

Comparison of motor patterns in the intact and deafferented flight system of the locust

I. Electromyographic analysis

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Summary. 1. Electromyographic recordings were made from a restricted set of flight muscles in *Locusta migratoria* during flight sequences in intact animals and in animals which had sensory input from the wings and wing-hinges removed. Recordings in intact animals revealed two new features of the flight motor pattern. The first was that the onset of elevator activity followed the onset of depressor activity at an almost constant interval which was independent of wingbeat frequency (Fig. 3), and the second was an almost synchronous activation of the elevator muscles in the two pterothoracic segments (Figs. 5, 6).

2. The most obvious and consistent effects of deafferentation were a decrease in wingbeat frequency and an increase in the magnitude and variability of the interval between the onset of depressor activity and the onset of the following elevator burst. This interval increased from about 20 ms in the intact animals to between 50 and 90 ms in the deafferented animals and became strongly dependent on wingbeat frequency (Fig. 10). The number of spikes/cycle in elevators became more variable and usually increased after deafferentation (Fig. 8). By contrast, deafferentation produced little change in the interval between the termination of the elevator activity and the onset of the following depressor activity (Fig. 10), nor did it produce any major changes in the pattern of discharge in the depressors (Fig. 7).

3. The relative timing of activity in homologous muscles in the two segments became more variable following deafferentation. However, in most deafferented preparations the hindwing depressor activity led the forewing depressor activity by 5 to 10 ms, this being similar to the intersegmental delay observed in intact animals. In all deafferented

preparations the elevator activity in the forewings led elevator activity in the hindwings rather than being almost synchronous as in intact animals (Fig. 6).

4. We conclude that deafferentation produces qualitative changes in the flight motor pattern to such an extent that the overall deafferented pattern is distinctly different from the intact motor pattern. In the following paper we show these changes to be due to major changes in the components of synaptic input to elevator motoneurons.

Introduction

The flight system of the locust was one of the first in which it was conclusively demonstrated that rhythmic motor activity resembling the normal flight motor pattern could be generated following deafferentation (Wilson 1961). Subsequently it was found that virtually all rhythmic motor systems have the capacity for generating rhythmic motor sequences in the absence of sensory feedback (Delcomyn 1980). The general nature of this phenomenon led to the concept of central pattern generation, that is, that the basic motor pattern is established by a central neuronal network now referred to as the 'central pattern generator' (Grillner 1985). Recently, however, the question has been raised of whether the concept of central pattern generation is applicable to systems with a large degree of sensory regulation (Pearson 1985). In the flight system of the locust, for example, it has been found that wing proprioceptors have strong phasic effects on the activity of the motor pattern and some proprioceptors appear to be intimately involved in the generation of the normal rhythmic activity (Wendler 1983; Möhl 1985a, b; Pearson et al.

1983). Furthermore, electrical stimulation of the wing stretch receptor afferents in deafferented preparations can radically alter the discharge patterns of identified flight interneurons (Reye and Pearson, in prep.). These observations indicate that the central oscillator active in deafferented preparations is modified substantially by sensory feedback in the intact animal. The question we are interested in is whether these modifications are such that the central oscillator functioning in deafferented preparations no longer provides the basis for generating the normal motor pattern in intact animals. One approach towards gaining an answer to this question is to compare the patterns of neural activity in intact animals and deafferented preparations, and the functional interactions of neurons in the two situations.

Rather surprisingly there has been no extensive analysis of the changes in the flight motor pattern following deafferentation. In his original study Wilson (1961) reported that the main effect of deafferentation was a slowing of the repetition rate of the cyclic activity without any major change in the overall motor pattern. Thus he concluded that sensory '...input...increases frequency as well as affecting small changes in pattern'. He did note, however, that the onset of elevator activity occurred later in the interval between successive depressor spikes and concluded that most of the decrease in repetition rate was due to an increase in the interval from depressor to elevator activity. More recently it has been reported that the usual motor pattern in deafferented preparations is one in which depressor activity follows elevator activity with a relatively constant latency independent of wingbeat frequency (Robertson and Pearson 1982; Hedwig and Pearson 1984). This differs from the situation reported in intact animals where the elevator to depressor interval is dependent on cycle time (Waldron 1967). Deafferentation also influences the burst patterns in individual motoneurons. In elevator motoneurons, for example, the number of spikes/cycle becomes more variable from animal to animal and this number often increases following deafferentation (Kutsch 1974).

Despite these previous observations there is still a considerable lack of knowledge about the changes in the flight motor pattern following deafferentation. For example, there has been no quantitative analysis of the dependence of the relative timing of elevator and depressor activity on flight frequency following deafferentation, nor has the pattern of individual motor units before and after deafferentation been analysed in detail. This lack of information prevents a clear assessment of the

extent to which the motor pattern in intact animals resembles the pattern in deafferented preparations. In this investigation we have compared in a quantitative manner the patterns of activity in a subset of flight muscles in intact and deafferented animals. We describe a number of consistent changes in the flight motor pattern following deafferentation, and conclude that sensory feedback must have a major role in the normal patterning of activity in elevator motoneurons.

