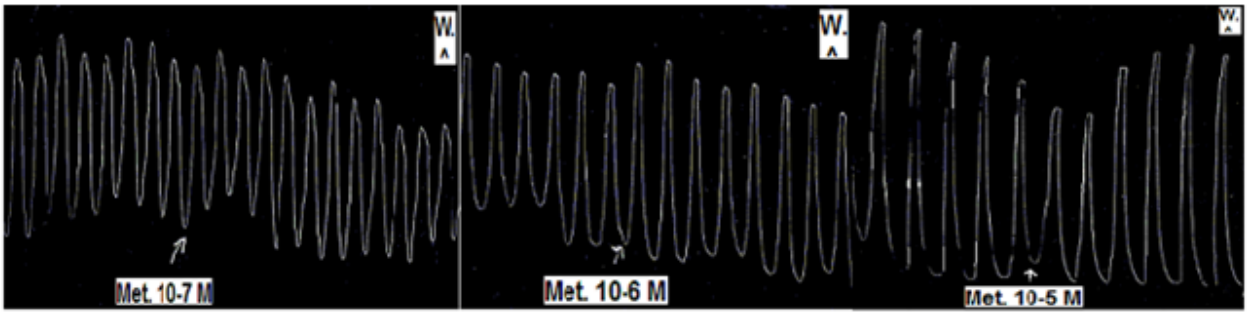

Fig. 2: The tracing showing the effects of the molar concentrations (10^{-7}, 10^{-6} and10^{-5} M) of metformin (Met.) on the spontaneous contractions of the isolated uterus of female non-pregnant rats (W. = washing the isolated uterus).

Glibenclamide or metformin, at 10^{-7} to 10^{-5} M was then added to the bath solution for 30 min. and with washing PSS in-between.

These experimental procedures were repeated on 5-8 isolated rat uteri for glibenclamide and on 5 isolated rat uteri for metformin.

STATISTICAL ANALYSIS

The parameters measured, the amplitude and frequency (per 30 min) of the spontaneous uterine contractions, are expressed as mean % ± SEM for the vehicle and drugs' doses of the control uterine contractions before each dose (the control is considered as 100%). Significance was tested using the paired-samples student's t tests were used to analyze the effects of the vehicle in comparison to the control. Also, the one-way analysis of variance (one-way ANOVA) followed by Post Hoc and least significant difference (LSD) tests (SPSS v16.0) were used to analyze and compare the effects and differences between the effects of the vehicle and different molar concentrations of the drugs. Differences were taken as significant if P values were <0.05 and *n* were the numbers in samples

(Ghosh, 1971; Aaronson *et al*., 2006; Downing and Hollingsworth, 1991).

RESULTS

The effect of glibenclamide on the amplitude and frequency of spontaneous uterine contractions

Table 1 depicts the effects of glibenclamide on the amplitude and frequency of spontaneous contractions of the isolated uterus of female non-pregnant rats. The ethanol vehicle had no significant effect on the amplitude of spontaneous uterine contractions over 30 min when compared to time matched control contractions (the mean effect of vehicle-4.42%±4.74). There were no significant differences between the mean effects on the amplitude of spontaneous myometrial contractions with glibenclamide at any concentrations 10^{-7}, 10^{-6} and 10^{-5} M (*n*=5-8); The means being: -18.05%±9.45,-24.24%±14.77 and - 29.34% ±9.87 respectively (table 1 and figure 1).

Ethanol had no significant (P<0.05) effect on the frequency of spontaneous uterine contractions when compared to control contractions (the mean effect of vehicle 3.43% ±2.15). Also, there were no significant (P<0.05) differences between the mean frequency of spontaneous contractions with glibenclamide concentrations 10^{-7}, 10^{-6} and 10^{-5} M being:- 1.33%±1.33, 3.18% ±3.39 and 0.03% ±3.29 respectively (table 1 and figure 1).

