THE SIGNIFICANCE OF THE CROSSED PHRENIC PHENOMENON

by

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To the memory of

BIRDSEY RENSHAW

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INTRODUCTION

The "crossed phrenic phenomenon" originated with an observation in 1895 by Porter during his investigations of the respiratory pathways in the spinal cord (18). He demonstrated physiologically the presence of crossed connections in this system between the descending respiratory fibers of one side and the phrenic motor nuclei of the opposite side. He observed that, in an animal with a spinal hemisection above the level of the phrenic nuclei, section of the phrenic nerve on the intact side resulted in the initiation of or increase in activity of the hemidiaphragm on the side of the hemisection. He showed in other experiments that the descending respiratory impulses crossed in the cord below the hemisection at the level of the phrenic nuclei and nowhere else. This is the experimental situation clarified by the diagram in Fig. 1. Hereafter, the side of the animal ipsolateral to the hemisection of the cord will be referred to as the crossed side. The side of the animal contralateral to the hemisection will be referred to as the direct side. Using this nomenclature, Porter's observation can be restated in this way: In an animal with a spinal hemisection above the level of the phrenic nuclei, interruption of the direct phrenic nerve resulted in the initiation of or increase in activity of the crossed hemidiaphragm. This is the essence of the "crossed phrenic phenomenon", a term which arose because of the continued inability of its numerous investigators to explain it satisfactorily in known physiological terms(22).

As early as 1885 Knoll(12) first made the observation described by Porter. Later, Girard(10) found that in addition to nerve section dyspnea was essential before crossing would occur. However, Langendorff(13)

believed that dyspnea was not essential and Schiff[23] too regarded the cutting of the phrenic nerve as a specific means of bringing about crossed activity. With this Porter agreed and, moreover, proposed an hypothesis to account for the initiation of function in the crossed pathway. He postulated that interruption of the direct phrenic nerve channeled the descending respiratory impulses away from the phrenic nucleus on the direct (intact) side into the phrenic nucleus on the crossed (hemisected) side.

In 1911, Beason and Robb[3] repeated Porter's observation and found that crossed activity also follows stimulation of the sciatic nerve, traction or mechanical stimulation of the direct phrenic nerve (on the intact side of the cord), and dyspnea. They conclude that since "these various conditions induce increased intensity of the bulbar respiratory impulses, the immediate crossing of the bulbar respiratory impulses on section of the phrenic nerve after previous hemisection of the opposite side of the cervical cord seems therefore to be a case of the spread of reflex (or automatic) responses pari passu with the increased intensity of nervous impulses. But since in the Porter experiment the crossing of the impulses appears to be permanent, additional factors are probably involved."[3]

In 1936 and again in 1938 Rosenblueth and his collaborators[22,23] repeated Porter's observation ".....with the purpose of obtaining an explanation of the crossed phrenic phenomenon in terms of the known properties of nerve fibers and synapses. That purpose was not achieved. The phenomenon apparently reveals new, previously unsuspected properties of nervous elements."[1] Their conclusions were that "1) crossed contractions occur directly as a result of cutting or blocking the motor

fibers (of the direct phrenic nerve), not as an indirect effect from relative asphyxia or from changes in the afferent impulses set up by respiratory movements; 2) the central changes are mediated by some process which does not require the conduction of nerve impulses; 3) once the crossed path has been opened--i.e., after a reversible block--subsequent crossings are more readily obtained."(21) In another publication they remark that "....the crossed phenomenon reveals properties of neurone paths differing in quality from the conduction of nerve impulses: the opening of a new, hitherto unused path, and the tendency for such a path to remain open for relatively long periods of time."(22)

In 1941 Seligman and Davis(24) and in 1948 Chatfield and Mead(2) while investigating Porter's observation found many procedures other than phrenic nerve interruption to be effective in producing crossed activity. They failed to explain their findings and "....emphasized that the essential problem of the crossed phrenic phenomenon remains to be solved."(2)

A recent review concludes that "some very fundamental yet unrecognized characteristic of central nervous integration must underlie the crossed phrenic phenomenon"(17). This hypothesis has been regarded as acceptable by several groups of investigators(1,2,6,15,21,22,24). "The phenomenon apparently reveals new, previously unsuspected properties of nervous elements."(1) If true, the proposal of a new property of central nervous system function bears important implications to neurophysiology in general(1) and evidence of similar properties should be sought in other neuronal systems.

However, there is evidence in the papers cited above which raises doubt as to the necessity of such an hypothesis in this regard. The assertion has been made that crossed activity does not occur unless dyspnea is present(3,10). In addition to phrenic nerve interruption, many other procedures have been found to initiate crossing. Some of these procedures were asphyxia or dyspnea(2,3,10,20,21,24), section of the vagi(2,21,22,24), electrical stimulation of the central cut end of the vagus(2), section of the sympathetic trunk(22), direct electrical stimulation of the respiratory center or the respiratory tract on the direct side(21), stimulation of the sciatic nerve(3), stimulation of the central end of the direct phrenic nerve(3), traction on the direct phrenic nerve(3), parenteral injection of nikethamide(2), prostigmine or eserine, acetylcholine protected by prostigmine, and strychnine(24). One characteristic common to these other procedures which induce crossed activity is that they augment activity in the uncrossed pathway as well. In the most recent of these studies(20), unimpeachable electromyographic recordings were used for the first time as indices of diaphragmatic activity. This marked an important advance over the mechanical methods of recording diaphragmatic activity which had been the best methods used up to that time. Not only was the crossed pathway shown to be active after hemisection alone, but crossed activity appeared to vary in proportion to the discharge from the respiratory center. "Crossed activity could be completely and differentially abolished by procedures which decrease respiratory effort (e.g. hyperventilation), and augmented by procedures which enhance respiratory effort (e.g. rebreathing, negative intratracheal pressure)."(20) In these experiments(20) the effects of

phrenic nerve interruption were not studied. The same authors, however, have recently presented evidence from experiments with different preparations that section of peripheral motor axons has no immediate retrograde effect upon the excitability of the parent cell bodies(20).

This summary of the literature reveals three significant facts. First, no group of experiments with this subject has been productive of uniform results; inter- and intra-species variations in the degree of respiratory hemiparesis produced by the spinal hemisection and in the amount of crossing produced by experimental manipulations are reported in every study. Indeed, most authors report variations in the ease with which crossing may be produced at various times in the same animal. Second, the majority of published recordings from both sides of the diaphragm reveal that crossed activity varies in the same direction and in response to the same manipulations which alter uncrossed activity. Third, the Porter experiment has never been demonstrated under conditions which would permit proof that similar alterations did not occur in the uncrossed pathway; in most cases in which crossing has been produced by manipulation of the phrenic nerve on the intact side, the manipulation itself has eliminated the index of respiratory activity in the uncrossed pathway. The single exception to this last statement consists of the brief undocumented description of action potential changes in the phrenic nerves of the two sides(22).

The above facts have prompted a re-investigation of the crossed respiratory activity in an attempt to test the proposed hypothesis of a new property of central nervous pathways in such a fashion as to establish its validity or to reject it entirely. This study was carried out using the most sensitive indicator of diaphragmatic activity available,

electromyographic recordings. In addition, care was taken to maintain a steady state of central respiratory activity throughout the experimental manipulations. Furthermore, a method was devised for recording from the uncrossed respiratory pathway during the period of interruption of the direct phrenic nerve. The results offer no evidence in support of the previously proposed hypothesis and indicate that increased activity in the crossed pathway during interruption of the phrenic nerve on the intact side is due to augmented respiratory center discharge.

METHODS

This study is based upon results derived from eleven cats and one dog. The animals were anesthetized with "Nembutal" (Sodium pentobarbital, Abbott) or "Dial-urethane" (Ciba), the latter being supplemented with ether which was discontinued for at least 30 minutes before the start of recording to allow its expiration and minimise its anesthetic effects during the experiment. Although dosage varied with each animal no alterations in experimental results could be attributed to the anesthetic drugs or to the manner of their use.

