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RESPONSE OF THE CAT'S UTERUS TO THE HORMONES OF THE POSTERIOR PITUITARY LOBE

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The specific changes in response to the oxytocic factor of the posterior pituitary lobe which take place in the rabbit's uterus under the influence of the ovarian hormones have not hitherto been shown to occur in any other species. Thus in the mouse, the response of the uterus to oxytocin is greatly increased by the administration of œstrin, but this cannot be counteracted by progestin which in the rabbit inhibits the response [Siegmund, 1930]. The guinea-pig's uterus is even more different from that of the rabbit since neither œstrone nor progesterone appear to alter appreciably its response to the oxytocic hormone when the measurements are made in the intact animal.

The changes in response to oxytocin which take place in the course of pregnancy, have been investigated quantitatively in the rabbit, the mouse, and the human. Again the rabbit is exceptional in showing two distinct phases, one of absence of response to oxytocin in the early stages of pregnancy, and one of increased response in the later stages of pregnancy and at parturition, whilst in the mouse and human there is a high sensitivity at parturition but no complete loss of sensitivity in the early stages of gestation.

In the present investigation we have studied the changes brought about in the activity of the uterus of normal and spayed cats injected with œstrone, progesterone and testosterone and of animals at various stages of pregnancy. Our chief aim has been to determine whether any changes in response to oxytocin take place during pregnancy and whether they can be correlated with the action of the ovarian hormones in the same way as in the rabbit [Knaus, 1930; Robson, 1933].

In each experiment we have investigated the effect of vasopressin and of adrenaline on the uterus as well as that of oxytocin. The effect of adrenaline on the cat's uterus affords a useful indication of the presence of luteal activity [Van Dyke & Gustavson, 1929]. The action of vasopressin is of interest because as Robson [1936] and Morgan [1937] have shown, it can inhibit the uterine muscle under certain conditions and act as an antagonist to oxytocin. The reaction of the uterus to these substances has been tested both *in vitro* and *in vivo*.

Methods

Experiments were performed in thirty cats. The non-pregnant animals were as a rule spayed by the dorsal route and injected for periods ranging from 1 to 2 weeks with œstrone (2–5 times daily with 0·01 mg. crystalline œstrone in solution in oil), or with œstrone followed by progesterone. Progesterone was given over a period of 4 days, the total dose being 6 mg. Daily doses of 2·0 mg. of testosterone were given over a period of 4–5 days, but this was not preceded by the injection of œstrone.

The experiments were done under chloralose anæsthesia. The animals were eviscerated (the stomach, intestine and spleen being removed), and one horn of the uterus was removed and cut in two, and the two strips were suspended in oxygenated Ringer-Locke solution in 100 c.c. containers for duplicate *in vitro* determinations. The contractions of the longitudinal muscle of the other horn were registered by means of a Cushny myocardiograph, which made it possible to obtain a record of the uterine movements without interfering with the natural position of the organ. The apparatus consists of a fixed and a movable arm, the movements of the latter being transmitted by a thread to a horizontal lever writing on the kymograph. The two lever arms were connected to the muscle of the uterine horn at a distance of 3–4 cm. apart. The abdominal cavity was closed by a celluloid screen which had a slit through which the recording instrument passed.

In a few experiments on pregnant animals the contractions of the circular muscle were registered by means of a method similar to that previously used by one of the authors [Robson, 1936].

We are indebted for the purified samples of oxytocin and vasopressin to Dr White of Parke, Davis and Co. Messrs Schering and Co. kindly supplied the progesterone (Proluton) and Ciba Ltd. the testosterone.

Results

All the results are summarized in Table I. Several points require more detailed description.

The spontaneous activity of the cat's uterus *in situ* varies greatly in different animals. Three types of activity can be clearly distinguished *in vivo*. These may be described as (i) the "irregular type", consisting of comparatively small and irregular movements; (ii) the "frequent" type in which there is a succession of rapid contractions and the relaxation is less complete than in (iii) the "intermittent" type in which the contractions are of large amplitude and are separated by relatively long intervals of rest. This type of activity corresponds with that described by Newton [1933] in pregnant guinea-pigs. The three types of spontaneous activity are illustrated in Fig. 1.