The effect of metformin on the amplitude of spontaneous uterine contractions

Table 2 showed the effects of metformin on the amplitude and frequency of spontaneous contractions of the isolated uterus of female non-pregnant rats. There were no significant (P<0.05) differences between the mean effects on the amplitude of spontaneous myometrial contractions in the different groups; the metformin molar concentrations 10^{-7}, 10^{-6} and 10^{-5} M (*n*=5); the means being: 0.79%±5.30,- 0.09%±8.07 and -3.37%±3.93 respectively (table 2 and figure 2).

There were no significant (P<0.05) differences between the mean effects on the frequency of spontaneous myometrial contractions in the different groups; the metformin molar concentrations 10^{-7}, 10^{-6} and 10^{-5} M (*n* = 5); the means being:- 0.64 %±4.03, -3.33% ±2.22 and - 1.05%±1.05 respectively (table 2 and figure 2).

DISCUSSION

The present study showed that glibenclamide (a KATP channel blocker) and metformin have no effects on the amplitude and frequency of the spontaneous contractions of the uterine strips from the female non-pregnant non-diabetic rats. In other words, these two oral antidiabetics do not change the spontaneous uterine contractions. The present study was performed on a small number of strips. The uterine strips were isolated from non-pregnant female rats, with their uteri and internal environments subjected to physiological, biochemical and hormonal stresses different from than in pregnant rats. The same should be considered concerning the uterine strips from non-diabetic female rats, which are different from diabetic female rats. These considerations are very impressive in explaining some agreements and disagreements with previous studies. Downing and Hollingsworth (Downing and Hollingsworth, 1991) reported that glibenclamide mechanism is by blocking ATP-dependent K^+-channels in pancreatic β -cells, leading to depolarization of membrane potential and as a result, calcium channels open leading to an increment in calcium entry that stimulates insulin release (Groop *et al*., 1991). Also, glibenclamide may act indirectly, by an action unrelated to its ATP-dependent K^+-channel blockade, to antagonize relaxin effect. This glibenclamide-relaxin interaction may be of a non-competitive allosteric nature (Gedeon and Koren, 2006).

Furthermore, relax in increased the myometrial cyclic AMP concentrations. The early stages of relaxin inhibition or of its low doses are possibly cyclic AMP-mediated and thus little affected by glibenclamide, but the maintenance of uterine quiescence or higher doses of relaxin may be K^+-channel opening-mediated events, and therefore were more affected by glibenclamide (Downing and Hollingsworth, 1991).

In accordance with the present observations about glibenclamide, Downing and Hollingsworth (Downing and Hollingsworth, 1991) indicated that glibenclamide reduced the cromakalim-induced inhibition of uterine contraction, which is consistent with the competitive antagonism of cromakalim observed *in vitro* uterine tissue (Piper *et al*., 1990; Rowan *et al*., 2008). In addition, it was reported that glibenclamide reversed the uterine inhibition induced by cromakalim and appears to stimulate uterine contractions above control. Moreover, our results are in agreement with Piper *et al*. (Piper *et al*., 1990) who stated that glibenclamide reduced the uterine sensitivity to cromakalim, which inhibits uterine contractions. Glibenclamide alone lacks an effect on uterine contractions. This is also in agreement with other authors (Piper *et al*., 1990; Rowan *et al*., 2008).

Aaronson *et al.* (2006) suggested that BKCa channels play little role in regulating the spontaneous contractions in myometrial rat strips, but inhibition of KV channels causes marked effects on spontaneous activity, thus these channels control the frequency and amplitude of spontaneous depolarizations in rats. Also, regarding glibenclamide, the present results are consistent with Shimano *et al*. who indicated that nitric oxide should control human myometrial relaxation during pregnancy, mediated by the activation of Ca^{2+}-activated K^+ channels.

