Differential activation among five human inspiratory motoneuron pools during tidal breathing

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Neural drive to inspiratory pump muscles is increased under many pathological conditions. This study determined for the first time how neural drive is distributed to five different human inspiratory pump muscles during tidal breathing. The discharge of single motor units (n = 280) from five healthy subjects in the diaphragm, scalene, second parasternal intercostal, third dorsal external intercostal, and fifth dorsal external intercostal was recorded with needle electrodes. All units increased their discharge during inspiration, but 41 (15%) discharged tonically throughout expiration. Motor unit populations from each muscle differed in the timing of their activation and in the discharge rates of their motor units. Relative to the onset of inspiratory flow, the earliest recruited muscles were the diaphragm and third dorsal external intercostal (mean onset for the population after 26 and 29% of inspiratory time). The fifth dorsal external intercostal muscle was recruited later (43% of inspiratory time; P < 0.05). Compared with the other inspiratory muscles, units in the diaphragm and third dorsal external intercostal had the highest onset (7.7 and 7.1 Hz, respectively) and peak firing frequencies (12.6 and 11.9 Hz, respectively; both P < 0.05). There was a unimodal distribution of recruitment times of motor units in all muscles. Neural drive to human inspiratory pump muscles differs in timing, strength, and distribution, presumably to achieve efficient ventilation.

METHODS

The present investigation is based on a complete reanalysis of motor unit recordings obtained during normal quiet breathing in two previous experiments (8, 17). They investigated the mean peak firing rates of single motor units in the costal diaphragm, scalenes, and parasternal intercostal muscles with and without increased chemical drive (17) and the mean peak firing rates in the dorsal external intercostal muscles during quiet breathing (8). This reanalysis allowed us to compare the firing times and frequencies of single motor units sampled in five major inspiratory muscles: the costal diaphragm, the scalenes, the second parasternal intercostal muscle, and the third and fifth dorsal external intercostal muscles. Studies were performed on five healthy men (aged 33–51 yr). Three subjects were studied in both tidal breathing and in voluntary breathing with different inspiratory flows and volumes (3), with little tonic activity during expiration. There is evidence that the timing of inspiratory activity varies systematically among intercostal muscles in animals (for review, see Refs. 9, 26) and humans (8, 18). Thus, for a particular set of intercostal muscles, motor units in muscles with the largest inspiratory mechanical advantage, i.e., those muscles that produce the largest change in pleural pressure per unit muscle mass and unit muscle tension (12, 41), discharge earlier and reach higher discharge frequencies. However, the recruitment differences between the various pump muscles have not been investigated.

We hypothesized that there would be differences in the timing of firing of single motor units in the various inspiratory pump muscles and that the firing frequencies would differ between muscles. This information provides insight into the descending respiratory “commands” that drive inspiration. Hence, the present study was conducted to determine the firing behavior of single human diaphragm motor units during quiet breathing and to compare this with the motor unit behavior of the scalenes, parasternal intercostal, and external intercostal muscles, all of which also discharge during quiet inspiration. Second, we looked for evidence that there is a bimodal distribution of distinct “early” and “late” firing times during inspiration, as has been reported for inspiratory motoneurons in animals (e.g., Refs. 13, 23). The data allowed us to make a comparison between the behavior of the five inspiratory pump muscles recently reported for the genioglossus, a major dilator of the upper airway (34). A preliminary report of some findings has been presented (35).

HUMAN BREATHING IS AN ESSENTIAL rhythmic activity that does not need volition, but it does require shortening of many inspiratory muscles that act on the chest wall (diaphragm, intercostal, and scalene muscles, the so-called “pump” muscles) and those that act on the upper airway (“valve” muscles). The upper airway must be maintained patent while the muscles acting on the chest wall generate forces to overcome airway resistance and the elastic recoil of the chest wall and lungs to allow inspiratory flow. Neural drive from medullary respiratory centers (14, 29) acting via central respiratory drive potentials depolarizes and thus recruits inspiratory motoneurons innervating chest wall muscles and derecruits them by active hyperpolarization (e.g., Refs. 1, 36). In contrast, the recruitment of upper airway motoneurons is achieved with inspiratory depolarization but without expiratory inhibition (32). The timing of drive to the different inspiratory muscles is thought to be controlled by one or more pontomedullary oscillators (16, 24, 30, 37).

