J. Physiol. (1977), **272**, *pp.* 367–382
With 7 text-figures
Printed in Great Britain

SYNAPTIC ACTIONS OF INDIVIDUAL VESTIBULAR NEURONES ON CAT NECK MOTONEURONES

BY S. RAPOPORT, A. SUSSWEIN, Y. UCHINO AND V. J. WILSON

From the Rockefeller University, New York, N.Y. 10021, U.S.A.

(*Received* 7 *February*, 1977)

SUMMARY

1. Unitary synaptic potentials evoked by the activity of single vestibulocollic neurones were recorded by means of spike-triggered signal averaging in neck extensor motoneurones of decerebrate cats. Properties of the vestibulocollic neurones which produced the potentials were examined.

2. Vestibulocollic neurones were first identified as projecting to the C3 grey matter by antidromic microstimulation within the C3 extensor motoneurone pool. The spontaneous or glutamate-driven activity of the vestibulocollic neurones was then used to trigger the averaging computer. In this way ten inhibitory and two excitatory neurones were identified (20 % of neurones tested).

3. Action potentials in local branches of vestibulocollic neurones were usually recorded in the vicinity of motoneurones. Mean orthodromic conduction time from the foot of the extracellular spike, recorded in the vestibular nuclei, that triggered the averager was 0·72 msec. Mean synaptic delay was 0·4 msec.

4. I.p.s.p.s had a mean time to peak of 0·81 msec and were readily reversed by injection of hyperpolarizing current. These data, together with the shape indices of i.p.s.p.s indicate that they are generated proximally on motoneurones.

5. All vestibulocollic neurones making synapses with motoneurones were monosynaptically driven by stimulation of the ipsilateral vestibular nerve. Four out of seven tested were inhibited by stimulation of the contralateral vestibular nerve (commissural inhibition).

6. Two excitatory neurones were located in Deiters' nucleus or on the Deiters'-descending border. Inhibitory neurones were found relatively medially in the vestibular complex in the medial, descending and Deiters' nuclei.

7. Vestibulocollic neurones acting on motoneurones were tested for

axon branching to more caudal levels of the spinal cord with electrodes placed at C5–7. Both of the excitatory and two out of nine inhibitory neurones branched.

INTRODUCTION

Many vestibulospinal neurones with axons in the lateral and medial vestibulospinal tracts (LVST and MVST) make monosynaptic connexions with neck motoneurones, and are part of a disynaptic pathway linking the labyrinth to these motoneurones (Wilson & Yoshida, 1969*a*, *b*; Akaike, Fanardjian, Ito & Ohno, 1973*b*). These vestibular neurones projecting to the neck segments (vestibulocollic neurones) are heterogeneous in many ways. Some are excitatory, others inhibitory (Wilson & Yoshida, 1969*a*, *b*; Akaike *et al.* 1973*b*); many receive short-latency input from the labyrinth, while others do not (Rapoport, Susswein, Uchino & Wilson, 1977). Some vestibulocollic neurones have axons projecting only as far as the neck segments while the axons of others (branching neurones) continue caudally to the cervical enlargement or even to the thoracic cord after giving off one or more collaterals to the upper cervical grey matter (Rapoport *et al.* 1977).

These observations raise several questions. For example, do both branching and non-branching vestibulocollic neurones make monosynaptic connexions with motoneurones? Is branching a property of excitatory and inhibitory neurones? Because of the intermingling of vestibulocollic neurones with different properties (Fig. 2 in Rapoport *et al.* 1977) it is difficult, perhaps impossible, to answer such questions even with very localized microstimulation in the vestibular nuclei. Therefore, we have chosen to use the technique first introduced by Mendell & Henneman (1971), developed by Jankowska & Roberts (1972) for use in the central nervous system, and already applied to descending connexions by Kirkwood & Sears (1973). Vestibulocollic neurones were identified by low-threshold antidromic activation and their activity, evoked by glutamate leakage or ejection from one barrel of a two-barrel electrode, was used to trigger an averaging program while recording intracellularly from neck motoneurones penetrated in the vicinity of the antidromic stimulation site. This procedure, which reveals monosynaptic connexions between the vestibulocollic neurone and the penetrated motoneurones, also allowed us to investigate a number of properties of the projection from the vestibular nuclei to motoneurones, such as the degree of divergence of a given vestibulospinal fibre, the location of synapses and the amplitude of unitary synaptic potentials.

Some of the results have been reported in abstract form (Wilson, Uchino, Susswein & Rapoport, 1976).

METHODS

Experiments were performed on twenty precollicularly decerebrated cats and on one cat anaesthetized with 50 mg/kg α-chloralose (I.V.). Before decerebration, preliminary surgery was performed under a halothane–nitrous oxide mixture. A dorsal laminectomy was performed from C1 to C6 and the medial portion of the cerebellum was removed by suction after occipital craniotomy to permit visualization of the vestibular nuclei. All animals were paralysed by I.V. administration of gallamine triethiodide (Flaxedil, Davis & Geck) and respired artificially. Bilateral pneumothorax was performed routinely. Mean arterial pressure was monitored from the femoral artery. When necessary, metaraminol bitartrate (Aramine, Merck, Sharp & Dohme) was administered by I.V. infusion to maintain an arterial pressure of 100 mmHg or more. Rectal temperature was maintained between 36 and 38 °C.

The vestibular nerve on both sides was stimulated by placing a 0·5 mm silver wire, insulated except for the spherical tip, on the nerve and an identical indifferent electrode on adjacent bone; stimuli were 100 μsec rectangular pulses. The C3 dorsal rami (DR) were dissected and placed on bipolar platinum hook electrodes. The motor axons in the DR mainly innervate biventer cervicis and complexus, synergistic extensors of the head (Reighard & Jennings, 1935; Richmond & Abrahams, 1975). Two stimulating electrodes were placed in the vicinity of the vestibulospinal tracts at the C5–C6 level.