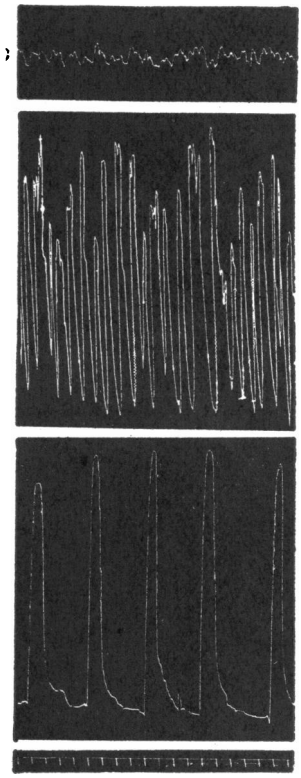

Fig. 1. Illustrating the three types of spontaneous activity observed *in vivo*. Upper tracing: irregular type. Spayed cat (C25). Middle tracing: frequent type. Spayed cat treated with œstrone and progesterone (C24). Lower tracing: intermittent type. Parturient cat (C14). Time intervals = min.

The "irregular" type of contraction is observed when the ovaries are not actively functional. It occurs in the spayed cats and in normal animals in the absence of either œstrus or pregnancy. The "frequent" type of contraction is observed in the uterus of animals treated with the ovarian hormones, œstrone, or œstrone followed by progesterone. It was also observed in one cat injected with transdehydroandrosterone, and in animals in the earlier stages of pregnancy. Lastly the slow "intermittent" type is found in the later stages of pregnancy, at parturition, and in the early puerperium.

There is thus a correlation between the type of contraction observed *in situ* and the functional state of the uterus. The *in vitro* experiments on the other hand show no such correlation. The shape and amplitude of the contractions observed *in vitro* have not the same characteristic differences which occur in the uterus *in situ*. The fre-

TABLE I. Showing the responses and spontaneous activities of the uteri of the experimental animals. Figures in brackets refer to response of circular muscle; all other figures to longitudinal muscle. Spontaneous activity: I = irregular; N = intermittent; F = frequent. Figures in italics denote inhibition. I.U. = international unit.

Condition	Injections	Response to oxytocin: *In vitro* I.U.	*In vitro* Av.	*In vivo* I.U.	*In vivo* Av.	Response to vasopressin: *In vitro* I.U.	*In vivo* I.U.	Response to adrenaline: *In vitro* γ	*In vivo* γ	Spontaneous activity: *In vitro*	*In vivo*
Spayed 15 days	Nil	0·05		0·5		>0·4	>0·5	>20	*30*	+ +	I
Spayed 14 days	,,	0·1	0·06	0·1	0·23	>0·5	>0·5	*20*	*10*	+ +	I
Dioestrous	,,	0·02		0·1		0·3	*0·3*	*10*	*20*	+	I
,,	Œstrone	>0·2		>0·1		>1·0	—	*100*	—	+ +	F
,,	,,	1·0		1·0		>1·0	*0·5*	—	<*50*	+	F
,,	,,	>1·0	< 0·9	0·2	0·27	>1·0	*0·02*	—	*10*	+ +	F
,,	,,	1·0		0·1		—	*0·1*	*100*	—	0	F
Spayed	,,	>1·0		0·2		>0·6	<*0·1*	—	*3*	0	F
,,	,,	>1·0		0·05		>1·0	*0·2*	—	*10*	+ +	F
Pseudo-pregnant	,,	0·02		0·02		0·1	—	*20*	*10*	0	F
Spayed	Œstrone and progesterone	0·05		—		>0·6	—	10	10	+	F
,,	,,	0·03		<0·05		0·1	>0·2	10	10	+ +	IF
,,	,,	0·1	0·06	0·1	0·06	>0·5	*0·5*	10	—	0	F
,,	,,	0·05		0·03		>0·6	*0·1*	10	30	0	F
,,	Testosterone	0·01		0·05		—	*0·1*	10	10	0	F
,,	,,	0·005	0·008	0·1		—	0·1	*10*	*10*	+ +	I
,,	,,	0·01		—		—	—	*10*	—	+ +	I
,,	Transdehydroandrosterone	0·01		0·05		—	—	*10*	*10*	0	F
Pregnant:											
Fœtuses = 4 mm.	Nil	>1·0		0·05		—	*0·1*	10	10	0	F
,, = 8 mm.	,,	0·1		0·05		0·5	>0·2	5	10	0	F
,, = 18 mm.	,,	0·2 (0·2)		0·2		—	—	20	—	+ +	F
,, = 20 mm.	,,		—	<0·1		—	*0·1*	—	10	+	F
,, = 10 cm.	,,	0·02		—		—	—	30	—	+ +	FN
,, = 13 cm.	,,	0·01		0·05		—	—	5	10–*10*	0	N
Parturient	,,	0·02		0·01 (0·01)		>0·1	>0·2	20	*10*	0	N
,,	,,	0·1	0·4	0·02 (0·01)	0·013	—	>0·1	10	*10*	0	N
Day after parturition	,,	0·005		(0·005) 0·01		>0·05	>0·05	20	*20*	0	N
Puerperium, 1st week	,,	0·05		<0·01		>0·6	>0·1	50	*10*	0	N
,, 13 days	,,	0·1		0·05		>0·6	*0·05*	>40	*10*	0	FN
,, 20 days	,,	0·01		0·02		>0·2	—	>10	*10*	0	I