Following tracheal cannulation, a dorsal laminectomy of parts of C_1 and C_2 was performed through a midline cervical incision and a hemisection of the cord completed between the roots of C_1 and C_2. In every case the cord lesion was examined by gross inspection of the formalin-fixed spinal cord and in some instances by microscopic examination of serial sections stained for myelin sheaths.

The diaphragm was exposed for recording by wide lateral traction on the edges of a long midline incision from the xiphoid to the umbilicus exposing the abdominal viscera. Further exposure was provided by caudal retraction on the liver and cutting of its cardinal ligament and also by upward traction on the xiphoid by means of a Kocher hemostat suspended from a rigid horizontal bar. Mineral oil on the diaphragm and saline-soaked cotton pads on the viscera helped keep drying of the tissues to a minimum.

For recording, two small hooked electrodes fashioned from insect pins were placed in each diaphragmatic leaf. The distance spanned by each pair varied from a fraction of a millimeter to several millimeters. Each pair

led by means of fine wire to one of two matched differential pre-amplifiers. Muscle action potentials from each diaphragmatic leaf thus amplified were visualised on twin oscilloscopes and photographed side by side on running film. Activity was recorded by this means from each diaphragmatic leaf.

The two tests described by Rosenbaum and Renshaw(20) were used to be certain that recordings from each diaphragmatic leaf were entirely independent of each other. In one test, a single electrical shock applied to the phrenic nerve on the intact side of the spinal cord produced action potentials which were simultaneously recorded with identical amplification from both diaphragmatic leaves. The large deflection recorded from the stimulated side was accompanied by only a minimal deflection recorded from the opposite side. Estimation of the amount of "crossed-recording" was provided by a comparison of the degree of amplification necessary to equilibrate the deflections from the two sides. This approximation provided a conservative estimate of the maximum "crossed-recording" at 3%. In a second test the phrenic on the side of the cord hemisection was interrupted. A large inspiratory discharge recorded from the intact side then produced no recorded deflection on the side of the cord lesion until recordings from that side were amplified considerably above the records from the active side. This test also demonstrated that the recording electrodes in each hemidiaphragm were recording localised activity only and were not being affected by distant events. Before the experimental procedures the recording apparatus was so adjusted that a calibration signal of known voltage produced deflections of identical amplitude on the two channels.

By means of the above-described recording technique it was possible to record only multiple unit activity in the diaphragm. This was difficult

to estimate in a quantitative sense. In an attempt to achieve a more quantitative determination of diaphragmatic activity, spike potentials occurring in individual motor units were recorded. Recording electrodes were fashioned from a pair of insulated fine steel needles positioned with cement on a small cardboard square so that the protruding uninsulated points were within a fraction of 1 mm. of each other(7). When thrust into the muscle, the electrode assembly remained stationary on the moving diaphragm without the need of additional fixation. In spite of the difficulty of maintaining a uniform relationship between recording electrode and motor unit on the constantly moving muscle over long periods of time, it was possible, in a few experiments, to obtain the desired action potentials of single motor units.

The validity of such a recording method in providing an index of total diaphragm activity is based upon the following considerations. Gesell, Atkinson and Brown(9) have found that the number of diaphragmatic single motor unit twitches per inspiration increased with increasing tidal air volume up to a plateau beyond which the number of twitches in any one unit remained uniform. Further increases in tidal air were brought about by recruitment of other units with higher thresholds whose behavior followed curves similar to those of the lower threshold units. Close correlation was observed between tidal air volume and the total number of twitches of all recorded units during any one inspiration. This included units some of which were maximally active, some of which were at threshold activity and some of which were in the process of increasing their number of twitches per inspiration. Although there was little correlation between tidal air volume and twitch number in a motor unit which was either maximally active or at threshold activity, there was

good correlation of tidal air volume with the twitch number of a motor unit during its period of increasing activity. Thus, it would seem that a reliable index of total diaphragm activity (and hence of respiratory center discharge) would be provided by the number of twitches per inspiration in a single motor unit so long as it is first proven that the unit chosen is capable of further increase and is not already maximally active during the control state. Such proof was obtained and described below (pages 16-17). Parallel activity in other units unintentionally recorded in the same experiment as well as similar results of other experiments tend to support the assumption that activity of other units behaves in the same fashion as in those being recorded. Assuming its validity as an index of total diaphragm activity, this quantitative method provided results which could be subjected to graphical representation and statistical analysis (see below Figs. 7 and 8, Table 1). The methods used in previous studies have not permitted such a quantitative treatment of the data.

Interruption of the direct phrenic nerve was accomplished by section or by cold block using a mixture of ice and salt. The cold block was more frequently used because of its reversibility and since it appeared necessary to avoid errors arising from nerve injury discharges(5). For this purpose the nerve was usually isolated in the open chest through a widely retracted intercostal incision which assured access to all contributing fibers. In some cases the nerve was isolated in the neck and interrupted below its most caudal contributing rootlets.

Ventilation was artificially controlled at will by a positive pressure pump which could be operated at a calibrated rate and stroke volume. This allowed a constant level of ventilation to be maintained over a prolonged period of time.

Using these methods, the effects of varying levels of ventilation, of direct phrenic nerve interruption, of certain drugs, or effects of combination of these on diaphragmatic activity were studied.

The general plan of the experimental situation is schematically indicated in Fig. 1. The side of the animal ipsalateral to the hemisection of the cord will be referred to as the crossed side. The side of the animal contralateral to the hemisection will be referred to as the direct side.

RESULTS

The "Crossed Phrenic Phenomenon": Initial experiments in this series dramatically demonstrated the ability of this experimental technique to reveal the increased activity in the crossed hemidiaphragm produced by interrupting conduction in the direct phrenic nerve. Typical recordings obtained from an animal following the indicated hemisection before, during and after cold-blocking the direct phrenic nerve are presented in Fig. 2.

In these records are shown muscle action potentials obtained from each leaf of the diaphragm, the direct side above, and the crossed side below. The slow waves seen in some of the records are artefacts due to gross diaphragmatic movement and could not be entirely eliminated from all records. In Fig. 2A is shown activity present after hemisection alone. On the crossed side, activity was present in spite of hemisection interrupting certain of its spinal pathways. This illustrates the extreme sensitivity of this method of recording since no independent mechanical activity could be seen in that diaphragmatic leaf. At "0" time the direct phrenic nerve was cold-blocked in the neck. Ten seconds after the block began, the increased crossed activity is indicated by the prolongation of the discharge and by the higher frequency of unit activity (Fig. 2B). This crossed activity continued to increase gradually up to the maximum shown in Fig. 2C which was recorded five minutes later. Recovery was slow after removal of the block, requiring about twenty minutes to return to the control level in Fig. 2E which approximates the activity in Fig. 2A before the block began. This same procedure has been repeated in other animals, has invariably produced identical results and has demonstrated that in spite of the different recording methods used the "crossed phrenic phenomenon"

was reproducible. The essence of this "phenomenon" is illustrated in Figs. 2B and 2C by the obvious increase in crossed activity following block of the direct phrenic nerve in an animal with a spinal hemisection.

Alterations of crossed activity in absence of block: Having reproduced the "crossed phrenic phenomenon" it appeared important to confirm previous findings(2,3,20,21,22,24) that manipulations other than interruption of the direct phrenic nerve are also capable of altering the amount of crossed activity. It has been suggested that crossed activity is directly proportional to the amount of central respiratory discharge(3,20). Accordingly, crossed activity was observed during several procedures which were calculated to affect the medullary respiratory center in a manner unrelated to manipulations of the direct phrenic nerve.