Fig. 1 shows the experimental arrangement and procedure. The motoneurone pool was located by recording antidromic field potentials evoked by stimulation of the C3 DR with glass micropipettes filled with 2 M-KAc, and having resistances of 1–3 MΩ (Fig. 1*A*). Once positioned within the motor nucleus the micropipette was used as a stimulating electrode for antidromic activation of vestibulocollic neurones projecting to the nucleus. Current-passing properties of the stimulating electrode were checked frequently. Vestibulocollic neurones were located by systematically tracking in the vestibular nuclei while applying 150–200 μsec constant-current cathodal pulses to the spinal micro-electrode. Responses were identified as antidromic by their fixed latency and by collision block with synaptic responses to stimulation of the labyrinth.

A double-barrel micro-electrode was used for extracellular recording in the vestibular nuclei. One barrel was filled with 2 M-NaCl saturated with Fast Green FCF (resistance 1–2 MΩ). The other was filled with a solution of 1 M-Na glutamate. During the search for vestibulocollic neurones a retaining anodal current was usually applied to the glutamate barrel.

Several criteria had to be met before a vestibulocollic neurone was used to search for connexions with motoneurones: (1) an antidromic threshold of less than 20 μA and preferably less than 10 μA; (2) spontaneous firing, firing evoked by glutamate leakage or iontophoresis, or firing evoked by polarization of the labyrinth; (3) reliable discrimination of the activity of the neurone from any other ongoing activity (Fig. 1*B*). Once a vestibulocollic neurone was found that met these criteria, it was tested for projection to the cervical enlargement, for response to stimulation of the ipsilateral vestibular nerve, and in some cases for the presence of commissural inhibition evoked by stimulation of the contralateral nerve. Commissural inhibition was studied with a PDP-11-45 computer programmed to display post-stimulus-time (PST) histograms and the integral of the neurone's discharge. After this study of the properties of the vestibulocollic neurone the spinal micro-electrode was once again used as a recording electrode, first for extracellular recording, and then to search for DR motoneurones in the vicinity of the location from which the vestibular neurone was first activated antidromically. Throughout, the search for

motoneurones and any intracellular recording of the activity of the vestibulocollic neurone was carefully monitored on a storage oscilloscope.

Action potentials of the vestibulocollic neurone were fed to a discriminator; the output pulses of this discriminator were used to trigger an averaging program of the computer. Two types of activity were averaged: (1) extracellular activity in

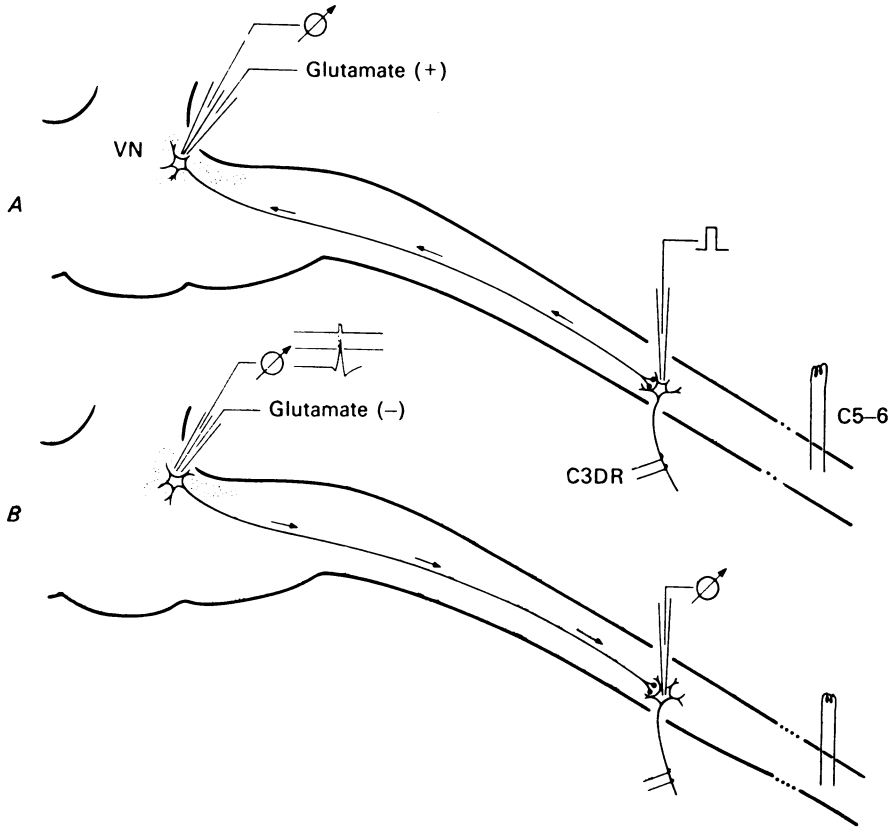

Fig. 1. Diagram of experimental arrangement. *A*, a vestibulocollic neurone in the vestibular nuclei (VN) is activated antidromically by micro-stimulation in or near the C3 DR motor nucleus. The C5–6 electrodes are used to test for a more caudal projection of the neurone's axon. A retaining current is applied to the glutamate barrel of the recording electrode. *B*, the vestibulocollic neurone is activated by a negative current applied to the glutamate barrel, while the spinal micro-electrode is used for intracellular recording from motoneurones. Averaging program is triggered by pulses from discriminator. In both *A* and *B* arrows indicate direction of impulse propagation. For further details, see text.

the vicinity of the original spinal stimulation position, to detect the local action potential in branches of the vestibulocollic neurone; (2) intracellular potentials in DR motoneurones to detect any post-synaptic potentials evoked in these motoneurones by the vestibulocollic neurone; recording time constant was 0·1 sec. As

few as 500 and as many as 2–3000 sweeps were averaged in different instances. Whenever any pre- or post-synaptic activity time-locked to activity of the vestibulocollic neurone was revealed by averaging, its latency was measured from the foot of the spike that triggered the averager. This was done by adding to the latency from the start of the discriminator pulse the time that elapsed from the foot of the spike to the start of the pulse; this measurement was made on the storage oscilloscope.

