before addition of 6 n*M* [^{3}H]diazepam, the amount of specifically bound [^{3}H]-diazepam was increased by approximately 30 percent. This increase was similar to that observed in brain membrane preparations (Fig. 1B).

Clonal lines of neuroblastoma and glioma cells have proved to be a suitable substrate for studying molecular mechanisms in the organization of catecholamine (*10*) and opiate receptors (*11*). The data reported here support the view that NB and, to a lesser degree, C6 cell lines are also adequate models for studying regulation of the GABA "receptor unit." This unit is composed of GABA and benzodiazepine receptors and GABA modulin. The physiological interaction of GABA modulin with GABA and benzodiazepine receptors in the membranes of NB and C6 glioma cells is indirectly suggested by studies with intact cells: (i) in intact NB cells (*12*), [^{3}H]clonazepam and [^{3}H]diazepam label a similar number of benzodiazepine receptors, with a K_d essentially identical to that shown in isolated neuroblastoma membranes; (ii) diazepam (10^{-6}*M*) added to NB or C6 cell cultures produces 50 percent release of the inhibitor into the medium and a concomitant 50 percent decrease of the inhibitor in the cell membranes.

In addition, the regulation of the GABA receptor units located on NB or C6 cell membranes is in many aspects similar to the regulation of those located on brain membranes. In NB and C6 clonal cell lines, as in brain, GABA binding is regulated by GABA modulin and its kinetics are modified by the activation of specific benzodiazepine receptors. Moreover, the GABA receptor can also modify the benzodiazepine receptor and as a result it can increase the affinity of the specific binding sites for benzodiazepines. Whereas the benzodiazepines act competitively with GABA modulin (*2*), it is not known whether GABA also regulates diazepam binding by acting on GABA modulin. One could speculate that the physiological agonist of the benzodiazepine receptor is GABA modulin. Conversely, one could also propose that an endogenous benzodiazepine agonist modulates the function of the GABA modulin, but such an agonist has not yet been found.

It is pertinent to discuss the physiological role linked to the interactions between GABA and GABA modulin and the pharmacological implications of the consequences of the activation of the benzodiazepine receptors. One possibility is that GABA modulates a Cl^- channel in NB and C6 cell lines. Initial results with NB cells indicate that the activation of GABA receptors causes an increase in the inward Cl^- flux (*13*); whether the cooperative interaction of the benzodiazepine and GABA receptors extends also to the GABA regulation of Cl^- channels remains to be investigated.

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9 January 1979; revised 17 April 1979

Menopausal Flushes: A Neuroendocrine Link with Pulsatile Luteinizing Hormone Secretion

Abstract. *Menopausal flush episodes were found to be invariably associated with the initiation of pulsatile pituitary release of luteinizing hormone. This was not accompanied by a significant change in circulating catecholamine or prolactin concentrations. Since pulsatile luteinizing hormone release results from episodic secretion of luteinizing hormone releasing factor by the hypothalamus, these findings suggest a link between the neuroendocrine mechanisms that initiate such episodic secretion and those responsible for the onset of flush episodes.*

Although estrogen withdrawal unquestionably plays a major role in the development of menopausal flushes, the physiological mechanism for the initiation of flushes and of transient physical changes during flush episodes (*1, 2*) remains elusive. It has been suggested that menopausal flushes are manifestations of vasomotor instability due to a transient increase in adrenergic activity (*3, 4*), but evidence to support this is lacking. The study described here was therefore designed to search for neuroendocrine correlates of spontaneous flushes in hypogonadal women.

A total of 55 flush episodes were studied in six postmenopausal women. All studies were carried out with the subjects at bed rest in a quiet room with a stable, ambient temperature. A normal diet with the exclusion of caffeine and nicotine was provided and all subjects were fully awake during the 8- to 10-hour studies. Onset of flush episodes was reported by the subjects and retrospectively confirmed by objective observation. Finger temperature was measured by using thermistors, and electrocardiogram and pulse rate were monitored. These parameters were continuously graphed by an eight-channel physiological recorder. Blood samples were obtained from an indwelling venous cannula at 2- to 15-minute intervals between flush episodes for determination of serum luteinizing hormone (LH), follicle stimulating hormone (FSH), and prolactin (PRL) concentrations by radioimmunoassay (*5*) and plasma dopamine (DA), norepinephrine (NE), and epinephrine (E) concentrations by radioenzymatic assay (*6*).