The marked influence that level of ventilation alone can have on crossed activity is shown in Fig. 3 from a typical experiment. After hemisection alone, with the chest closed, activity was present on both sides as shown in Fig. 3A. Before opening the chest, artificial ventilation was begun and was maintained constant at four separate levels of respiratory minute volume. Figures 3B to 3E inclusive were recorded after stable activity was reached at each of these levels of respiration. It deserves to be emphasized that after such changes equilibrium could not be attained in less than 5-10 minutes. At a respiratory minute volume of 600 cc/min. (Fig. 3B) activity had disappeared on the crossed side and was all but gone on the direct side. At 480 cc/min. (Fig. 3C) increased tonus was evident in the direct hemidiaphragm and isolated sporadic unit twitches were present in the crossed hemidiaphragm. Following the reduction of respiratory

minute volume to 360 cc/min. (Fig. 3D) activity in the direct hemidiaphragm increased still more and became rhythmic while evidence of tonic activity appeared in the crossed hemidiaphragm. At 240 cc/min. respiratory minute volume (Fig. 3E) which approximated the normal state (cf. Fig. 3A), rhythmic activity was obvious in both leaves of the diaphragm. In other experiments, direct activity as well as crossed activity was completely abolished by even higher levels of ventilation, and still further increased activity than this figure shows was produced by more severe hypoventilation. Since the amount of crossed activity varied in a predictable manner with alterations in respiratory minute volume (always in a direction opposite to the change in ventilation), it thus appeared as if the amount of activity in the crossed hemidiaphragm were responding, in part at least, to the changes in drive from the respiratory center.

Under conditions of constant controlled respiratory minute volume, alterations in crossed activity were also produced by procedures which affect the medullary respiratory center activity independently of changes in ventilation.

The effect of a central respiratory depressant, pentothal sodium, upon both crossed and uncrossed activity recorded during constant pulmonary ventilation is illustrated in Fig. 4. Obvious depression of diaphragmatic activity on both sides resulted, as shown in Figs. 4B and 4C, beginning within thirty seconds after the injection. Return to the control state required approximately thirty minutes.

The effect of respiratory center excitation is illustrated in records C, D and E of Fig. 5. Artificial ventilation was carefully adjusted to a level just sufficient to abolish crossed activity and held constant at this

same level throughout the experiment. A small amount of potassium cyanide rapidly injected intravenously (Fig. 5C) was followed within thirty seconds by the appearance of crossed activity along with an increase in direct activity (Fig. 5D). This rapidly returned to the control state in Fig. 5E which approximates the activity at the time of injection (Fig. 5C).

These results confirm previous observations(2,3,20,21,22,24) that activity in the crossed pathway to the phrenic nerve may be modified even in the presence of an intact conduction pathway over the direct phrenic nerve. They also indicate that the amount of crossed activity varies in a fashion similar to the amount of direct activity, in direct proportion to the intensity of the discharge from the medullary respiratory center.

Alterations of crossed activity in the presence of direct phrenic block: Having seen how crossed activity varies with alterations in respiratory drive brought about by experimental variables, the effects of interruption of the direct phrenic nerve were next studied 1) while all known variables were held constant and 2) while certain of these variables were altered in a controlled manner.

Fig. 5 presents results of this nature. As already stated, artificial ventilation was adjusted to a level just sufficient to abolish activity in the crossed hemidiaphragm and held constant at this same level throughout the experiment. Under these conditions (Fig. 5A), block of the direct phrenic nerve alone (Fig. 5B) failed to produce any crossed activity. During the second direct phrenic block (Fig. 5F), which again failed to produce any crossed activity, a small amount of KCN was rapidly injected intravenously. Once more crossed activity appeared within thirty seconds

after the injection, presumably due to reflex respiratory center excitation from the cyanide. It should be noted that the amount of crossed activity produced by cyanide in the presence of block (Fig. 5C) is not appreciably different from that produced by cyanide in the absence of block (Fig. 5D). The rapid return to the control state is illustrated by a comparison of records 5A and 5E.

Further evidence that alterations of crossed activity in the presence of direct phrenic block may be due to alterations in the discharge from the respiratory center is illustrated in Fig. 6. In these records it is seen that under conditions of constant controlled artificial ventilation, direct phrenic nerve section produced no obvious change in crossed activity (cf. Figs. 6A and 6B). Subsequently induced alterations in respiratory exchange were accompanied by alterations in crossed activity which were in the same direction as the presumed changes in respiratory center activity, increasing with hypoventilation (Fig. 6C) and decreasing with hyperventilation (Fig. 6D). These changes were entirely reversible.

The failure of direct phrenic nerve section to produce an obvious change in crossed activity may have been due to our inability to detect minor changes in the massive barrage of potentials from the diaphragm. This prompted the use of the quantitative method (described above pp. 9-10) in which single motor unit action potentials were recorded as the index of total diaphragm activity and hence of respiratory center discharge.

One such experiment is illustrated in the graph of Fig. 7. The clarity with which individual motor units were recorded from each diaphragmatic leaf is shown in the inserts in the graph. That neither of these units was maximally active during the control period was shown 1) by an increase in their activity which accompanied hypoventilation (not illustrated)

and 2) by the gradual increase in unit activity throughout the run (see graph, Fig. 7) which was due to some non-specific alteration in the state of the preparation unrelated to the blocking of the direct phrenic nerve. The graph itself presents counts of the number of these twitches during each inspiration plotted against time, measured by the number of respirations. During constant controlled artificial ventilation, the direct phrenic nerve was cold-blocked. The period of application of cold to the nerve, as indicated by the black bar, is understandably briefer than the period of functional block as indicated by the cessation of activity on the direct (Rt.) side. During this period of interruption of the direct phrenic nerve, the number of twitches per inspiration on the crossed side shows no significant change. This impression was confirmed by statistical analysis of the data represented in the graph. Comparison of the mean of the pre- and post-block control periods with the mean of the period of block reveals no significant change during the block(4).

The results of a similar experiment are presented in Fig. 8. The upper records of this figure (8A, 8B and 8C) constitute proof that the "crossed phrenic phenomenon" was demonstrable in this animal while respiration remained spontaneous and uncontrolled. During the period of direct phrenic block (Fig. 8B), altered crossed activity was evidenced by: 1) increased duration of the total inspiratory discharge, 2) increased frequency in several of the single motor units, and 3) recruitment of previously inactive units. Controls before and after block may be compared in Figs. 8A and 8C. In the lower set of records (Figs. 8D, 8E and 8F), obtained from the same animal, the experiment was repeated in exactly the same way except for the use of constant, controlled artificial ventilation

throughout the run. No change in the amount of crossed activity whatsoever (Fig. 8E) occurred with this block, during constant ventilation. In these records, one single motor unit from each hemidiaphragm stood out with sufficient clarity under enlargement to permit counting. The number of these motor unit twitches per inspiration during the entire run is presented in the graphs below the records and also reveals the failure of the crossed unit to increase its activity during the period of the direct phrenic block.

Results of these quantitative recordings (Figs. 7 and 8) bear out the findings in previous qualitative records (Fig. 6) and prove that interruption of the direct phrenic nerve during constant artificial ventilation failed to produce even minor alterations in the amount of crossed activity. Furthermore, this was demonstrated with both methods a total of seventeen times in eleven cats and one dog.

It has been seen that alterations in crossed activity during interruption of the direct phrenic nerve occurred only 1) when ventilation was uncontrolled (e.g. Figs. 2 and 8) or 2) as a result of some procedure which was calculated to alter the activity of the respiratory center (e.g., Figs. 5 and 6). Since crossed activity always changed in the same direction as the presumed alteration in respiratory center drive, it would be simplest to explain the "crossed phrenic phenomenon" in the following terms: Interruption of the direct phrenic nerve in a spontaneously breathing animal produces increased crossed activity by intensifying the discharge from the respiratory center.

