EFFECTS OF CATECHOLAMINES ON THE SMOOTH MUSCLE OF THE FEMALE REPRODUCTIVE TRACT[1]

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This review focuses primarily on certain aspects of the actions of the adrenergic amines epinephrine and norepinephrine and of isoproterenol on the smooth muscle of the uterus, oviduct, and ovary. The adrenergic innervation of this muscle is mentioned briefly as background for the consideration of the effects of adrenergic nerve stimulation. The literature citations are selective, not encyclopedic, and cover work published primarily during the past ten years. More extensive analyses of the pharmacology of the myometrium can be found in numerous excellent reviews (1–6).

MYOMETRIUM

Most uteri contain both alpha (excitatory) and beta (inhibitory) adrenoceptors, and the direction of the response of the myometrium to catecholamines depends upon the specific amine and on the relative dominance of one or the other of the adrenoceptors. This relative dominance is apparently under hormonal control and may vary from species to species. For example, in the rabbit and rat norepinephrine excites an estrogen-dominated uterus but relaxes a progesterone-dominated one, while just the opposite occurs in the cat (8–12). Although the mechanisms underlying these hormonal effects on the adrenoceptors within the muscle are unclear, the effects are of interest because they probably represent the only examples of endocrinological regulation of adrenoceptors in autonomic effector cells.

Studies on the actions of catecholamines on the uterus have been mainly concerned with effects on the excitability and contractility and with explanations of these effects in terms of ionic and metabolic changes within the muscle.

Alpha-excitatory effects.—Stimulation of alpha-receptors by epinephrine and norepinephrine results in a transient depolarization of the cell membrane which initiates action potentials in quiescent muscles or increases their frequency in spontaneously active ones. As a result, the force and frequency of contractions are elevated. In higher doses these amines produce an initial burst of action potentials followed by a sustained depolarization and contracture (11, 13, 14).

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The ionic mechanisms underlying these excitatory actions are not precisely known, although it is assumed that an increase in the membrane permeability to one or several ion species might occur (15). An increase in sodium permeability with a resultant influx of sodium across the cell membrane would seem the most logical explanation. Such an increase has never been convincingly demonstrated because measurements of sodium flux in smooth muscle are complicated by the extensive binding of this ion to extracellular sites and by the rapidity of its exchange between extracellular and cellular compartments (16). A delayed (about 30 min), transient increase in potassium efflux has been observed during the stimulatory action of norepinephrine in the pregnant rat myometrium, but this could hardly be related to the membrane depolarization and contractions which begin within seconds after the drug is administered (11, 16).

A promising approach to the study of the ionic mechanisms of drug action involves the measurement of changes in membrane conductance during the drug effects in solutions of different ionic compositions. This can be done by recording the amplitude and time-course of electrotonic potentials produced by externally applied pulses of constant current (14, 17–19). Because small segments of myometrium exhibit cable properties when stimulated electrically under appropriate conditions (14), changes in the electrotonic potentials during drug action reflect alterations in membrane conductance, assuming that the longitudinal resistance of the tissue remains constant (17). Using this technique, Szurszewski & Bülbring (19) have shown in the guinea-pig uterus that the membrane depolarization caused by norepinephrine is due to an increase in chloride conductance, and that caused by acetylcholine to an increase in sodium conductance. However, the increase in action potential frequency produced by both agents is sodium-dependent. These results substantiate an earlier suggestion (20) that an increase in chloride conductance is responsible for the alpha-excitatory actions of catecholamines in the pregnant cat uterus. Whether an increase in chloride conductance is universally responsible for alpha effects on the excitability of the myometrium remains to be determined. With the availability of techniques for measuring electrotonic potentials and for voltage clamping the myometrium (21), it is hoped that the ionic mechanisms subserving the membrane potential changes caused by the catecholamines in a variety of species will soon be clarified.

A normally polarized membrane is not an absolute prerequisite for the actions of epinephrine and norepinephrine. Muscles depolarized by exposure to isotonic potassium solutions still contract in response to these amines, although no change in membrane potential occurs (22, 23). These effects are prevented by alpha blockers and indicate sites of action somewhere beyond the electrically polarized membrane.

Calcium is absolutely essential for the contractile response to the catecholamines in both normally polarized and depolarized uteri (4, 20, 22). In the absence of extracellular calcium, contraction is abolished, and in the presence of a limited increase in extracellular calcium it is augmented.