If a vestibulocollic neurone evoked post-synaptic potentials in one motoneurone, as many other motoneurones as possible were studied while the trigger spike could still be discriminated satisfactorily. If no post-synaptic potentials were revealed at least five motoneurones were studied, whenever possible, before the vestibulocollic neurone was discarded and the search for another was begun.

The location of every successful vestibulocollic neurone was marked by ejecting Fast Green dye at the completion of recording (Thomas & Wilson, 1965). At the end of each experiment the positions of the tips of the metal electrodes used for stimulation at C5–C6 in the spinal cord were marked by lesions made by passing 20 μA of cathodal current through the electrodes for 15 sec. The animal was sacrificed and the brain stem and spinal cord were removed and fixed in 10% formaldehyde in saline; 100 μm frozen sections were then cut in the plane of the electrode tracks. After location of dye marks the brain stem sections were stained according to the technique of Klüver & Barrera, and histological reconstruction was used to determine the location where the vestibulocollic neurone had been recorded. The locations of lesions in the spinal cord, recovered in thionin-stained sections, showed whether the electrodes were well placed in the vicinity of the LVST and MVST.

RESULTS

1. *Action potentials in branches of vestibulocollic neurones*

When a vestibulocollic neurone was fired antidromically at a low threshold of a few microamperes it was usually possible, by recording at the stimulating site while triggering with the neurone's activity, to extract from the background noise the action potential in a nearby branch of its axon. This is illustrated in Fig. 2 for a neurone that was activated antidromically with a threshold of 7 μA. Recording was from a local axonal branch of the vestibulocollic neurone, and not at a distance from either of the vestibulospinal tracts, as is shown by the changes that result from movement of the recording electrode. At the original stimulating site there is a positive-negative potential with a predominant negative phase. Movement of the electrode dorsally or ventrally approximately 100 μm, with concomitant threshold increases to approximately 30 μA, leads not only to changes in the wave form but to great attenuation of the potential. With a further movement of 100–150 μm in either direction the potential disappears.

It was routinely observed that the latency for antidromic activation, measured from the stimulus artifact to the foot of the action potential recorded in the vestibular nuclei, was longer than orthodromic conduction time. The latter was measured from the foot of the vestibulocollic

neurone action potential to the reversal point, from positivity to negativity, of the action potential recorded in the spinal cord. For fifteen neurones the mean difference was 0·39 msec ± 0·15 S.D. This rather long time difference must be due in part to the latency to antidromic spike initiation which, however, probably does not exceed 0·2 msec (Blair & Erlanger,

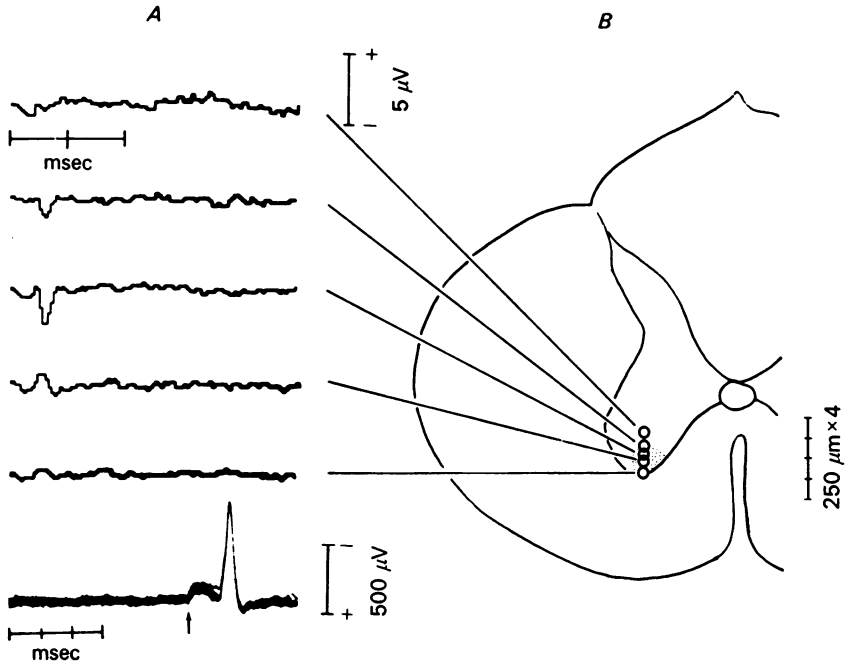

Fig. 2. Action potentials in a branch of a vestibulocollic neurone. *A*, five averaged extracellular records (1200–3300 sweeps) recorded at different positions in the grey matter. The action potential seen in the averaged records was revealed when the averager was triggered by the activity of the vestibular neurone shown activated antidromically in the bottom trace of *A* (stimulus at upward arrow). *B*, recording location for the five traces in *A*. Stippled area outlines motoneurone pool.

1935; Jankowska & Roberts, 1972). Additional delays are likely to be due to a low safety factor at branch points as the impulse propagates from smaller into larger branches, and to slowing of the antidromic impulse as it approaches the cell body (cf. Ito, Hongo, Yoshida, Okada & Obata, 1964).