Influence of phrenic block upon respiratory center discharge. Throughout the experiments dealing with interruption of the direct phrenic nerve it could only be assumed that respiratory drive behaved in the expected fashion during the period of the block. This assumption was necessary because the paralysis of the direct hemidiaphragm during the block period prevented its use at that time as an index of respiratory drive. To circumvent this difficulty and to eliminate the need for such an assumption, an index of respiratory drive was required which would allow its measurement during the block period as well as during the control periods. Records from portions of the internal intercostals active during inspiration should afford as valid an index of respiratory center activity as records from the diaphragm. Indeed, repeated observations(8) on such muscles indicate that they behave in a manner identical to the behavior of the diaphragm. Accordingly, action potentials recorded from bilaterally symmetrical portions of these muscles provided a continuous measure of direct as well as crossed activity during block of the direct phrenic nerve, although proof of the completeness of the block had to be sacrificed.

Data from such an experiment are presented in Table 1. The figures are derived from recordings of single motor units in bilaterally symmetrical interchondral portions of internal intercostal muscles. Counts of the number of these single unit twitches per inspiration on each side were made before, during and after direct phrenic nerve block. In the first run, the animal was allowed to breathe spontaneously; while in the second run, ventilation was controlled artificially. In the upper portion of the table are shown the mean number of twitches per inspiration on each side during spontaneous respiration comparing the control periods (left) with the block

period (right). A statistically significant increase in the amount of activity on both sides during the period of the block is indicated by the large value of t for the difference between these means(4). After artificial ventilation was began, the crossed intercostal unit was lost. Another unit was found which increased its activity markedly with slight decreases in ventilation and this unit was used thereafter as the index of crossed activity. Repetition of the direct phrenic block under these conditions of constant artificial ventilation produced no significant change in activity on either side during the period of the block as shown by the low t values.

Microscopic examination of Weil stained sections of the spinal cord lesion in this experiment showed incomplete hemisection of the cord such that a small portion along the medial edge of the ventral column remained unsectioned. In the light of the findings of Pitts(16) and of Rosenbaum and Renshaw(20) the functional capacity of this ventral column remnant is believed to have been insignificant.

In this type of experiment the intensity of central respiratory discharge, indicated by the amount of direct activity, was shown to increase significantly with direct phrenic block while respiration was uncontrolled. But during constant artificial ventilation, direct phrenic block failed to produce any change in respiratory drive whatsoever.

DISCUSSION

The results derived from these experiments provide the basis for a simple and logical explanation of what has been termed a "phenomenon".

In confirmation of Rosenbaum and Renshaw(20) these results indicate that the crossed respiratory pathway is normally active under the conditions of these experiments. No specific manipulation of the direct phrenic nerve was required to initiate conduction through crossing neurons. The number of phrenic motor neurons activated by crossed connections was small. This was apparent from the fact that it was often necessary to search for electromyographic evidence of activity in the crossed hemidiaphragm. The paucity of crossed activity made it impossible, in many instances, to rely upon visual evidence of the lack of independent muscular action. Mechanical recordings, such as those used previously, were not sufficiently sensitive to detect the small amount of movement and were inevitably affected by movements transmitted from the direct hemidiaphragm. For these reasons, evidence based upon mechanical recordings is unreliable and has led to the formation of erroneous conclusions.

Although the number of active motor units in the crossed hemidiaphragm was relatively small, the evidence indicates that the crossed pathway behaved in a fashion identical to the behavior of the direct pathway and of other inspiratory muscles(8). Augmentation of respiratory center discharge resulted in an increased rate of firing of crossed phrenic units and recruitment of new units into the pattern of each inspiratory cycle. The results again reveal that crossed activity is abolished more readily than direct activity by influences which reduce central respiratory discharge(20), suggesting that the crossed phrenic neurons require a greater

intensity of presynaptic bombardment to initiate their firing. This is probably the basis for the variability in the amount of respiratory hemiparesis produced by cord hemisection, and for the variability in the ease with which crossing could be produced at different times in the same animal, which are such outstanding features of the results of previous investigators.

With the use of adequate means of controlling ventilatory rate, and with attention to the general condition of the preparation, it was possible to obtain the necessary steady state of central respiratory activity upon which to superimpose the effects of interruption of conduction in the direct phrenic nerve. The recognition of such a steady state requires a highly sensitive index of central respiratory activity which has not been used in previous studies. The production of such a steady state requires relatively protracted periods of uniform artificial ventilation before equilibrium is reached; a uniform level of anesthesia; a stable circulatory condition; and the avoidance of alterations of afferent activity from general somatic afferents in the vicinity of wounds, etc. The appreciation of the importance of maintaining a uniform discharge from the respiratory center during direct phrenic interruption is evident in the reports of the earlier investigators. However, no evidence that such a condition was actually achieved is available from the published protocols or records. When a steady level of central respiratory discharge was produced in the present experiments, no detectable alteration of crossed activity accompanied interruption of conduction over the direct phrenic nerve.

On the other hand, when alterations secondary to direct phrenic interruption were permitted to affect changes in central respiratory discharge,

changes in crossed phrenic activity invariably occurred. In previous studies of this nature, activity in the direct hemidiaphragm was the only uncomplicated index of the intensity of the central respiratory discharge. When this index was eliminated by manipulation of the direct phrenic nerve, the experiments became uncontrolled and difficult of interpretation. The use of intercostal muscles, instead of the diaphragm, obviated this difficulty by affording an index of central respiratory discharge which could be observed continuously throughout the period of direct phrenic nerve block. These intercostal recordings offer conclusive evidence that interruption of direct phrenic conduction during spontaneous ventilation produced an increase in central respiratory discharge reflected as an increase in both direct and crossed intercostal activity. Thus, the increase in crossed phrenic activity, under similar conditions, can be ascribed to alterations in central respiratory drive.

The augmentation of central respiratory discharge following direct phrenic block during spontaneous ventilation is probably due to at least two important factors. The immediate increase in activity, appearing during the first inspirations following the block, may result from paralysis of the direct hemidiaphragm and failure of the normal amount of lung inflation. The resulting alteration in afferent vagal activity from pulmonary stretch receptors would be sufficient to bring about increased discharge from the respiratory center(11,14). This is not inconsistent with the findings of Chatfield and Mead(2). The slow, gradual increase following the initial change could very well be due to altered blood gas tensions resulting from inadequate ventilation. Such changes have been demonstrated to occur simultaneously with the appearance of increased crossed activity(25).

This may very well be the basis for the delayed development of crossed activity and its persistence after removal of the direct phrenic block which appears so strikingly in some of the records of earlier investigators as well as in this study.

Of widespread interest in this regard has been the work of Fleisch and his collaborators(6) who recorded action potentials from the phrenic nerves themselves and found increased activity in both nerves following interruption of one nerve (spinal cord intact). They suggest that the increased phrenic activity is due to a proprioceptive reflex initiated by interruption of the nerve and that the afferent impulses arrive centrally in the motor roots of the phrenic nerve itself. An alternative explanation favored by these authors is that "a nerve cell in discharging an impulse along a fiber is informed by this same fiber whether or not the impulse arrives at its destination"(6), ..."the intensity of a nervous discharge is reinforced if that discharge is prevented from reaching its goal"(5). Certain features in the experimental data they present, however, make it difficult to accept their interpretations. First, the condition of their experimental animals seemed precarious inasmuch as out of 50 animals only 5 lived long enough for records to be made, a duration of approximately 3 minutes(15). Second, their method of quantitating results depended upon the assumption that any increase in activity occurring in the nerve would summate with that already existing. Such records of the action potentials should show an increase in amplitude corresponding to the addition of the new responses to the pre-existing basal level. Published records(5,p.84,Fig. 3) show that this did not always occur thus proving this method of quantitation unreliable. And finally, augmentation of inspiratory discharges occurred

only after the interruption of one phrenic. If either of the two hypothetical explanations of this observation were true, augmentation would be predicted as a result of interruption of the second phrenic as well as the first. This did not occur. No significant change at all occurred in the phrenic nerves following this second block(5). In view of these inconsistencies and departure from predicted results, another explanation for the observations of Fleisch and his collaborators must be sought.