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experiments. Each subject gave informed, written consent to the procedures approved by the human ethics committee of the University of New South Wales. Key methods are given below, and further details of the recordings are available (8, 17).

General procedures. Each muscle was studied on a separate occasion. With the exception of the scalene, ultrasonography assisted placement of needle electrode in the other muscles. Subjects were seated comfortably, unrestrained, and breathed quietly through a mouth piece (Fig. 1). Tidal volume was obtained by integration of the flow signal. Inspiratory time (TI) was measured from the airflow signal. To compare between subjects and between motor units, the details of the recordings are available (8, 17).

Experiments. Each subject gave informed, written consent to the experiments. Each subject was able to maintain a minimal firing frequency that exceeded 2.0 Hz. The onset time, relative to the onset of inspiratory airflow, was taken as the time that the unit first began to discharge or the time the unit increased its discharge frequency above the background tonic firing rate determined visually. The end discharge time was taken as the time of the last discharge of the unit in each breath or when the discharge frequency returned to tonic values. The preinspiratory activity occurred when activity began before inspiratory flow (<0% TI). Likewise, postinspiratory activity occurred when activity continued after 100% TI. The onset discharge frequency was calculated from the frequency of the first interspike interval within each breath. For tonic units, it was measured at the first increase of the instantaneous frequency above the tonic level. The end discharge frequency was determined with a similar procedure. The peak discharge frequency was calculated with a running average of over 200 ms for each breath and then averaged across the three breaths. This process minimizes the effect of any motor unit firing missed in the spike discrimination analysis. All spike analyses were performed by a single observer.

In addition, to assess the initial recruitment time for the whole population of motor units sampled at each site, multiunit EMG was rectified and integrated (decay time constant, 50 ms), and signals were monitored online with the data acquisition system. The EMG signals were amplified (×1,000–10,000), filtered (53 Hz to 3 kHz), and stored on computer (Spike2 with 1401 interface, CED) for offline analysis. Spikes from motor units were selected based on a threshold crossing, and they were manually sorted using individual “templates” based on their size and morphology.

Measurements of discharge properties. Measurements of the discharge properties of the single motor units were based on the instantaneous frequency plots for each motor unit over three consistent consecutive breaths. The three consecutive breaths chosen had consistent respiratory timing and changes in inspiratory flow and volume. All the units for each muscle were classified into tonic or phasic categories, depending on whether they discharged throughout both inspiration and expiration. To be classified as “tonic,” units had to maintain a minimal firing frequency that exceeded 2.0 Hz. The onset time, relative to the onset of inspiratory airflow, was taken as the time that the unit first began to discharge or the time the unit increased its discharge frequency above the background tonic firing rate determined visually. The end discharge time was taken as the time of the last discharge of the unit in each breath or when the discharge frequency returned to tonic values. The preinspiratory activity occurred when activity began before inspiratory flow (<0% TI). Likewise, postinspiratory activity occurred when activity continued after 100% TI. The onset discharge frequency was calculated from the frequency of the first interspike interval within each breath. For tonic units, it was measured at the first increase of the instantaneous frequency above the tonic level. The end discharge frequency was determined with a similar procedure. The peak discharge frequency was calculated with a running average of over 200 ms for each breath and then averaged across the three breaths. This process minimizes the effect of any motor unit firing missed in the spike discrimination analysis. All spike analyses were performed by a single observer.

In addition, to assess the initial recruitment time for the whole population of motor units sampled at each site, multiunit EMG was rectified and integrated (decay time constant, 50 ms), and the onset time was measured for each breath at the increase in activity above tonic or baseline levels and expressed relative to the onset of inspiratory flow (0% TI). Figure 2 shows examples of the integrated selective multiunit EMG records for each muscle. The initial increase in multiunit EMG activity from each muscle was sorted based on its onset time, and then each data point was joined by an unbroken line (see Fig. 3). This gives the cumulative data for the entire population of onset times.