2. *Connexions between vestibulocollic neurones and neck motoneurones*

In a preliminary experiment, neurones within the vestibular nuclei were selected at random without first activating them antidromically

from the motor nucleus. Using this procedure only one of many neurones was found to have a connexion with a neck motoneurone. In later experiments, fifty-four vestibulocollic neurones were studied following local antidromic identification. Only eleven (20%), with antidromic thresholds of 1–18 μA ($7 \leqslant 5\ \mu$A) evoked synaptic potentials, although many of the remainder were tested on many motoneurones. It was sometimes possible to test successful vestibulocollic neurones on several motoneurones within a radius of approximately 300 μm from the original antidromic stimulation site. In such cases connexions were sometimes observed with more than one motoneurone, but never with all motoneurones tested. For example the vestibulocollic neurones whose actions are illustrated in Figs. 3 and 4 made connexions with 3/7 and 2/7 motoneurones respectively. Of the twelve vestibulocollic neurones that we studied two were excitatory, ten inhibitory.

Fig. 3 illustrates the actions of an excitatory neurone (antidromic threshold 8 μA) on two motoneurones. Very small unitary (Jankowska & Roberts, 1972) e.p.s.p.s, with simple rising phases, were evoked in both. The motoneurones had a tendency to fire spontaneously, and hyperpolarizing current had to be passed through the recording electrodes to prevent this firing: 3·5 nA in *A*, 10 nA for the lower e.p.s.p. in *B*. The upper e.p.s.p. in *B*, however, was recorded without such current injection. Note that the presynaptic volley is visible in both intracellular and extracellular records, particularly in *A*.

The actions of an inhibitory neurone are shown in Fig. 4. This neurone, which was fired antidromically at a threshold of 4 μA, evoked unitary i.p.s.p.s in the two target motoneurones (Fig. 4*C*, *D*). In both instances the i.p.s.p.s were easily reversed by injection of hyperpolarizing current, as is illustrated for a 10 nA current injection in Fig. 4*D*.

3. *Synaptic delay*

It was usually possible to detect evidence of activity in presynaptic branches of vestibulocollic neurones in both intracellular and extracellular records (for example, Figs. 3, 4). By assuming that the presynaptic activity was in the branch that made synaptic contact with the motoneurone we were able to measure synaptic delay. Whenever the presynaptic impulse had clear positive and negative phases (Fig. 3) measurement was from the point of reversal (from positivity to negativity) to the start of the synaptic potential (Jankowska & Roberts, 1972). Not infrequently the presynaptic impulse was purely positive or negative, or even had a more complex shape, indicating that recording was from more than one branch (Jankowska & Roberts, 1972). In such cases we measured from the positive or negative peak; if the shape was too complex, we

did not attempt a measurement. Mean synaptic delay was 0·4 msec ($\pm$ 0·15 S.D., n = 15). This value is similar to earlier less direct measurements (Eccles, 1964) and very close to the precise measurements for the

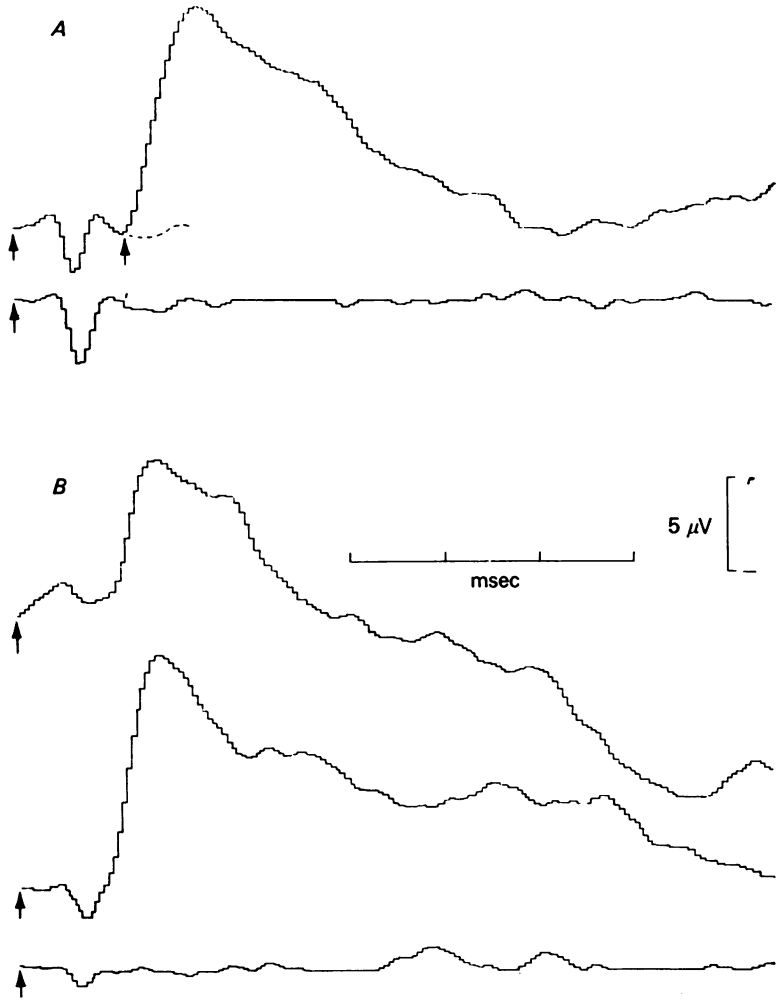

Fig. 3. Unitary e.p.s.p.s in a C3 DR motoneurone. *A* and *B* show e.p.s.p.s evoked in two different motoneurones by activity of one vestibulocollic neurone. Upper trace in *A* recorded intracellularly while injecting a hyperpolarizing current of 3·5 nA into the neurone; lower trace is extracellular record. Second arrow in upper trace points to divergence between intracellular and extracellular records. In *B*, upper trace recorded without current injection, while middle trace recorded during injection of 10 nA hyperpolarizing current. Lower trace is extracellular record. In all traces the upward arrow at the start of the sweep indicates time of output pulse of discriminator (see Fig. 1). All records are averages of 1000–2500 sweeps.