We thus arrive at the following conclusion. In a spontaneously breathing animal with a hemisection of the spinal cord above the phrenic motor nucleus, interruption of conduction in the direct phrenic nerve results indirectly in an augmentation of central respiratory discharge which is reflected as an increase in activity in the crossed respiratory pathway. Furthermore, the results offer no evidence that any additional, subtle, obscure alteration takes place in the crossed respiratory pathway, for when adequate control of experimental conditions permits the achievement of a steady state, the "crossed phrenic phenomenon" vanishes.

SUMMARY

1. In barbiturate anesthetized animals with spinal hemisections at C_1-C_2, muscle action potentials were recorded simultaneously from each hemidiaphragm during various procedures designed to explain the appearance of increased crossed activity (ipsilateral to the hemisection) following interruption of the direct phrenic nerve (contralateral to the hemisection).

2. The activity of the crossed hemidiaphragm was always less than the activity of the direct hemidiaphragm.

3. Using activity on the direct side as an index of central respiratory discharge, activity on the crossed side was found to vary in a fashion parallel to activity on the direct side during procedures which altered the intensity of the central respiratory discharge.

4. Activity on the crossed side increased following interruption of the direct phrenic nerve during spontaneous uncontrolled ventilation. Qualitative and quantitative recordings revealed that such a change did not occur during controlled artificial ventilation.

5. Central respiratory discharge increased following interruption of the direct phrenic nerve during spontaneous uncontrolled respiration. Such a change did not occur during controlled artificial ventilation.

6. It is concluded that the amount of crossed respiratory activity is directly proportional to the intensity of the central respiratory discharge and is entirely independent of the state of conduction in the direct phrenic nerve. The "crossed phrenic phenomenon" occurs only when experimental conditions permit an augmentation of central respiratory discharge to take place as a secondary result of interruption of the direct phrenic nerve.

Figure 1

From the respiratory center shown in the medulla, nervous pathways descend on each side of the spinal cord to the phrenic motor nuclei from whence nerve impulses traveling in the phrenic nerves activate each leaf of the diaphragm. Descending spinal pathways were experimentally interrupted, as shown, by hemisection of the spinal cord between C_1 - C_2 and by section or cold block of the direct phrenic nerve, as indicated by the arrow.

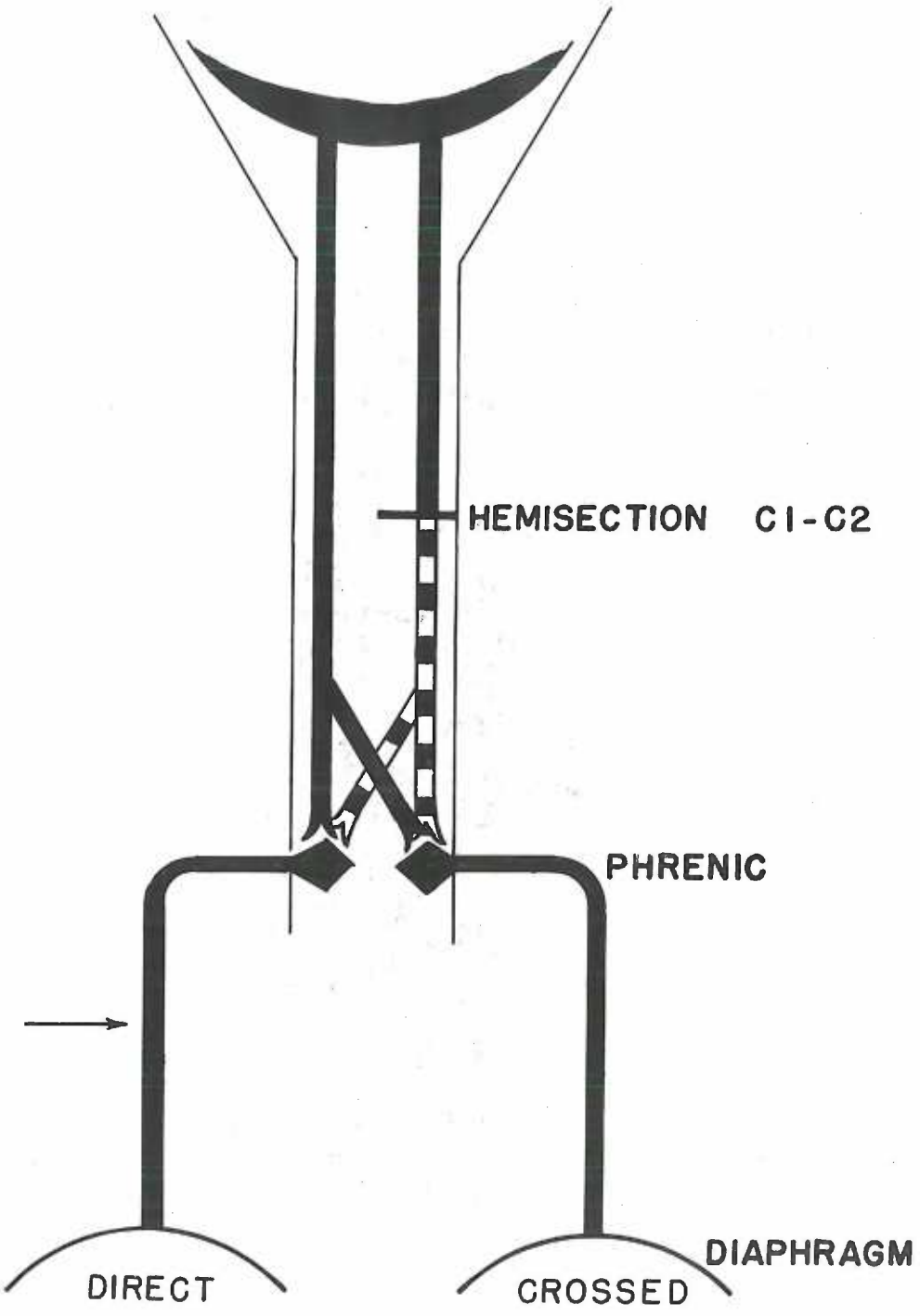

HEMISECTION C1-C2
PHRENIC
DIAPHRAGM
DIRECT
CROSSED

Figure 2

The "crossed phrenic phenomenon". Cat. Dial. Left hemisection. Chest closed. Muscle action potentials simultaneously recorded from each leaf of the diaphragm. Right phrenic nerve cold-blocked in neck from "0" time (between A and B) to 5' 15" (just after C) during spontaneous uncontrolled respiration (5-10-50).