Statistical analysis. The time of discharge (onset, peak, and end) and discharge frequencies of single motor units, tidal volume, and TI were compared between the five testing sessions using one-way ANOVA tests with Student-Newman-Keuls post hoc analysis. If data
were not normally distributed, then a Kruskal-Wallis test was applied with Dunn’s post hoc analysis. For units in each muscle, Pearson product-moment correlations were performed on the onset time (%TI), the duration of firing, and the peak discharge frequencies. The proportion of phasic and tonic units and the presence of preinspiratory/postinspiratory activity in each muscle were analyzed with $\chi^2$ tests. In addition, cross correlations between the signal of lung volume (from the summed signal of the inductance bands) and instantaneous firing frequency (smoothed over 200 ms) were computed for all potential phase variations between the two signals. The strength of each correlation was calculated with a linear coefficient of determination ($r^2$). The respiratory phase of the maximal $r^2$ determined whether the unit was an inspiratory discharging unit (31, 34). Statistical significance was set at $P < 0.05$. Unless indicated, values are given as means $\pm$ SE.

RESULTS

Data were obtained from a total of 280 single motor units in five subjects with recordings from the diaphragm (40 U), scalene (56 U), second parasternal intercostal (69 U), third dorsal external intercostal (67 U), and fifth dorsal external intercostal (48 U). There was no significant difference for inspiratory flow, Ti, or tidal volume when recordings were made from different muscles (see Table 1).

Typical recordings for the five muscles are shown in Fig. 2. Overall, 239 (85%) of the motor units discharged phasically during inspiration, whereas 41 motor units (15%) discharged throughout inspiration and tonically during expiration. Figure 3 plots the timing of discharge of each motor unit in each muscle. All motor units that discharged tonically increased their discharge frequency during inspiration (Fig. 2F). The degree of tonic firing differed between the inspiratory muscles ($P < 0.001$). In the diaphragm, all units were phasically active with no tonic activity, and in the third dorsal external intercostal the units were predominately phasic, with only one motor unit tonically active. In contrast, 29% of scalenes ($n = 16$), 13% of parasternal intercostal units ($n = 9$), and 31% of fifth dorsal external intercostal units ($n = 15$) discharged tonically.

Timing of inspiratory discharge. There were significant differences between the inspiratory muscles in the timing of inspiratory discharge. In addition to the discharge timing for each single motor unit, Fig. 3 illustrates the onset timing determined from the selective multiunit EMG recordings. The thin dotted horizontal lines indicate those units that discharged tonically, and the thick horizontal lines show the time that the discharge frequency increased phasically during inspiration. The solid ascending curves represent the cumulative onset times of the integrated multiunit EMG from each recording site. The progressive recruitment of the single units and the multiunit EMG shows that there was a unimodal distribution of recruitment times throughout inspiration for all muscles (see Fig. 4). Figure 4 illustrates the distribution of recruitment times in absolute time (ms), whereas the times quoted elsewhere are expressed as a %TI.

The mean onset time for phasically active inspiratory units was earliest for the diaphragm units at $26 \pm 3\%$ (mean $\pm$ SE)}
of Ti, and latest for the fifth dorsal external intercostal units (43 ± 4% Ti). The diaphragm and the third dorsal external intercostal muscle (29 ± 2% Ti) were recruited significantly earlier than units from the fifth dorsal external intercostal (P < 0.05) but were not recruited significantly earlier than scalene (32 ± 4% Ti) and second parasternal intercostal (34 ± 2% Ti) units. This difference was confirmed by measurement of the timing of the peak correlation between discharge frequency and lung volume (see Table 2). The peak correlation for the phasic units in the diaphragm (−0.69 ± 0.14 s; r² = 0.78), third dorsal external intercostal (−0.38 ± 0.03 s; r² = 0.84), scalene (−0.17 ± 0.14 s; r² = 0.78), and second parasternal intercostal (−0.44 ± 0.31 s; r² = 0.82) inspiratory units occurred before the end of inspiration, whereas the mean peak correlation for fifth dorsal external intercostal inspiratory phasic units occurred just after the end of inspiration (0.59 ± 0.13 s; r² = 0.82). These times differed significantly (P < 0.05).