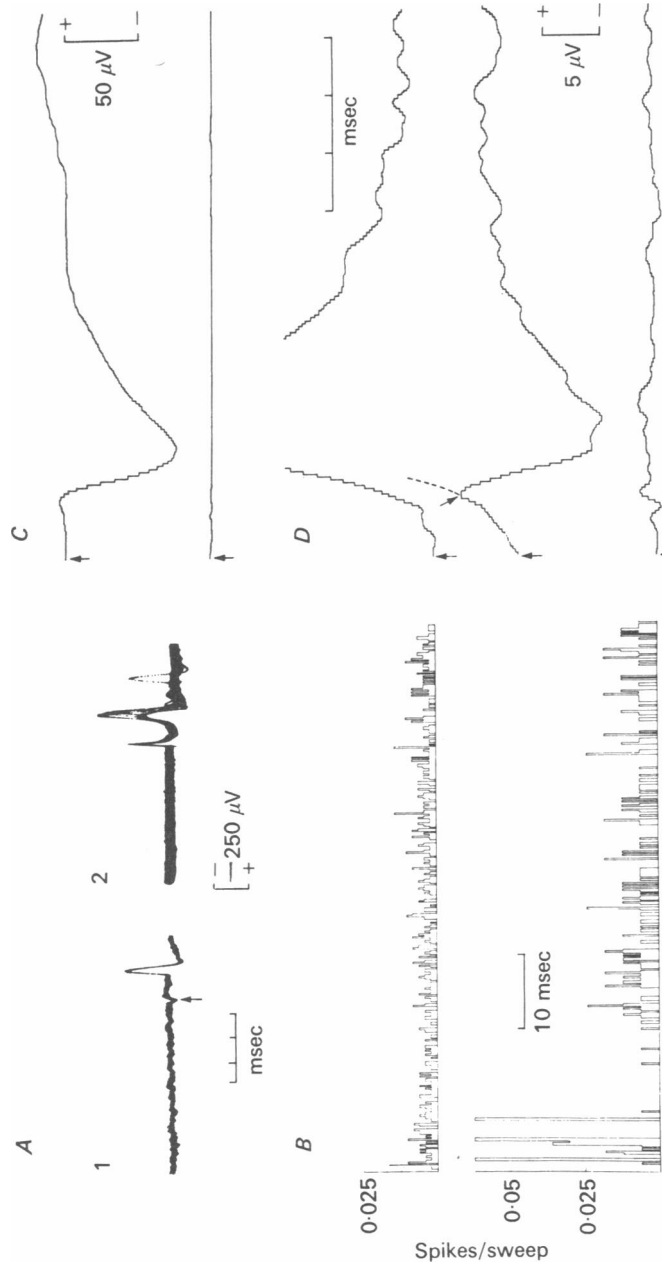

Fig. 4. Properties of an inhibitory neurone in the vestibular nuclei and of unitary i.p.s.p.s evoked by activity of this neurone in two motoneurones. *A*1, antidromic spike of neurone evoked by an 8 μA stimulus (at upward arrow) to the electrode in C3 DR motoneurone pool. *A*2, monosynaptic response of the neurone to stimulation of the ipsilateral vestibular nerve at 1·4 times N_1 threshold. *B*, upper trace shows spontaneous activity of the neurone as a PST histogram (440 sweeps). Lower trace shows effect of a 150 μA triple shock to the contralateral vestibular nerve (150 sweeps). *C*, unitary i.p.s.p. evoked in one motoneurone by activity of the inhibitory neurone; lower trace, extracellular record. *D*, unitary i.p.s.p. (middle trace) evoked in another motoneurone. I.p.s.p. was reversed by injection of 10 nA hyperpolarizing current and part of this reversed i.p.s.p. is shown in the upper trace. Downward arrow in middle trace shows divergence between i.p.s.p.s recorded with and without current injection. Lower trace, extracellular record. All records in *C* and *D* are averages of 600–1000 sweeps; upward arrows indicate time of discriminator output pulses.

synapses between Ia inhibitory interneurones and motoneurones (0·33 msec ± 0·04 S.D., Jankowska & Roberts, 1972) and Ia afferents and motoneurones (most frequent value 0·4 msec, Watt, Stauffer, Taylor, Reinking & Stuart, 1976).

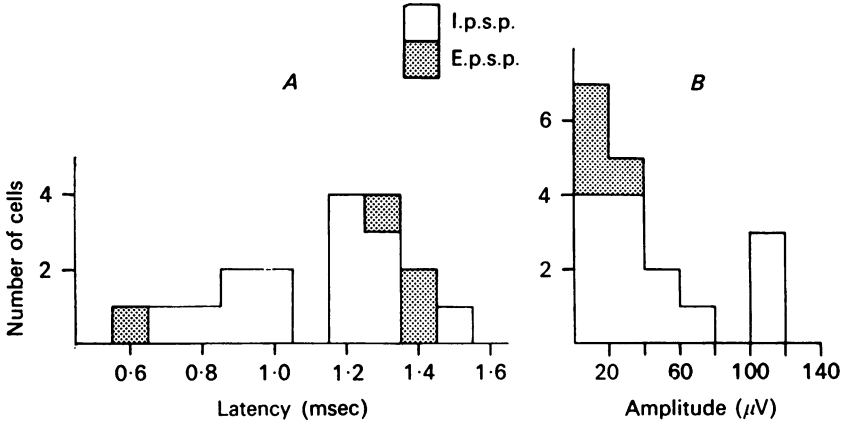

Fig. 5. Latency and amplitude of unitary synaptic potentials.

