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The Pyloric Pattern of the Lobster (*Panulirus interruptus*) Stomatogastric Ganglion Comprises Two Phase-Maintaining Subsets

SCOTT L. HOOPER
Neurobiology Program, Department of Biological Sciences, Ohio University, Athens, OH 45701
Hooper@ohiou.edu

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Abstract. The pyloric pattern approximately maintains phase over a three- to fivefold frequency range when the pattern is defined by the pacemaker burst beginning. However, in this reference frame certain pattern elements maintain phase better than others, which suggests phase-maintaining subgroups might exist. Reanalysis of these data in reference frames defined by each element shows the pattern contains two groups of pattern elements within which phase is well maintained but between which maintenance is relatively poor. A third element shows intermediate maintenance with each group. If ventricular dilator neuron burst beginning (VDB) is chosen as pattern beginning, all members of one group occur early in the pattern, all members of the other occur late in the pattern, and the intermediate element occurs between the groups. Thus, at least for phase maintenance, VDB is a "natural" pyloric pattern beginning. These results suggest full description of complex patterns is best achieved by analysis in many reference frames.

Keywords: stomatogastric system, pyloric neural network, crustacea (lobster, *Panulirus interruptus*), motor pattern, phase maintenance

Introduction

I have shown that, when the beginning of pyloric dilator (PD, PDB) neuron activity is chosen to define pattern beginning, the pyloric network maintains phase fairly well (i.e., delays from PDB to the other pattern elements change proportionally with period) over a three- to fivefold frequency range. However, it was apparent that, in this reference frame, certain of the pattern's elements maintained phase considerably better than others (Hooper, 1996). This could arise because the network is divided into subsets within which phase is well maintained but between which it is not. I therefore desired to fully characterize the response of this network as frequency changes, and to identify any constant delay or constant phase subsets within it, by determining the delay changes with respect to all pyloric pattern elements, not just PDB. This characterization is difficult because rhythmic patterns appear different in different reference frames, and certain interrelationships among neuronal activities are most easily discerned in certain reference frames.

In order to fully appreciate this issue, it is useful to provide a theoretical example. Figure 1 shows a rhythmic pattern comprising three neurons A, B, and C with the following relationships (for simplicity, only the beginnings of neuronal activities are considered):

- Neuron A is an endogenous oscillator (it rhythmically depolarizes and fires bursts of action potentials).
- Neuron B fires with a constant delay after neuron A's activity regardless of cycle period (neuron B shows no phase compensation as cycle period is altered).
- Neuron C's delay after neuron B varies proportionally with cycle period (the time from the beginning