It is generally agreed that the "final common event" in the action of all smooth muscle stimulants involves the release of calcium ions into the cytoplasm

with the resultant activation of the contractile elements (4). By analogy with skeletal muscle, activation of the contractile elements begins when free intracellular Ca^{2+} rises above 10^{-7}M and reaches a maximum at 10^{-6}M. The amount of calcium required for maximal activation of the contractile proteins is 1.3μ mole/g actomyosin and an ionic calcium concentration of 10^{-6}M. (24). The pregnant myometrium needs about 8mμ moles of calcium per gram of muscle for complete activation of its contractile proteins, assuming an average actomyosin content of around 6mg/g myometrium (25, 26). Activator calcium may come from extracellular sources, from intracellular storage sites, e.g., sarcoplasmic reticulum, mitochondria, inner surface of sarcolemma, or from all three (4, 27). Depolarization of the cell membrane presumably increases calcium permeability. Hence drugs which depolarize the membrane may also enhance calcium influx (4). Experimental proof of this idea has been difficult to obtain, since such a small net gain of calcium is needed for maximal activation of the contractile elements. It is hard to measure accurately this small increase in tissue calcium content because more than 90% of the total Ca exchange occurs in the extracellular space. The exchange of calcium in the extracellular compartment therefore obscures the much smaller flux of this ion across the cell membrane (28).

Recently a new method has been developed for measuring calcium uptake and exchange in smooth muscle (29). This method uses lanthanum to distinguish membrane calcium flux from total calcium exchange. The trivalent lanthanum ion has a higher affinity than calcium for tissue binding sites and apparently does not enter the cell to an appreciable extent. Thus lanthanum displaces extracellular calcium and blocks calcium movement across the cell membrane, effectively "locking in" cellular calcium.

With this technique van Breemen (29) found, in vascular muscle, that tissue calcium content and uptake of Ca^{45} were significantly elevated during a K-induced contraction but not during one caused by norepinephrine. Furthermore, if lanthanum was given before the muscle was exposed to the potassium solution (127mM) the K-contraction was abolished while the response to norepinephrine persisted for one contraction. These results are in substantial agreement with earlier suggestions (5) that in vascular smooth muscle norepinephrine at least during its initial action releases calcium from intracellular stores. During subsequent contractions these stores are probably replenished by calcium from extracellular sources. A similar situation might exist in the myometrium in view of the finding that epinephrine-induced contractions of the rabbit uterus in calcium-free solutions declined too slowly to be accounted for solely on the basis of extracellular calcium mobilization (22).

One storage site for intracellular calcium may be the sarcoplasmic reticulum. The reticulum is sparse in the myometrium and therefore its physiological importance has been questioned (30). Elements of the reticulum have recently been isolated from the cow myometrium, and although their calcium binding ability is lower than that of striated muscle it may be sufficient to play a significant role in calcium release and sequestration during contraction and relaxation of the uterus (27).

Beta-inhibitory effects.—Inhibition is characterized by cessation of spontaneous action potentials followed by muscle relaxation and a gradual hyperpolarization of the cell membrane (11, 31). The reduction in pacemaker discharge is characteristic of a number of myometrial relaxants but membrane hyperpolarization is not (32). Unlike inhibition in the intestine where the suppression of action potentials is a beta and hyperpolarization an alpha effect (33, 34), both actions in the myometrium are mediated by beta receptors (35).

These effects could result from alterations in passive ion permeabilities, stimulation of action ion pumps, changes in calcium movements across the cell membranes, or from combinations of all three (15, 16, 36). In the rat myometrium, the magnitude of the hyperpolarization caused by epinephrine and isoproterenol varies with $[K^+]_o$ in a manner suggesting an increase in potassium permeability (36). However, ion flux data suggest that a decrease in sodium permeability may also play a role (16). Electrogenic sodium pumping seems an attractive candidate for mediating hyperpolarization because an electrogenic sodium pump can be demonstrated under special circumstances in the rat myometrium (37). However the hyperpolarization is unchanged in K-free, Na-free, or Cl-free media (38), conditions that usually abolish electrogenic sodium pumping in most excitable cells (39, 40). Therefore the contribution of such a pump to the beta inhibitory actions in the myometrium seems unlikely. Ouabain (10^{-3}M) and low temperature (10°C) reduce the hyperpolarization by 50%, however, indicating that some metabolic process is involved (38). An electrogenic calcium pump has been suggested since the hyperpolarization caused by isoproterenol in the rat myometrium is abolished in Ca-deficient or lanthanum-containing solutions (lanthanum is believed to block Ca-flux across the cell membrane, p. 21), and reduced in elevated $[Ca^{2+}]_o$ (38).

