cially helpful in experiments on ganglia like that from *Aplysia* with relatively large numbers of neurons.
We plan to further characterize the cells participating in the behavior. One way in which we hope to do this is by monitoring the direction of spike propagation in the peripheral nerves and matching their timing with soma recordings. Activity of cells with peripheral nerve spikes proceeding outward (centrifugal) are of possible motoneuron origin; activity of axons spikes proceeding inward (centripetal) may be from sensory neurons. With improvements in cell characterization and identification, and with the recording of synaptic potentials we hope that optical recording will become a useful tool in working out the neuron interactions that underlie simple behaviors.

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Switching among functional states by means of neuromodulators in the lobster stomatogastric ganglion

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Summary. The lobster stomatogastric ganglion contains the central pattern generators (CPGs) for the pyloric and gastric mill rhythms. All of the neurons and their synaptic connections have been identified for each rhythm and serve as the basis for understanding the mechanisms by which chemical neuromodulators are able to alter the functional state of each CPG. Using examples of different amines and peptides, I show how these substances can be found within specific neurons and how their application to the CPG can alter the motor patterns in specific ways. I also discuss what changes in cellular and synaptic properties occur as a result of bath application and particularly in the case of proctolin, how these changes may have behavioral correlates. The various outputs appear to be the result of a functional 'rewiring' of anatomically defined neural circuits and this may be a widespread mechanism for the production of closely-linked but behaviorally distinct movement patterns.
Key words. Lobster stomatogastric system; neuromodulation.

Many classes of rhythmic behaviors do not exist as discrete entities but as a continuum of various more or less stable states. An example is vertebrate locomotion. Separate functional modalities such as walking, trotting, galloping etc. can exist as recognizable behaviors but are not completely regular due to cycle by cycle changes imposed on each state by sensory feedback and descending and coordinating fibers extrinsic to the particular pattern generator involved. Given this complexity, an important consideration in the neuronal analysis of networks producing rhythmic movements is whether there is a separate central pattern generator (CPG) for each variation of a fundamental movement, or whether different functional states are carved out of one large network which has the potential for producing all possible rhythmic movements a particular effector system is capable of. The two possibilities are not meant to be mutually exclu-

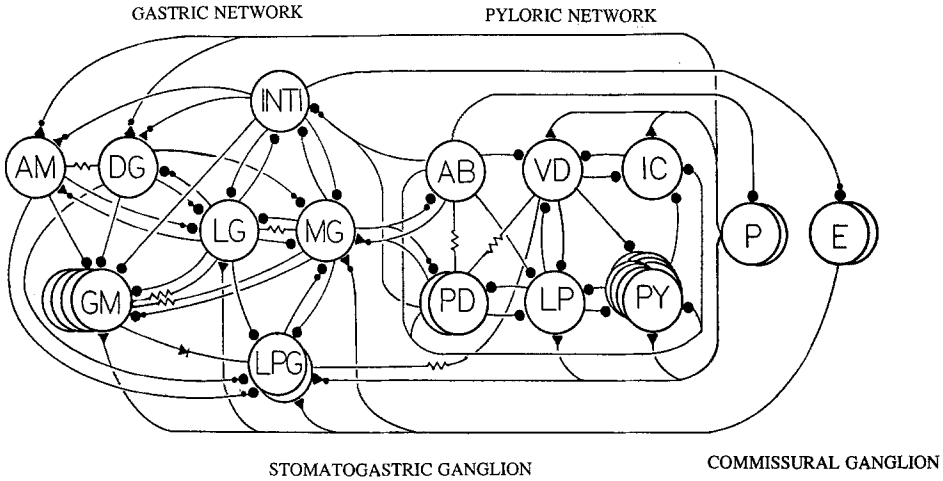

Figure 1. Neural circuitry of the gastric and pyloric central pattern generating networks. All of the neurons are in the stomatogastric ganglion except the P and E cells which are in commissural ganglia. Black dots represent chemical inhibitory synapses, black triangles represent chemical excitatory synapses. The resistors and diodes represent electrotonic junctions. The smaller black dots signify a functional synapse, one which has not yet been fully proven to be monosynaptic. Abbreviations of neurons are: AM, anterior median; DG, dorsal gastric; GM, gastric mill; Int 1, interneuron 1; LG, lateral gastric; MG, median gastric; LPG, lateral posterior gastric; AB, anterior burster; PD, pyloric dilator; VD, ventricular dilator; LP, lateral pyloric; IC, inferior cardiac; PY, pyloric.

sive nor to imply one mechanism would be more advantageous than the other. There is the option, however, that numerically large nervous systems might favor a separate CPG for each pattern or an overlap of elements from different but related CPGs[42], options that 'simpler' nervous systems could not afford.