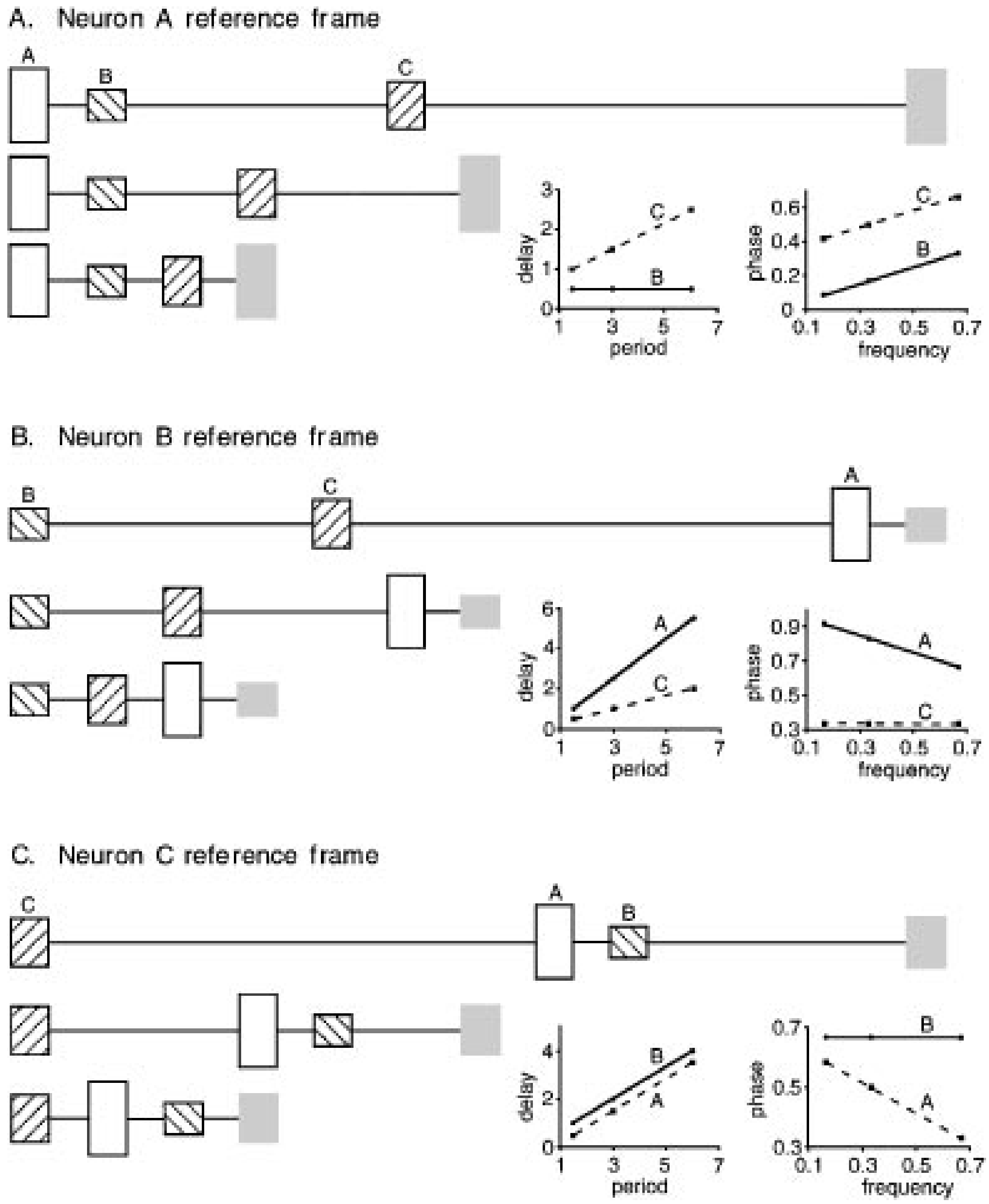

Figure 1. Delay and phase relationships are more apparent in certain reference frames. The three panels show the same pattern at three frequencies in, respectively, neuron A, neuron B, and neuron C reference frames (gray boxes represent the first burst of the next cycle). Note that the pattern appears different in different reference frames; this illusion persists even when several cycles are drawn. Insets are the delay and phase plots associated with each panel. The pattern's phase relationships as cycle frequency changes are as follows: the delay from the beginning of neuron A activity to the beginning of neuron B activity is constant; the delay from the beginning of neuron B activity to the beginning of neuron C activity varies proportionally with changes in neuron A's (the network's pacemaker) period (neuron C maintains phase perfectly with neuron B). These relationships are most apparent in certain reference frames. For instance, the horizontal line for neuron B's delay in neuron A's reference frame (A) shows neuron B maintains constant delay with respect to neuron A, and the horizontal line for neuron C's phase in neuron B's reference frame (B), and for neuron B's phase in neuron C's reference frame (C), show that neurons B and C maintain phase perfectly with each other.

of neuron B's activity to the beginning of neuron C's activity doubles when neuron A's period doubles, and hence neuron C maintains perfect phase with neuron B).

Figure 1 shows the pattern, and the associated delay and phase plots (insets), at three frequencies (each trace in a given panel represents one frequency) using each neuron's beginning (A, neuron A; B, neuron B; C, neuron C) as pattern reference point. There are two points to make about this figure. First, even though the same pattern is shown in each panel, choosing different pattern definers results in patterns that appear different (this illusion persists even when several cycles of the pattern are drawn). Second, the pattern's phase relationships are easier to discern in certain reference frames. For example, it is immediately apparent from the delay plot in A that neuron B maintains constant delay after neuron A, and from the phase plots in B and C that neurons B and C maintain constant phase. In a small, idealized pattern such as this, these relationships can be deduced in any reference frame (e.g., the equal slopes of the neuron B and C phase lines in neuron A's reference frame imply neurons B and C maintain perfect phase). However, in a larger pattern (the pyloric pattern has ten elements) with natural variability, it would be extremely difficult to deduce the pattern's timing relationships from analyses in a single reference frame.

These considerations suggested that the pyloric phase data should be reanalyzed using each pattern element as pattern definer. This reanalysis shows that the pyloric pattern appears to contain two groups of pattern elements within which phase is well maintained but between which phase is comparatively poorly maintained. These results thus suggest that, unless phase maintenance within a pattern is nearly perfect (DiCaprio, 1997), a complete description of phase relationships in complex, multiunit patterns is best obtained using all pattern elements as reference frame definers.

Some of this work has been previously published in abstract form (Hooper, 1995).

Methods

General

All dissections, electrophysiology, and data acquisition were performed as is standard for phase analysis in this preparation (Hooper, 1996). In brief, stomatogastric nervous systems were removed from the animal and continuously superfused with chilled *Panulirus* saline. Extracellular nerve recordings and intracellular neuron recordings were used to identify the pyloric neurons (Selverston et al., 1976), and pyloric frequency was altered by current injection into the anterior burster (AB) neuron. Data were recorded on chart recorder and digital tape. The delays between PDB and the other pattern elements (beginnings and endings of the activities of the pyloric neurons) were measured either by hand or using Spike II (Cambridge Electronic Design) scripts. These data in the PDB reference frame were then transferred to a Macintosh Quadra 950. All subsequent analyses (transformation to other reference frames, phase calculation) were performed using scripts in Kaleidagraph (Synergy Software). Statistical analyses (two-tailed Student's t-test) were performed using JMP Statistical Visualization (SAS Institute, Inc.).

Transformation to Other Reference Frames

Figure 2 shows a diagram of two pyloric cycles in the PDB reference frame. In this frame the pattern's order within a single cycle is (1) PD neuron activity beginning (first trace; PDB), (2) PD neuron activity ending (PDE), (3) nearly simultaneous beginnings of lateral pyloric (LP) and inferior cardiac (IC) neuron activity (second and fourth traces; LPB, ICB), (4) pyloric (PY) neuron activity beginning (third trace; PYB), (5) the endings of LP and IC neuron activity (LPE, ICE), (6) ventricular dilator (VD) neuron activity beginning (fifth trace; VDB), and (7) the endings of PY and VD neuron activity (PYE, VDE). These data can be transformed to the other possible reference frames (PDE, LPB, LPE, PYB, PYE, VDB, VDE, ICB, and ICE) by appropriate algebraic manipulation of the PDB data. Figure 2 shows the transformation into the VDB reference frame. For example, VDB to PDB time equals PDB to PDB time (cycle period in the PDB reference frame) minus PDB to VDB time. These manipulations were used to calculate delays of all pattern elements in all reference frames. Phase was then calculated as phase = delay/period.

The data shown here were drawn from 13 preparations and comprise some 16,000 points in the PDB reference frame. The database in all reference frames comprises some 160,000 values (in certain preparations not all neurons were active). PDB reference frame data had already been extensively checked (Hooper, 1996).

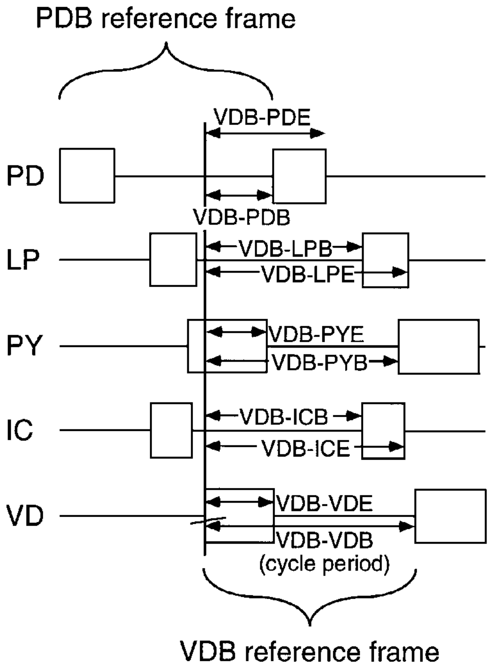

Figure 2. Transformation from PDB to VDB reference frame. Two cycles of the pyloric pattern in the PDB reference frame are shown. The pyloric pattern element ordering is PDB, PDE, LPB and ICB, PYB, LPE and ICE, VDB, PYE, and VDE, after which the pattern repeats. PDB reference frame data can be transformed into any other reference frame by algebraic manipulation. For example, VDB to PDB delay equals PDB to PDB delay (period in the PDB reference frame) minus PDB to VDB delay.