Beta-mediated relaxation is also reduced by elevated $[Ca^{2+}]_o$ in spontaneously contracting as well as electrically stimulated muscles (32, 41). These effects are seen in both normally polarized and K-depolarized muscles (32, 41–43]. Thus the removal of calcium from the contractile elements is probably the final common mediator of the inhibitory as well as the excitatory actions of the catecholamines on the uterus.

Relaxation occurs when intracellular Ca^{2+} is reduced below 10^{-7}M as a result of inhibition of calcium influx, re-uptake into intracellular stores, active extrusion across the cell membrane, or a combination of all three processes (24). Unlike the local anesthetics, which prevent myometrial contractions by blocking calcium influx, epinephrine and isoproterenol have little effect on influx (41, 43). Stimulation of re-uptake of calcium into intracellular storage sites has been suggested as a mechanism for the action of isoproterenol (42). However, recent experiments with the "lanthanum trapping" technique (p. 21) suggest that extrusion of calcium across the cell membrane might be the principle action of this catecholamine (38). Extrusion is energy dependent because the electrochemical gradient for calcium ions across the cell membrane is overwhelmingly inward (44). Hence the inhibitory effects of the catecholamines involve an active, metabolic process.

Relation to tissue cAMP.—A prominent metabolic component of the effects of beta adrenergic agents in smooth muscle is an increase in tissue levels of cyclic 3′, 5′-adenosine monophosphate, cyclic AMP (45). Cyclic AMP and the enzymes controlling its intracellular concentration may participate in the regulation of uterine motility (46). The concentration of cAMP at any given moment is a balance between the amount synthetized by adenyl cyclase and that degraded by 3′, 5′-cyclic mononucleotide phosphodiesterase (45, 47). Epinephrine, norepinephrine, and isoproterenol all stimulate adenyl cyclase activity and thus increase cAMP in the myometrium. The order of potency for this action in the rat is isoproterenol > epinephrine > norepinephrine, and is the same as that for relaxation (46, 48–50). There is a dose-response relationship between the increase in cAMP or relaxation and the concentration of catecholamine (46, 48). The increase in cAMP and relaxation are both potentiated by inhibition of phosphodiesterase (50, 51) and prevented by beta blockers (46–51). The dibutyryl derivative of cAMP mimics the relaxant effect of catecholamines (48, 51). Thus most of the criteria for establishing cAMP as the "second messenger" for the effects of beta-adreno-ceptor stimulation in the myometrium have been satisfied at least in the rat, the species most extensively studied (47).

The increase in tissue cAMP produced by the stimulation of beta receptors usually correlates temporally with relaxation (38, 46, 48). Papaverine, an inhibitor of phosphodiesterase (52), also elevates tissue cAMP, the increase paralleling the relaxant action of this agent on the rat uterus (38). One group of workers could not find a correlation between tissue cAMP concentration and relaxation (53, 54). However, their observation that in epinephrine-treated uteri the addition of propranolol restored contractions before cAMP had decreased to control levels may be explained by a competition between the relaxant effects of cAMP and the alpha-stimulatory actions of epinephrine.

What is the link between cAMP and relaxation? Perhaps the increase in cAMP initiates a series of cellular reactions that provides the energy for calcium extrusion and thus the removal of calcium from the contractile proteins. Papaverine, dibutyryl cAMP, and isoproterenol, in addition to increasing tissue cAMP concentration, also reduce tissue calcium content in the rat uterus (38). Cyclic AMP is known to affect calcium movements in bone and liver (49) and to stimulate a calcium pump in heart muscle (55).

Of course cAMP is not the exclusive regulator of uterine motility. For example the relaxant effects of norepinephrine on the rat uterus are potentiated by nitroglycerine, which has no effect on tissue cAMP, as well as by theophylline, an inhibitor of phosphodiesterase (56). Furthermore, D-600, a derivative of verapamil, a nonadrenergic relaxant (57), does not alter uterine cAMP nor calcium content (as measured with the lanthanum technique) (38). Since lanthanum cannot delineate intracellular calcium distribution, these results imply that D-600 acts primarily by stimulating reuptake of calcium into tissue stores.