Materials and methods

The aim of this investigation was to determine the major changes that occur in the flight motor pattern following deafferentation. To do this we studied the patterns of activity in a restricted number of flight muscles from which electromyographic (EMG) recordings could be made reliably in intact and deafferented preparations. Although no attempt was made to record the activity from all the major flight muscles, our intracellular analysis (Wolf and Pearson 1986) revealed that the major effects of deafferentation we report here are apparent in all flight motoneurons. The muscles selected for study were the anterior tergo sternals and the first basalars (muscles 83 and 97 of the forewings and 113 and 127 of the hindwings). There were three reasons for this choice: 1) each muscle is innervated by a single fast motor axon (Tyner and Altman 1974), 2) they are direct flight muscles which only function to move the wings, and 3) electromyograms can be easily and reliably recorded from these muscles. Although recordings were often made simultaneously from all four muscles on one side, the more usual procedure was to record pair-wise from different combinations of muscles.

Figure 1 shows the experimental arrangement for recording the patterns of activity in both intact and deafferented animals. After removal of the legs, each animal was tethered by the pronotum to a rigid rod and held upside down facing the opening of a wind tunnel. The size of the opening of the wind tunnel was 10×8 cm. The wind velocity was kept within the range of 2 to 3 m/s; in most experiments it was set at 2.5 m/s. Mounting the animal in an inverted position had the advantage that the attachment sites of the tergo sternals and first basalar muscles on the sternum were accessible for placement and repositioning of the electromyographic recording electrodes. Single recording electrodes (100 μ m copper wires insulated except for the tips) were placed just through the cuticle of the sternum in the region of muscle attachment (Fig. 1B). The indifferent electrode was placed in the abdomen. To demonstrate that the EMG patterns we recorded in this investigation were not influenced significantly by our method of tethering, we implanted recording electrodes in the same muscles and recorded the EMG patterns with animals flying in their normal flight posture in front of the wind tunnel, and also flying on a device which allowed flight in circles (50 cm diameter) with an almost normal flight velocity (up to 2 m/s). In both situations the EMG patterns were the same as those described in this paper.

Almost all afferent input from the wings and wing-hinges of locusts enters the thoracic nerve cord via nerves 1 of the meso- and metathoracic segments (Altman et al. 1978). Thus the flight system could be extensively deafferented simply by cutting these nerves. To do this, small flaps of ventral cuticle were removed above the meso- and metathoracic ganglia and the underlying tracheae deflected to expose the nerves 1 for transection. Occasionally the tracheae were damaged but provided they were not pulled from the ganglia this damage did

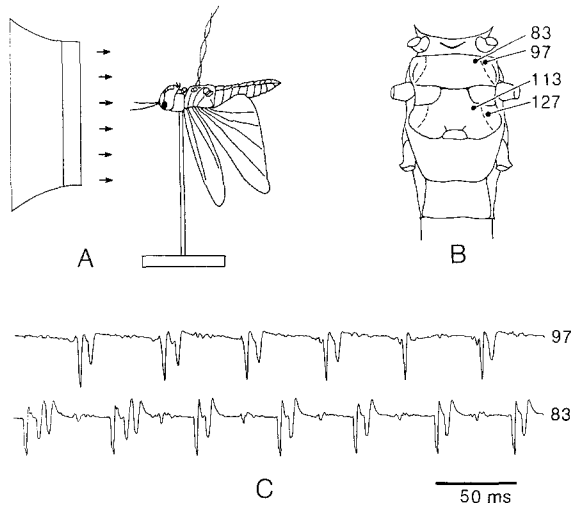


Fig. 1 A–C. Experimental arrangement used to record the electromyographic (EMG) activity in flight muscles of tethered locusts. **A** Animals were attached by the pronotum to a rigid rod and held inverted facing the opening of a wind tunnel. **B** Ventral view of the thorax showing the sites of electrode insertion for recording from the anterior tergothoracic muscles (83 and 113) and from the first basalar muscles (97 and 127). The indifferent electrode for all recordings was placed in the abdomen. **C** Example of EMG pattern recorded in the forewing anterior tergothoracic (83) and first basalar (97) muscles near the beginning of a flight sequence

not influence flight performance in our animals. This was judged by the fact that the motor patterns in undissected animals were similar to those in animals in which the nerves 1 were exposed but not cut. Thus data from both preparations have been combined in the Results. In some instances EMG recordings were made in the same animal before and after transection of the thoracic nerves.

In addition to the afferents in nerve 1, some wing afferents are contained in nerves 6 of the pro- and mesothoracic ganglia. The mesothoracic nerves 6 were routinely cut in our deafferented preparations, but the prothoracic nerves 6 were not always cut because they were sometimes not accessible through the small opening in the cuticle above the mesothoracic ganglion (the size of this opening was minimized so as not to damage the attachment of elevator muscles and to retain maximum rigidity of the thorax). We were not concerned with any afferent input via the prothoracic nerves 6 because we were unable to detect any consistent differences in the motor output patterns in animals with complete deafferentation and in animals with prothoracic nerves 6 intact.

