Effects of Magnesium, Oxytocin, and Prostaglandin F$_{2\alpha}$ on the Generation and Propagation of Excitation in the Longitudinal Muscle of Rat Myometrium during Late Pregnancy

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Abstract In order to find out the changes in myometrial properties towards parturition, effects of Mg, oxytocin, and prostaglandin F$_{2\alpha}$ (PGF$_{2\alpha}$) were investigated on longitudinal muscle strips taken from rat uteri on Days 20 and 22 of pregnancy. With intracellular recording by microelectrode, membrane potential was 50.5±1.4 mV on Day 20, and 49.9±1.0 mV on Day 22 in the Mg-free Krebs solution. The slow potential was larger, and the spike potentials during a burst discharge exhibited variable amplitude on Day 20, whereas these were regular on Day 22. With extracellular recording by the rubber gap method, spike potentials discharged in a synchronized manner on Day 22, suggesting a better coordination between cell groups. When 1.2 mM Mg was applied, the spontaneous activity was depressed more strongly on Day 22. The excitatory effect of PGF$_{2\alpha}$ faded sooner, whereas that of oxytocin persisted. When the fading of PGF$_{2\alpha}$ had occurred, the excitatory effect of oxytocin was weaker than when it was given alone. Because propagation of spontaneous activity, occurring either in the Mg-free solution or in the presence of oxytocin and PGF$_{2\alpha}$ in normal Krebs solution, did not show a definite direction from ovarian to vaginal side of the longitudinal muscle, it was concluded that generation of spontaneous activity was not localized in a given site of the muscle, but originated anywhere in Days 20 and 22 preparations.

Key Words: magnesium, oxytocin, prostaglandin F$_{2\alpha}$, longitudinal muscle.
cells in vitro deprived of the nervous influences exhibit spontaneous activity which is in some cases rhythmic, organized activity is definitely dependent on the intrinsic neural elements. The classic thesis proposed by Alvarez and Mahoney (1922), i.e. the gradient of the intestine, has been proved to occur owing to recent elaborate investigations (for review, see Holman, 1981).

By contrast, uterine motility depends mostly on myogenic control under the influence of ovarian and other hormonal factors. It has long been proposed that uterine muscle is kept quiescent during pregnancy, and brought to activity at term (CsaPo, 1956; Reynolds, 1965). The achievement of delivery is thought to depend on pacemaker activity which is initiated at the tubo-uterine junction. This proposal is teleologically acceptable, especially in the multiparous animals such as rat, because fetuses should move from the ovarian to vaginal end. A similar concept has also been assumed to occur in monoparous animals such as monkey and human (Reynolds, 1965). It has been shown in rats that the uterus in vivo is not necessarily inactive during pregnancy (Fuchs, 1978). Thus, it was argued that the contractions remain local during pregnancy, and the whole uterus participates in a synchronized activity at term (Fuchs, 1978). In support for this, gap junctions are found in the myometrium during a very limited period around delivery (Garfield et al., 1978). On the other hand, in vitro experiments indicate that spontaneous activity and cable properties of muscle strips occur at nearly all stages of pregnancy (Kuriyama and Suzuki, 1976; Kanda and Kuriyama, 1980).

Such a dilemma between in vitro and in vivo experimental results would probably be introduced because the surrounding media of muscle cells and size of preparations are different, and furthermore some artificial effects may be brought about by stretching the muscle strips in order to ensure the impalement of microelectrodes. The present study was undertaken primarily to know the point of initiation and propagation of spontaneous activity, keeping in mind the assumption that the spontaneous activity may proceed from ovarian to vaginal end of a muscle strip. The experiments were carried out on in vitro longitudinal muscle strips of rat myometrium during late pregnancy. The mechanical responses and electrical activities recorded intracellularly and extracellularly were obtained. In addition the effects of Mg, oxytocin, and prostaglandin F\(_{2\alpha}\) were also studied.