quency of the contractions has therefore been taken as the sole measure of spontaneous activity *in vitro*. In all the groups of animals included in Table I totally quiescent uteri were occasionally found. No correlation could be detected between the frequency of movements *in vivo* and *in vitro*. Thus in the spayed animals small irregular movements were seen *in vivo* when large and frequent spontaneous contractions occurred *in vitro*, whilst in the œstrone and in the progesterone treated animals an intensive spontaneous contractility *in vivo* was frequently associated with complete quiescence *in vitro*.

In conclusion it may be stated that the contractions observed *in vivo* give a good indication of the functional state of the uterus, whilst the spontaneous contractions in the isolated organ vary in an apparently arbitrary manner.

Response to oxytocin. The average minimal effective doses of oxytocin in the various groups of animals are shown in Table I. In cases where no effect was obtained the maximal ineffective dose was used in the calculation of the averages.

The response of the uterus to oxytocin increases during pregnancy. Both *in vivo* and *in vitro* there is a maximum sensitivity towards the end of term and in the early puerperium.

Striking results were obtained *in vitro* in the hormone-treated animals. The uteri of œstrone-treated cats were practically insensitive to oxytocin *in vitro*, whilst after testosterone the response *in vitro* was of the same order as that obtained in parturient animals. The average difference in response between the œstrone and the testosterone groups is approximately a hundredfold.

No consistent differences in response were obtained *in vivo* between cats injected with œstrone, with œstrone followed by progesterone or with testosterone. Progesterone did not diminish the response to oxytocin either *in vivo* or *in vitro*.

Response to vasopressin. The characteristic action of vasopressin in cats injected with œstrone or œstrone and progesterone and in animals in the early stages of pregnancy consists of a prolonged inhibition of spontaneous movements and tone. This effect is, however, only obtained in the intact animal and in no experiment did vasopressin inhibit the uterus *in vitro*. The inhibitory effect of vasopressin on the uterus *in situ* persists after suprarenalectomy. Illustrations of the effects of vasopressin *in vivo* are shown in the next paper.

No vasopressin inhibition was obtained in the spayed animals and cats in the later stages of pregnancy. Inhibition by vasopressin seemed to be associated with the "frequent" type of contractions.

Occasionally vasopressin caused a small contraction of the uterus *in vitro*, and we are not satisfied that this effect may not have been due to some oxytocic substance present even in the purified samples of pitressin.

Response to adrenaline. Adrenaline caused a contraction of the uterus *in situ* in the progesterone-treated cats and during pregnancy. In one animal investigated during the latest stages of pregnancy a short contraction was followed by a prolonged inhibition, whilst during parturition and the puerperium adrenaline always caused a pure inhibition *in vivo*. Inhibition was also obtained in the spayed animals and marked inhibition was found in the œstrone-treated cats.

The results with testosterone were contradictory. In one of the spayed cats injected with testosterone (C31) there was a clear adrenaline reversal (contraction of the uterus), whilst in the other cases adrenaline inhibited the uterus. The histological examination of the endometrium revealed a progestational reaction in the uterus where the adrenaline reversal occurred (the animal had been castrated 4 days before the injections of testosterone were begun, and 6 days before the experiment). In the other two cats there was no progestational reaction of the endometrium.