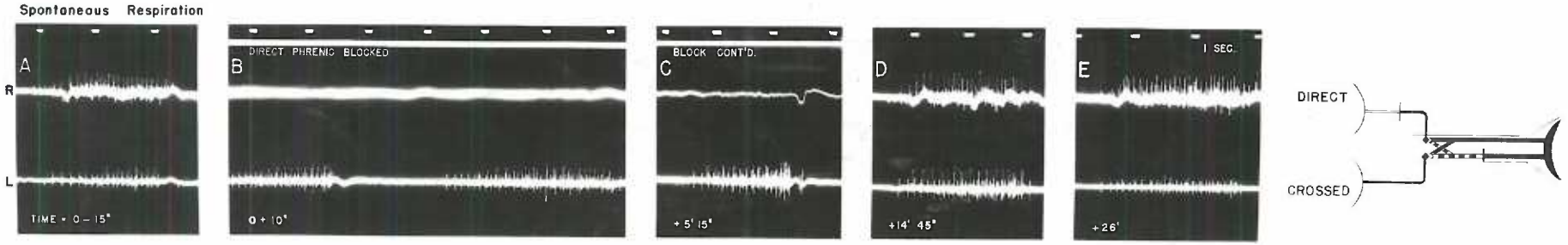
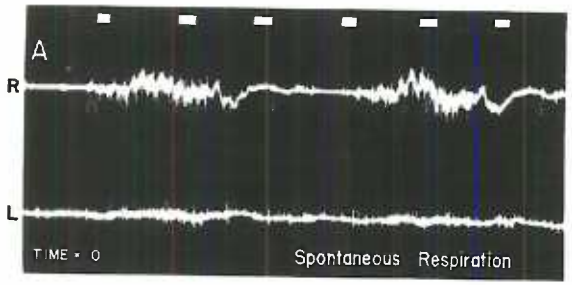
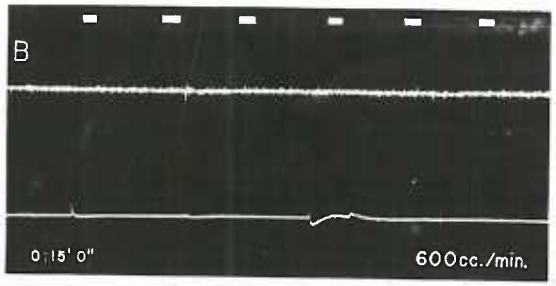
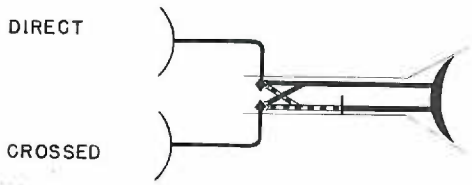

Spontaneous Respiration
A
R
L
TIME = 0 – 15"
DIRECT PHRENIC BLOCKED
B
0 + 10"
BLOCK CONT'D.
C
+ 5' 15"
D
+14' 45"
1 SEC.
E
+26'
DIRECT
CROSSED

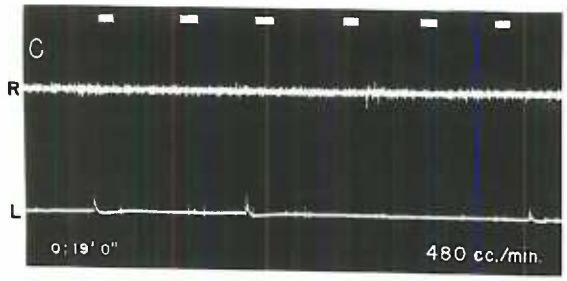
Figure 3

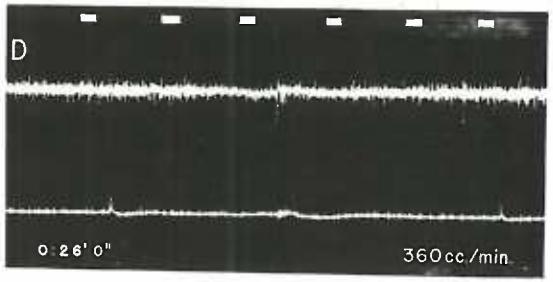
Effect of artificial ventilation of varying levels upon diaphragmatic activity. Cat. Dial. Left hemisection. A: closed chest control during spontaneous respiration. B to E inclusive: open chest, controlled artificial ventilation decreased in four stages from 600 cc/min. (B), to 480 cc/min. (C), to 360 cc/min. (D), to 240 cc/min. (E). The records shown were taken after stable activity at each stage was reached, requiring from four to ten minutes after each change in ventilation (11-29-49).


A
R
L
TIME = 0
Spontaneous Respiration


B
0:15' 0"
600cc./min.


DIRECT
CROSSED


C
R
L
0:19' 0"
480 cc./min.


D
0:26' 0"
360cc/min.

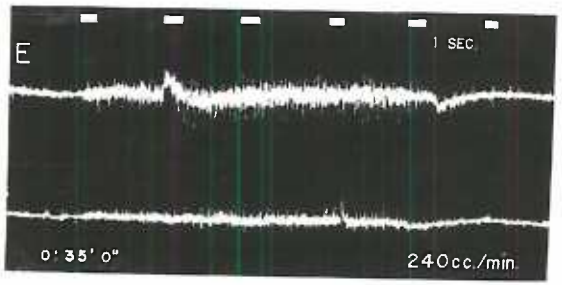

E
1 SEC
0:35' 0"
240cc./min.

Figure 4

Effect of central respiratory depression on diaphragmatic activity. Cat. Dial. Left hemisection. Open chest. During constant controlled artificial ventilation (240 cc/min.), 10 mgm. pentothal sodium rapidly injected intravenously. B and C recorded at indicated time after injection (11-29-49).

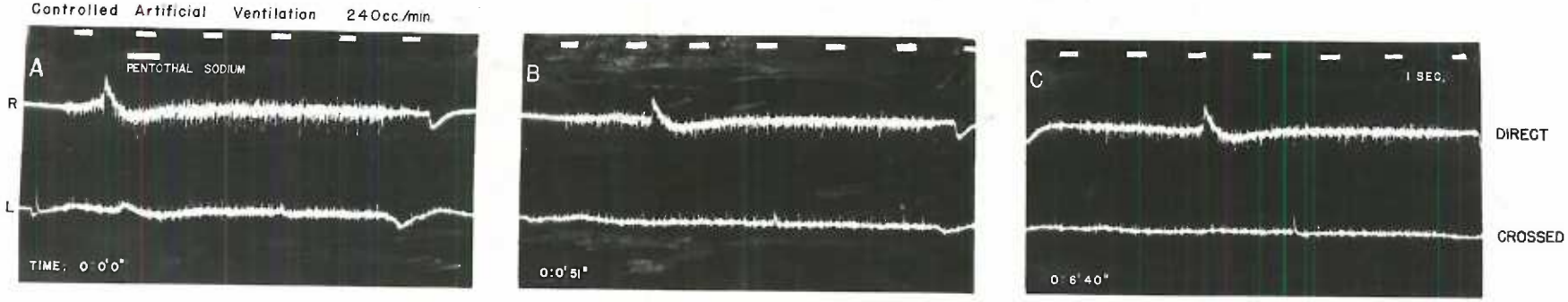

Controlled Artificial Ventilation 240cc./min.
A
PENTOTHAL SODIUM
R
L
TIME. 0:0'0"
B
0:0'51"
C
1 SEC.
0:6'40"
DIRECT
CROSSED

Figure 5

Effects of central respiratory excitation upon diaphragmatic activity with and without direct phrenic nerve interruption. Cat. Dial. Left hemisection. Open chest. Ventilation adjusted to level of disappearance of crossed activity and held constant throughout. A: Control state. B: Direct phrenic blocked in chest. Note absence of crossed activity. C, D and E: Response to 0.13 mgms. KCN intravenously. F and G: Response to 0.13 mgms. KCN intravenously during maintained block of direct phrenic nerve. H: Return to control state. Note equivalence of crossed activity in D and G (2-20-50).