The onset of each of the 55 flushes was characterized by a sudden intense sensa-

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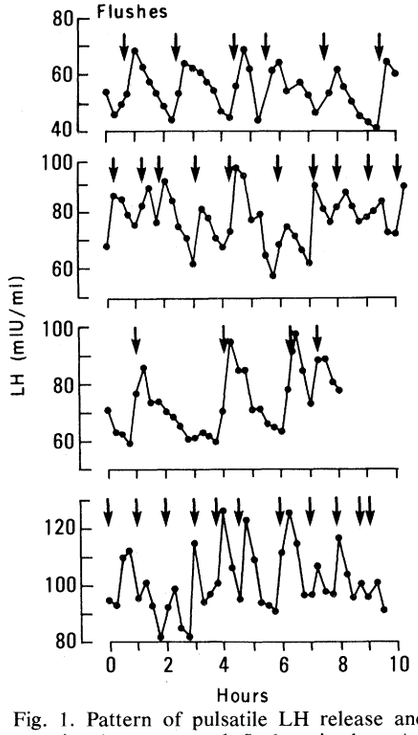


Fig. 1. Pattern of pulsatile LH release and associated menopausal flush episodes. Arrows indicate flush onset. Each part illustrates a separate 8- to 10-hour study in which blood samples were obtained at 15-minute intervals. Note that each flush is synchronized with an LH pulse.

tion of heat centered on the face, then moving to the neck and chest, and finally becoming generalized. All flushes were accompanied by perspiration of variable degree. The duration of a flush was 2.7 ± 0.1 minutes [mean ± standard error (S.E.)]. Flushes were associated with a mean increase in finger temperature of 7.5°F (from 85.5° ± 1.2° to 92.9° ± 0.73°F) and a mean increase in pulse rate of nine beats per minute (from 78.2 ± 1.4 to 87.3 ± 1.7 beats per minute).

Analysis of the data revealed a close synchrony between flushes and LH pulses (Fig. 1). The mean frequency of LH pulses was 73.5 minutes (range, 54.5 to 120 minutes) and the women with the most frequent pulses had the highest frequency of flushes. A total of 66 LH pulses occurred during the course of our study and these were associated with 55 flushes. Thus, LH pulses were not always accompanied by flushes. Conversely, however, a flush was never seen to occur in the absence of an LH pulse.

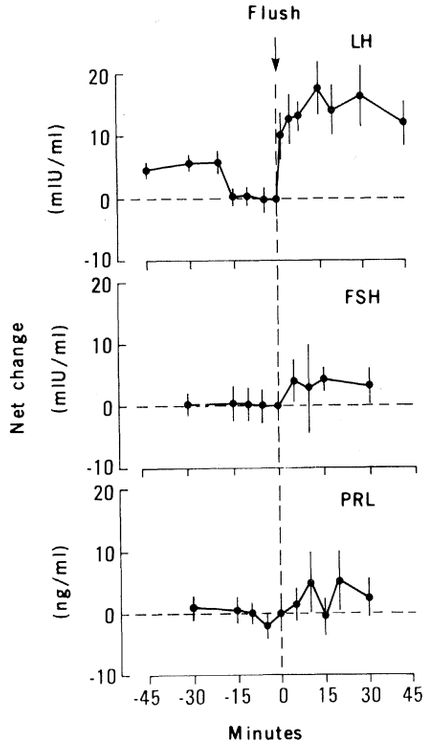
To determine more precisely the temporal relationship between flushes and hormonal changes, composite data for the 55 flushes were examined, including 2- to 5-minute sampling during 15 flushes. Serum concentrations of LH, FSH, and PRL before and after these 55 flush episodes are shown in Fig. 2. A prominent feature of these data is the coincidence of the onset of the flushes with the initial rise in circulating LH at the time of an LH pulse. A small but significant (P = .016) parallel rise in serum FSH was also found. No associated change occurred in serum PRL concentrations.