The mean onset of activity derived from the integrated multiunit EMG provided an estimate of the earliest onset of activity for a population of motor units in each muscle. The diaphragm (−2.5 ± 0.6% Ti) was recruited first (earlier than scalenes and fifth dorsal external intercostal; P < 0.05), followed by the third dorsal external intercostal (−1.0 ± 2.6% Ti), earlier than fifth dorsal external intercostal (P < 0.05), second parasternal intercostal (4.4 ± 3.0% Ti), and scalene (7.1 ± 5.0% Ti) muscles (P = not significant). The fifth dorsal external intercostal was recruited last (14.5 ± 3.3% Ti; Fig. 3). Motor unit activity before inspiratory airflow was recorded from each muscle in the multiunit recordings. There were differences

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**Table 1. Ventilatory data during electromyographic recordings**

<table>
<thead>
<tr>
<th>Muscle</th>
<th>Inspiratory Flow, l/s</th>
<th>Inspiratory Time, s</th>
<th>Tidal Volume, liter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diaphragm</td>
<td>0.47 ± 0.06</td>
<td>1.7 ± 0.21</td>
<td>0.8 ± 0.01</td>
</tr>
<tr>
<td>Scalene</td>
<td>0.49 ± 0.07</td>
<td>1.8 ± 0.23</td>
<td>0.8 ± 0.04</td>
</tr>
<tr>
<td>2nd parasternal intercostal</td>
<td>0.38 ± 0.03</td>
<td>2.0 ± 0.10</td>
<td>0.7 ± 0.05</td>
</tr>
<tr>
<td>3rd dorsal external intercostal</td>
<td>0.37 ± 0.04</td>
<td>2.1 ± 0.39</td>
<td>0.7 ± 0.03</td>
</tr>
<tr>
<td>5th dorsal external intercostal</td>
<td>0.42 ± 0.08</td>
<td>2.1 ± 0.36</td>
<td>0.8 ± 0.11</td>
</tr>
</tbody>
</table>

Values are means ± SE for the mean inspiratory flow, inspiratory time, and tidal volume for each muscle. For each muscle, the mean is derived from the mean for each subject. There were no significant differences for the ventilatory data across the 5 recording sessions (P > 0.05).
between muscles in the likelihood of “preinspiratory” activity (before inspiratory airflow, <0% Ti). The diaphragm multiunit recordings usually had preinspiratory activity (84% of sites; P < 0.001). Preinspiratory activity was less common in the scalene (35%), second parasternal intercostal (41%), and third dorsal external intercostal (46%), and it occurred least of all in the fifth dorsal external intercostal muscle (19%).

Postinspiratory activity (after inspiratory airflow, >100% Ti) also occurred in all muscles but was most obvious in the fifth dorsal external intercostal muscle. In early expiration (measured at 120% Ti), 77% of the fifth dorsal external intercostal motor units were still active, fewer units were active in the other muscles (P < 0.05): diaphragm (50%), third dorsal intercostal (49%), scalenes (45%), and second parasternal in-

<table>
<thead>
<tr>
<th>Muscle</th>
<th>Number</th>
<th>Onset Time, ms</th>
<th>Onset Time, % Inspiratory Time</th>
<th>r²</th>
<th>Lag Time, s</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Phasic units</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diaphragm</td>
<td>40</td>
<td>446 ± 60*</td>
<td>25.5 ± 3.2*</td>
<td>0.78 ± 0.02</td>
<td>-0.69 ± 0.14*</td>
</tr>
<tr>
<td>Scalene</td>
<td>40</td>
<td>628 ± 93*</td>
<td>31.8 ± 4.3*</td>
<td>0.78 ± 0.02</td>
<td>-0.17 ± 0.14*</td>
</tr>
<tr>
<td>Parasternal intercostal</td>
<td>60</td>
<td>712 ± 45†</td>
<td>34.4 ± 2.1</td>
<td>0.82 ± 0.01</td>
<td>-0.44 ± 0.31*</td>
</tr>
<tr>
<td>3rd dorsal external intercostal</td>
<td>66</td>
<td>593 ± 58*</td>
<td>28.7 ± 2.1*</td>
<td>0.84 ± 0.01†</td>
<td>-0.38 ± 0.03*</td>
</tr>
<tr>
<td>5th dorsal external intercostal</td>
<td>33</td>
<td>1170 ± 130</td>
<td>43.0 ± 4.0</td>
<td>0.82 ± 0.02</td>
<td>0.59 ± 0.13</td>
</tr>
<tr>
<td><strong>Tonic units</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Diaphragm</td>
<td>0</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Scalene</td>
<td>16</td>
<td>-60.3 ± 161</td>
<td>-5.4 ± 10.4</td>
<td>0.64 ± 0.05</td>
<td>-0.07 ± 0.46</td>
</tr>
<tr>
<td>2nd parasternal intercostal</td>
<td>9</td>
<td>-165 ± 129</td>
<td>-7.8 ± 6.4</td>
<td>0.64 ± 0.07</td>
<td>-0.18 ± 0.51</td>
</tr>
<tr>
<td>3rd dorsal external intercostal</td>
<td>1</td>
<td>-92.3</td>
<td>-5.4</td>
<td>0.86</td>
<td>3.97</td>
</tr>
<tr>
<td>5th dorsal external intercostal</td>
<td>19</td>
<td>684 ± 174</td>
<td>44.3 ± 10.9</td>
<td>0.62 ± 0.04</td>
<td>0.86 ± 0.49</td>
</tr>
</tbody>
</table>