Although it may seem logical for alpha stimulatory amines to reduce tissue cAMP levels, they have no effect on this nucleotide (53). Oxytocin acetylcholine,

and prostaglandin $F_{2\alpha}$ also do not influence cAMP levels yet they are potent myometrial stimulants (46, 48).

Because at least three processes regulate $[Ca^{2+}]i$ (uptake from extracellular sources, exchange with intracellular binding sites, extrusion across the cell membrane), it would be unrealistic to think that all drugs influencing uterine motility would act on the same process. However, it is tempting to speculate that the beta-effects of the catecholamines are mediated via an increase in cAMP, which in turn initiates the energetic reactions needed for the extrusion of calcium. The recent suggestion of Triner et al (59) that the physiological antagonism between acetylcholine and beta-adrenergic agents in the uterus may be mediated through an interaction at the level of cAMP deserves serious consideration.

Cyclic AMP may also be an intermediary in the action of estrogens on the uterus. Cyclic AMP and GMP stimulate protein synthesis in isolated uterine segments in a manner similar to that caused in vivo by the administration of estradiol-17-beta. These actions are common to a number of nucleotides, however (60). More pertinent is the finding that the uterine content of cAMP is markedly reduced in ovariectomized rats and promptly restored by giving estrogens (61, 62). This latter effect is potentiated by theophylline and prevented by propranolol but not by adrenalectomy (61–63), implicating an adrenergic ingredient in the action of estrogen, possibly the release of catecholamines from cellular or subcellular binding sites. However, an early (within 5 min) increase in cAMP concentration in the chick oviduct after estrogen administration was not an absolute requirement for the estrogen-stimulated protein synthesis in this tissue (64). Progesterone caused a delayed (3 hr) and progressive (up to 24 hr) increase in tissue cAMP concentration (64). The relation of this observation to the mechanism of action of progesterone is not clear. A systematic study of uterine cAMP concentration at different times in the estrous cycle or pregnancy may help to clarify the relation between the ovarian hormones and uterine cAMP.

Hormonal influences on uterine adrenoceptors.—Since the original observation of "pregnancy reversal" in the cat by Dale in 1906 (65), a number of speculations have been advanced to explain the influence of the ovarian hormones, estrogen and progesterone, on the uterine response to catecholamines. Although none of these provides a satisfactory answer, three recent contributions will be mentioned.

Uteri from nonpregnant or estrogen-dominated cats have a predominance of beta receptors while those from pregnant or progesterone-treated animals have mostly alphas (66). An aqueous extract of a pregnant or progesterone-proliferated uterus can change the normal inhibitory response of the estrogen-dominated uterus to a contraction (9, 67). Furthermore, if a uterine segment from a progesterone-treated or pregnant cat is suspended in the same organ bath with that from an estrogen-treated animal, there is a reversal of the response of the estrogenized muscle to epinephrine and norepinephrine. This effect is prevented by alpha-blockers (9). These findings suggest that something is present in the pregnant or progesterone-proliferated uterus that can be extracted by water and that

can diffuse in an isolated organ bath. The substance does not behave like progesterone or oxytocin (67), but as yet it has not been identified chemically.

A similar series of experiments has recently been done on the rat uterus (68). In this species, the uterus from an animal at the end of pregnancy or during estrous is stimulated by norepinephrine and epinephrine, while the mid-term or diestrous uterus is inhibited. When an aliquot of the medium bathing a strip of pregnant myometrium stimulated by epinephrine was applied to a diestrous strip, the muscle contracted. This "motor substance" is believed to be prostaglandin E_2, and its liberation has been implicated in the "adrenaline reversal" (68). Unfortunately, the influence of alpha blockers on the response of the uterus to PGE_2 was not tested. This should be checked, because the excitatory effects of norepinephrine and epinephrine on the rat myometrium are prevented by alpha blockers (11). If PGE_2 is involved in "adrenaline reversal" it would be interesting to measure the uterine content of PGEs at different times in pregnancy and during the estrous cycle.