Plasma concentrations of DA, NE, and E associated temporally with 15 flushes are shown in Fig. 3. No significant changes were observed in any of the catecholamines from 30 minutes before to 30 minutes after the flush.

A temporal relationship between pulsatile release of LH and initiation of menopausal flushes is clearly demonstrated by these data. The LH release by the pituitary per se can be excluded as the cause of flushes, since flushes do occur in hypophysectomized women (*7*). It therefore seems likely that a suprapituitary mechanism must initiate both the pulsatile release of LH and flush episodes.

Studies with rhesus monkeys suggest that the site governing the pulsatile release of LH is within the arcuate nucleus (*8*) and involves pulsatile LRF secretion (*9*). In humans, the concordance of LH and FSH pulses in postmenopausal women (*10*) also suggests a temporal relationship between episodic LRF secretion (*11*) and pituitary pulsatile LH release. However, it is unlikely that LRF pulsatile release itself is responsible for the initiation of flushes. Yen *et al.* (*12*) demonstrated that the pulsatile pattern of gonadotropin release in hypogonadal women with gonadal dysgenesis closely resembles that in postmenopausal women. These individuals, who have never been exposed to normal female levels of endogenous estrogen, do not experience flushes even at advanced age. However, when estrogen is administered for several months and then discontinued, these women experience classical menopausal flushes for the first time (*4*). This observation suggests that menopausal flushes are a manifestation of a classical withdrawal syndrome, mediated through functional changes of estrogen-sensitive neurons, which is linked to the control of LRF pulsatile release.

That the secretion of LRF is, at least in part, under the control of catecholaminergic neurons is suggested by the finding that pulsatile LH release is promptly abolished in ovariectomized monkeys by the administration of α-adrenergic (but not β-adrenergic) blocking agents (*13*). Attempts to delineate the α-adrenergic control of pulsatile LH re-



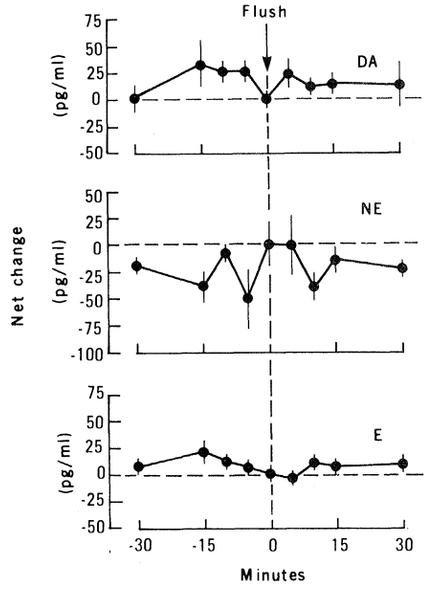


Fig. 2 (left). Mean (± S.E.) serum LH, FSH, and PRL concentrations measured before, during, and after the 55 episodes of flushes. The onset of each flush is set at time t = 0 and the data are expressed as the difference from this reference point. Serum LH concentration is significantly elevated (P < .0001) immediately after the onset of the flush and the elevation lasts for at least 45 minutes. A small but significant (P = .016) parallel increase in serum FSH occurred with no associated change in serum PRL concentration. Data analysis was by analysis of variance and Student's t-test for paired data. Fig. 3 (right). Mean (± S.E.) plasma DA, NE, and E concentrations during 15 flushes. The data are normalized around the onset of each flush (at t = 0) and expressed as net change in picograms per milliliter from this reference point. No significant change is seen in any of the three catecholamines from 30 minutes before to 30 minutes after the flush episodes.