Values are means ± SE. Table shows the mean correlation and lag time for units in each muscle along with the mean time for the onset of inspiratory firing. The coefficients of determination are higher for phasic than tonic units. Negative lag times indicate that the peak correlation between firing frequency and flow occurred prior to the end of inspiration (see METHODS). *Significantly different from 5th dorsal external intercostal (P < 0.001). †Significantly different from diaphragm (P < 0.05).
tercostal (45%). As a consequence, the mean end firing time for the fifth dorsal external intercostal units (146 ± 4% Ti) was significantly later than for the diaphragm (124 ± 4% Ti), third dorsal external intercostal (125 ± 4% Ti), scalene (113 ± 4% Ti), and second parasternal intercostal (127 ± 4% Ti; P < 0.05). For each muscle, there was a significant negative linear correlation between onset time and duration of firing (Fig. 5).

Motor units that were recruited early discharged for the longest duration (P < 0.001). The strength of the peak coefficient of determination (r²) between lung volume and each unit’s discharge frequency during quiet breathing were assessed (see Table 2). Phasic motor unit had higher r² values than the tonic units (P < 0.001).

Discharge frequencies. As shown in Fig. 6A, the onset and peak firing frequencies for each phasic motor unit differed significantly between muscles. The mean onset discharge frequencies for the phasic units in the diaphragm (7.7 ± 0.3 Hz) and third dorsal external intercostal (7.2 ± 0.2 Hz) were significantly higher than for the scalene (6.0 ± 0.2 Hz), second parasternal intercostal (6.6 ± 0.2 Hz), and fifth dorsal external intercostal (6.0 ± 0.2 Hz; P < 0.05). The mean peak frequency for the diaphragm (12.6 ± 0.5 Hz) was also higher than for the scalene (9.3 ± 0.4 Hz), second parasternal intercostal (10.8 ± 0.3 Hz), and fifth dorsal external intercostal (10.1 ± 0.4 Hz; P < 0.05). The peak frequency for the third dorsal external intercostal (11.9 ± 0.4 Hz) was higher than the peak frequency for scalene and fifth dorsal external intercostal (P < 0.05). The change in frequency during inspiration from onset frequency to peak frequency was largest for the diaphragm (4.9 ± 0.4 Hz) and third dorsal external intercostal (4.8 ± 0.4 Hz) motor units compared with the scalene (3.3 ± 0.3 Hz), the second parasternal intercostals (4.2 ± 0.3 Hz), and the fifth dorsal external intercostal (4.1 ± 0.4 Hz; P < 0.05). For the tonically active motor units in scalene, parasternal intercostal, and fifth dorsal external intercostal, there was no difference in either the onset or peak firing frequencies (Fig. 6B). The change between the onset and peak firing frequencies for the tonic units (~4.2 Hz) was not significantly different from that of the phasic units in the same muscle.

To determine whether the inspiratory drive peaked simultaneously for each muscle, the time at which single units reached their maximal firing frequency during inspiration was compared (Fig. 7). Peak firing occurred significantly later for the fifth dorsal external intercostal, at 90 ± 4% Ti, than for the diaphragm (74 ± 2% Ti), the third dorsal external intercostal (72 ± 2% Ti), and the scalenes (76 ± 3% Ti; all P < 0.05), with the peak for the second parasternal intercostal occurring at 81 ± 3% Ti.