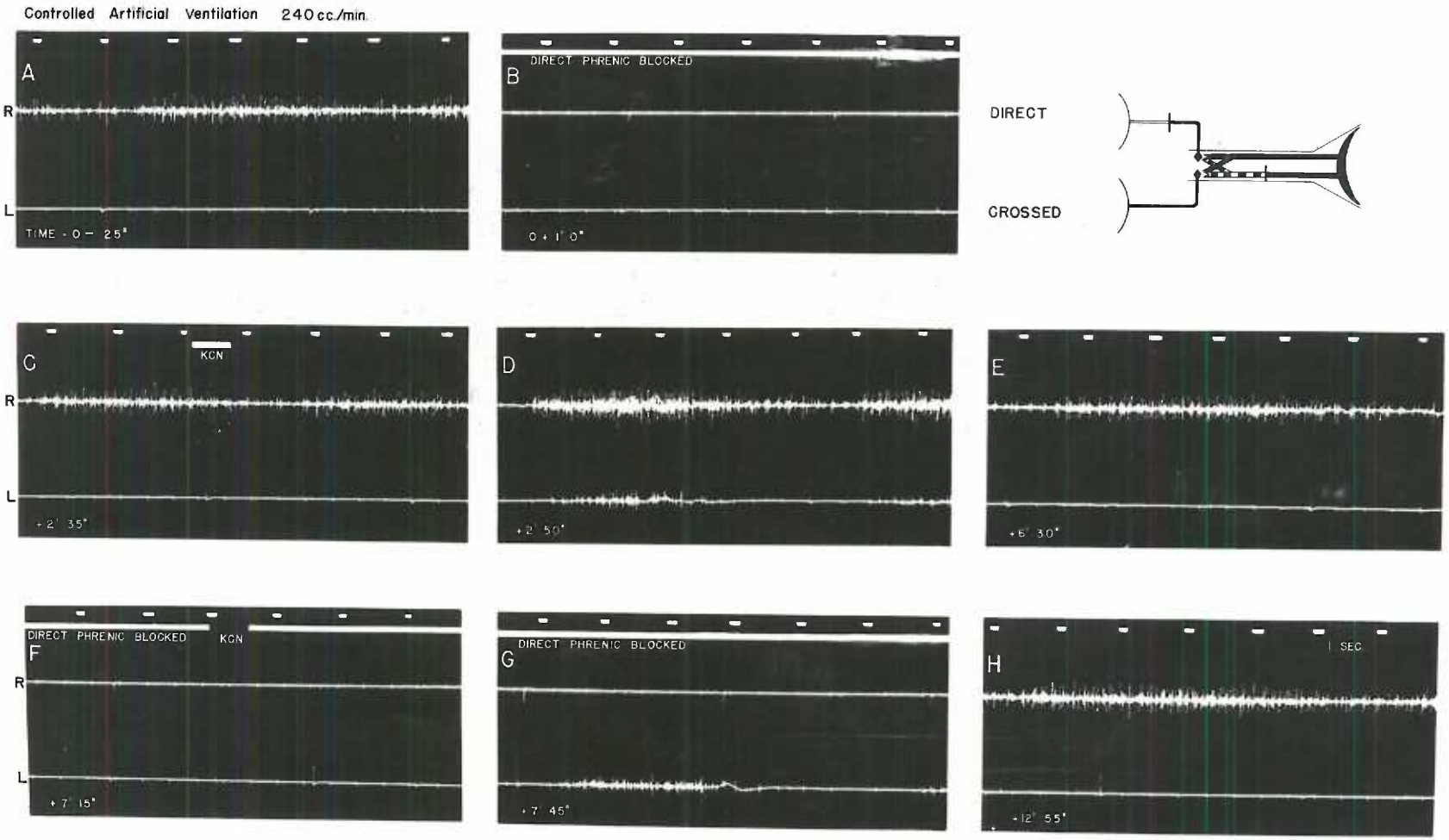

Controlled Artificial Ventilation 240 cc./min.
A
R
L
TIME - 0 - 25"
DIRECT PHRENIC BLOCKED
B
0 + 1' 0"
DIRECT
CROSSED
C
KCN
+2' 35"
D
+2' 50"
E
+5' 30"
DIRECT PHRENIC BLOCKED
KCN
F
+7' 15"
DIRECT PHRENIC BLOCKED
G
+7' 45"
1 SEC
H
+12' 55"

Figure 6

Effects on diaphragmatic activity of altering pulmonary ventilation after direct phrenic nerve section. Cat. Nembutal. Right hemisection. Open chest. Uniform artificial ventilation in A and B. A: Control state before direct phrenic nerve section in the chest. B: One minute after section. C: Artificial ventilation stopped for 20 sec. from 4' 25" to 4' 45". D: Apnea during hyperventilation. Results illustrated in C and D were entirely reversible (8-26-49).

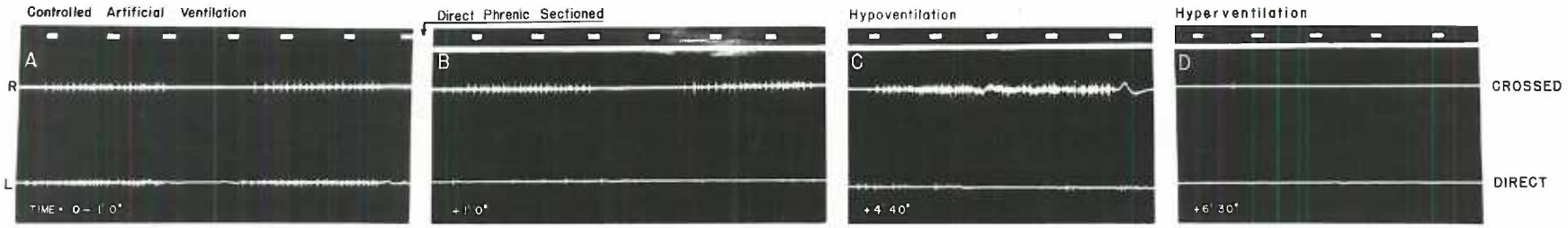

Controlled Artificial Ventilation
Direct Phrenic Sectioned
Hypoventilation
Hyperventilation
A
B
C
D
R
L
CROSSED
DIRECT
TIME - 0 - 1' 0"
+1' 0"
+4' 40"
+6' 30"

Figure 7

Effect of direct phrenic nerve interruption under controlled conditions recorded quantitatively. Cat. Dial. Left hemisection. Open chest. During constant controlled artificial ventilation the direct phrenic nerve was cold blocked in the chest. Graph illustrates number of discharges per inspiration of a single motor unit on each side before, during and after the block. Inserts illustrate clarity of single unit recordings (4-10-50).