lease in humans (*10*) have been inconclusive because cardiovascular side effects preclude the use of doses of α-blockers comparable to those used in rhesus monkeys. In addition to its role in pulsatile LH release, increased adrenergic activity has been implicated in the initiation of flushes (*1-4*). The finding of increased finger temperature, as a result of vasodilatation during flushes (*2, 14*), would seem to be inconsistent with increased adrenergic activity, since finger vessels are under the exclusive control of a tonic α-adrenergic vasoconstrictor mechanism (*15*). Further, the present finding that circulating catecholamine concentrations do not change during flush episodes suggests that a peripheral adrenergic mechanism is probably not causally involved in flush initiation. However, central adrenergic activation could occur, resulting in local release of vasoactive substances such as prostaglandin or histamine (*16*), which may explain why vasodilatation outlasts the flush episode. This is consistent with the clinical observation that administration of clonidine, an α-adrenergic agonist that acts centrally to decrease adrenergic activity, provides symptomatic relief of flushes (*17*). Thus, our results do not rule out the involvement of the central catecholaminergic system in the initiation of flushes. On the contrary, they suggest a link between central neuroendocrine mechanisms that initiate episodic LRF neuronal activity and those determining the onset of flush episodes. We anticipate that this finding will provide an important clue for future research into the neuroendocrine mechanism (*7*) involved in menopausal flushes.

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2 April 1979; revised 27 June 1979

By Dawn's Early Light: Matutinal Mating and Sex Attractants in a Neotropical Mantid

Abstract. *Females of the neotropical mantis* Acanthops falcata *adopt a special posture at dawn which is maintained for about 20 minutes. During the same period, males fly strongly, even in the absence of females. Our studies show that in this posture females are secreting a pheromone that acts as a sex attractant. All sexual activity in this species normally occurs between dawn and sunrise. It can be triggered by any dark-to-light transition, irrespective of real time. This sexual periodicity is probably an antipredator adaptation.*

Tropical praying mantises often have complex defensive adaptations (*1-4*). Several species are highly specialized mimics of leaves, sticks, or flowers (*1, 2*). Such species tend to be rare or widely dispersed; the females are usually larger than the males and have reduced wings. Females of the neotropical dead-leaf mantid *Acanthops falcata* Stal are completely flightless, whereas the smaller males can fly. In order to mate, the males have to find the widely dispersed females. Females strike indiscriminately at moving objects up to the size of males, so the latter run the risk of being treated as prey. Although it has been shown that males of some mantid species can copulate while being eaten (*5-7*), this adaptation has no effect on fitness if the female doing the eating is sexually unreceptive and copulation cannot occur. The male has two major problems, it must find a female of the right species and must find one that is ready to mate. These problems could be solved if females that were ready to mate released a sex attractant pheromone. Our studies show that they do.

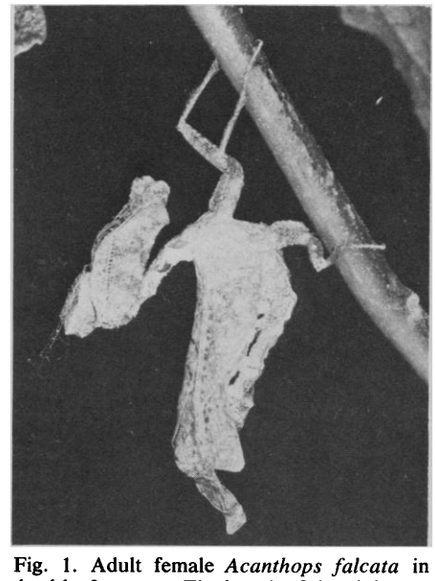

Fig. 1. Adult female *Acanthops falcata* in dead-leaf posture. The length of the abdomen is about 25 to 30 mm. The anterior legs fold around the head and conceal it.

Unmated females release a pheromone for a limited period each day until they have mated. The pheromone release occurs during the short period after dawn during which males fly. We have shown that both the pheromone release and the onset of flight activity are triggered by the dark-to-light transition. Both appear to be independent of any internal clock.

Our attention was first drawn to this problem when, very early in the morning, we transferred hand-reared virgin females to a large outdoor cage (*8*) and found wild males resting on the outside of the cage and mated pairs within. We later found that at dawn (*9*) all the males within the cage became unusually active. After a brief period of rocking movements, they flew or walked with wings raised, eventually settling close to a female. From this position they jumped the last few centimeters onto her dorsal surface and there grasped the leading edge of the female's wings with their raptatory anterior legs. During the same period the females exposed the dorsal surface of their abdomen by raising the wings and

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