For units in the diaphragm, third dorsal external intercostal and second parasternal intercostal, the duration of discharge was positively correlated with the peak firing rate (P < 0.05), and the onset time of firing was negatively correlated with the peak firing rate (P < 0.05). In these muscles, the earliest recruited motor units fired for the longest time and reached the

![Fig. 5. Relationship between onset firing time and the duration of firing. A–E: onset time against the duration of discharge of inspiratory motor units. Each phasic unit is represented once by the average times derived from three breaths. Pearson correlations show a significant relationship, with the earliest recruited units in each muscle discharging for the longest duration (P < 0.001). The 3 square data points on the x-axes of the scalene, 3rd dorsal external intercostal, and 5th dorsal external intercostal each represent a unit where the onset time was earlier than ~20% Ti.](https://jap.physiology.org/doi/10.1152/jappl.00171.2006)
intercostal only (P higher than corresponding frequency from scalene and 5th dorsal external

TI, or tidal volume to differ when recordings were made from
times.

throughout inspiration with a unimodal distribution of onset
in the other muscles. Inspiratory motor units were recruited
significantly later and reached their peak firing later than units
fifth dorsal external intercostal motor units were activated
were tonically active in the fifth dorsal external intercostal. The

diaphragm and third dorsal external intercostal muscles, whereas the onset times were
later and firing frequencies were lower for the scalene units
than for the diaphragm units. Thus these differences are not
likely to have affected our major conclusions.

As for diaphragm motor units in volitional inspiratory tasks
in humans (3), all five pump muscles progressively recruited
new units throughout inspiration, with no evidence for the
bimodal pattern of recruitment seen in animal studies (e.g.,
Refs. 13, 23). The lack of evidence for a large population of
early recruited units is interesting. There are several possible
explanations, including a sampling bias. However, the graded
recruitment of motor units in all five inspiratory muscles is
evident from the onset times of firing of the single motor units
as well as the integrated multiunit EMG recordings. Therefore,
we do not think that the differences are due to sampling bias.

Based on studies of central respiratory drive potentials in
animals (15, 36), the prevailing view suggests that descending
drive is temporally uniform across inspiratory motoneuron
pools (see also Ref. 39) and that the timing of activation
depends on the motoneuron resistance and related intrinsic
properties (Ref. 1, for review, see Ref. 33). This view is
incompatible with one of our findings for tonically active units.
Many of the tonically active units increased their discharge late
in inspiration (particularly in the fifth dorsal external intercostal;
see Fig. 3). Thus these tonically active motoneurons had
consistently responded to an effective suprathreshold excitatory
drive, and so the late increase in inspiratory firing modulation
suggests late arrival of descending inspiratory drive at some motoneurons that were already active. We argue
that the motoneurons receive inspiratory drive that is unevenly
distributed within and between each muscle.

Gradients of inspiratory drive have been observed within the
intercostal muscles in humans (8, 18) and animals (11, 26) and
are functionally linked to local gradients of mechanical advan-
tage across and along intercostal spaces (for review, see Ref.
9). This suggests that control of the timing and amount of
different muscles, there was higher inspiratory flow during
recordings from the diaphragm compared with the third dorsal
external intercostal. However, inspiratory flow was the same
during the experiments in which data were collected from the
scalene and diaphragm muscles, whereas the onset times were
later and firing frequencies were lower for the scalene units
than for the diaphragm units. Thus these differences are not
likely to have affected our major conclusions.

Fig. 6. Onset and peak discharge frequencies for single motor units in the 5
inspiratory muscles. Mean onset (○) and mean peak discharge frequencies (▲)
for each phasic inspiratory motor unit (A) and each tonic inspiratory motor unit
recorded during quiet breathing (B) for the 5 inspiratory muscles. Mean onset
(●) and mean peak discharge frequencies (▲) are shown for each of the 5
inspiratory muscles (phasic and tonic). The diaphragm had no tonic activity.
Only one tonic motor unit was recorded in the 3rd dorsal external intercostal.
For tonic units, the onset firing rate was taken when rate increased. *Signifi-
cantly higher than corresponding frequency from scalene, 2nd parasternal
intercostal, and 5th dorsal external intercostal (P < 0.05). **Significantly
higher than corresponding frequency from scalene and 5th dorsal external
intercostal only (P < 0.05).

highest peak frequencies. There was no relationship between
recruitment time and firing frequency for scalene or fifth dorsal
external intercostal muscles.