Data were recorded on tape and analyzed using a general purpose computer (DEC LSI 11/23). This time resolution for the analysis of events was 100 μ s. The unit EMG recordings were first passed through trigger circuits with thresholds adjusted so as to generate pulses corresponding as closely as possible to the negative peaks of the unitary muscle potentials. Usually the patterns of activity during different flight sequences in a single preparation were very similar and it was not necessary to record for long periods of time or for a large number of trials. Flight sequences were evoked for about 1 min in preparations with sensory afferents intact. These sequences were terminated by removal of the wind stimulus. Sequences of rhythmic activity in deafferented preparations usually did not last longer than 30 s even with continuous wind on the head.

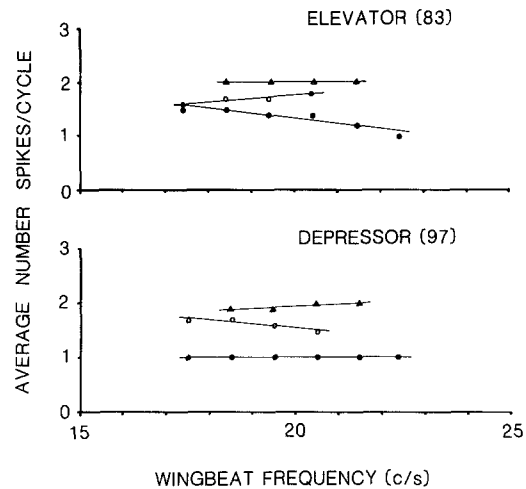


Fig. 2. Average number of spikes/cycle in muscle 83/elevator (top) and muscle 97/depressor (bottom) as a function of wingbeat frequency. Bin-width for averaging was 1 cycle/s. Data points were obtained by averaging from between 5 and 256 measurements for each point (the number of measurements was more than 40 for all but four points). Each set of data was obtained from a single trial in each of 3 different animals. Corresponding symbols indicate the same animal. Lines are best-fitting

All experiments were done on adult male *Locusta migratoria* aged between 3 and 5 weeks after the final moult.

Results

1. Patterns of activity in intact preparations

Figure 1C shows the motor pattern typically recorded from the mesothoracic muscles 83 and 97 (anterior tergothoracic, elevator, and first basalar, depressor, respectively) in an intact preparation. The number of spikes/cycle generated in each muscle was somewhat variable from animal to animal but within individual animals the average number per cycle was fairly constant and largely independent of wingbeat frequency. Figure 2 shows the average number of spikes/cycle for muscles 83 and 97 as a function of wingbeat frequency for single flight sequences in three different animals. During ongoing flight neither muscle was observed to generate more than 2 spikes/cycle. However, larger numbers of spikes were often generated near the beginning and the termination of flight activity, particularly in the elevator muscles. As the wingbeat frequency slowed following termination of the wind stimulus there were usually more spikes generated per cycle in the elevator (up to 5/cycle) but little change occurred in the number of depres-

sor spikes. It appears, therefore, that elevator activity is more subject to variation than depressor activity. This conclusion was supported by observations on deafferented animals (see below).

Because of the relative constancy of the depressor bursts we used the onset of depressor activity as the reference in quantifying the motor patterns. Two parameters were analyzed: a) the timing of the onset of elevator activity following the onset of depressor activity (we term this the D_1 to E_1 interval) and b) the timing of the onset of depressor activity following the *last* spike in an elevator burst (we term this the E_L to D_1 interval). Our reason for analysing the D_1 to E_1 interval was that Wilson (1961) had reported that the main effect of deafferentation was to increase this interval. The E_L to D_1 interval was measured for a different reason. An earlier study on deafferented preparations had shown that the generation of a depressor burst was strongly linked to the generation of a preceding elevator burst (Hedwig and Pearson 1984). This finding suggested that a basic feature of the flight motor pattern was a central coupling of depressor to elevator activity. If this is true we would expect the E_L to D_1 interval to be similar in intact and deafferented preparations. Because the characteristics of the depressor bursts changed little with wingbeat frequency in both intact and deafferented preparations, these two parameters also specify the timing of the elevator burst within the period of consecutive depressor bursts when the cycle time is known and thus provide a fairly complete description of the timing of the motor pattern.

Figure 3(top) shows two examples of the relationship between wingbeat frequency and the D_1 to E_1 interval for the forewing muscles 83 and 97. In one of these animals the latency was almost independent of frequency (slope not significantly different from zero) while in the other the latency progressively decreased with increasing frequency (slope differed significantly from zero). In the latter case the dependency of the D_1 to E_1 interval on frequency resulted in an almost phase-constant pattern. For an individual animal there was little variability in the intervals at any one frequency as indicated by the standard deviation bars in Fig. 3 (top), but we did find some variation between different animals. Figure 3(bottom) shows the interval-frequency relationship for the forewing muscles 83 and 97 for all 15 animals we studied. The slopes of the interval-frequency relationship were fairly evenly distributed, and there was no indication that animals could be divided into separate populations on the basis of the slope of this relationship. Despite the variation from animal to ani-