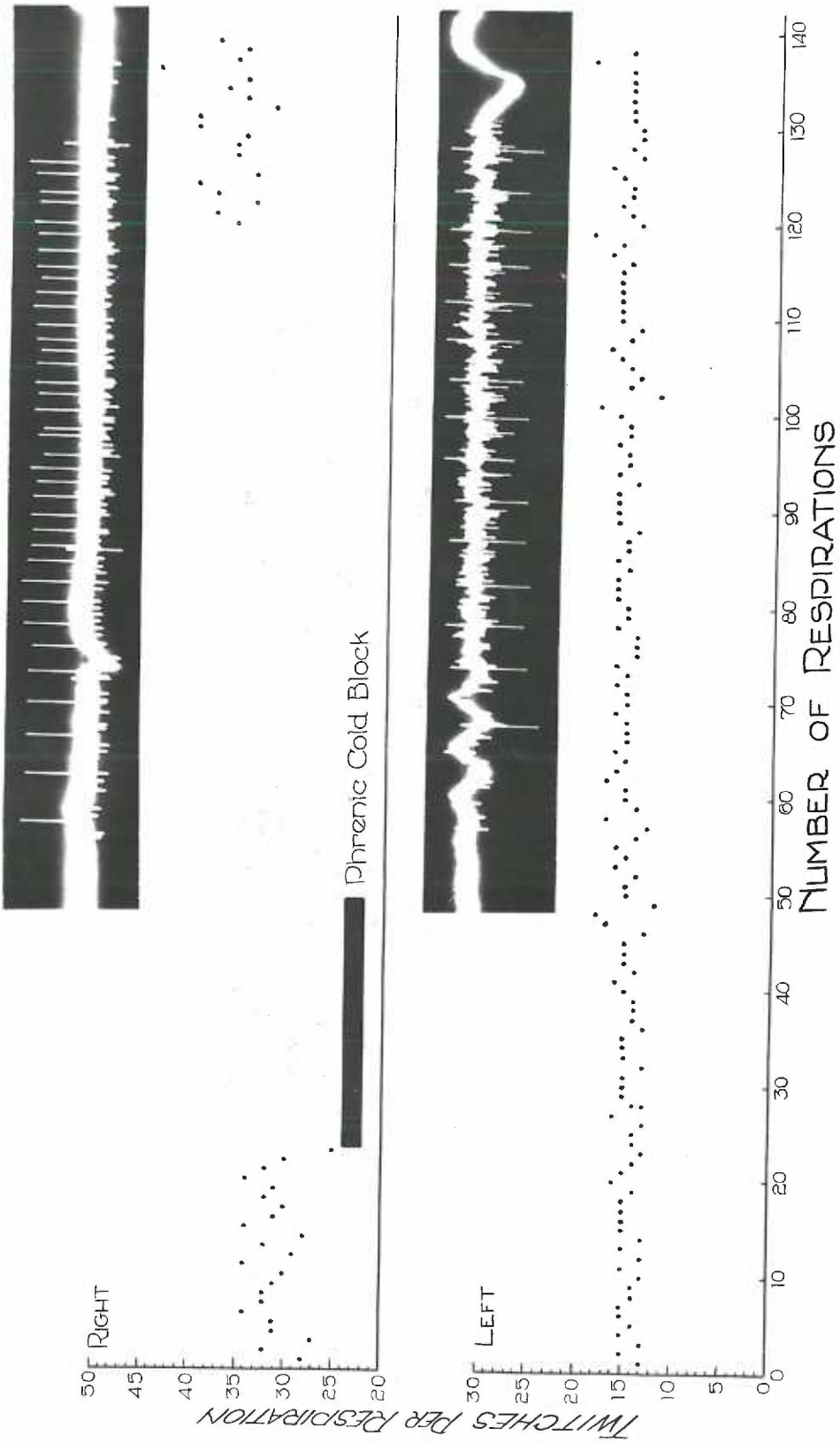

Phrenic Cold Block
RIGHT
LEFT
TWITCHES PER RESPIRATION
NUMBER OF RESPIRATIONS
50
45
40
35
30
25
20
30
25
20
15
10
5
0
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140

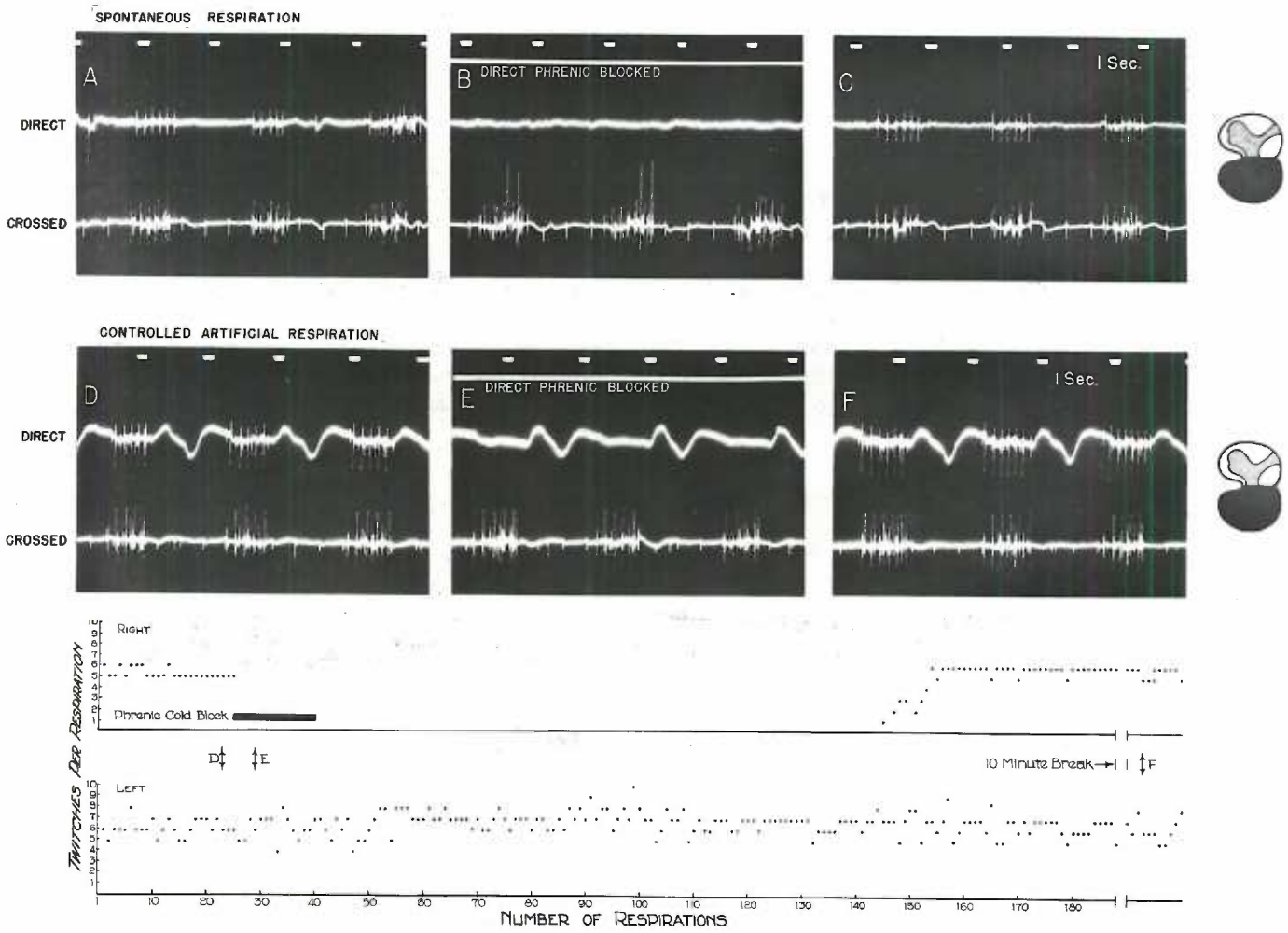
Figure 8

Effect of direct phrenic nerve interruption under variable and under controlled conditions. Cat. Dial. Left hemisection.

Above: Closed chest, spontaneous respiration. A and C: Before and after block of direct phrenic nerve in the neck. B: During direct phrenic cold block. Note augmentation of crossed activity.

Middle: Closed chest, controlled artificial ventilation. D and F: Before and after block of direct phrenic nerve in neck. E: During direct phrenic block.

Below: Graph showing the number of discharges per inspiration of a single motor unit on each side during entire period from which D, E and F were selected. Lettered arrows indicate periods illustrated in records. Note uniformity of crossed activity throughout run (5-17-50).


SPONTANEOUS RESPIRATION
A
B DIRECT PHRENIC BLOCKED
C
1 Sec.
DIRECT
CROSSED
CONTROLLED ARTIFICIAL RESPIRATION
D
E DIRECT PHRENIC BLOCKED
F
1 Sec.
DIRECT
CROSSED
RIGHT
Phrenic Cold Block
D
E
10 Minute Break
F
LEFT
TWITCHES PER RESPIRATION
1 10 20 30 40 50 60 70 80 90 100 110 120 130 140 150 160 170 180
NUMBER OF RESPIRATIONS

SINGLE MOTOR UNITS OF INTERCHONDRAL PORTIONS OF
INTERNAL INTERCOSTALS IN 5TH INTERSPACE

	PRE AND POST BLOCK CONTROL PERIODS			PERIOD OF DIRECT PHRENIC BLOCK			
	No. of Resps.	Twitches per resp. mean	S. D.	No. of Resps.	Twitches per resp. mean	S. D.	t for difference between means
SPONTANEOUS RESPIRATION							
Direct	68	9.79	.761	35	15.31	1.884	14.8
Crossed	68	1.81	.709	35	3.43	.698	7.75
ARTIFICIAL VENTILATION							
Direct	60	4.17	.679	51	4.196	.873	0.126
Crossed	56	6.50	.818	51	6.82	.74	1.35

t for α .01, d.f. 120 = 2.62

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