**METHODS**

**Preparations.** Daily vaginal smears were taken from Wistar-strain female rats. The females on proestrus were caged with males overnight, and mating was confirmed by the presence of sperm in the vaginal smear on the following morning. When successful mating was proved, the day was counted as Day 1 of pregnancy. Parturition usually occurred in the afternoon of Day 22 of pregnancy. The present work was carried out on uteri taken from animals sacrificed in the morning of Days 20 or 22 of pregnancy. Longitudinal muscle strips were dissected with
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fine scissors from antiplacental parts in the ovarian portion of the uterus.

The control solution was Krebs solution containing (mM): NaCl 121.9, NaHCO3 15.5, KCl 4.7, KH2PO4 1.2, CaCl2 2.5, MgCl2 1.2, and glucose 11.5. It was equilibrated with a gas mixture of 95% O2+5% CO2 (pH of 7.3). Because the spontaneous activity was infrequent or absent in normal Krebs solution, Mg ion was omitted unless otherwise stated. Several muscle strips were taken from the same uterine horn and kept at room temperature (about 25°C), for use within the day. The preparation was equilibrated with warm Krebs solution for 30–60 min before the experiment began. The temperature of the bathing solution was 36°C.

Drugs used were synthetic oxytocin (Pituitan S, Nihon-zoki Pharm. Co.) and prostaglandin F2α (Ono Pharm. Co.). Concentration of the agents is given in U/ml for oxytocin, and g/ml for prostaglandin F2α (PGF2α).

Mechanical recording. The in vivo length of the uterine horn was measured, and a muscle strip 3 cm long and 1 mm wide was set up horizontally in a recording chamber. Spontaneous activity was recorded by a force displacement transducer, and the effects of ligation were studied (for detail, see Fig. 7).

Electrical recording. Intracellular recording of the membrane activity was made on a longitudinal muscle strip 1 mm wide and 5 mm long by conventional microelectrodes filled with 3M KCl, in the partition chamber as described by Abe and Tomita (1968). Extracellular recording of the membrane activity was made by a rubber gap method. In the present experiments, a muscle strip of 2 cm in vivo length (0.3–0.5 mm wide) was separated in the middle by a thin rubber film, through a small hole (approximately 0.2 mm in diameter) in which the muscle strip was passed, and the solution having the same composition was added in each chamber (0.8 ml capacity) separated by the rubber film. A pair of silver electrodes coated by AgCl was dipped in each chamber, and the electrical activity across the rubber film was fed to a DC amplifier. A negative electrical signal on the ovarian side of the muscle strip was recorded as an upward deflection on an inkwriting oscillograph (cf. Fig. 2).

RESULTS

Experiments in Mg-free Krebs solution

The present experiments were carried out on the uterine muscle strips taken from rats pregnant for 20 or 22 days. The day of pregnancy is shown in parenthesis in the following description.

Burst discharges occurred spontaneously in the solution, where the external Mg ion was omitted. Figure 1 shows the mechanical record (upper trace), and electrical activity recorded by an intracellular microelectrode (lower trace). Records A and B were taken on Day 20. The interval between activity was regular, and the burst discharge was accompanied by contraction in a one-to-one manner.

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Records to the right show the responses on a fast sweep speed. In A, the successive spontaneous burst discharges exhibit a variable pattern, even though recorded from the same cell. The electrical activity is probably composed of spike potentials and slow potentials, which appear responsible for the generation of the former. When the amplitude of the slow potential was larger, the amplitude of the spike potential was smaller. The slow potential was sometimes interrupted, but resumed within the burst (middle of the fast sweep). The record shown in B (Day 20) exhibited nearly the same features, but the discharge pattern was less variable, i.e. the amplitude of the spike potential was almost constant.