4. *Properties of synaptic potentials*

(*a*) *Latency.* The latency of synaptic potentials, measured from the foot of the vestibulocollic neurone spike to the start of the e.p.s.p. or i.p.s.p., ranged from 0·6 to 1·5 msec (Fig. 5*A*). These values are the sum of orthodromic conduction time and synaptic delay: mean conduction time was 0·72 msec (± 0·15 S.D.), so that the latencies are obviously monosynaptic. Because of the difference between orthodromic conduction time and antidromic latency of the vestibulocollic neurone (part 1, above) the latency of the synaptic potential was often identical or close to the antidromic latency measured at the original stimulating location or near the motoneurone. The range of latencies overlaps almost completely with the range of latencies of monosynaptic e.p.s.p.s and i.p.s.p.s evoked in C3 by strong stimulation of the vestibular nuclei (Wilson & Yoshida, 1969*a*, *b*).

(*b*) *Amplitude.* The amplitudes of e.p.s.p.s and i.p.s.p.s recorded in eighteen motoneurones are shown in Fig. 5*B*. Because these are amplitudes of averaged unitary synaptic potentials they do not necessarily give the true range of unitary potential amplitudes.

The resting potentials of the motoneurones from which recordings were made were usually 30–50 mV, and this may partly account for the small amplitude of e.p.s.p.s. The i.p.s.p.s include some with averaged amplitudes of 100 μV or more, but they are in general smaller than the averaged Ia unitary i.p.s.p.s recorded in a population of motoneurones with similar resting potentials by Jankowska & Roberts (1972, their Fig. 4).

(*c*) *Time to peak and shape index.* Most synaptic potentials had relatively brief times to peak (Fig. 6*A*). For thirteen unitary i.p.s.p.s mean peak time was 0·81 msec ± 0·15 S.D., almost identical to the 0·85 ± 0·18 S.D. measured for unitary Ia i.p.s.p.s by Jankowska & Roberts (1972). The p.s.p.s can also be described by their shape index, obtained by plotting time to peak against half width (Rall, Burke, Smith, Nelson & Frank,

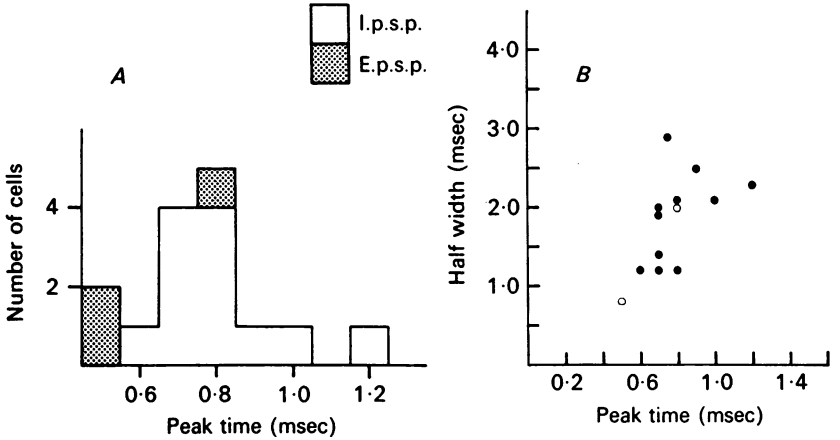

Fig. 6. Time to peak (*A*) and shape index (*B*) of unitary synaptic potentials. In *B* open circles are e.p.s.p.s, filled circles i.p.s.p.s.

1967). This was done for eleven i.p.s.p.s and two e.p.s.p.s that had smooth falling phases, and the result is shown in Fig. 6*B*. Comparing this plot with Fig. 4 of Rall *et al.* (1967) shows that the p.s.p.s are generated mainly in compartment 3 of the motoneurones if a fast conductance transient is assumed, or in compartment 1 if a slow transient is assumed. These results, and the easy reversibility of both unitary i.p.s.p.s (this paper) and vestibular nucleus or labyrinth-evoked compound i.p.s.p.s (Wilson & Yoshida, 1969*b*; Wilson & Maeda, 1974) all agree in placing vestibulocollic inhibitory synapses, like Ia inhibitory synapses (for example Smith, Wuerker & Frank, 1967; Jankowska & Roberts, 1972) very proximally on motoneurones.

5. *Properties of inhibitory and excitatory vestibulocollic neurones*

(*a*) *Location of neurones.* Fig. 7 shows the locations at which activity of twelve vestibulocollic neurones was recorded. There is a tendency for inhibitory neurones to be medial in the nuclei. They are found not only in the medial nucleus or on its borders (Wilson & Yoshida, 1969*b*) but also in the descending nucleus and even in Deiters' nucleus. Of the two excitatory neurones one was in Deiters' nucleus, the other on the border between Deiters' nucleus and the descending nucleus.