Perhaps the most attractive theory proposed for the reversal phenomena suggests that the adrenoceptive sites regulate the ionic permeability and/or the ion gradients across the myometrial cell and in this way control the direction of the response to catecholamines. Evidence for this idea has been presented for the cat uterus where the chloride content of the pregnant uterus is nearly twice that of the virgin (20). Thus in the pregnant uterus the chloride equilibrium potential is well below the membrane potential of the myometrial cell. Under these conditions, if epinephrine selectively increased chloride permeability it would depolarize the cell and have an excitatory effect. On the other hand if epinephrine produced a selective increase in potassium permeability, which apparently it does in the virgin uterus, the membrane would hyperpolarize with resultant inhibition (20). In the rat myometrium, although there are no changes in ionic gradients during pregnancy, potassium permeability is highest and sodium permeability lowest at mid-term while at term the sodium permeability rises significantly (69). If the beta inhibitory effects of catecholamines are related to an increase in potassium permeability (p. 9) they would be more pronounced at mid-pregnancy, which is indeed the case (11).

Adrenergic innervation; *effects of nerve stimulation.*—Information about the adrenergic innervation of the female reproductive tract has been greatly advanced within recent years by the application of histochemical, biochemical, and morphological techniques to studies on the musculature of this tract. More detailed accounts of the findings appear elsewhere (66, 70, 71). Only major points will be summarized here.

The adrenergic neurons innervating the myometrium arise from ganglia located in or near the uterus. Therefore the short, post-ganglionic fibers coming from these ganglia do not degenerate when the preganglionic (hypogastric) nerves are cut or the spinal cord is sectioned. Hence some degree of local adrenergic activity remains after section of the extrinsic nerves or spinal cord. The density of

innervation varies from species to species as well as within different regions of the myometrium in any one species. The adrenergic innervation of the cat uterus is greater than that of the rabbit, guinea pig, or human, while in the rat the innervation is limited almost entirely to the vasculature. The only species examined thus far that shows regional variations in the density of uterine innervation is the human, where the density is highest in the cervix and lowest in the corpus. All of these morphological findings correlate well with biochemical determinations of tissue norepinephrine, the principal component of the adrenergic nerves.

The norepinephrine content of the nerves, as well as the density of their distribution, is altered during pregnancy or after the administration of estrogen and progesterone. In the guinea pig and rabbit, the only species studied so far, there is little change in the transmitter content or nerve distribution in the myometrium during the first part of gestation. The innervation patterns at this time in pregnancy resemble those in estrous animals or in spayed animals given estrogen. After midpregnancy there is a sharp decline both in distribution and transmitter content so that by the end of gestation scarcely any nerves are visible within the myometrium. This reduction is mimicked by the administration of progesterone or by the selection of animals in metestrus (70, 71). Thus the amount and distribution of adrenergic transmitter liberated during nerve activity depends upon the gravid or hormonal state of the animal.

Several preparations of hypogastric nerve-uterine muscle have been devised, and the effects of nerve stimulation can be studied both in vivo and in vitro (72–76). No work on the effects of stimulation of the uterine nerves (postganglionic, "short" adrenergic) has been reported, although the electrical activity in these nerves has been recorded (77).

Electrical stimulation of the hypogastric nerves causes the uterus of the estrogen-dominated rabbit to contract and that of the progesterone-dominated rabbit to relax (73, 74). These actions are mimicked by norepinephrine, prevented by alpha and beta blockers respectively, and are accompanied by changes in the membrane potentials of the myometrial cells. In estrogen-dominated muscles, nerve stimulation depolarizes the membrane and initiates spike discharge, while in progesterone-dominated muscles the membrane is hyperpolarized (78). Junction potentials, the localized changes in membrane potential characteristic of neuro-effector transmission in certain parts of the intestine and in the male reproductive tract (1), are not seen in the rabbit myometrium. Since the innervation density is so much less in the myometrium than in the intestine or vas deferens (1, 70), the number of myometrial cells in close proximity to nerve terminals is undoubtly quite small. Probably certain "key" cells are directly activated by transmitter and then excitation (or inhibition) spreads from cell to cell over the myometrium via myogenic conduction, because the uterine muscle behaves like a physiological syncytium (14). As a result the uterus could give a full sized contraction in response to the localized stimulation of a few "key" cells by the neurotransmitter.