The validity of the transformed data was checked in two ways. First, the delays calculated by each of the nine transformation scripts were verified by hand for all elements in one cycle of one preparation. Second, for each preparation the data of each type (e.g., ICB delay) in every reference frame (a total of some 2,160 graphs) were plotted over the entire frequency range and visually checked for outliers; the transformation of these outliers was then verified.

In the PDB reference frame, phase maintenance of certain pattern elements was anomalous in four preparations in which the PY or IC neurons were silent or only weakly active, and in one preparation in which PD neuron burst duration did not change as period was altered (Hooper, 1996). The data from these preparations were closely examined in the other reference frames. Data for a pattern element from these preparations were excluded from grouped analyses (Figs. 7 and 8) if (1) their delays did not vary linearly with period or (2) the slope or intercept of linear fits to the data in either the delay or phase representations were extremes, and were more than twofold different from the group average. Excluded data are shown as fine dashed lines on Figs. 3 to 6.

Results

Figures 3 through 6 show phase plots of four (ICB, LPB, PYE, and PDE) pattern elements in all reference frames (PDB, PDE, LPB, LPE, PYB, PYE, VDB, VDE, ICB, and ICE). In each plot solid lines are linear fits to the data from individual preparations, fine dashed lines are data from preparations excluded from grouped analysis (see Methods), and the coarse dashed lines with open diamonds, solid squares, and solid circles are, respectively, the average of the fits and the perfect phase compensation and constant delay lines calculated from the average of the fits (see Hooper, 1996).

It is apparent from these figures that the various pyloric elements maintain phase better (the average fit line is closer to horizontal) in certain reference frames. ICB (Fig. 3) maintains phase well (panels surrounded by solid rectangles) in ICE, LPB, LPE, and PYB reference frames (average slope $\leq$0.04), maintains phase moderately (panel surrounded by dashed rectangle) in the PDE reference frame (average slope 0.13), and maintains phase comparatively poorly in PDB, PYE, VDB, and VDE reference frames (average slope $>$0.2 for all). LPB (Fig. 4) maintains phase well in ICB, ICE, LPE, and PYB reference frames, moderately in the PDE reference frame, and comparatively poorly in PDB, PYE, VDB, and VDE reference frames. PYE (Fig. 5) maintains phase well in PDB, VDB, and VDE reference frames, moderately in the PDE reference frame, and comparatively poorly in ICB, ICE, LPB, LPE, and PYB reference frames. PDE (Fig. 6) maintains phase moderately (average slope approximately 0.1) in all reference frames except VDE (average slope 0.18).

This dependence of phase maintenance on reference frame can be shown more succinctly by plotting the average (dark bars) and constant delay (light bars) phase slopes for all elements in all reference frames (Fig. 7) (reference frames (ordinate) have been ordered to group frames with similar average slopes). There are two points to make about this figure. First, as was also seen when phase was analyzed in the PDB reference frame alone (Hooper, 1996), most elements show moderate to large phase compensation (compare light and dark bars. Negative slopes are overcompensation (delays