For the purposes of this paper we can operationally define a CPG to be that collection of central nerve cells which, when isolated from all sources of rhythmic input, are capable of producing a rhythmic motor pattern similar in its basic form to that produced in the intact animal. A very large number of rhythmic behaviors will fit under this umbrella[7], and no serious challenge to the existence of such a concept has been suggested[33]. The mechanisms underlying the generation of rhythmic motor patterns in several invertebrate preparations can now be qualitatively described[39]. Such descriptions are remarkable for their lack of common neuronal mechanisms despite the similarities between the motor patterns they generate. One can consider possible mechanisms to be composed of three principal types: cellular (bursting pacemaker potentials), synaptic (postinhibitory rebound, strength, delayed excitation, etc.), and circuit (reciprocal inhibition). Each of these types can be further broken down into specific properties which may be considered the 'building blocks' of any pattern generating system[11]. It is clear that chemical neuromodulatory substances are present that can come into contact with neurons that compose CPGs, and that such modulators interact with CPGs in such a way that both cellular and synaptic properties can be modified. As a result the circuit can change in a way that alters the motor pattern significantly for a protracted period of time[25].

The control of rhythmic motor networks can thus be viewed as being under the influence of both neural (sensory feedback and descending control fibers), and chemical mechanisms, the role of both being to start and stop the pattern, to modify it continuously and possibly to make significant changes to its form, i.e., its functional state. Here we will examine the mechanisms by which such state changes are produced in the neural networks of the lobster stomatogastric ganglion.

The stomatogastric system

The stomatogastric nervous system produces two rhythmic motor patterns which operate the striated muscles of the gastric mill and pyloric regions of the lobster stomach[40]. The neural circuit for producing the basic pyloric and gastric patterns is shown in figure 1. These connections were determined by stimulating and recording from pairs of cells in isolated ganglia, often with the rhythms in a quiescent state. Most of the synaptic connections were also tested for monosynapticity[41] but it should be made clear that the neuronal connections were in a somewhat artificial state and that in the fully intact system certain synaptic parameters, such as strength, may be quite different.

Pyloric rhythm

The pyloric CPG is composed of fourteen neurons, of which thirteen are motor neurons and one is an interneuron. There is an additional pair of interneurons called P cells[35] located in the commissural ganglia (fig. 2) which are phasically inhibited by bursts of activity in the AB interneuron. The P cells provide bursts of EPSPs to all of the pyloric motorneurons except the PDs and AB. This pathway, and possibly others from the commissural ganglia, must be intact for the pyloric rhythm to operate. The pyloric pattern, illustrated in

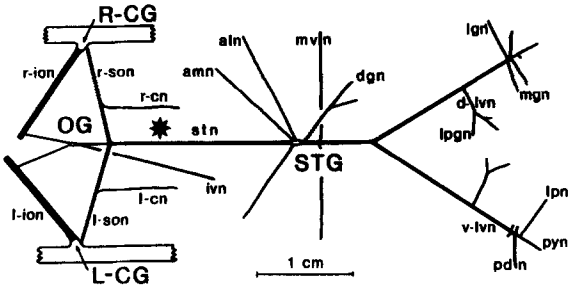

Figure 2. Diagram of the dissected stomatogastric nervous system as pinned out. The star indicates the site of the stomatogastric nerve block. Abbreviations: R & L-CG, right and left commissural ganglion; son, superior oesophageal ganglion; ion, inferior oesophageal ganglion; OG oesophageal ganglion; cn, chemoreceptor nerves; stn, stomatogastric nerve; amn, anterior median nerve; aln, anterior lateral nerve; mvn, median ventricular nerve; lgn, lateral gastric nerve; mgn, medial gastric nerve; d-lvn, dorsal branch of lateral ventricular nerve; lpgn, lateral posterior gastric nerve; v-lvn, ventral branch of lateral ventricular nerve; lpn, lateral pyloric nerve; pyn, pyloric nerve; pdn, pyloric dilator nerve.

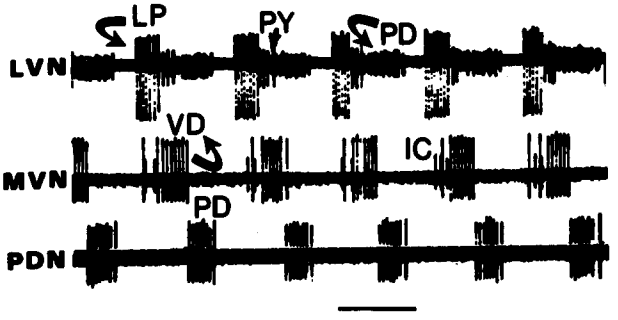

Figure 3. Extracellular nerve recordings of the pyloric rhythm from a combined preparation, i.e. with the commissural ganglia intact. Abbreviations as in figs 1 and 2. Time bar is 0.5 s.

figure 3, consists of a three-phase burst cycle occurring at a frequency of about 2 Hz. When the electrotonically coupled PD-AB group fire, they strongly inhibit all of the other pyloric neurons. When the PD and AB cells stop firing the LP and IC cells fire and then the VD and PY cells fire in bursts to complete the cycle.