Records shown in C, D, and E are from muscle strips of different rats pregnant for 22 days. The rats were sacrificed at 10 a.m., several hours prior to parturition. Different patterns are apparent from those of Day 20. The amplitude of slow potentials is smaller and more uniform, and the spike potential was regular. Aborted generator potentials can be observed at the end of burst discharges in records D and E. The membrane potential in the Mg-free media was $50.5 \pm 1.4$ mV (mean $\pm$ S.D., $n=27$, 10 rats) and $49.9 \pm 1.0$ mV (mean $\pm$ S.D., $n=35$, 13 rats) for 20 and 22 days, respectively. They were not significantly different from each other in $t$-test ($P>0.05$).

Records shown in Fig. 2 illustrate the electrical activity in muscle strips (Day 20), obtained by bipolar recording (see METHODS). The main difference between
the microelectrode recordings described previously and the bipolar ones is that the former were from single cells and the latter from muscle bundles. Under the binocular microscope, small muscle bundles about 0.2 mm wide were observed to attach together and to branch from each other, and an excised muscle strip was composed of about four such bundles. Because of the bipolar recording of the burst discharge, the electrical activity can be complicated, for instance in a burst such as shown in A. Nevertheless, the initial downward deflection indicates that the origin of the spontaneous activity was located in the vaginal side of the muscle strip. In B and C, the pacemaker site is located in the ovarian and vaginal side of the muscle strip, respectively. In D, the electrical activity was more or less regular and the spike potentials within a burst discharged in groups. This tendency was more pronounced in record E. Presumably, a bundle could be a unit of conduction of excitation. If so, the complicated pattern would mean an asynchronous discharge in a muscle strip, in addition to any irregularity of the membrane activity. Twin type of spike discharge in a burst (D, E) may imply some delay of membrane response in muscle bundles. In F, the spike potentials across the rubber plate discharged in a one-to-one manner. In this case, it is supposed that spike activity in the bundles is completely synchronized. Among above electrical activities, discharge types shown in A, B, and C were most usually observed. A comparison of records suggests that the pacemaker site is not fixed on either the ovarian or

Fig. 2. Spontaneous membrane activity of longitudinal muscle strips (2 cm long) recorded by rubber gap method. A rubber film separated the muscle strip in the middle. Each record is from a different rat pregnant for 20 days. Experiments were carried out in Mg-free Krebs solution. Upward deflection occurred when the ovarian side became negative.
vaginal side of the muscle strip of rats pregnant for 20 days.

Figure 3 illustrates spontaneous discharges in muscle strips of different rats pregnant for 22 days. Records A and B show complicated patterns, reminiscent of those mostly observed in rats pregnant for 20 days. In the muscle strip shown in C, the amplitude of compound action potential was larger and the spike activity which originated on the vaginal side was conducted into the ovarian side in a one-to-one manner. Such one-to-one conduction of spike potentials was observed in most of the muscle strips (D–G). In D, the initial spike potential came from the vaginal side, while later ones within the burst were initiated on the ovarian side. It is interesting to speculate whether such changes in direction of propagation were due to the re-entry of spike activity from a muscle bundle on the other side of the separating rubber film. However, propagation of spike potentials was from ovarian to vaginal side throughout in the case of E. As far as the polarity of action potentials indicates, the propagation initially invaded from the ovarian side, then from the vaginal side, and again from the ovarian side in the case of F. In G, at the beginning of the burst the amplitude of generator potential progressively increased on the ovarian side until a spike potential was generated. Then the spike potential was conducted into the vaginal side. Finally, the direction of spike propagation reversed, i.e. from vaginal to ovarian side. Thus, together with the microelectrode recording, it can be concluded that not only the membrane property of muscle cells, but also the cell groups differ between the longitudinal muscles from rats pregnant for 20 and 22 days. The largest amplitude of spike potential

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in the burst discharge of different muscle strips was measured, and the average was 4.4 ± 2.7 mV (± S.D., n = 94; ranging between 19.3 and 1.2 mV) for Day 20, and 10.5 ± 6.7 mV (n = 83; ranging between 28.0 and 1.1 mV) for Day 22, respectively.