In order to elicit a clear cut response of the uterus (either excitation or inhibition) it is often necessary to stimulate the hypogastric nerve at frequencies be-

tween 15 and 50 pulses per second (72, 74, 76), a range above the physiological frequency of the uterine nerves (77). When the hypogastrics are stimulated at frequencies below 5 p/sec no overt response of the uterus usually occurs (72, 74, 76). However, in the guinea pig, hypogastric stimulation between 1 and 5 p/sec markedly increases uterine sensitivity to oxytocin (76). The potentiation is most striking in estrous animals and is absent in the parturient uterus (79). These findings correlate well with the results of Sjoberg and colleagues (70, 71) who showed an increase in innervation density and catecholamine content in the estrous myometrium but a marked decrease at the end of pregnancy.

OVIDUCT

With the current interest in reproductive biology the "era of indifference to the fallopian tube has passed" (page v. in 80). and excellent reviews of the anatomy, physiology, pharmacology, and pathology of this region of the female reproductive tract are now available (81–82).

The smooth muscle of the oviduct contains both alpha and beta adrenoceptors prompting the suggestion that adrenergic influences play a role in the transport of ova and sperm and in fertilization (66). The effects of catecholamines have been investigated most frequently in the rabbit and human, where epinephrine and norepinephrine stimulate and isoproterenol relaxes the smooth muscle of the oviduct both in vivo and in vitro. These actions are prevented or reversed by the appropriate adrenoceptive blocking agents (81, 83–88). Earlier reports that the response to epinephrine varied in the different parts of the tube and with the ovarian cycle (66), have been partially substantiated. In a careful, quantitative evaluation Levy & Lindner (89) found no difference in the response of the ampulla or isthmus of the rabbit oviduct to either isoproterenol or phenylephrine, both agents causing relaxation and contraction respectively and equally at either end of the oviduct. On the other hand, the isthmic region of the fallopian tube is said to be more sensitive to catecholamines than the ampullary region in the nonpregnant woman. This regional difference disappears during pregnancy when all parts of the tube are equally sensitive to catecholamines (84). The stimulatory effects of epinephrine and norepinephrine are also reduced during the luteal phase of the menstrual cycle in the human female (86, 90). Administration of estrogens to castrated rabbits increases the alpha receptor sensitivity of their oviducts, while progesterone reduces alpha and enhances beta sensitivity (87). Hence, the excitatory actions of epinephrine and norepinephrine are most pronounced in the estrogen-dominated animal and the inhibitory actions of isoproterenol are most effective in the progesterone dominated. Alterations in the sensitivity or activation of adrenoceptors may influence ovum transport in the tube and thereby regulate the fate of the fertilized ovum. A systematic analysis of the regional sensitivity of the oviduct to catecholamines in a variety of species under different hormonal conditions would be of value.

Although modern biophysical techniques have been used for studies on the electrical and mechanical activity of the oviduct (87, 91–94), they have not been

exploited to investigate the cellular mechanisms mediating the actions of catecholamines in this tissue. Until definitive experimental evidence proves otherwise we may assume that these mechanisms are similar to those outlined for the myometrium. A recent study reported that beta adrenergic agents increased in short circuiting current and potential difference across an isolated segment of rabbit oviduct (95). These effects were potentiated by dibutyryl cAMP and theophylline, and were thought to result from an increase in the active transport of chloride ions across the cell membrane, stimulated by an elevation of tissue cAMP. Since the segment of oviduct used in these experiments contained primarily the mucosa with its secretory epithelium, the results may not be applicable to the smooth muscle of the oviduct.

The adrenergic innervation of the oviduct and the effects of adrenergic nerve stimulation have received considerable attention during the past few years. The adrenergic nerves to the oviduct are primarily postganglionic, the "short" neurons whose cell bodies lie in or near the muscle (70, 83). The innervation density and catecholamine content are higher in the oviduct than in the myometrium, although the density progressively decreases from the ampulla to the isthmus (70, 71, 83). At the junction of the ampullary and isthmic regions there is an abrupt increase in density, confined chiefly to the circular muscle layer, suggesting an adrenergically-controlled sphincter (66, 93). This pattern of innervation is the same in all species thus far examined.