An analysis of the pyloric system using a sucrose block of the stomatogastric nerve to reversibly remove the influence of the descending commissural fibers [35] and selective cell ablations [27] during ongoing activity has shown there are two fundamental mechanisms involved in burst and pattern formation [28, 29]. Most of the pyloric neurons appear to be 'conditional bursters', i.e., under the influence of the commissural transmitters, they display intrinsic bursting pacemaker potentials which cause the membrane potentials of the cells to vary cyclically (slow wave). Each cell can be considered to be an oscillator and the entire pyloric network a system of coupled oscillators. In addition, the connectivity of the network itself gives rise to resonant properties which also contribute to burst formation. The principal underlying connections responsible appear to be reciprocal inhibition, a class of circuit which has often been suggested as the basis for alternate bursting. Computer simulations of such networks have robust characteristics especially when the property of postinhibitory rebound is incorporated into the model.

A problem separate from the generation of bursts, although sharing some of the same mechanisms, is how the correct phase relationships between the bursts are established. We know there are a variety of straightforward mechanisms which can account for the observable behavior. The PD-AB neurons are strongly coupled to each other and strongly inhibit all of the other pyloric neurons so that when this group is active, the other pyloric neurons are hyperpolarized. When the PD-AB group ceases to be active, the remaining pyloric cells will rebound from their inhibitory state with different time constants, the LP and IC being faster than the VD and PYs [16]. This simple mechanism causes the LP and IC to fire their bursts before the VD and PYs, the overall effect being to create a three phase rhythm – PD/AB $\gg$ LP/IC $\gg$ VD/PY.

An important feature of the burst and pattern generating mechanism is the fact that the inhibitory transmitters are released both in pulsatile and continuous fashion, the former following action potentials and the latter following the membrane potential of the presynaptic terminal [14, 34]. The absence of a threshold function for pyloric neurons is an important characteristic when postulating a computational explanation of the pyloric rhythm since all available simulation algorithms rely on discrete events (action potentials) to indicate neuronal activity.

Gastric rhythm

The gastric central pattern generator produces a basic four phase rhythm when the commissural ganglionic pathways are intact. The bursts of impulses in the motor nerves are arranged in two antagonistic pairs: LG & MG, which open the two lateral teeth of the gastric mill, alternate with bursts in the LPGs, which close them. The four GM neurons fire bursts which pull the single medial tooth forward in a power stroke and the DG & AM fire out of phase with the GMs as a return stroke (fig. 4). The neural circuit responsible for the generation of this pattern (fig. 1) differs in an important way from the circuit which generates the pyloric rhythm. Instead of having many neurons which are capable of intrinsic bursting, only one neuron has this proclivity [36, 40]. As a result, the mechanism for both burst generation and pattern formation appears to reside almost entirely within the network of synaptic connections making up the circuit. Cell killing experiments have been performed on the four GM cells and on the DG/AM pair with very little alteration of the ongoing pattern. However, it is possible to see preparations in which DG is the only cell bursting, a phenomena which demonstrates this cell is intrinsically capable of generating bursts and that this property can act in parallel with network burst forming properties. This evidence points to the fact that the medial tooth motorneurons must be driven by the neurons of the lateral teeth subsystem, by Interneuron 1 (Int 1) and by the burstiness of DG when the commissurals are present.

The lateral teeth network contain one pair of strong reciprocally inhibitory synapses which could, in principle, serve as the basis for the production of alternate bursts (fig. 5). This

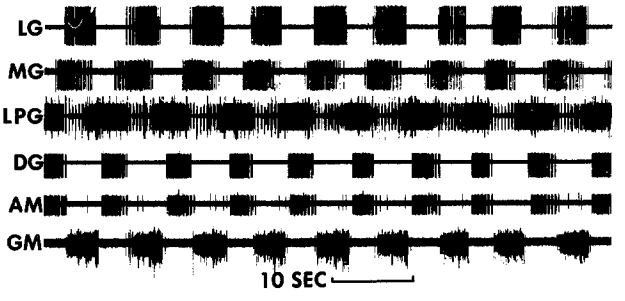

Figure 4. Burst pattern of gastric mill motorneurons in vitro from a combined preparation.