Figure 4 shows a recording of successive burst discharges in a muscle strip (Day 20). The interval between the bursts ranged from 1.5 to 2 min. Patterns were irregular and full of variation. It is normally not possible to find out the direction of conduction of spike potentials, except for the initial responses and sometimes those at the end of a burst. Inspection of the deflection of initial responses indicates that some were conducted in the vaginal to ovarian direction (a, c, e, f), while others were the opposite way (b, d). This indicates that the pacemaker site is not located in a certain area in the same muscle strip, but the spontaneous activity originates multifocally.

Records of successive burst discharges in a muscle strip for Day 22 are shown in Fig. 5. In Aa and Ac, the propagation was from ovarian to vaginal side, whereas propagation was in the opposite direction in Ab. In this particular case, an abortive generator potential preceded the burst, then conducting spike potentials were produced. It can also be observed in the succeeding discharge that the direction was in the ovarian to vaginal direction.

The experimental results shown in Fig. 5A were steady state responses recorded after 1 hr equilibration in the warm bathing solution. Record Ba (Day 22) was
taken after 20 min equilibration, then the following ones every 15 min (Bb–Bd). Every record indicates the direction of burst discharge to occur from the ovarian to vaginal side. During the early period of incubation (Ba, Bb), spike potentials discharged in groups. As the time proceeded, the amplitude of spikes grew larger (Bc, Bd). The interval between the spike activities remained almost constant throughout. Taking into consideration that a muscle strip is composed of several bundles, it may be assumed that the increase in the amplitude of spikes was due to the synchronization of activity among the bundles. The detailed nature is unknown.

To summarize how the pacemaker site is located in a given site in the longitudinal muscle strips, 10–30 successive bursts were recorded as shown in Figs. 4 and 5. Discharges directed from ovarian to vaginal side were estimated as a per cent of total bursts for Day 20 (open bar) and Day 22 (filled bar, Fig. 6). The muscle strips were subdivided into 5 groups according to this percentage (abscissa). In the figure, zero on the abscissa means that propagation occurred totally from the vaginal to ovarian side. Approximately one third of muscle strips exhibited unidirectional propagation either from ovarian to vaginal side or vice versa. The rest showed bidirectional propagation. There was in the relationship no significant difference between Day 20 and Day 22 muscle strips. Apparent unidirectional propagation does not necessarily mean that the pacemaker site is localized at a given site, because the separating rubber plate was so thin and the muscle strip was about 2 cm long (see METHODS). Thus, it can probably be concluded that the spontaneous activity can originate anywhere, and is conducted along the whole length of a muscle strip.

Most of muscle strips (3 cm long) were spontaneously active, and the contract-
Ile amplitude and rhythm were regular as shown in Fig. 7A (Day 20) and B (Day 22). In some muscle strips contractions were periodic (C, Day 22), and the quiescent period between the contractions ranged from 20–30 min. A stepwise increase in the magnitude of contraction was a common observation in such cases. Ligation by thread every 1 cm (x, xx) caused an increase in the frequency of contractions (A and B). It was found under a binocular microscope that when a portion of muscle strip contracted, another part beyond the ligation stretched or contracted. There is a possibility that contraction of one part stretches another which is then stimulated to contract so that the contraction will be the same frequency but out of phase. On the other hand, because observation with the microscope indicated that the initial contraction started from either part across the ligation, it seems likely that each part was contracting independently. In C, the ligations appeared to initiate contractions. However these did not last long. Therefore, injury may cause the contractions, but this does not fully account for the increase in the frequency of contractions seen in A and B. The pacemaker activity can be generated anywhere in a muscle strip (cf. Fig. 6). Therefore when a muscle strip is functionally isolated by ligation each part can independently generate spon-

Fig. 6. Histogram showing the direction of impulse propagation of spontaneous discharge in longitudinal muscle strips of pregnant uteri (open bar, 20 days (n=65); filled bar, 22 days (n=73)). In each muscle strip, the % ratio of propagation from ovarian to vaginal sides for 10–30 spontaneous burst discharges was estimated and divided into 5 groups (abscissa), and percentages of muscle strips belonging to each group against total muscle strips for Day 20 and Day 22 are plotted in ordinate. Experiments were carried out in Mg-free Krebs solution.
spontaneous contractions. The question arises why an intact uterine muscle strip does not exhibit a tetanic tone like the intestinal smooth muscle tissues. This remains to be clarified.