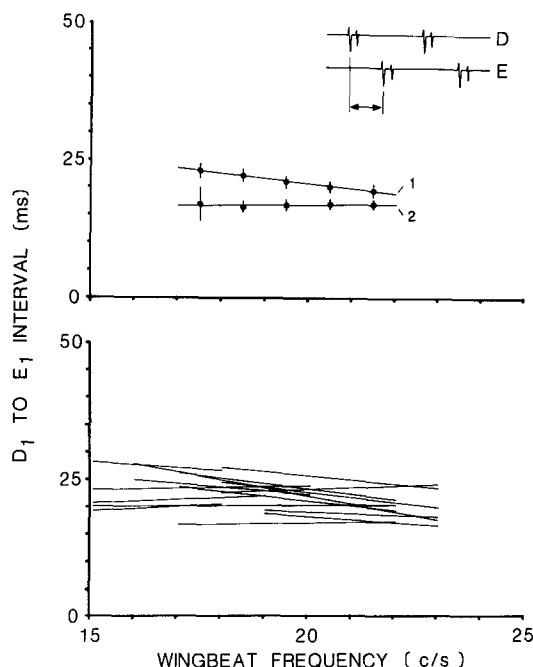


Fig. 3. Relationship between the wingbeat frequency and the interval between the first spike in a depressor burst and the first spike in the following elevator burst for muscles 83 and 97 in intact tethered animals. The inset at the top right indicates the measured interval (termed the D_1 to E_1 interval). The two relationships shown in the top graph (1 and 2) are for two different animals. In one animal the D_1 to E_1 interval was virtually constant at about 17 ms for all frequencies. In the other animal the D_1 to E_1 interval decreased progressively as frequency increased. Each point indicates the average interval for frequencies within 0.5 cycles/s on either side of the data point. Vertical bars represent standard deviations. The data points in the frequency range from 18 to 21 c/s were obtained by averaging between 78 and 325 measurements for each point. The lower set of graphs shows the best fitting lines for the relationships between the D_1 to E_1 interval and wingbeat frequency for all 15 animals studied

mal it was clear that, in general, the D_1 to E_1 interval was not strongly dependent on wingbeat frequency. This is apparent when the data from all animals were pooled (Fig. 10). Thus a characteristic feature of the intact flight pattern was that elevator activity began about 20 ms following the onset of depressor activity and this interval was relatively independent of wingbeat frequency. This relative constancy of the D_1 to E_1 interval was also observed in the recordings from hindwing anterior tergosternal and first basalar muscles. The only difference to the pattern in the forewing muscles was that the interval on average was about 8 ms longer for the hindwing pair. The reason for this was that the onset of elevator activity in the two segments occurred almost synchronously whereas hindwing depressor activity led forewing depressor activity by about 8 ms (see below).

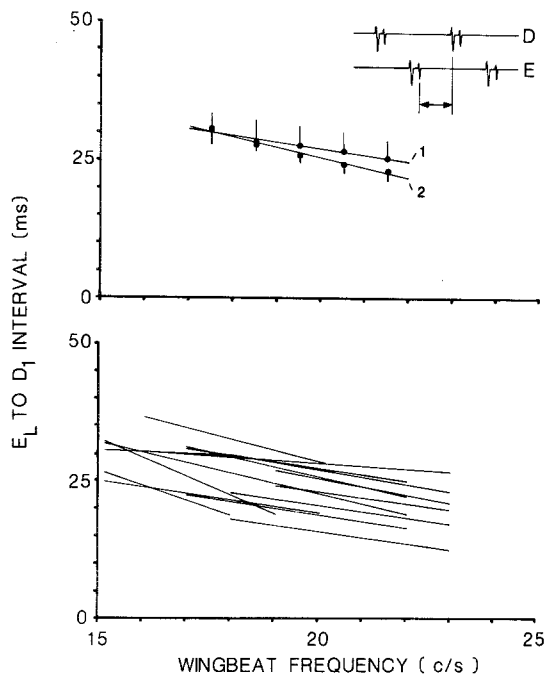


Fig. 4. Relationship between wingbeat frequency and the interval between the last spike in an elevator burst and the first spike in the following depressor burst. Recordings were made from muscles 83 and 97 in intact tethered animals. The two plots in the top graph (1 and 2) were derived from data obtained from the same two animals yielding the plots in Fig. 3 (top). Inset at the top right indicates the measured interval (termed the E_L to D_1 interval). Each point indicates the average interval for frequencies within 0.5 cycles/s on either side of the data point. Vertical lines represent standard deviations. Note the larger variation in the E_L to D_1 intervals compared to the D_1 to E_1 intervals in Fig. 3. The data points in the frequency range of 18 to 21 c/s were obtained by averaging between 78 and 325 measurements for each point. Lower set of graphs shows the relationships between the E_L to D_1 intervals and the wingbeat frequencies for the same 15 intact tethered animals yielding the plots in Fig. 3 (bottom)

In contrast to our observations on the D_1 to E_1 interval, the interval between the last spike in the elevator burst and the onset of the following depressor activity, the E_L to D_1 interval, was consistently dependent on wingbeat frequency in all 15 animals (Fig. 4). Furthermore there was more variability in this interval in individual animals and between animals. This variability can be seen by comparing the magnitudes of the standard deviations in Figs. 3(top) and 4(top), and the variation in all the interval-frequency relationships of the different animals in Figs. 3(bottom) and 4(bottom). When the data from all the animals were pooled the dependency of the E_L to D_1 interval on frequency was quite apparent (Fig. 10).