However, the present observations disagreed with Villar *et al*. (1986) who reported that sulfonylureas (tolbutamide and glibenclamide) are capable of relaxing contractions of rat uterus induced by various agents, e.g. oxytocin, in a medium with Ca^{2+}. The capacity of sulfonylureas to relax also oxytocin-induced Ca-free contraction indicates the possibility that the action of these drugs could be due, above all, to a decrease in the intracellular free calcium level. The likely mechanism of sulfonylurea-induced uterine relaxation is not due to an increase in the efflux of calcium but to an increase in the Ca -uptake by intracellular organelles. Also, glibenclamide antagonized the uterine contractions evoked by CaCl2 and did not affect the spontaneous contractions of rat uterus (Villar *et al*., 1986). This later effect is in accordance with the present study. Also in agreement with the present study, Aaronson *et al*. (2006) reported that BKCa channels inhibitors- which suppress BKCa currents (Knock *et al*., 1999) and/or inhibit the relaxing effects of agents which activate these channels in isolated tissues- have no or only minor effects on the spontaneous contractions of rat myometrial strips under basal conditions.

The present results are in accordance indirectly with Vedernikov *et al.* (1986) who reported that both forskolin and 1,9-dideoxyforskolin inhibit uterine contractions. Glibenclamide and other K^+- channel blockers attenuated this inhibitory effect of forskolin, whereas glibenclamide was less effective against 1,9-dideoxyforskolin. This inhibition of uterine contractions by 1, 9-dideoxyforskolin and forskolin in pregnant rats' uterine rings involves activation of adenylatecyclase and calcium-dependent-potassium channels and, with forskolin ATP-dependent potassium channels (Vedernikov *et al*., 2000).

Metformin is able to cross the placenta and enter fetal circulation and rated by FDA as pregnancy risk category B (Anonymous, 2003; Rowan *et al*., 2008). However, one trial has shown that metformin alone or with supplemental insulin in women with GDM is not associated with higher perinatal complications as compared with insulin. In addition, this trial showed that women used the combined treatment of insulin and metformin required less insulin compared with women used insulin alone. The same study reported a similar rate of macrosomia and CS in metformin and insulin groups (Wray, 2007). However, in women with GDM, many studies reported that there was no difference in the rate of Cs between insulin and glibenclamide use (Langer *et al*., 2000; Jacobson *et al*., 2005; Bertini *et al*., 2005).

Glibenclamide is considered as a safe drug to be used to achieve euglycemia during pregnancy in women with type-2 or GDM. Compared to metformin and other sulfonylureas, glyburide does not cross the placenta in significant amount (Koren, 2001; Escande *et al*., 1989) and has no effect on the rate of fetal anomalies. It has been reported that glyburide is as effective as insulin to achieve euglycemia in patients with GDM (Koren, 2001).

CONCLUSION

The present results suggest that the two oral antidiabetics, glibenclamide and metformin, do not affect the spontaneous uterine contractility in the non-pregnant non-diabetic female rat strips, or at least, they do not inhibit the spontaneous myometrial contractions in these rat strips. Therefore, we suggest that these two drugs may be useful for achieving the euglycemia in diabetic pregnant women, without interfering with uterine contractions during labour, unlike insulin. However, the effects of the oral antidiabetics on the uterine contractility need further investigations in the diabetic pregnant female rats. Also, further studies regarding oral hypoglycemic medications safety in pregnancy is recommended to open a new era in the management of type-2 and GDM.

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REFERENCES

Aaronson PI, Sarwar U and Gin S *et al* (2006). A role for voltage-gated, but not Ca^{2+}-activated, K^+ channels in regulating spontaneous contractile activity in myometrium from virgin and pregnant rats. *Br. J. Pharmacol.,* **147**(7): 815-824.

Anonymous (2003). Gestational diabetes mellitus. *Diabetes Care.,* **26**(Suppl 1): S103-105.

Balsells M, Corcoy R, Adelantado JM, Garcia-Patterson A, Altirriba O and de Leiva A (2000). Gestational diabetes mellitus: metabolic control during labour. *Diabetes. Nutr. Metab.,* **13**: 257-262.