Experiments in normal Krebs solution

When 1.2 mM Mg was added to the bathing solution, making the solution into normal Krebs solution, the membrane activity was stabilized (Fig. 8), i.e. the interval between spontaneous activity became longer and the duration of discharges shorter. The resting membrane potential was not changed, and the contractile activity was slightly depressed (A, Day 20). In other preparation, spontaneous activity ceased, and responses could be evoked to electrical stimulation (Ba, Day 20). Record Bc shows evoked responses when the muscle strip had been exposed to Krebs solution for 10 min and shows further shortening of the burst. Record Bb is a spontaneous burst discharge recorded on fast sweep in the Mg-free solution, and Bd is an evoked burst in normal Krebs solution. It should be noticed that the maximum amplitude of spike potential was not changed by the application of Mg ions. On Day 22 the effects of adding 1.2 mM Mg were essentially the same but stronger (C, D) than on Day 20.

Figure 9 illustrates the effects of $3 \times 10^{-4}$ U/ml oxytocin and $10^{-7}$ g/ml PGF$_{2\alpha}$ on the electrical activity obtained by bipolar recording. Records shown in A are
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from a Day 20 preparation, and B from Day 22. Record Aa shows spontaneous activity in the Mg-free Krebs solution, and Ab the activity in normal Krebs solution. When Mg was present, the duration of burst discharge became shorter, and in this particular record the direction of propagation became opposite. Ac and Ad are responses to oxytocin and PGF$_{2a}$ in normal Krebs solution, respectively. Nearly equal effects were caused by these doses of agents, i.e., the burst became longer, and the amplitude of compound action potentials larger. The direction of propagation was from ovarian to vaginal side in the presence of either agent. Records Ba–Bd, taken with the same experimental procedure, show similar findings for strips from Day 20. In the case of B, spontaneous activity was also generated in normal Krebs solution (Bb). With microelectrode method, the doses of oxytocin and PGF$_{2a}$ were found to induce depolarization of 3–8 and 4–10 mV on preparations of Days 20 and 22, respectively.

In a given muscle strip, 10–30 burst discharges were recorded in the presence of either 3 x 10^{-4} U/ml oxytocin or 10^{-7} g/ml PGF$_{2a}$ contained in the normal Krebs solution. The per cent of total propagations which travelled from the ovarian to vaginal side was determined for each agent. The value for PGF$_{2a}$ was plotted in the ordinate, and that for oxytocin in the abscissa for the same muscle strip (Fig. 10).

Fig. 8. Effects of 1.2 mM Mg application on the contractile and membrane activity in the longitudinal muscle of pregnant rat uteri. A and B were from Day 20 preparations, and C and D Day 22. At the time indicated by arrows, 1.2 mM Mg was applied. Filled triangles show external electrical stimulation (200 msec). Bb shows a control burst in Mg-free solution on fast sweep, and Bd the response in normal Krebs solution. About 10 min elapsed between Ba and Bc. Except for Bb and Bd, the sweep speed is the same throughout (shown in bottom).
Again, 0% means that all burst discharges were directed from the vaginal to ovarian side in a given muscle strip. PGF$_{2\alpha}$ (10$^{-7}$ g/ml) was incapable of inducing spontaneous activity in some muscle strips on Day 20 (bottom). Neither oxytocin nor PGF$_{2\alpha}$ caused a preferential direction of propagation in either Day 20 or Day 22. No close correlation of direction of propagation was noticed in the presence of oxytocin and PGF$_{2\alpha}$. Therefore, it can be concluded that all portions of the longitudinal muscle are equally sensitive to the agents.