Although it is well known that movements of the hindwings lead those of the forewings by between 5 and 10 ms (Wilson and Weis-Fogh 1962),



Fig. 5. Example of EMG recordings from the forewing and hindwing first basalar (fore - 97; hind - 127) and anterior tergosternal (fore - 83; hind - 113) muscles in intact tethered animals. The arrowheads under the traces labelled 97 and 83 indicate time of occurrence of the negative peak of the spikes in the two traces labelled 127 and 113, respectively. Note that the onset of activity in depressor muscle 127 precedes the onset of activity in depressor muscle 97 by about 9 ms. By contrast the onset of activity in the two elevator muscles occurs with muscle 113 leading muscle 83 on average by less than 2 ms

and correspondingly hindwing depressor activity leads the forewing depressor activity, there exists no detailed analysis of the relative timing of elevator activity in the two segments. Wilson (1961) shows one figure (his Fig. 6) in which hindwing elevator activity leads forewing activity by about 7 ms in *Schistocerca gregaria*, and Altman (1975) shows a similar shift in adult *Chortoicetes terminifera*. In our analysis of the timing of elevator activity in the two segments, we have never observed latency shifts of this magnitude. Figure 5 shows EMG recordings made simultaneously from the forewing and hindwing anterior tergosternal (elevator) and first basalar (depressor) muscles. In this set of records, which were typical of those seen in all preparations, the interval between the onset of depressor activity in the two segments is about 9 ms whereas the interval between the onset of elevator activity was about 2 ms. Thus the onset of elevator activity occurred almost synchronously in the two segments. This occurred in all 7 animals in which intersegmental coordination was examined. For these 7 animals the mean intervals between the onset of depressor activity in the two segments were 6.7, 6.8, 7.1, 7.8, 8.3, 8.3 and 10.6 ms. In the same animals the mean intervals between the onset of elevator activity in the two segments were 1.8, 1.1, -0.7, 1.0, 1.0, 2.5 and 1.2 ms, respectively. Note that in one animal the forewing elevator became active on average slightly before the hindwing elevator. In the intact animals there was little variation in the interval between the onset of activity in homologous units of the

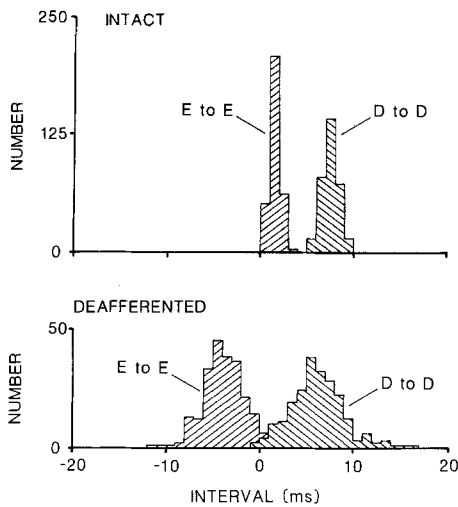


Fig. 6. Intersegmental timing of the onset of elevator and depressor activity. Top: Intact tethered animal. Bottom: Deafferented preparation. The two histograms in each panel show the distributions of the intervals between the onset of activity in hindwing and forewing anterior tergo-sternal muscles (*E* to *E*) and between the onset of activity in hindwing and forewing first basalar muscles (*D* to *D*). Negative values of the interval mean that forewing activity leads hindwing activity. Each pair of histograms represents data obtained from a single animal. Distributions in other animals were similar but the means varied from animal to animal (see text for values)

two segments. This is shown by the small variations in the histograms in the top of Fig. 6.

2. Patterns of activity in deafferented preparations

Figure 7 shows examples of the EMG recordings from the forewing anterior tergo-sternal (83) and first basalar muscles (97) of an animal in which all sensory input from the wings and wing-hinges had been removed. By comparison with the EMGs recorded from the same muscles in intact animals two differences, initially reported by Wilson (1961), were obvious. The first was a slowing of the repetitive activity to about half the frequency observed in intact animals, and the second was a shift in elevator activity so that it occurred later in the interval between depressor bursts. One feature which did not change significantly was the characteristics of the depressor activity. In most of our deafferented animals the first basalar discharged twice each burst (Fig. 7) with the interval between the two spikes being about 30% longer than in intact animals. The activity of wing eleva-