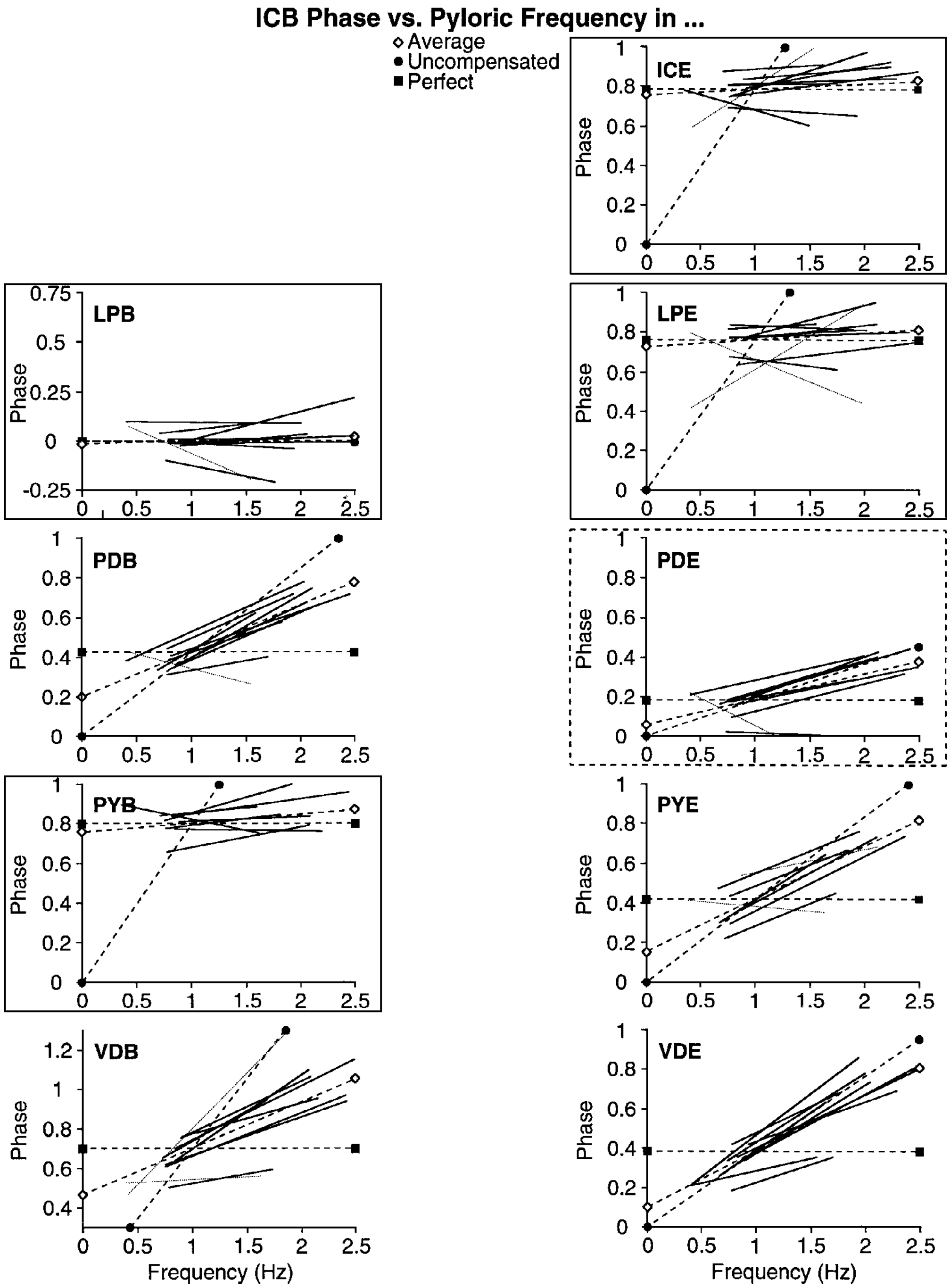

Figure 3. ICB maintains phase well (average phase lines are close to horizontal) in ICE, LPB, LPE, and PYB reference frames (solid rectangles), moderately in PDE's reference frame (dashed rectangle), and comparatively poorly in PDB, PYE, VDB, and VDE reference frames. Each plot shows (in the reference frame indicated in the plot's upper left corner) linear fits to the ICB phase (phase = delay/period) data from individual preparations (solid lines), the average of these fits (coarse dashed line with open diamonds), and the perfect phase (coarse dashed line with solid squares) and constant delay (coarse dashed line with solid circles) lines calculated from the average fit. Also shown (fine dashed lines) are data from preparations that were excluded from group analysis (see Methods).

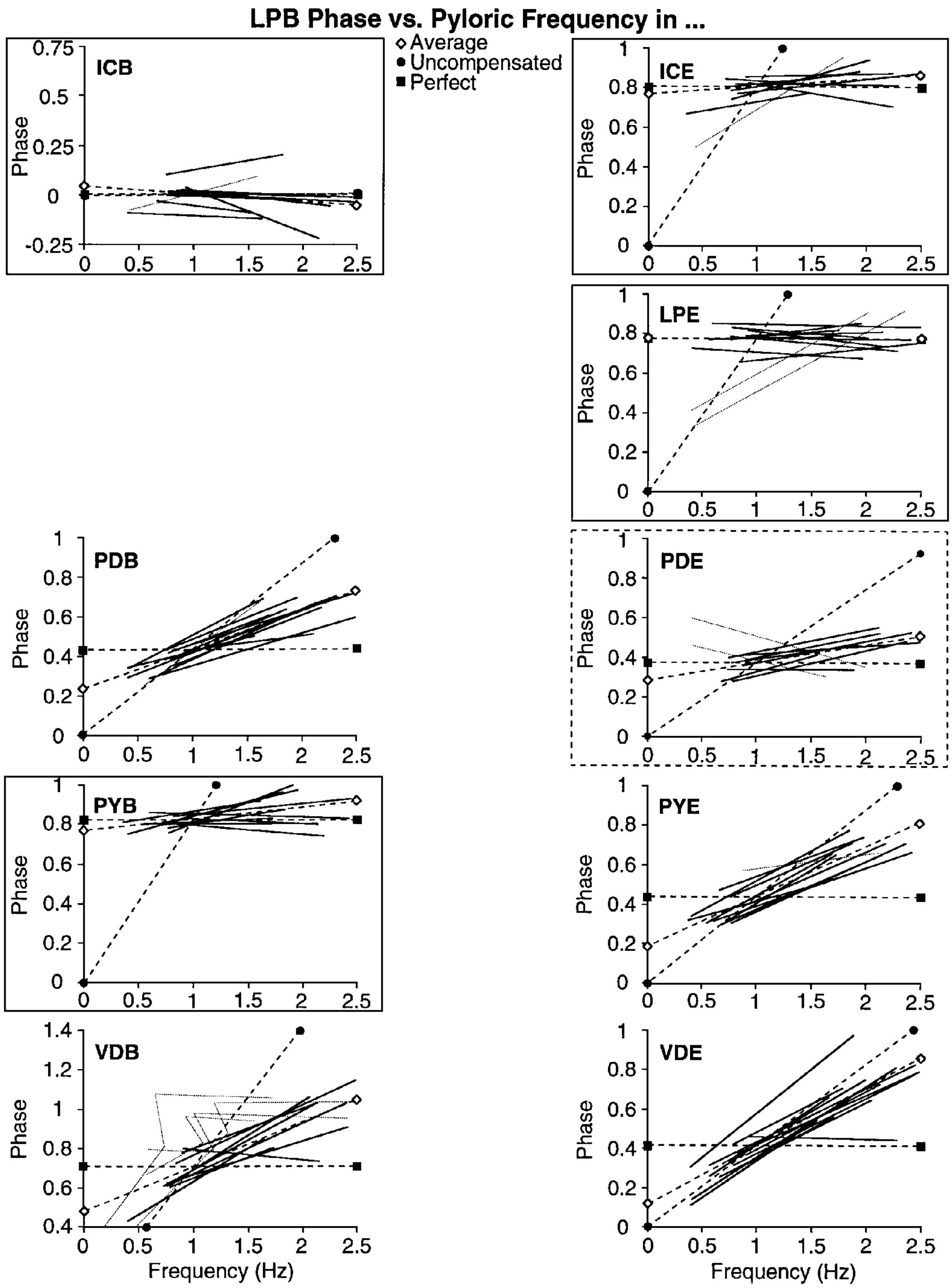

Figure 4. LPB maintains phase well (average phase lines are close to horizontal) in ICB, ICE, LPE, and PYB reference frames (solid rectangles), moderately in PDE's reference frame (dashed rectangle), and comparatively poorly in PDB, PYE, VDB, and VDE reference frames. See Fig. 3 legend for definitions of lines and symbols.