(*b*) *Branching.* Nine inhibitory and two excitatory neurones were tested for branching to the cervical enlargement. Both excitatory neurones branched, but only 2/9 inhibitory neurones did so. None of the inhibitory neurones in the medial nucleus branched. We reported earlier (Rapoport

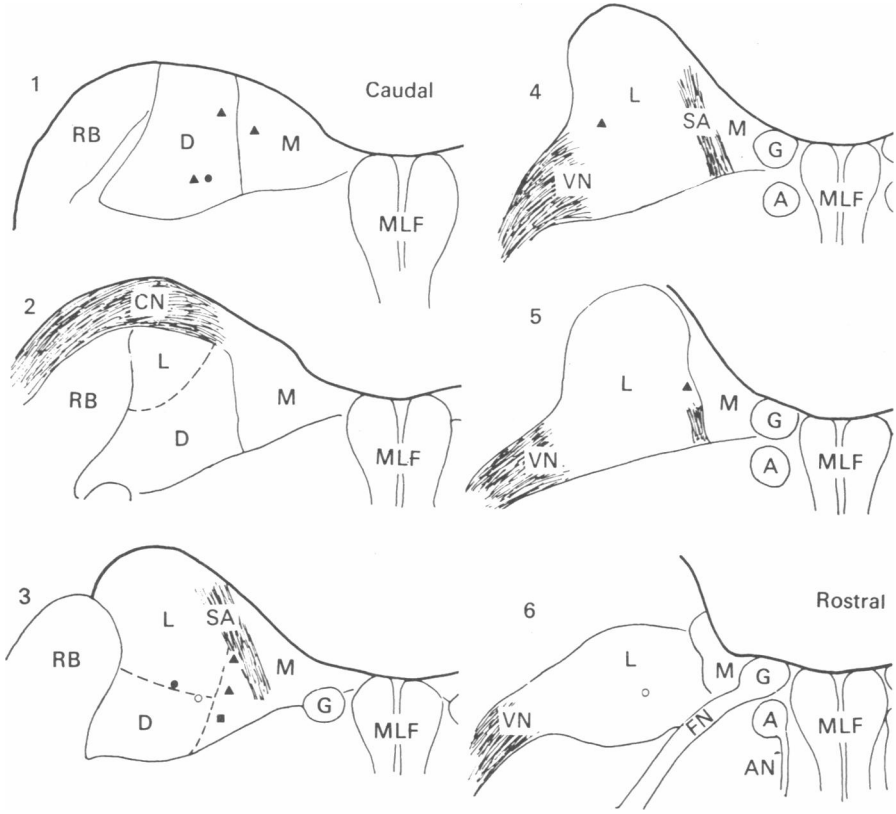

Fig. 7. Locations of inhibitory and excitatory vestibulocollic neurones in the vestibular nuclei. Filled and open symbols represent inhibitory and excitatory neurones respectively. Triangles, non-branching neurones; circles, neurones branching to C5–6; square, neurone not tested for branching. The six transverse sections are through different levels of the vestibular nuclei, with one the most caudal. Abbreviations: *A*, abducens nucleus; AN, abducens nerve; CN, cochlear nerve; D, descending nucleus; FN, facial nerve; G, genu of facial nerve; L, lateral (Deiters') nucleus; M, medial nucleus; MLF, medial longitudinal fasciculus; RB, restiform body; SA, stria acoustica; VN, vestibular nerve.

et al. 1977) that medial nucleus vestibulocollic neurones projecting ipsilaterally do not branch. The medial nucleus inhibitory neurones described in this paper are part of this larger (n = 11) sample.

(*c*) ***Ipsilateral and commissural input from the labyrinth.*** Vestibulocollic

neurones projecting to the ipsilateral spinal cord are, as a group, highly likely to receive monosynaptic input from the ipsilateral labyrinth: monosynaptic firing is seen in at least 60% of Deiters' neurones, and in 82% of a combined population of descending and medial nucleus neurones (Rapoport *et al.* 1977). It is therefore not surprising that all tested vestibulocollic neurones (10/10) acting on motoneurones responded monosynaptically to stimulation of the ipsilateral vestibular nerve (for example, Fig. 4*A*2); some also gave a later response that was presumably disynaptic. The vestibulocollic neurones we studied are part of a population relaying labyrinth activity directly to motoneurones.

Seven vestibulocollic neurones making synapses with motoneurones were tested for the presence of commissural inhibition (Shimazu & Precht, 1966) by stimulation of the contralateral vestibular nerve. Inhibition was seen in four cases, three inhibitory neurones and one excitatory. Commissural inhibition of an inhibitory vestibulocollic neurone is illustrated in Fig. 4*B*: three stimuli delivered to the contralateral vestibular nerve caused a depression of the neurone's spontaneous activity that lasted approximately 15 msec. The threshold of commissural inhibition was typically low, between 1 and 2 times N_1 threshold, within the threshold range observed by Shimazu & Precht (1966).

DISCUSSION

The technique of studying synaptic actions of identified peripheral or central neurones, first used by Mendell & Henneman (1971) is a powerful tool for the study of connexions in the central nervous system. It is particularly applicable when the presynaptic neurones each make monosynaptic connexions with many target neurones, as is the case for the Ia projection to motoneurones. Many triceps surae Ia fibres have synapses on almost all homonymous motoneurones (Mendell & Henneman, 1971; Scott & Mendell, 1976) although the degree of divergence of some Ia fibres is substantially smaller (Scott & Mendell, 1976); there are fewer connexions with heteronymous motoneurones (Scott & Mendell, 1976). Likewise, Ia inhibitory interneurones connect with most of the motoneurones in close proximity of their terminals, and Jankowska & Roberts (1972) have estimated that each interneurone makes synapses with approximately 20% of the pool of target motoneurones. Our results on the vestibulocollic system are in striking distinction to these examples.