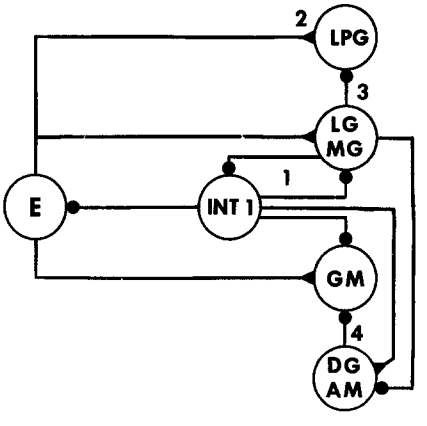

Figure 5. A hypothetical scheme for the production of the gastric mill rhythm is illustrated in this simplified diagram of the gastric mill CPG. (1) Reciprocal inhibition between LG/MG and Int 1 provides the basic rhythmicity; (2) E cell activation keeps the LPG in a high state of tonic firing; (3) rhythmic inhibition from the LG/MG pair periodically inhibits the LPGs; (4) delayed excitation from Int 1 periodically fires the DG/GM pair which inhibits the GMs.

pair connects the LG/MG group with Int 1. These two groups could alternate with one another setting up the basic alternation for the circuit. The LPGs are normally (when the system is not bursting) extremely active and fire continuously at high frequencies. Bursting in LG and MG would periodically interrupt this ongoing activity. The bursting in Int 1 would act as a driver to DG and as an inhibitor to the GMs. We cannot, at this time say that this is how the gastric system actually works, but it does serve as a starting hypothesis for how it might.

As with the pyloric pattern generator, two interneurons in the commissural ganglia, in this case called E cells, are rhythmically inhibited by Int. 1. The two cells feed bursts of excitation back to the gastric pattern generator in a way analogous to the way the P cells feed excitation back to the pyloric, the result being the same in both cases – enhancement of the rhythm.

Switching among functional circuits

The circuits for the gastric and pyloric network were determined in isolated ganglia or combined preparations (i.e., with the commissural and esophageal ganglia attached). The circuitry, under these conditions, produces the two fundamental patterns just discussed. But in the intact animal the regularity of the bursts disappears and gives way to quite complex variations in both rhythms. What are the factors responsible for such imposed variations? I believe we can consider four different processes:

1. Phasic input – conventional synaptic input to elements of the CPG having an immediate but short-lived effect on the pattern. The source may be peripheral sensory receptors or central loops such as the P and E cell loops just mentioned. In addition coordinating fibers from other CPGs which are used to maintain particular phase relationships are also present. There is a rich literature on the mechanisms by which phasic feedback can alter or entrain an oscillator, most of which stems from the work of von Holst [19].
2. Tonic input – again conventional synaptic input, but not bursting or patterned in a particular time ordered sequence. An example might be the command fiber inputs commonly found in Crustacea [46] which are capable of eliciting distinct and sometimes rhythmic stereotyped behavior patterns. Since it is practically impossible to record from individual command fibers in situ, one cannot say with certainty that such fibers actually do fire tonically during normal behavior, but experimentally tonic stimulation can emulate normal behavioral activities.
3. Hormonal input – chemical input delivered systemically and acting on target neurons with appropriate receptors. The time course of the activity is over a much longer time scale which is what differentiates this activity from the rapidly acting conventional synaptic activity. The active substances can be released from neurons (neurosecretory type cells) or from glandular tissue.
4. Local hormonal input – chemical neuroactive substances released from nerve terminals and acting over short distances (hundreds of μ) such as the neurophil of a single ganglion. The cell bodies of such neurons can be in different ganglia from the one where the responsive neurons are. Many of the neurons which act this way may contain more than one type of neuroactive chemical.

The first two types of input are useful in interfacing a CPG with its external environment, being capable of altering the pattern on a cycle-by-cycle basis and triggering particular CPGs. The second two types, referred to as neuromodulatory, appear to alter a particular CPG so that many functional states can be obtained from one basic circuit. While the general concept of hormonal control of neuronal state is not new, the actual mechanisms of how such alterations occur first at the cellular level and then at the systems level, has not been explained. The 'black box' approach to this question only supplies input/output relationships. The problem of how these four kinds of control devices are used in situ and how they interact with one another is fundamental to understanding the neural control of behavior.

The stomatogastric system can provide a useful model for the analysis of how neuromodulatory substances, both local and systemic, act by themselves and how they interact with conventional synaptic inputs. There are at least four reasons why this system is especially useful.