Bell R, Bailey K, Cresswell T, Hawthorne G, Critchley J and Lewis-Barned N (2008). Trends in prevalence and outcomes of pregnancy in women with pre-existing type I and type II diabetes. *Bjog.,* **115**: 445-452.

Bertini AM, Silva JC and Taborda W *et al* (2005). Perinatal outcomes and the use of oral hypoglycemic agents. *J. Perinat. Med.,* **33**: 519-523.

Catalano PM, Drago NM and Amini SB (1995). Maternal carbohydrate metabolism and its relationship to fetal growth and body composition. *Am. J. Obstet. Gynecol.,* **172**: 1464-1470.

Crowther CA, Hiller JE, Moss JR, McPhee AJ, Jeffries WS and Robinson JS (2005). Effect of treatment of

gestational diabetes mellitus on pregnancy outcomes. *N. Engl. J. Med.,* **352**: 2477-2486.

Downing SJ and Hollingsworth M (1991). Antagonism of relaxin by glibenclamide in the uterus of the rat *in vivo*. *Br. J. Pharmacol.,* **104**: 71-76.

Dunne F, Brydon P, Smith K and Gee H (2003). Pregnancy in women with Type 2 diabetes: 12 years outcome data 1990-2002. *Diabet. Med.,* **20**: 734-738.

El Mallah KO, Narchi H, Kulaylat NA and Shaban MS (1997). Gestational and pre-gestational diabetes: Comparison of maternal and fetal characteristics and outcome. *Int. J. Gynaecol. Obstet.,* **58**: 203-209.

Escande D, Thuringer D and Le Guern S *et al* (1989). Potassium channel openers act through an activation of ATP-sensitive K+-channels in guinea-pig cardiac myocytes. *Pfluigers. Arch.,* **414**: 669-675.

Evers IM, de Valk HW and Visser GH (2004). Risk of complications of pregnancy in women with type 1 diabetes: Nationwide prospective study in the Netherlands. *B.M.J. (Clinical research ed.)* **328**: 915.

Franchi AM, Chaud M, Gonzalez ET, Gimeno MA and Gimeno AL (1988). Effects of experimental diabetes on spontaneous contractions, on the output of prostaglandins and on the metabolism of labelled arachidonic acid, in uteri isolated from ovariectomized rats. *Influences of estradiol. Prostaglandins,* **35**: 191-205.

Gedeon C and Koren G (2006). Designing pregnancy centered medications: Drugs which do not cross the human placenta. *Placenta,* **27**: 861-868.

Ghosh MN (1971). Fundamentals of Experimental Pharmacology.Scientific book agency. Calcutta 1, pp.16, 17, 39, 40, 43, 46.

Goldraij A, Sterin AB, Gimeno MF and Gimeno AL (1979). Effects of insulin on the oxytocin-induced motility of rat isolated uterine horns. *Acta. Physiol. Lat. Am.,* **29**: 337-340.

Groop LC, Barzilai N and Ratheiser K *et al* (1991). Dose-dependent effects of glyburide on insulin secretion and glucose uptake in humans. *Diabetes. Care,* **14**: 724-727.

Jacobson GF, Ramos GA, Ching JY, Kirby RS, Ferrara A and Field DR (2005). Comparison of glyburide and insulin for the management of gestational diabetes in a large managed care organization. *Am. J. Obstet. Gynecol.,* **193**: 118-124.

Jawerbaum A, Catafau JR, Gonzalez ET, Novaro V, Gomez G, Gelpi E and Gimeno MA (1996). Eicosanoid production, metabolism and contractile activity in the isolated uterus from non-insulin-dependent diabetic rats during late pregnancy. *Prostaglandins.,* **51**: 307-320.