Unlike the myometrium, the oviduct does not show a decrease in innervation density or catecholamine content during pregnancy (96). However, in the rabbit the catecholamine content of the isthmic portion of the tube is markedly decreased during egg transport, a time when the circulating estrogen levels are low. Injection of estrogen raises the catecholamine content above that of the control animals and at the same time blocks egg transport by constricting the ductal lumen (97). It will be recalled that estrogen also sensitizes the alpha receptors in the oviduct to the actions of norepinephrine. Thus the tubal arrest could be due to local adrenergic influences, because it can be counteracted by the administration of an alpha blocker (98). Progesterone accelerates egg transport, and increases norepinephrine content and concentration in the isthmus (97). Local adrenergic influences have also been implicated in the periodic bursts of motility that are seen in the oviducts of humans and rabbits (86, 99). The intensity and periodicity of the bursts are greater during the proliferative than during the luteal phase of the cycle in the human (86). At all phases of the cycle they are abolished by alpha blockers, suggesting the bursts may be due to a local release of norepinephrine (86). Collectively these findings point to a prominent adrenergic component in the regulation ot tubal motility.

The effects of adrenergic nerve stimulation on tubal contractions have been studied in the rabbit and human (83, 84, 100). Stimulation of the perivascular nerves to the oviduct at supramaximal frequencies and intensities causes the isolated human fallopian tube to contract (100). This response is unchanged in the presence of atropine or ganglion blockade and is converted to inhibition by an alpha blocker. The inhibition is abolished by a beta blocker. The nerve-

mediated response is potentiated by elevation of extracellular calcium ion concentration and reduced by low calcium concentrations, by magnesium, and by low temperature (84, 101). The isthmus of the human oviduct is more responsive to nerve stimulation than the ampulla (84) correlating with the greater innervation density at the isthmus (83). It is possible that a sphincter, controlled by adrenergic nerves, operates at the isthmic end of the oviduct and regulates the retention of ova in the ampulla. Functional blockage of ovum transport resulting from adrenergic nerve activity could occur in individuals undergoing stress or fear and affect fertility. This "tubal locking" mechanism should be especially prominent during estrogen-domination (cf. pp. 27, 28).

Prostaglandins of the E type (PGE_1, PGE_2) have recently been shown to inhibit adrenergic neurotransmission in a variety of organs including the oviduct (102, 103). An intravenous injection of PGE_1 reduces the response of the rabbit oviduct to adrenergic nerve stimulation and to exogenously-administered norepinephrine. If an analogy to similar actions in the spleen and vas deferens is permissable (103–105), we might assume that PGE_1 is acting both pre- and post-junctionally; to depress the release of norepinephrine from the nerve terminal and (at low doses) to decrease the sensitivity of the smooth muscle cell to the transmitter. In view of the finding that adrenergic nerve stimulation releases endogenous prostaglandins of the E series from autonomic effector cells (103, 104) it is tempting to speculate that the ubiquitous prostaglandins act as endogenous local regulators of adrenergic neural activity. This fascinating possibility is worthy of careful, systematic investigation in the female reproductive system.

OVARY

Interest is currently directed to the earlier observations of anatomists that the perifollicular region of the ovary contains abundant smooth muscle elements. Recent histochemical and ultra-structural studies on the ovaries from the rat, rabbit, cat, guinea pig, monkey, and human, confirm the presence of smooth muscle cells and in addition indicate an adrenergic innervation whose density is especially striking in the human and cat but less marked in the rat and rabbit (70, 196–111). Unmyelinated nerves containing dense core vesicles lie within 2000–3000 Å of the smooth muscle cells (110). Since smooth muscle cells are influenced by release of neurotransmitter within a distance of 200–10,000 Å (1), it seems likely that the smooth muscle in the perifollicular region of the ovary is partially or entirely under neural control. There is indirect, pharmacologic evidence for adrenergic influences on ovarian smooth muscle. In the hen, intrafollicular injections of dibenzyline, an alpha blocker, prevent ovulation (112). Intra aortic injections of dibenzyline in rabbits pretreated with human chorionic gonadotropin reduce the number of hemorrhagic and ruptured follicles (113).

The presence of muscular activity in the ovary has been substantiated by experiments on ovaries from cats and rabbits (114, 115). The ovaries show spontaneous contractions in an isolated organ bath as well as in situ. Contractions are stimulated by epinephrine and norepinephrine and inhibited by isoproterenol. These

actions are prevented by appropriate adrenoceptive blocking agents, thus establishing the presence of alpha and beta receptors within the ovarian smooth muscle. The influences of hormonal environment or of adrenergic nerve stimulation on these contractions have not yet been described.

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