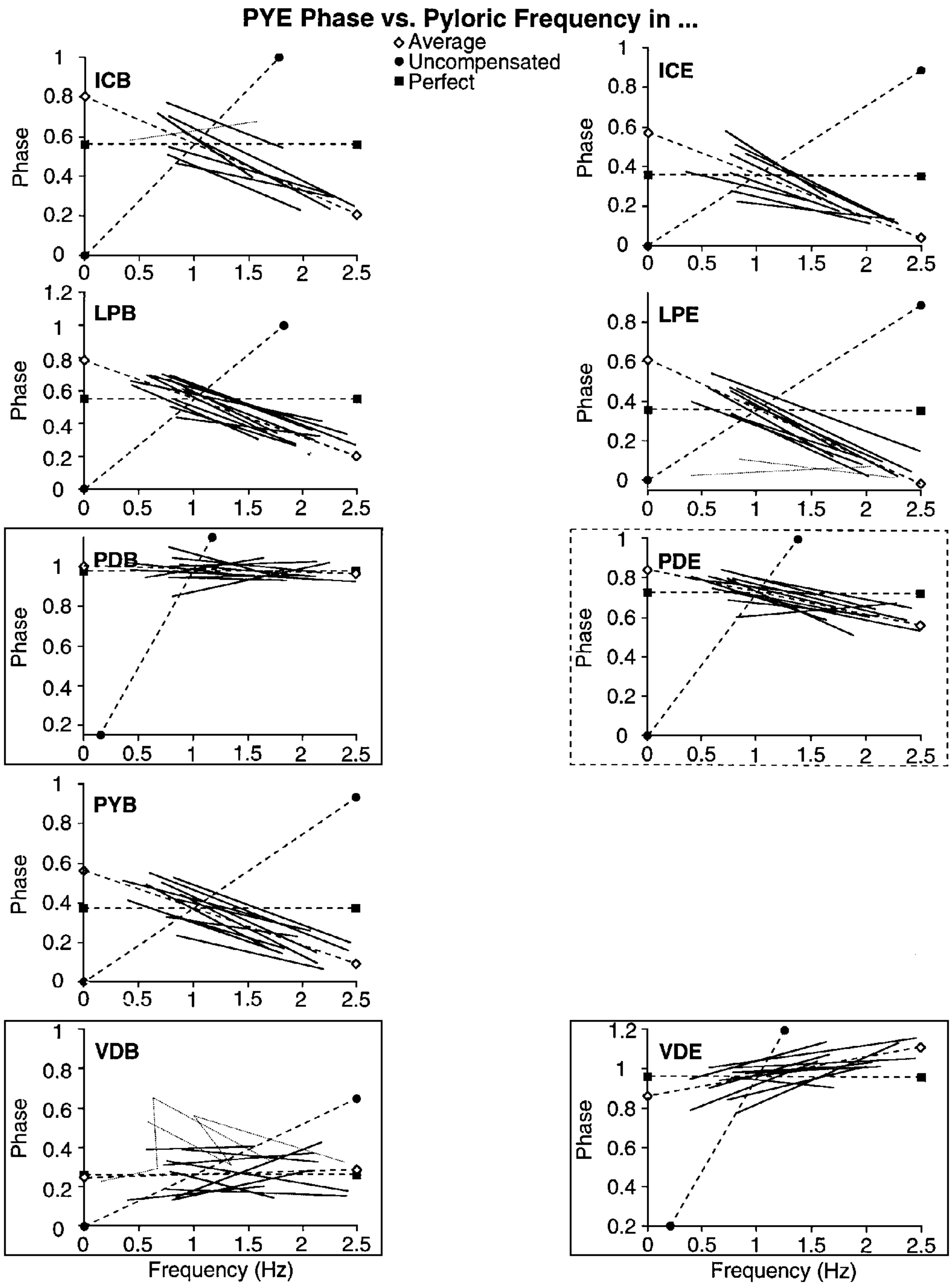

Figure 5. PYE maintains phase well (average phase lines are close to horizontal) in PDB, VDB, and VDE reference frames (solid rectangles), moderately in PDE's reference frame (dashed rectangle), and comparatively poorly in ICB, ICE, LPB, LPE, and PYB reference frames. See Fig. 3 legend for definitions of lines and symbols.