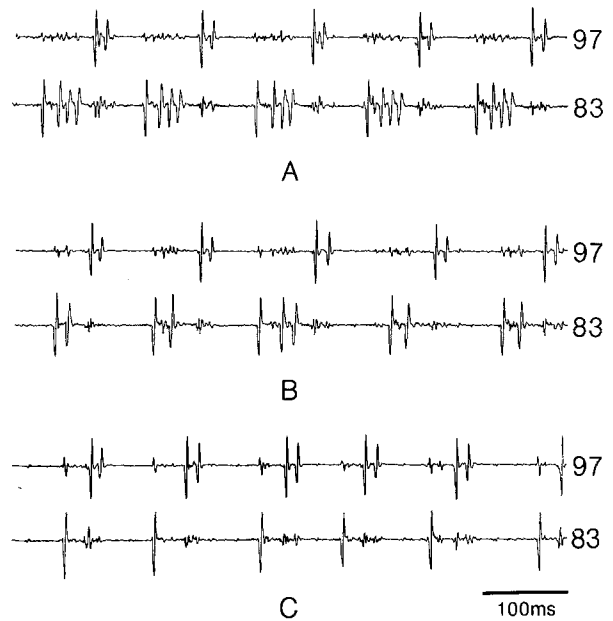


Fig. 7A-C. EMG recordings from forewing first basalar (97) and anterior tergo-sternal (83) muscles in a deafferented preparation. All sets of records are from the same flight sequence in one animal. **A** Near the beginning of the sequence the onset of elevator activity occurred approximately midway between depressor bursts and up to 4 elevator spikes were generated in each cycle. **B** Later in the same sequence fewer spikes were generated in each cycle. **C** Near the end of the sequence only a single elevator spike was generated in each cycle and this occurred later in the interval between successive depressor bursts. Note that depressor activity remained relatively constant throughout the sequence

tor motoneurons, however, was strongly affected by deafferentation. The influence of deafferentation on the burst activity in the forewing anterior tergo-sternal (elevator muscle 83) is shown in Fig. 8. The plots in this figure show the relationship between the average number of spikes per cycle versus wingbeat frequency for 11 deafferented preparations (left) and 14 intact animals (right). For intact animals the average number of spikes/cycle for each animal was fairly constant and within the range of 1 to 2 spikes/cycle. By contrast, in deafferented preparations the number of spikes/cycle showed considerably more variation from animal to animal, and in some cases this number was strongly dependent on frequency. Again it should be emphasized that deafferentation had little influence on the number of depressor spikes/cycle (remaining in the range of 1 to 2/cycle).

Despite the fact that the pattern of activity in the anterior tergo-sternal muscles was variable within single animals (Fig. 7) and between animals (Fig. 8), the one very consistent feature was a marked increase in the D_1 to E_1 interval. In Fig. 9 (top) the dependence of this interval on wingbeat

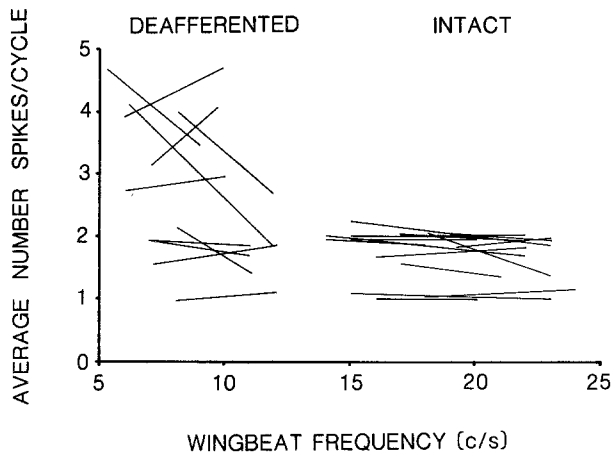


Fig. 8. Average number of spikes/cycle as a function of wingbeat frequency in elevator muscle 83 in 11 deafferented preparations (set of lines on left) and 14 intact preparations (set of lines on right). Each line is the best fitting line through the data points (not shown) for a single trial in each animal

frequency is shown for the 9 deafferented animals we examined. This should be compared with the data in Fig. 3 (bottom) (note the difference in the ordinate scale between these two figures). The comparison shows that the D_1 to E_1 interval in deafferented preparations became strongly dependent on wingbeat frequency, was increased by a factor of between 3 and 4, and was variable between different animals. This variability resulted from the fact that the elevator produced different numbers of spikes in different preparations (see Fig. 8). The greater the number of spikes for a given wingbeat frequency the shorter the D_1 to E_1 interval. In some preparations the elevator discharged up to 5 spikes per cycle while in others it discharged only a single spike late in the depressor cycle. Usually the pattern for an individual animal remained fairly constant. However, in a few animals the pattern changed from one with multiple elevator spikes (Fig. 7A) to one with only a single elevator spike (Fig. 7C).

One feature of the motor pattern which was little affected by deafferentation was the E_L to D_1 interval (Fig. 9 bottom). The magnitude of this interval was similar to that observed in intact animals (compare with Fig. 4 bottom – note difference in scales of the ordinates) and the interval decreased slightly as wingbeat frequency increased.

Apart from changes in the pattern of motor output within a single segment, deafferentation also influenced the coordination of activity between segments. The main effect of deafferentation on intersegmental coordination was to cause an increase in the variability of the relative timing of activity in homologous muscles. The histograms