Figure 11 shows the effects of oxytocin and PGF$_{2\alpha}$ on the bipolar recording of electrical activity (Day 22). The muscle strip was quiescent in Krebs solution. When 3 x 10$^{-4}$ U/ml oxytocin was applied, burst discharges occurred (a, b). Oxytocin at 10$^{-5}$ U/ml caused continuous spike activity (c), and the effects did not fade by longer exposure (d). The record on fast sweep indicates that the spike was either monophasic or biphasic. PGF$_{2\alpha}$ at 3 x 10$^{-5}$ g/ml initiated burst discharges (e). However they ceased spontaneously about 10 min after the beginning of drug application even in the continued presence of the agent (f).

Figure 12 shows experiments carried out on the same muscle strip as in Fig. 11. PGF$_{2\alpha}$ at 10$^{-7}$ g/ml generated spontaneous bursts (a, b), which ceased spontaneously 15 min after application of the agent (c). After the cessation of burst discharge in the continued presence of PGF$_{2\alpha}$, 10$^{-3}$ U/ml oxytocin was applied (d). Again, oxytocin caused initiation of burst discharges. However the effects were far weaker.
than when it was given alone (cf. Fig. 11, c, d). Burst discharges ceased when oxytocin was withdrawn (e), and they appeared again when oxytocin was readministered (f). This sort of observation shown in Figs. 11 and 12 was made on 3 other muscle strips. Therefore, to summarize, the muscle strips became desensitized to PGF$_{2a}$, but the effects of oxytocin persisted. In the muscle desensitized to PGF$_{2a}$, effects of oxytocin were no longer additive, but the responses were depressed.

DISCUSSION

Regarding the changes in morphological and physiological property of myometrial cells towards parturition, several important findings have been reported. Gap junctions are formed around the time of parturition in rats, guinea-pigs and sheeps, suggesting that cell-to-cell connections play a prerequisite role in the performance of parturition (GARFIELD et al., 1978, 1979). Oxytocin receptors have also been reported to increase (ALEXANDROVA and SOLOFF, 1980). Noradrenaline reversal occurs in the longitudinal muscle (MARSHALL, 1970) as well as in the circular muscle in rats (OSA and WATANABE, 1978; KISHIKAWA, 1981). The
The physiological role of these phenomena is hard to evaluate at present. The membrane becomes depolarized at the end of pregnancy (Casteels and Kuriyama, 1965; Kanda and Kuriyama, 1980; Kishikawa, 1981), and this was taken as an indication of progesterone withdrawal. In the present experiments on the longitudinal muscle of rats exposed to the Mg-free Krebs solution, the rest-

![Figure 11](image1)

**Fig. 11.** Effects of oxytocin and PGF$_{2\alpha}$ on the membrane activity of the longitudinal muscle recorded by rubber gap method. The records show the response to $3 \times 10^{-4}$ U/ml oxytocin (a, b), $10^{-8}$ U/ml oxytocin (c, d), and $3 \times 10^{-8}$ g/ml PGF$_{2\alpha}$ (e, f). The preparation was taken from a rat uterus on Day 22 of pregnancy. The muscle strip was quiescent in normal Krebs solution. In d, membrane activity is shown on a fast sweep (right).

![Figure 12](image2)

**Fig. 12.** Effects of oxytocin and PGF$_{2\alpha}$ on the membrane activity of the longitudinal muscle recorded by rubber gap method. The preparation was the same muscle strip as in Fig. 11 (Day 22). The application of $10^{-7}$ g/ml PGF$_{2\alpha}$ is shown in records a, b, and c. Records (d-f) are the continuation after record c. Oxytocin ($10^{-5}$ U/ml) was applied at the time indicated by the arrow (d) in addition to the prior presence of $10^{-7}$ g/ml PGF$_{2\alpha}$ throughout. Oxytocin was withdrawn (e) and readmitted (f), leaving $10^{-7}$ g/ml PGF$_{2\alpha}$ present.