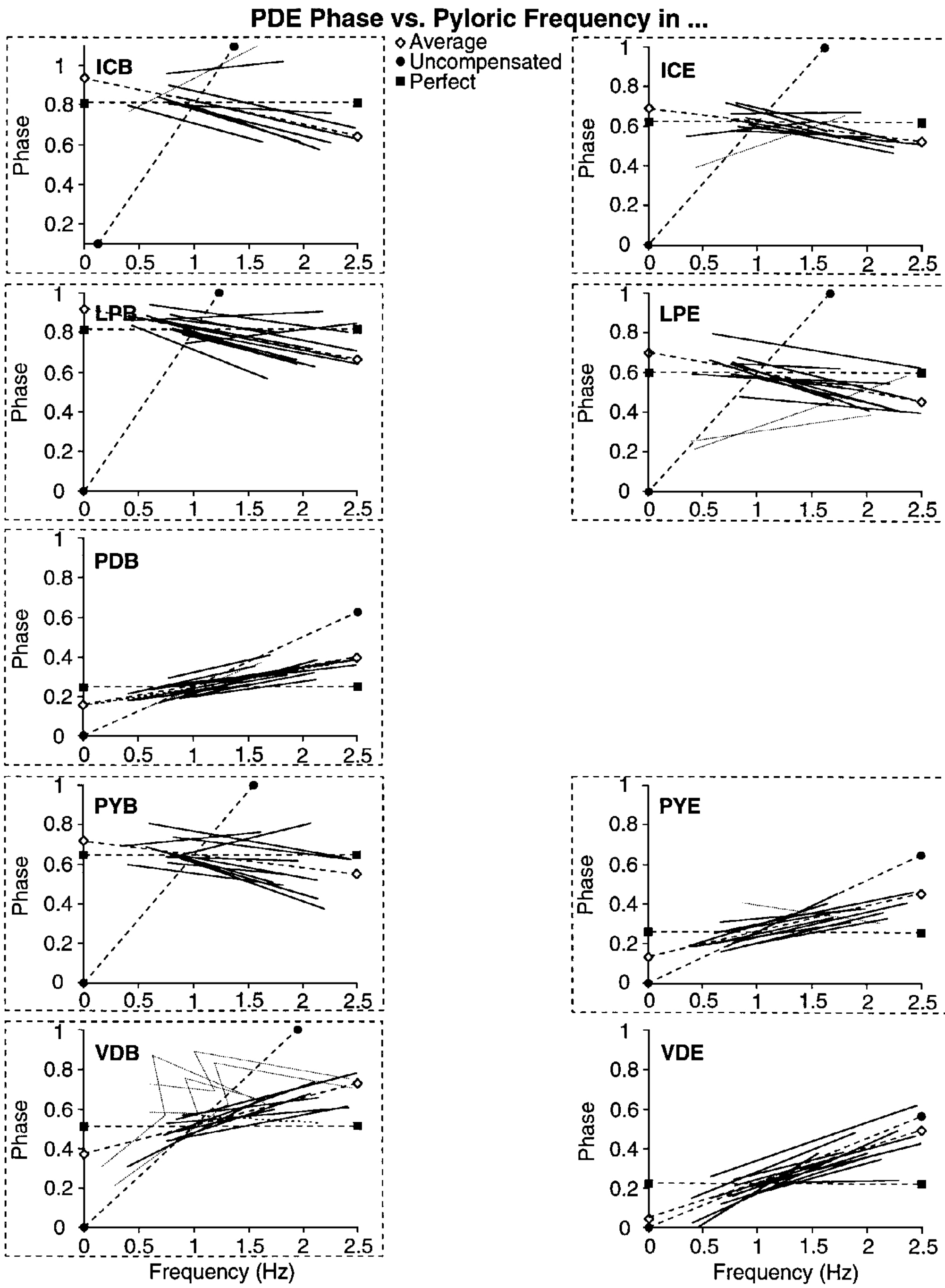

Figure 6. PDE maintains phase moderately in almost all reference frames (dashed rectangles). In all plots except VDE the average phase slope (coarse dashed line with open diamonds) is moderate, and there is no segregation between good and comparatively poor phase maintainers similar to that shown in Figs. 3 to 5. See Fig. 3 legend for definitions of lines and symbols.

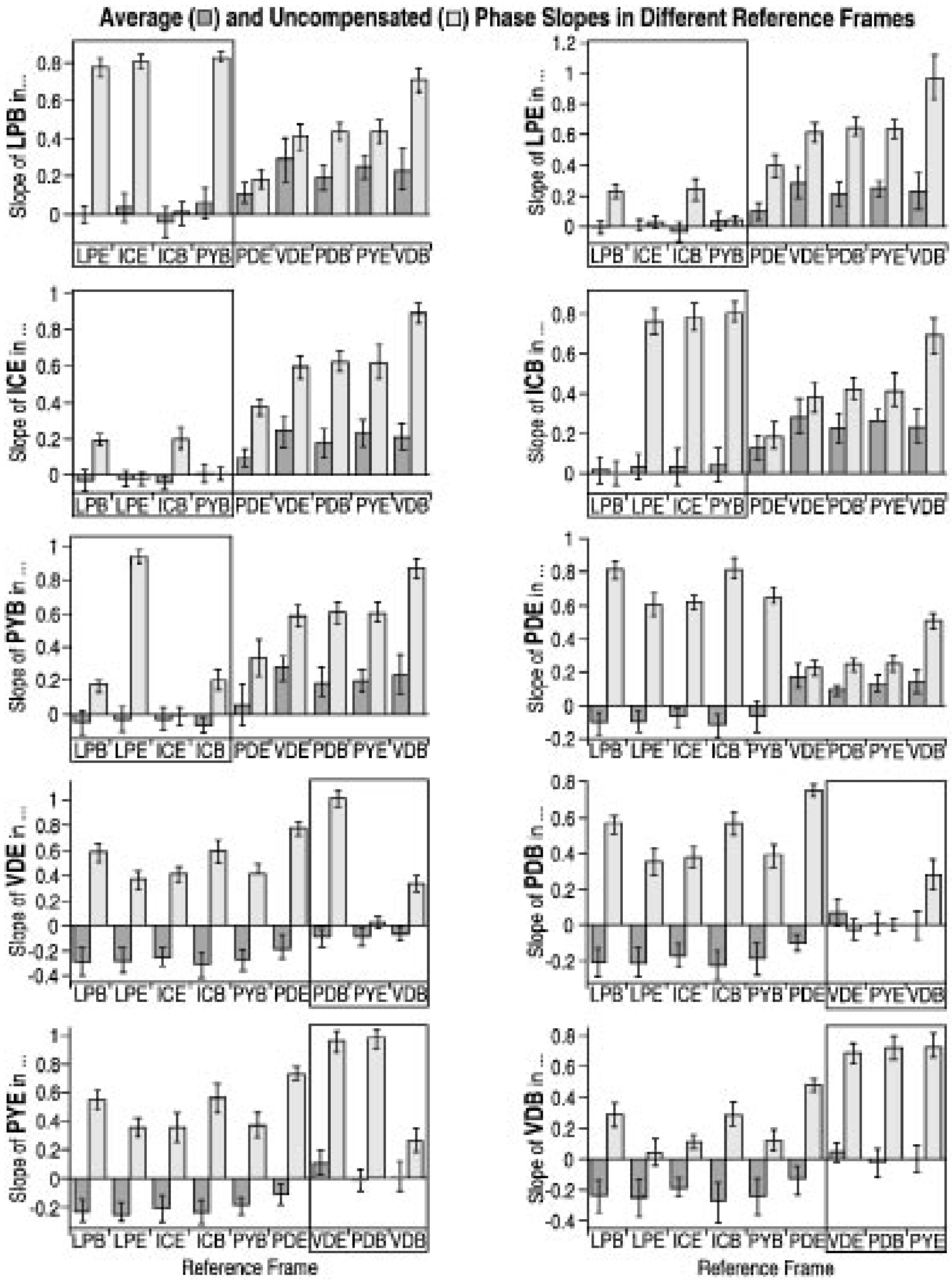

Figure 7. Plots of average (dark bars) and constant delay (light bars) phase slopes of all pattern elements in all reference frames. Each plot shows phase slopes of the noted pattern element in all reference frames. Almost all pyloric elements, regardless of reference frame, show moderate to large phase compensation (compare dark and light bar amplitudes). Rectangles surround reference frames in which the pattern element maintains phase well. Reference frame order has been arranged to maximize these groupings and is the same in all plots (except that each plot's own pattern element, of course, is lacking as a reference frame). Two groups (left rectangles for LPB, LPE, ICE, ICB, PYB; right rectangles for VDE, PDB, PYE, and VDB) in which phase is well maintained are apparent. PDE shows moderate phase maintenance in all reference frames. Constant-delay phase slopes are averages of constant-delay phase lines calculated individually for each preparation. Error bars are standard deviations of the mean.