Jensen DM, Damm P and Moelsted-Pedersen L *et al* (2004). Outcomes in type 1 diabetic pregnancies: A nationwide, population-based study. *Diabetes. Care.,* **27**: 2819-2823.

Kaya T, Cetin A, Cetin M and Sarioglu Y (1999). Effects of endothelin-1 and calcium channel blockers on contractions in human myometrium. A study on myometrial strips from normal and diabetic pregnant women. *J. Reprod. Med.,* **44**: 115-121.

Knock GA, Smirnov SV and Aaronson PI (1999). Voltage-gated K^+ currents in freshly isolated myocytes of the pregnant human myometrium. *J. Physiol.,* **518**: 769-781.

Koren G (2001). Glyburide and fetal safety: Transplacental pharmacokinetic considerations. *Reprod. Toxicol.,* **15**: 227-229.

Kuznetsova L, Plesneva S, Shpakov A and Pertseva M (2005). Relaxin adenylyl cyclase system of pregnant women with diabetes: Functional defects in insulin and relaxin adenylyl cyclase signaling systems in myometrium of pregnant women with type 1 diabetes. *Ann. NY. Acad. Sci.,* **1041**: 446-448.

Kuznetsova LA, Fedin AN, Chistiakova OV, Plesneva SA, Shpakov AO and Pertseva MN (2006). Involvement of adenylyl cyclase signaling mechanisms in relaxing effect of relaxin and insulin on the rat uterus, trachea and human myometrium. *Ross. Fiziol. Zh. Im. I M Sechenova.,* **92**: 863-871.

Langer O, Conway DL, Berkus MD, Xenakis EM and Gonzales O (2000). A comparison of glyburide and insulin in women with gestational diabetes mellitus. *N. Engl. J. Med.,* **343**: 1134-1138.

Martindale (2010). The complete drug reference, 35th ed., Pharmaceutical Press.

McMurtrie EM, Ginsberg GG, Frederick GT, Kirkland JL, Stancel GM and Gardner RM (1985). Effect of a diabetic state on myometrial ultra structure and isolated uterine contractions in the rat. *Proc. Soc. Exp. Biol. Med.,* **180**: 497-504.

NICE (2008). Diabetes in pregnancy. Management of diabetes and its complications from preconception to the postnatal period. In London: CG63 - RCOG Press.

Piper I, Minshall E, Downing SJ, Hollingsworth M and Sadraeli H (1990). Effects of several potassium channel openers and glibenclamide on the uterus of the rat. *Br. J. Pharmacol.,* **101**: 901-907.

Rowan JA, Hague WM, Gao W, Battin MR and Moore MP (2008). Metformin versus insulin for the treatment of gestational diabetes. *N. Engl. J. Med.,* **358**: 2003-2015.

Shimano M, Nakaya Y, Fukui R and Kamada M *et al* (2000). Activation of Ca2+-activated K+ channels in human myometrium by nitric oxide. *Gynecol. Obstet. Invest,* **49**(4): 249-54.

Stenninger E, Lindqvist A, Aman J, Ostlund I and Schvarcz E (2008). Continuous subcutaneous glucose monitoring system in diabetic mothers during labour and postnatal glucose adaptation of their infants. *Diabet. Med.,* **25**: 450-454.

Vedernikov YP, Syal AS, Okawa T, Saade GR and Garfield RE (2000). Adenylate cyclase and potassium

channels are involved in forskolin- and 1, 9-dideoxyforskolin-induced inhibition of pregnant rat uterus contractility. *Am. J. Obstet. Gynecol.,* **182**(3): 620-624.

Villar A, Ivora MD, D'Ocon MP and Anselm E (1986). Effects of sulphonylureas on spontaneous motility and induced contractions in rat isolated uterus. *J. Pharm. Pharmacol.,* **38**(10): 778-780.

WHO (2007). WHO Model Lists of Essential Medicines, 15th edition.

Wray S (2007). Insights on the uterus. *Exp. Physiol.,* **92**(4): 621-631.