The response to adrenaline *in vitro* agreed on the whole with the *in vivo* effects. The chief difference was seen in parturient animals in which a slight motor effect of adrenaline usually persisted *in vitro* but not *in vivo*.

Discussion

The uterus of the cat, like that of other species investigated, becomes more reactive to oxytocin in the course of pregnancy. It differs, however, from the uterus of both the rabbit and the mouse in revealing no increased response to oxytocin after treatment with œstrone. It further differs from the rabbit's uterus and is similar to that of the mouse, the guinea-pig and the dog in showing no loss of response to oxytocin after treatment with progesterone. It is improbable therefore that either œstrone or progesterone are solely responsible for the changes in response which take place in the cat's uterus during pregnancy. Testosterone causes sensitization of the uterine muscle to oxytocin, and it is of interest to note that Parkes [1937] has extracted a substance with androgenic properties from ovarian material.

The possibility that the alterations in the reactivity of the uterus which are obtained under various functional conditions may, in different species, be produced by different hormones has to be considered. Thus in the rabbit the sensitivity to oxytocin *in vitro* is increased by œstrone and decreased by testosterone, whilst in the cat the opposite is true.

There is no evidence at present that vasopressin plays any physiological role in the control of the uterine activity in pregnancy and parturition. Such a possibility cannot, however, be excluded, especially since, according to Starling & Verney [1925] the anti-diuretic principle, which appears to be identical with vasopressin [Fraser, 1937] is constantly secreted into the blood. The inhibition of uterine activity by vasopressin observed in the cat's uterus in the earlier stages of gestation, and also demonstrable in the rabbit's uterus under the influence of œstrone and progesterone [Robson, 1936], may possibly be a factor in maintaining the quiescence of the uterine muscle during pregnancy. The uterine quiescence which, as Reynolds [1932] has shown, follows the injection of gonadotropic hormone in the rabbit even in the absence of luteal function may also possibly be due to a vasopressin inhibition.

A direct comparison of effective doses in experiments *in vitro* and *in vivo* should be permissible, since drugs were added *in vitro* to 100 c.c. containers and injected intravenously into a blood volume of 100–150 c.c. Table I shows that in several instances the response of the uterus *in situ* differs from that of the organ suspended in Ringer-Locke solution. Thus, in cats treated with œstrone the response of the isolated uterus to oxytocin was greatly reduced as compared with that of the spayed animals, whilst the response of the organ *in situ* was not appreciably altered. A similar effect *in vitro* has previously been obtained in the bitch [Robson & Henderson, 1936].

These results in the cat and in the bitch stand in contradiction to the sensitizing effects produced by œstrin *in vitro* in certain other species, e.g. the rabbit and the mouse. The responses to vasopressin *in vitro* and *in vivo* will be discussed more fully in the next paper.

The discrepancies between the *in vivo* and *in vitro* results obtained in the present experiments suggest that in certain species it is necessary to investigate the spontaneous activity and the responses of the uterus in the intact animal in order to assess in a satisfactory manner the effects of hormones on the uterus.

SUMMARY

1. The action of oxytocin, vasopressin and adrenaline on non-pregnant, normal and spayed cats injected with œstrone, progesterone and testosterone and on pregnant, parturient and puerperal cats has been investigated.

2. Three types of spontaneous movements *in vivo* are described, associated with spaying, the action of the ovarian hormones and the later stages of pregnancy respectively.

3. The response to oxytocin of the uterus examined both *in vivo* and *in vitro* increases during pregnancy and is at a maximum at parturition and during the early puerperium.

4. Œstrone causes no increase in response and progesterone no decrease in response to oxytocin.

Œstrin decreases and testosterone increases the response to oxytocin *in vitro*.

5. Vasopressin inhibits the uterus of cats treated with the ovarian hormones *in situ*, but it has no effect on the uterus *in vitro*. Vasopressin produced no inhibition in spayed animals or in the later stages of pregnancy.

6. Adrenaline contracts the uterus of the progesterone treated, and of the pregnant cat. It relaxes the uterus of the spayed and œstrone-treated animal.

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