A. Pattern Element Phase Slopes in All Reference Frames

Reference Frame (columns); Pattern element (rows)

	LPB	LPE	ICE	ICB	PYB	PDE	VDE	PDB	PYE	VDB
LPB		.00	.04	-.04	.06	.11	.30	.20	.25	.23
LPE	-.01		.01	-.04	.03	.10	.29	.21	.25	.23
ICE	-.03	-.02		-.04	.01	.09	.25	.18	.24	.21
ICB	.01	.03	.03		.04	.13	.28	.23	.27	.24
PYB	-.06	-.04	-.03	-.07		.05	.28	.18	.20	.23
PDE	-.10	-.10	-.07	-.12	-.07		.18	.09	.13	.14
VDE	-.29	-.28	-.25	-.31	-.27	-.19		-.09	-.08	-.07
PDB	-.21	-.21	-.17	-.23	-.20	-.10	.07		.02	.01
PYE	-.23	-.25	-.21	-.24	-.19	-.11	.10	-.01		.02
VDB	-.23	-.25	-.19	-.27	-.25	-.12	.05	-.02	.01	

B. With Respect to Phase Maintenance, VDB Is a "Natural" Pattern Defining Element

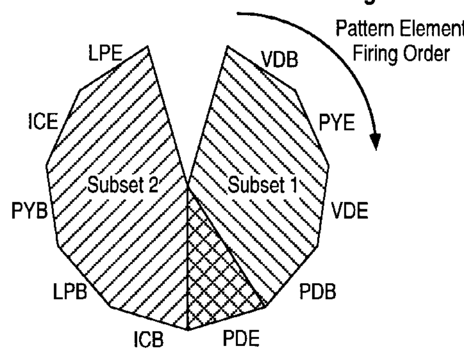

Figure 8. A: Matrix of pattern element average phase slopes in all reference frames. Columns are the reference frames and rows are the pattern elements (e.g., LPB's average phase slope in LPE's reference frame is 0.00, in ICE's reference frame is 0.04, etc.). Pattern element and reference frame order are the same as in Fig. 7. Two phase-maintaining groups (LPB, LPE, ICE, ICB, PYB (subset 1, upper left dark rectangle) and VDE, PDB, PYE, VDB (subset 2, lower right dark rectangle); all phase slopes $<\pm 0.1$, average of magnitudes ≤ 0.04 for both groups) are immediately apparent. The two groups maintain phase comparatively poorly with each other (best slope -0.17, worst 0.30, average of magnitudes 0.24 ± 0.04). PDE (gray) maintains phase moderately with each group (best slope 0.05, worst 0.18, average of magnitudes 0.10 ± 0.03). Statistical significance of groupings (using values above the diagonal): the LPB, LPE, ICE, ICB, PYB and the VDE, PDB, PYE, VDB groups each differ from PDE at $p < 0.001$. Each subset and PDE differ from the outgroup (the values in the upper right box) at $p < 0.001$. B: If VDB is chosen as pattern definer, these groups can be associated with temporally related groups of pyloric activity. When VDB begins the pattern (see Fig. 2), the next three elements are PYE, VDE, and PDB. These four elements also form the phase-maintaining group (subset 2) in the lower right of Fig. 8A. PDE is the next pattern element and also maintains phase moderately with both phase-maintaining groups. ICB, LPB, PYB, ICE, and LPE constitute both the upper left phase-maintaining group (subset 1) and the next pattern element sequence. LPE and VDB do not maintain phase well, which is represented by the gap between them.

more than double with period doubling)). Second, there appear to be two distinct subsets (rectangles) of the pattern within each of which phase is well maintained but between which phase is comparatively poorly maintained. The first subset consists of LPB, LPE, ICE, ICB, and PYB and second of VDE, PDB, PYE, and VDB.

These relationships can be more clearly seen in a matrix of pattern element average phase slope versus reference frame (Fig. 8A). In this matrix reference frames are the columns and pattern elements are the rows (e.g., LPB has a slope of 0.00 in the LPE reference frame, a slope of 0.04 in the ICE reference frame, etc.). In theory, the corresponding values above and below the diagonal (e.g., LPB in the LPE reference frame and LPE in the LPB reference frame) in this matrix should be equal in magnitude and opposite in sign. In fact, the magnitudes of all corresponding pairs are very close (maximum difference 0.03), and none are statistically different even at $p < 0.2$.

The dependence of pyloric element phase maintenance on reference frame is now immediately apparent. LPB, LPE, ICE, ICB, and PYB (dark rectangle, upper left corner) all maintain phase in each other's reference frames within a slope of ± 0.07 (average of magnitudes, 0.03 ± 0.02). VDE, PDB, PYE, and VDB (dark rectangle, lower right corner) maintain phase in each other's reference frames within a slope of ± 0.10 (average of magnitudes, 0.04 ± 0.03). Pattern elements in one group maintain phase comparatively poorly in reference frames defined by the other group's elements (upper right and lower left rectangles; best slope -0.17, worst 0.30, average of magnitudes 0.24 ± 0.04). PDE phase slopes in the various reference frames, and the slopes of the other pyloric elements in the PDE reference frame (gray), are intermediate between good and relatively poor phase maintenance (best 0.05, worst 0.18, average of magnitudes 0.10 ± 0.03), and thus, at least on an observational level, PDE links the two group's activities.

Consideration of these data and the pattern element orders obtained using different pattern definers reveals that if the pattern is defined by the beginning of VD neuron activity (VDB), the phase-maintaining subsets in Fig. 8A can be associated with temporally related groups of pyloric activity. That is, as is shown in Figs. 8B and 2, if VDB is chosen to start the pattern, the next three pattern elements are PYE, VDE, and PDB. These four elements also form the lower right phase-maintaining group (subset 2) shown in Fig. 8A. The next pattern element is PDE, which maintains phase moderately with both groups in Fig. 8A. ICB, LPB, PYB, ICE, and LPE constitute both the next sequence of pattern elements, and the upper left phase-maintaining group (subset 1) in Fig. 8A. LPE and VDB do not maintain phase well (slope 0.23), which is represented in Fig. 8B by the gap between them.