1. A large number of putative chemical substances have been found in the lobster and in some cases localized to particular somata and neuropil areas.
2. The bath application of such substances at physiological concentrations can alter both the gastric and pyloric motor patterns.
3. A dye-sensitized photoinactivation method permits the rapid analysis of the effects of neuromodulatory substances at different concentrations and combinations on single isolated cells or synapses.
4. It is now possible to observe behavioral consequences of modulatory action either directly in the case of the gastric mill or indirectly for the pyloric system.

The anterior pyloric modulator (APM)

One of the first neurons in the stomatogastric system which was found to have a modulatory effect was the APM. The APM is a small cell located in the oesophageal ganglion which when stimulated produces long-term, neuromodulation of the pyloric rhythm [8, 31]. Although this cell has not been shown to contain acetylcholine, the plateau potentials which are produced in the pyloric cells after APM stimulation can be mimicked by the application of muscarinic agonists such as oxytremorine or pilocarpine suggesting the APM is cholinergic. One effect of pilocarpine is the turning on of bursting pacemaker conductances, which occurs also in the presence of TTX [1]. This phenomenon may be due to a decrease in voltage-dependent potassium conductances [30]. APM activity or the presence of cholinergic agonists elicits major changes in an ongoing pyloric pattern. The overall burst period decreases while the number of spikes per burst is increased. There are also changes in the phase relationships between some bursting units (fig. 6). Such changes are typical of those which result from the action of neuromodulatory substances – effects on the membrane properties of a relatively large number of units which are established slowly and which act over a long duration [31]. Compilation of the various actions reveals significant changes at all levels; bursting pacemaker and plateau potentials are strongly modified

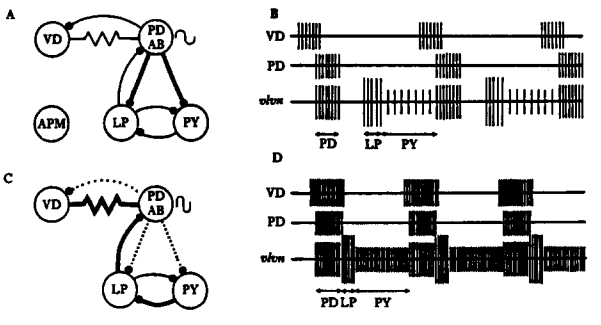

Figure 6. Modification of the pyloric pattern by the muscarinic action of the APM neuron. Changes in the circuit are shown in *A* and *C* with three synapses becoming very weak and three becoming very strong. Changes to the pyloric rhythm are shown in *B* and *D*. (Modified from Nagy and Dickinson [27]).

in some cells, electrotonic junctions between VD and the PD/AB cells have increased coupling coefficients, synaptic connections from the PD/AB cells to VD, LP and PY are weakened while those from LP to PD and PY to LP are strengthened. In short, the neuromodulator produces a fundamental functional 'rewiring' of the pyloric network which results in a switching of the functional state of the system.

Neuromodulators affecting the stomatogastric system

A large number of putative neuromodulatory substances have now been tested on the stomatogastric system. The overall rationale is to demonstrate that a particular substance exists in the animal and, if possible, to localize it by immunocytochemical techniques. When cells containing these substances can be located, one then tries to demonstrate whether their stimulation leads to both measurable release and physiological changes that are identical to those produced by bath application. When these criteria have been successful, it is then possible to experimentally determine exactly how the basic pyloric or gastric pattern is modified and what cellular mechanisms are responsible. This means finding out how the binding of the chemical substance to the receptors of a particular cell eventually leads to the changes in conductance or transmitter release that are responsible for the changes in functional state.

It might be useful at this point to compile the data presently available. Table 1 lists the chemicals which have been tested so far. They can be roughly categorized as amines or peptides and all have been shown to be contained in crustaceans by either radioimmunoassay or immunohistochemistry. Not all of the substances have been characterized biochemically so the immunologic reagents may be recognizing epitopes of closely related compounds. For example, proctolin extracted from the stomatogastric system comigrates with synthetic proctolin on HPLC and is therefore a true proctolin [24]. However, it is likely that FMRFamide-like peptides found in lobster are not FMRFamide [26]. Not all of the substances shown have been extracted from lobster or crab tissue and shown rigorously to have the same chemical composition as the true substance.

Proctolin

Table 1 shows clearly that there are many gaps in our knowledge of how neuromodulators act on the stomatogastric rhythms, where they are located and how they operate in the intact animal. However, proctolin has received much attention and is a good example of a substance which interacts with both the gastric and pyloric neural networks to alter their overall functional states.