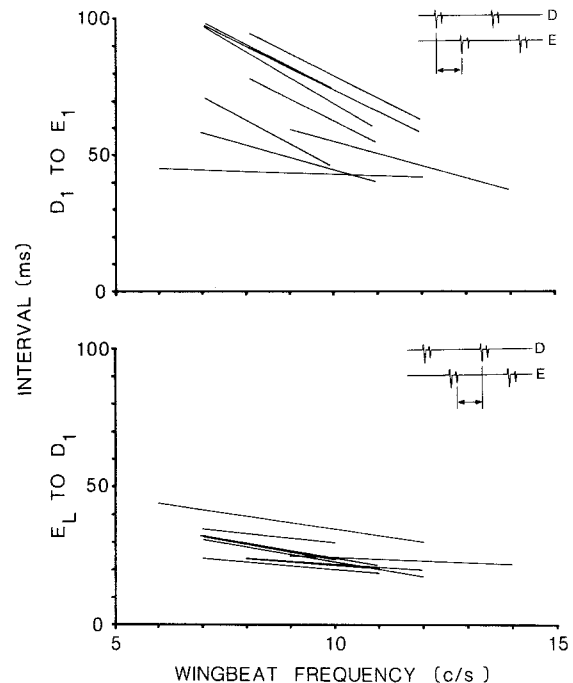


Fig. 9. Timing of activity in muscles 83 and 97 in deafferented preparations. The top graph shows plots of the relationship between wingbeat frequency and the D_1 to E_1 interval in 9 deafferented preparations. Each line is the best fitting line to the data points (not shown) for a single trial in one animal. The bottom graph presents plots of the relationship between wingbeat frequency and the E_L to D_1 interval for the same 9 animals. These plots should be compared with similar plots for the intact tethered animal (Figs. 3 and 4 – note differences in ordinate scales)

at the bottom of Fig. 6 show this variability for both the elevator and depressor muscles in one animal. The variability in other animals was similar. Another change produced by deafferentation was that the onset of activity in the forewing elevators preceded the onset of hindwing elevator activity. The mean for the preparation shown in Fig. 6 (bottom) was -2.8 ms. Mean values for other deafferented preparations were -7.4 , -5.4 , -1.7 , -1.7 , -0.8 , -7.6 and -5.9 ms. The coordination of the depressors, on the other hand, usually remained qualitatively similar to that occurring in the intact animal, i.e. hindwing depressor activity preceded forewing activity by between 5 and 10 ms. The corresponding mean intervals for the lag between the onset of hindwing and forewing depressor activity were 7.2, 11.5, 10.2, 9.5, 8.7, 0, 9.5 and 0 ms. Note that the values of most of these intervals are close to those occurring in intact animals, although in two deafferented preparations the onset of depressor activity occurred on average synchronously in the two segments.

Discussion

In this investigation we have obtained a quantitative description of the changes in the flight motor pattern of the locust following removal of sensory input from the wings, and gained more information on the motor output pattern in intact animals. The motor patterns we recorded in intact tethered animals showed that the onset of elevator activity occurred at an almost constant interval following the onset of depressor activity (Fig. 3). Since this interval was nearly independent of flight frequency it follows that the phase of the onset of elevator activity in the depressor cycle advanced as the wingbeat frequency decreased. In the only other detailed analysis of the timing of elevator and depressor activity in flying locusts, Waldron (1967) reported in *S. gregaria* that the phase of elevator activity remained relatively constant for changes in wingbeat frequency. We cannot account for the difference of our result with that of Waldron, but it should be noted that in some of our animals the D_1 to E_1 interval increased with decreased frequency to also yield an almost phase constant pattern. Because of variation from animal to animal we conclude that it is not appropriate to characterize the intact motor pattern in *L. migratoria* as strictly phase-constant or as strictly latency-constant. Nonetheless an important feature was that the time between the onset of depressor activity and the subsequent elevator burst was, in general, almost independent of wingbeat frequency as can be seen when the data from all animals were pooled (Fig. 10). This characteristic has not been reported in previous studies on the flight motor pattern.

Another characteristic of the intact motor pattern not previously reported is the almost synchronous activation of the forewing and hindwing elevators (Figs. 5 and 6). There are very few published data on the activity patterns of elevator motor units and none allow a direct comparison with our observations. Nevertheless in other species of locusts it has been reported that hindwing elevator activity precedes forewing elevator activity by 5 to 10 ms (Wilson 1961; Altman 1975). There are at least three possible explanations for the apparent discrepancy of our observations with those of Wilson and Altman. The first is that there are species differences. The second is that the recordings in the earlier studies were done using pairs of elevator muscles other than the pair we studied, and that there is an intersegmental delay in the activation of members of these other pairs. We regard this as unlikely because we did examine intersegmental latencies in a variety of elevator muscles

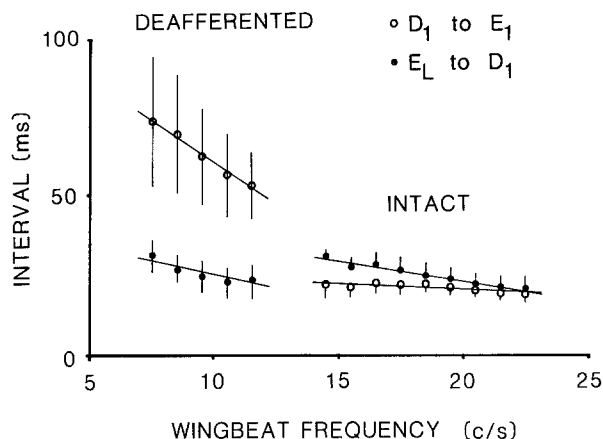


Fig. 10. This graph summarizes the major effects of deafferentation on the timing of activity of forewing muscles 83 and 97. Each plot was derived by averaging the values at each frequency for all preparations active at that frequency. Bars are standard deviations. Open and closed circles show relationships between wingbeat frequency and the D_1 to E_1 interval and the E_L to D_1 interval, respectively

and always found almost synchronous activation (see also Wolf and Pearson 1986). Finally there is the possibility that the published records of Wilson and Altman are not typical of the normal pattern in homologous elevator muscles. Until the patterns are reexamined in *S. gregaria* and in *C. terminifera* no firm conclusions can be drawn to explain the difference between our observations and those of Wilson and of Altman.