The membrane becomes depolarized at the end of pregnancy (Casteels and Kuriyama, 1965; Kanda and Kuriyama, 1980; Kishikawa, 1981), and this was taken as an indication of progesterone withdrawal. In the present experiments on the longitudinal muscle of rats exposed to the Mg-free Krebs solution, the rest-
The electrical activity recorded extracellularly by the present method is far less quantitative than that by the sucrose gap method (Bennett and Burnstock, 1966), because the size of spike potentials is affected by the short-circuiting factor. However, it has some advantage in representing the membrane properties in terms of cell groups. It has been proposed that visceral smooth muscle tissues belong to a single-unit type and a group of cells composes a unit of propagation (Nagai and Prosser, 1963; Tomita, 1970). If it is assumed that the properties of each muscle cell become homogenous, and there is closer connection of cells (or cell groups) in the longitudinal as well as the transverse axis of the uterus, the whole uterus would exhibit a synchronized pattern of contraction. As a whole, although there were variations among individual rats, a synchronized pattern was remarkable on Day 22, and the amplitude of spike potentials recorded extracellularly was generally larger on Day 22 than on Day 20. A unit size of propagation of excitation has not been determined in uterine tissue. Csapo (1962) described in rabbit uterus muscle bundles that conjugate and branch from each other. Provided that a bundle conforms to a unit of excitation, closer connection between bundles would assure synchronization, thereby a uterus can undergo a propulsive movement as a whole at the end of pregnancy.

As in other laboratories (Fuchs, 1972; Porter and Downing, 1978; Anderson et al., 1981), parturition usually occurred in our Wistar-strain colony in the afternoon of Day 22 of pregnancy. The present experiments were not concerned with the uterus during the delivery. Nevertheless, the ultimate behavior of the uterus during parturition would be an extension of changes occurring from Day 20 to Day 22. It has been reported, from in vivo studies, that ovarian and distal end contracts independently and asynchronously on Day 21, and the uterine activity becomes synchronized on Day 22 (Fuchs, 1978). In other reports, the uterus in vivo becomes quiescent on Day 21, a period immediately before the parturition (Porter and Downing, 1978). In accordance with this, it was found in the present experiments, that the spontaneous activity of longitudinal muscle strips was more strongly depressed in the presence of 1.2 mM Mg, i.e. in normal Krebs solution, on Day 22 than Day 20 (Fig. 8). The composition of the circulating blood including hormonal components and the vascularization are far more complex than the artificial bathing solution, hence some discrepancy between in vivo and in vitro results would be expected.

How the contractile activities of the longitudinal and circular muscles participate in the performance of delivery, and whether the intrauterine pressure measurements represent the activity of the longitudinal or circular muscle layers are undetermined problems. It was also found in vitro that application of 1.2
mM Mg more strongly depressed the spontaneous activity of the circular muscle strip near term (unpublished observation). Taking the spontaneous quiescence in vivo and in vitro into consideration, it could be speculated that the muscle cells are ready for the synchronized activity of the whole uterus near term, but are kept quiescent by some means. Mg, relaxin, and/or catecholamines could be candidates for the stabilizing factor, although detailed mechanisms have to be investigated. The uterus becomes active when initiating factors of excitation exert their actions, either PGF₂α, oxytocin or others. As far as the present results indicate, the excitatory effect of PGF₂α was not long-lasting, a finding which is parallel to that found by in vivo studies (FUCHS, 1972). The present results, therefore, point to a physiological role of oxytocin for the performance of parturition in rats. It is puzzling that oxytocin became less effective when the uterus was desensitized to PGF₂α (Fig. 12).

The pacemaker activity was not localized, but appears to occur anywhere in the uterine longitudinal muscle strip in late pregnancy exposed to Mg-free Krebs solution, and to normal Krebs solution containing PGF₂α and oxytocin (Figs. 6 and 10). The assumption that prerequisite for the performance of delivery depends on stronger excitability and/or sensitivity to excitatory agents at the tubal end of the uterus deserves further investigation. The assumption might be illusive, but in order to find out if it is true, a more elaborate in vitro system would be required in future experiments.

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