Where, in the stomatogastric system, is proctolin found? Using a polyclonal antibody, stomatogastric systems from three species, *Cancer, Panulirus* and *Homarus* were examined by Marder [24]. Although there were some important variations, in general, proctolin-like staining could be seen broadly dispersed throughout the neuropil of the stomatogastric ganglia of each species. Stained cell bodies could be observed in the commissural ganglia, and while their axons could not be traced to the stomatogastric ganglia, there were always some fibers stained in the SON and STN nerves, the most direct pathway between the CG and the STG. Radioimmunoassays (RIAs) of stomatogastric ganglia showed approximately 100–300 fmoles of proctolin-like material [26] in each. In addition two different solvent systems were used to fractionate crude extracts of stomatogastric tissue on C18 Sephadex columns. When run with synthetic proctolin, the peaks were indistinguishable, suggesting the material obtained from the animals was authentic proctolin.

The effects of proctolin on the gastric mill could be demonstrated in vivo and in vitro. To examine these effects, one must first consider the behavior of the three teeth of the gastric mill. When the mill is not cycling, the two lateral teeth are held apart and the medial tooth is in a dorsal-caudal position [17]. When the mill does turn on, either spontaneously or as a result of removing the eyestalks, two kinds of behav-

Neuromodulatory substances in the lobster stomatogastric system

Substance	Presence	Location (see fig. 2)	Pyloric	Gastric	References
ACh	+	OG	↑ freq-shift ↑ burst freq.	+ + some	8, 31
Histamine	+	IVN	Excitatory	Excitatory	6
Dopamine	+ HC BC	DG, STG neuropil (L-cell)	Inhibits PD & VD, excites AB, tonic activity in isolated LP, PY, IC		2, 10, 13, 21, 22, 38
Serotonin	IHC, HPLC	STG neuropil CG pericardial organ	Bursting in AB, tonic activity in isolated IC, inhibits isolated VD & LP neurons		4, 10, 43
Octopamine	BC	STG neuropil	Excitatory to most cells, conc. dependent bursting in AB	Bursting in DG	3, 9, 10, 25, 45
GABA	IHC	STG neuropil cells in OG	$\uparrow_g K^+$, $\uparrow_g Cl^-$ suppresses activity		5
Proctolin	IHC, RIA, HPCL	STG neuropil cells in OG (crab) & CGs	Starts bursting ↑ #'s LP-APs, ↑ freq & amplitude of AB bursts	Initiates rhythm, two different dose-dependent patterns	18, 20, 23, 24
FMRF-amide-like	IHC, RIA	STG neuropil, STN fibers, 4 cells in OG, cells in CGs, CG neuropil	Initiates rhythm, ↑ freq, ↓ LP activity, ↑ PY activity		20, 25, 26
Substance P	IHC	STN fibers, STG neuropil	Changes freq. of ongoing rhythm	Some neurons activated	12
CCK/ Gastrin	IHC	Cells in CG, STG neuropil	Turns on at 10^{-4} M	Alters bursting, can initiate rhythm at 10^{-4} M	44
RPCH	IHC	STG neuropil, 2 cells in OG cells & NP of CG		Initiates rhythm	32
AKH				Initiates rhythm	32

HC, histochemical evidence; BC, biochemical evidence; IHC, immunohistochemistry; HPLC, high performance liquid chromatography; RIA, radioimmunoassay.

ior are observed, each having different states of coordination and using functionally different parts. In the squeeze mode, only the cusps of the three teeth come together simultaneously. In the cut and grind mode, the lateral teeth come together not only with their cusps, but along the entire serrated length of each tooth while the medial tooth remains in its resting position. After this 'cut' phase, the medial tooth is pulled forward in a kind of a power stroke while the lateral teeth grind backwards along the medial tooth file. Many modifications of this basic pattern can be observed.

Both the lateral teeth and the medial tooth can show oscillatory movements alone. Sometimes there are additional openings during the cut and grind mode of activity but we never observe transitional states between these two basic modes. When there are transitions between the two, they occur as all-or-none transitions on a cycle-by-cycle basis.

The effects of proctolin on the behavior of the stomach in the intact animal are quite remarkable in that at different dosages, the proctolin produces movements analogous to either the squeeze or the cut and grind mode which are observed spontaneously. The proctolin can be injected into the semi-open circulatory system while observing the tooth movements with an endoscope. A dose of 0.0001 mg, which we estimate results in a blood level of 10^{-8} M, is the threshold dosage for eliciting chewing, starting after a 10–100-s latency. Injections of 0.001 mg produce chewing in the squeeze mode while a dose of 0.1 mg triggers cut and grind chewing.