The most important results of this study are those concerned with the effects of deafferentation. Although some of these results confirm previously reported qualitative features such as the slowing of the flight rhythm and a delay in the onset of elevator activity relative to depressor activity, our quantitative description has shown clearly for the first time that the motor pattern in deafferented preparations is quite different from that occurring in intact animals. Thus we do not agree with the conclusion of Wilson (1961) that afferent input produces only minor changes of the deafferented pattern, and that the latter can be regarded as a more or less slowed-down version of the intact pattern. The most obvious and consistent effect of deafferentation was to delay the onset of activity in elevators, that is, to increase the D_1 to E_1 interval. In addition, the D_1 to E_1 interval in deafferented preparations showed a strong dependence on frequency and was highly variable between animals (Fig. 9) and within single animals (Fig. 7). This is in contrast to the intact preparations where the D_1 to E_1 interval is constant at approximately 20 ms. When the data from all the animals are

pooled this difference in the characteristics of the D_1 and E_1 interval is very obvious (open circles in Fig. 10). This change in the D_1 to E_1 interval is not simply a result of a shift of the entire elevator burst relative to the depressor bursts, nor a simple consequence of the slowing of the flight rhythm. If the former occurred we would expect a decrease in the E_L to D_1 interval or the duration of the elevator burst following deafferentation, and if the latter occurred we would expect the phasing of the elevator activity to remain the same following deafferentation. Neither of these events was observed, however. Deafferentation increased the phase of occurrence of elevator spikes in the depressor cycle and had little influence on the E_L to D_1 interval. Pooling the data from all animals shows that the quantitative features of the E_L to D_1 interval were similar in intact and deafferented animals (filled circles in Fig. 10). It should be noted that our finding that deafferentation produces a qualitative change in the flight motor pattern does not depend on the parameters we used for describing this pattern. For example measurement of the phase of elevator activity in the depressor cycle (or vice versa) instead of determining burst intervals would also have revealed that deafferentation alters the flight motor pattern.

Another important change in the motor pattern following deafferentation was an increase in the variability of the discharge pattern of elevators (Fig. 8). Normally in intact animals the anterior tergothoracic muscle was activated twice per cycle, whereas in deafferented preparations anywhere from 1 to 5 spikes/cycle were recorded from this muscle and there was not necessarily any consistent relationship between the number of spikes and frequency (Fig. 8). This variability in elevator activity was also noted by Kutsch (1974) and is apparent in the records of Wilson and Gettrup (1963). By contrast, deafferentation led to little change in the activity of the first basalar. It was this fact that allowed us to use the first basalar spike as the reference in this investigation. Thus from our observations on intact and deafferented animals we conclude that the depressor bursts are less subject to variation than elevator bursts, and that one function of afferent input is to stabilize elevator activity. The latter conclusion is of some interest because previous studies have suggested that most of the control of wing dynamics is produced by changes in the relative timing of activity in different depressors (Zarnack and Möhl 1977; Baker 1979). We would expect therefore a significant influence of wing proprioceptors on depressors. This afferent control of depressor activity has indeed

been demonstrated (e.g. Möhl 1985b) but the magnitudes of these effects are relatively small when compared to the very large influence we found deafferentation to have on elevator activity.

One feature of the motor pattern which was not qualitatively altered following deafferentation was the timing of depressor activity in the meta- and mesothoracic segments (Fig. 6). Although deafferentation did lead to an increase in the variability of the interval between the onset of hindwing depressor activity and the onset of forewing depressor activity, it was clear in the majority of our deafferented preparations that the mean value of this interval remained in the range of 5 to 10 ms. Thus the intersegmental delay in depressors must be determined primarily by central circuits with afferent input functioning to stabilize this interval to within a narrow range. A similar conclusion was reached by Hedwig and Pearson (1984) in their analysis of synaptic input to flight motoneurons in deafferented animals. The variability in the timing of elevator activity in the two segments was also increased by deafferentation but, in addition, forewing elevator activity usually commenced before hindwing elevator activity instead of almost synchronously as it does in intact animals. In the following paper (Wolf and Pearson 1986) we show that this shift in the timing of elevator muscle activity is not due to a shift in the timing of phasic synaptic input to the elevator motoneurons in the two segments but rather to a difference in the time during the phasic depolarization at which the first elevator spike is generated.

In summary, we have observed a number of consistent changes in the flight motor pattern following deafferentation. These changes are of such magnitude that we conclude that the motor patterns in intact and deafferented animals are qualitatively different. Three questions we are left with are: 1) why is the D_1 to E_1 interval short and constant in intact animals, 2) why is this interval prolonged and more variable after deafferentation, and 3) why is there more variability in the pattern of elevator activity in deafferented compared to intact animals? We have been able to obtain plausible answers to all three of these questions by intracellularly recording from flight motoneurons in intact and deafferented animals. These recordings are presented in the following paper (Wolf and Pearson 1986).

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