DISCUSSION

This study provides evidence for differential distribution of
neural drive to the diaphragm and other human inspiratory
pump muscles during quiet breathing in humans. The onset,
peak, and end motor unit discharge characteristics are similar
for the diaphragm and the third dorsal external intercostal
muscle (Fig. 7). These motor units discharged earlier and at
higher rates than those in the scalene, second parasternal
intercostal, and fifth dorsal external intercostal muscles. Unlike
the diaphragm and third dorsal external intercostal muscles,
which had minimal or no tonic activity, a third of the units
were tonically active in the fifth dorsal external intercostal. The
fifth dorsal external intercostal motor units were activated
significantly later and reached their peak firing later than units
in the other muscles. Inspiratory motor units were recruited
throughout inspiration with a unimodal distribution of onset
times.

Although there was no systematic trend for inspiratory flow,
Ti, or tidal volume to differ when recordings were made from

Fig. 7. Onset, peak, and final firing frequencies for motor units in each of the
5 inspiratory muscles. Discharge frequencies for phasic motor units during
quiet breathing are plotted against %TI (means ± SE). *Compared with the
timing of diaphragm motor units, those in the 5th dorsal external intercostal
discharged later during inspiration (see text).
inspiratory activity is programmed at a “pre-”motoneuronal level and may involve the activity of respiratory-related interneurons (6). These gradients of neural drive are centrally determined and persist after the removal of all afferent input in the dog (for review, see Ref. 9). Such a control mechanism when applied to all the inspiratory pump muscles could explain our findings. One potential contributor could be differences in the degree of monosynaptic excitation of inspiratory motoneuron pools from medullary respiratory centers (6, 20, 29, 38).

The firing rates of single motor units differed between the inspiratory pump muscles. They were ~2 Hz higher in the diaphragm and third dorsal external intercostal muscles. This elevated rate may reflect increased descending drive to the motoneuron pools or differences in the intrinsic properties of the motoneurons (for review, see Ref. 33). There were also large differences in the incidence of tonically firing units across the muscles, with no tonic firing in the diaphragm. The presence of tonic firing may reflect a specific descending tonic drive or separate local tonic activity arising either via persistent inward currents in the specific inspiratory motoneuron dendrites or related excitatory interneurons driving the motoneuron (22). On the other hand, there may be a graded distribution of inhibitory central respiratory drive potentials, which supports tonic firing in expiration.

There are major differences between the behavior of motor units innervating the pump muscles and the upper airway muscles such as the genioglossus (34). First, the peak firing frequency of pump muscles (range 8–13 Hz across the five muscles) is well below that of the peak frequencies in genioglossus during quiet breathing in the supine posture (range 15–25 Hz). Second, there is a high population of genioglossus motor units that discharge tonically with no inspiratory modulation (34). Third, the mean onset of firing is earlier for phasic inspiratory motor units in genioglossus (~20% Ti) than for the earliest recruited pump muscle (diaphragm, 26% Ti, P < 0.001). Such a difference in timing would prepare the airway for inspiratory flow.

Although the method of sampling motor units from humans in vivo to obtain data on samples of inspiratory motoneurons is established (2, 3, 17, 19, 34), it has limitations. First, only single units whose potentials were identifiable from the raw EMG were extracted. Therefore, activity in the lowest threshold units can be missed. To circumvent this problem, we used selective multiunit recordings to assess the earliest onset of inspiratory activity in the vicinity of the electrode. These selective recordings reveal that, in the diaphragm (rather than the other inspiratory muscles), there is early activity in most costal recording sites, with 84% of sites showing multiunit activity before inspiratory airflow (Fig. 3). Nevertheless, new units also continue to be recruited throughout inspiration. Second, an increase in force may be achieved by an increase in firing rate of motor units (rate coding) as well as recruitment of additional units. The diaphragm responds to an increased chemical drive with a larger increase in rates than for muscles such as the scalene (17). The population responses described here provide no information about the balance of rate coding and recruitment for the different muscles. Nevertheless, there were differences in the timing of firing and in the firing frequencies across the different muscles. Our comparison of motor unit behavior between the various inspiratory muscles is made on the assumption that the recordings were acquired during quiet breathing generated by pontomedullary respiratory centers. Electrophysiological and imaging studies in humans support this view (e.g., Refs. 5, 27, 28).

In conclusion, our results suggest that the neural drive is differentially timed to human inspiratory pump muscles. Functionally, this may be important so that the activation of breathing muscles is coordinated to achieve efficient ventilation (7). We propose that this is achieved via a premotoneuronal network that organizes the timing of recruitment of the inspiratory muscles. It is important to determine this because the operation of this system must change in pathological conditions (21, 25, 40, 42).

GRANTS

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