It is possible that proctolin injected into live animals could be acting on inputs to the stomatogastric ganglion or at neuromuscular junctions rather than at the level of the CPG. It was therefore of considerable interest to see how various dosages of proctolin applied directly to the STG might effect the gastric motor output pattern. The response of the isolated CPG to bath applied proctolin indicated that the action was primarily at the level of the pattern generating elements. Proctolin had a general excitatory effect on the ongoing rhythm when the stomatogastric ganglion was attached to the commissural and esophageal ganglia. Changes in the ongoing pattern could be observed as early as five seconds after the onset of the perfusion although it was usually 5 min before the effect had become stable. In general there were large increases in burst duration as well as increases in spike frequency in all of the neurons except the LPGs (fig. 7). The strongest effect was on the DG and LG neurons and was accompanied by a phase change in the DG burst. DG, which normally stops at the onset of the LG burst, instead continued firing well into the LG and GM bursts, two neurons which are normally antagonists of DG.

The bath-applied threshold was approximately 10^{-10} M and fully reversible. Increasing dosages of proctolin produced higher amplitudes and steeper rise times of the membrane potentials of LG, MG and DG which also had higher firing frequencies during the burst. Increased doses of proctolin also led to an increased overlap of the DG burst with the LG and GM bursts.

LG and DG appear to be the primary targets for the action of proctolin. For LG, the principal effects was the induction of regenerative plateau properties as revealed by the presence of long-lasting plateau potentials, sensitivity to triggering inputs and the occurrence of oscillatory prepotentials. In DG, however, the effect was to induce endogenous bursting. Under the influence of proctolin, DG, which normally cannot alter the gastric rhythm much, can, when directly depolarized, accelerate the pattern up to three times normal.

In addition to inducing significant changes to the ongoing rhythm, proctolin can trigger gastric mill activity in ganglia which are not cycling. If a combined preparation is perfused with proctolin and then the stomatogastric nerve is blocked with sucrose, the gastric bursting will continue unabated. If the nerve is blocked in the absence of proctolin, the gastric cycling will cease. If proctolin is added to the bath, cycling will resume after 20–30 min of perfusion.

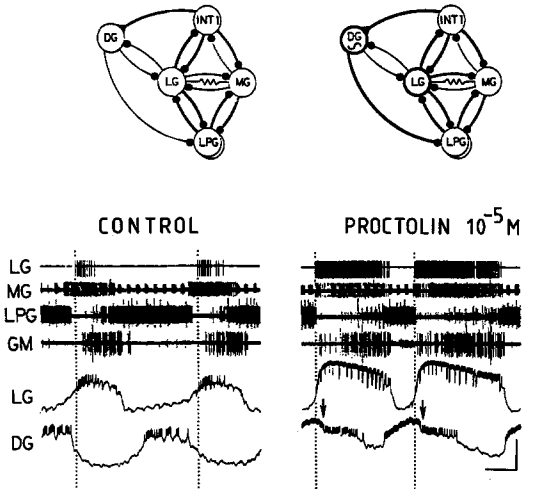

Figure 7. Changes in the in vitro combined gastric pattern as a result of the bath application of proctolin. See text for details.

The overall effects induced by changes in the burst parameters are in agreement with the changes observed in the behavior of the teeth following proctolin injections in vivo. Two of the most prominent changes in the gastric network observed in vitro were the induction of endogenous bursting in DG and plateauing in LG. In addition the apparent synaptic strength of these neurons was augmented after proctolin application.

The squeeze mode

The normal activity pattern for the 'squeeze' mode of chewing in terms of teeth movements, is that the tips of the three teeth come together simultaneously, which, based on the biomechanics of the system would be consistent with short LG/MG bursts and a short gap in LPG firing. This is accompanied by a forward movement of the medial tooth following the closing with only a short delay, and a small backwards movement of the lateral teeth.

The cut and grind mode

Under the influence of proctolin, the lateral teeth remain closed for a longer period of time as a result of the long burst duration of LG/MG. What accounts for the subsequent delay in the forward movement of the medial tooth and backward movement of the lateral teeth? Based on the intracellular data, this delay can be expected since DG and GM are coactivated, thus holding the medial tooth back until the force generated by the GMs overcomes that of the DG. These movements are consistent with the cut and grind mode of chewing elicited by high doses of proctolin injected into the animal. This chewing mode is not only characterized by a different pattern of motorneuron firing and a different trajectory for movements of the teeth, but also demonstrates the use of functionally different parts of the three teeth compared to the squeeze mode.

Effects of proctolin on the pyloric rhythm

The pyloric system is strongly influenced by the application of proctolin [20, 24, 25]. Like the gastric system, proctolin can initiate bursting in the pyloric system of isolated stomatogas-

tric ganglia. There are major changes to the burst pattern, the most conspicuous being an increase in the number of spikes per burst. Proctolin also increases the frequency and amplitude of AB firing, the probable driving source for the increase in frequency of slowly cycling preparations. In the pyloric system the threshold concentration was approximately 10^{-10} M proctolin and the effects were dose-dependent.