Discussion

These data show that the pyloric pattern contains two subsets within which phase is well maintained (phase slope $\leq$0.04). Relative to the maintenance observed within a subset, each subset maintains phase moderately (slope approx. 0.1) with another pattern element, PDE, and maintains phase with the other subset comparatively poorly (slope approx. 0.2). These conclusions are consistent with data analyzed only in the reference frame defined by the pyloric pacemaker (PDB column in Fig. 8A; Hooper, 1996). However, they would have been difficult to draw, and much less firm, without performing analyses in all reference frames.

Considerable efforts were made to identify trivial explanations of these results. One such explanation would be that the differing degrees of phase maintenance are related to the differing pattern element orders in the various reference frames (e.g., perhaps elements that occur earlier in a given frame always maintain better phase). Although this is true when VDB is chosen as pattern definer, it is not true in general (e.g., VDB is late in the PDB reference frame and maintains phase well, whereas LPE is early and does not; ICB is early in the LPB reference frame and maintains phase well, whereas VDE is late and does not).

Tight phase maintenance also "trivially" arises if the pattern element used to define the reference frame inhibits a pattern element with zero delay, since the ending of the inhibited neuron's activity will then necessarily maintain phase perfectly in that frame (zero delay gives zero phase regardless of frequency). This explanation is probably correct for some "ending-beginning" pattern element pairs (e.g., PYE phase maintenance in the PDB reference frame). However, it cannot explain most phase maintenances because generally there is a significant overlap or delay (see Fig. 2) between the ending of a pyloric neuron's activity and the beginning of the activity of the neurons that inhibit it (for example, there is a significant overlap between LPE and PYB, and a significant delay between LPE and VDB, even though the PY and VD neurons inhibit the LP neuron). Furthermore, it cannot explain why the beginnings of neuronal activities maintain phase well in "beginning" reference frames (e.g., LPB maintains phase well in ICB and PYB time). The most parsimonious conclusion is thus that the pyloric pattern actually contains two phase-maintaining subsets.

Is the Correlation between Phase Maintaining Subsets and Pattern Element Order Significant?

The correlation between the two phase-maintaining groups and pyloric pattern element order when VDB defines the pattern is striking, but to correctly interpret this observation it is necessary to calculate the likelihood this association arose by chance. Consider the 10-element sequence ABCDEFGHIJ. The correlation between grouping and order noted in Fig. 8B is equivalent to all permutations of this sequence in which ABCD and FGHIJ are kept together (ABCD-E-FGHIJ, ADCB-E-FGHIJ, ADCB-E-GHJIF, etc.) plus (since the sequence does not have a defined beginning, i.e., it could also be BCDEFGHIJA) all permutations in which BCDE and GHIJA are kept together plus all permutations in which CDEF and HIJAB are kept together, etc. The number of such permutations is 10 (the number of sequence beginnings) times 4! (the number of permutations of 4 objects taken 4 at a time) times 5! (the number of permutations of 5 objects taken 5 at a time). There are 10! possible permutations of the sequence, and hence the likelihood of the observed correlation is $(10)(4!)(5!)/10! = 1/126$ or 0.8%.

This correlation is thus unlikely to be due to chance, and hence, at least with respect to phase maintenance, VDB represents a "natural" pattern defining element. When the pattern is observed from this perspective, it consists of early (VDB, PYE, VDE, and PDB) and late (ICB, LPB, PYB, ICE, and LPE) phase-maintaining groups joined by PDE.

Implications for Pyloric Pattern Generation

The mechanisms underlying pyloric phase maintenance are only beginning to be explored (Hooper, 1994), but three points should be made here. First, there is no obvious correlation between the observed phase-maintaining subsets and the network's synaptic connectivity. Second, the separation of the beginning and ending of PD and PY neuron activities to different subsets may initially be surprising. However, pyloric neuron activity beginning and ending are induced by different neurons (Selverston et al., 1976) and involve different mechanisms (Russell and Hartline, 1982). It is thus not unexpected that neuron activity beginning and ending could be relatively uncorrelated.

Third, the fact that PDE is an observational link between the subsets is *not* evidence that PD neuron activity mechanistically links them, and the division of the pyloric pattern into different phase-maintaining groups is *not* evidence that pattern elements in one phase-maintaining group are not mechanistically involved in determining the timing of the other group's pattern elements (e.g., the VD neuron is almost certainly involved in determining IC neuron timing (Hooper and Moulins, 1990)). The pyloric network has a highly interconnected synaptic connectivity pattern, and pyloric activity is a distributed function of the entire network (Warshaw and Hartline, 1976; Hartline, 1979; Miller and Selverston, 1982b; Selverston et al., 1983a, 1983b; Marder, 1984; Selverston and Moulins, 1985; Hooper and Marder, 1987; Hooper and Moulins, 1990; Kepler et al., 1990; Marder et al., 1993). Consequently, causal mechanisms cannot be derived from simple observations of whole network activity.

Implications for Other Systems

Two aspects of this work are of particular concern with respect to the description and analysis of systems in general. First, the pyloric network has been intensively studied over the past twenty-five years and is among the best understood of all neural networks. It is striking that despite this work the possible presence of two subsets within the network has not been noted before. This, of course, is solely because the particular alteration of the system needed to reveal these subsets (changing frequency by current injection into the AB neuron) had not been previously applied, but it does highlight the importance of (1) altering system activity in all possible ways in investigations of neural network function, (2) our inability to predict all aspects of network function even in extremely well-described networks, and (3) the possibility that other apparently unitary networks may similarly contain as yet undescribed substructure.

Second, it is important to again note that it would have been extremely difficult to arrive at the interpretations presented here without analyzing the data in multiple reference frames, and that describing multiphase patterns (that do not maintain phase perfectly) in only one reference frame could introduce perceptual biases. For instance, there is a strong temptation in pacemaker-driven networks to use this element as pattern definer and to identify pacemaker activity as the central aspect of network activity. The data shown here, however, show no clear hierarchical ascendancy of the pacemaker ensemble (represented by the PD neuron) in pyloric phase maintenance (Fig. 8), even though the pacemaker ensemble strongly inhibits all other network neurons (Selverston et al., 1976; Eisen and Marder, 1982) and is generally the network's rhythmic driver (Selverston, 1977; Miller and Selverston, 1982a). This work thus suggests that multiphase patterns are best described by analysis in many reference frames, and that it is necessary to guard against possible perceptual biases when such analyses are not performed.

Acknowledgments

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