Effects of amines on the pyloric system

Perhaps the most elegant series of studies on the effects of neuromodulators has been on the effects of dopamine, octopamine and serotonin on the pyloric rhythm [15]. All three of these substances produce unique alterations of the pyloric rhythms, some of which are dose-dependent. After relatively low concentrations of dopamine (10^{-6} M), the polyoric rhythm slows down with some units stopping entirely. At higher concentrations (10^{-4} M), some units will shut off and others burst at a higher than normal frequency. Serotonin and octopamine both cause complex effects on the pyloric rhythm [9,10]. Each neuron can be synaptically isolated from other neurons of the pyloric circuit by using the dye-activated photoinactivation technique [27], and individual responses to the amines determined. While all of the cells respond differently to each amine, a common feature is the activation of rhythmic activity in the AB neuron. While there is no method at present to synthesize the cooperative effects of all the pyloric neurons, this work does show clearly that the receptors for each of the amines and the cellular processes which are responsible for the electrical manifestations are different and complex.

Conclusions

Our work on the lobster stomatogastric ganglion central pattern generators has shown that the basic neuronal circuitry can produce stable motor patterns of different types when modulated by chemical substances which are the same, or very similar, to those found in the animal. Such different output states continue to be influenced by other shorter-acting neural inputs to the CPGs such as those from proprioceptors and other CPGs. Each output state, however, appears to be a significant variation of the basic pattern and our work with proctolin indicates that the new pattern has behavioral significance for at least this substance. We do not as yet understand what is the importance of all of the other chemically-induced states, nor do we know the physiological relationship between conventional fast synapses and the slower-acting neuromodulatory inputs. Even the results of more than one substance being present at the same time or the interactions of different concentrations has not been investigated yet. We do know that neuromodulatory substances can effect a single CPG so as to alter its output state in a stable and characteristic way by inducing changes in membrane and synaptic properties. This has the effect of functionally 'rewiring' the circuit for as long as the modulator is present in a high enough concentration.

The idea that the neural networks which comprise the CPGs of invertebrates are rigid 'hard-wired' neural circuits needs to be replaced with the notion of extremely flexible circuits which can be 'sculpted' out of the anatomical networks by the actions of modulators. The key to understanding how modulators are able to switch a network into various states will be to understand how the release of the neuromodulatory substances is controlled by the nervous system, by other hormones and perhaps by the action of the genes which manufacture them.

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Multifunctional interneurons in behavioral circuits of the medicinal leech

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Summary. We are using the medicinal leech to study the neuronal basis of behavioral choice. In particular, we are recording from neurons, both extracellularly and intracellularly, in preparations that can express three different behaviors: the shortening reflex, crawling and swimming. We have found that particular mechanosensory neurons can elicit any of the behaviors, and that the movements are produced by just four sets of muscles, each controlled by a small number of motor neurons. Hence, there must be three different pattern-generating neuronal circuits, each of which can be activated by the same set of sensory neurons. We are studying how the choice is made among the three behaviors by recording, while one behavior is being performed, from neurons known to be involved in the initiation of the other two. We have found that an interneuron, cell 204, which is known to initiate and maintain swimming, is also active during shortening and crawling. The activity level in this interneuron can influence whether a mechanosensory stimulus produces shortening or swimming. The neuronal mechanisms by which this choice is normally effected awaits further elucidation of the circuits that elicit and generate shortening and crawling.
Key words. Behavioral state; command neuron; gating neuron; leech; mechanosensory neuron; motor neuron; pattern-generating neuron; rhythmic motor pattern; shortening reflex; trigger neuron.

Introduction

It is commonly observed that an individual animal can respond very differently to a well-defined and controlled stimulus when it is presented on two different occasions. This variability has been attributed to such factors as the animal's motivational state, its ontogenetic development, or the environmental context in which the stimulus is presented. For instance, a dog's response to a meaty bone may depend on whether it is hungry or sated, whether it is newborn or mature, and whether or not there is a receptive mate nearby. A variety of terms have been used to describe these variations in behavioral state: "mood", "motivation" (Eibl-Eibesfeld[9]), "behavioral set" (Evarts et al.[10]), or "drive" (von Holst[27]). Such states are deduced from experiments monitoring an animal's response to a particular stimulus under different conditions (McFarland & Sibly[20]) or at different times (von Holst[27]), or to simultaneous presentation of two different stimuli that when presented individually evoke different behaviors (Davis[5,7,8]). Behavioral threshold can be