The target motoneurones in our experiments were those of biventer cervicis and complexus, synergistic muscles that are linked monosynaptically (Wilson & Maeda, 1974; Anderson, 1977). All motoneurones in this population are excited and inhibited monosynaptically by vestibulocollic

neurones (Wilson & Maeda, 1974). While we may have missed some very small synaptic potentials (however, see Fig. 3), only 20% of our tested vestibulocollic neurones evoked measurable synaptic potentials in this rather homogeneous motoneurone population, even though we always sampled within approximately 300 μm of the original antidromic stimulation point. Some vestibulocollic neurones may not make synapses with motoneurones. Nevertheless, our results suggest that the vestibulocollic projection is less divergent than the Ia projection. The difficulty in finding connexions between vestibulocollic neurones and motoneurones restricted our sample to ten inhibitory and two excitatory neurones. However, the data are sufficient for drawing some conclusions about both synaptic potentials and vestibulocollic neurones.

As described in Results, unitary vestibulocollic synaptic potentials were produced in motoneurones after a mean synaptic delay of 0·4 msec, comparable to earlier measurements in the central nervous system. The potentials, whose shape index suggests a proximal location on or near motoneurone somata, were small: i.p.s.p.s ranged in amplitude from less than 10 to over 100 μV with a mean amplitude of 40–60 μV. I.p.s.p.s evoked by stimulation of the whole vestibular nerve and recorded in a population of DR motoneurones under similar conditions may be as large as 1 mV. This shows that there is a reasonable amount of convergence of vestibulocollic neurones on a neck motoneurone, in agreement with earlier observations (Wilson & Maeda, 1974).

Ipsilaterally projecting vestibulocollic neurones are located in Deiters' nucleus and in the medial and descending vestibular nuclei (Rapoport *et al.* 1977). The two excitatory neurones we studied were both in, or on the border of, Deiters' nucleus, which is well known to give rise to the excitatory LVST. Stimulation experiments have already suggested that there are inhibitory vestibulocollic neurones in the medial nucleus (Wilson & Yoshida, 1969*b*). Our small sample confirms this result. Of ten inhibitory neurones five were in the medial nucleus, particularly in the area under the stria acoustica, or on its borders. While concentrated in the medial part of the vestibular nuclei (Fig. 7), inhibitory neurones are found in nuclei other than the medial. Three inhibitory neurones were in the descending nucleus, one on the Deiters' descending border and one in Deiters' nucleus. All three of these nuclei contribute to the MVST (Wilson & Yoshida, 1969*b*; Akaike, Fanardjian, Ito, Kumada & Nakajima, 1973*a*; Rapoport *et al.* 1977). Since transection of this tract abolishes disynaptic inhibition evoked in neck motoneurones by stimulation of the ipsilateral labyrinth (Akaike *et al.* 1973*b*; Wilson & Maeda, 1974), the inhibitory neurones in our sample must project in the MVST. It may be that all ipsilateral vestibulocollic MVST neurones, whatever their location, are

inhibitory. Parenthetically, our results provide the first evidence about the nature of any descending nucleus neurones.

Our results show that vestibulocollic neurones making synapses with motoneurones are subject to commissural inhibition; the inhibitory vestibular nucleus neurones in the commissural systems may be located ipsi- or contralaterally to their target neurones (Shimazu, 1967; Wilson, Wylie & Marco, 1968). Ampullary nerve stimulation that inhibits neck motoneurones is often followed by a period of disinhibition (Fig. 2 in Wilson & Maeda, 1974). Inhibition of ipsilateral or contralateral inhibitory neurones in the commissural pathway could provide the substrate for this disinhibition, which does not involve cerebellar circuits (V. J. Wilson & M. Maeda, unpublished observations).

Although our sample is small, the results show that 'branching' to more caudal spinal levels cannot be related exclusively to either excitatory or inhibitory neurones, and that both branching and non-branching neurones may act monosynaptically on neck motoneurones. A more comprehensive study could reveal particular sub-populations of excitatory or inhibitory neurones that, for example, branch or do not branch as a group. On the other hand the difference between branching and non-branching neurones may relate to some inputs that they receive or to some as yet unknown features of their projection.

We thank Professor M. Yoshida for participating in some experiments. This work was supported in part by grant NS 02619 from the National Institutes of Health. A. Susswein was the recipient of N.I.H. postdoctoral fellowship no 5 F32 NS 05011.

REFERENCES

AKAIKE, T., FANARDJIAN, V. V., ITO, M., KUMADA, M. & NAKAJIMA, H. (1973*a*). Electrophysiological analysis of the vestibulospinal reflex pathway of rabbit. I. Classification of tract cells. *Expl. Brain Res.* **17**, 477–496.

AKAIKE, T., FANARDJIAN, V. V., ITO, M. & OHNO, T. (1973*b*). Electrophysiological analysis of the vestibulospinal reflex pathway of rabbit. II. Synaptic actions upon spinal neurones. *Expl. Brain Res.* **17**, 497–515.

ANDERSON, M. E. (1977). Segmental reflex inputs to motoneurones innervating dorsal neck musculature in the cat. *Expl. Brain Res.* **28**, 175–187.

BLAIR, E. A. & ERLANGER, J. (1935). On the process of excitation by brief shocks in axons. *Am. J. Physiol.* **114**, 309–316.

ECCLES, J. C. (1964). *The Physiology of Synapses*. Berlin, Gottingen, Heidelberg: Springer.

ITO, M., HONGO, T., YOSHIDA, M., OKADA, Y. & OBATA, K. (1964). Antidromic and trans-synaptic activation of Deiters' neurones induced from the spinal cord. *Jap. J. Physiol.* **14**, 638–658.

JANKOWSKA, E. & ROBERTS, W. J. (1972). Synaptic actions of single interneurones mediating reciprocal Ia inhibition of motoneurones. *J. Physiol.* **222**, 623–642.

KIRKWOOD, P. A. & SEARS, T. A. (1973). Monosynaptic excitation of thoracic expiratory motoneurones from lateral respiratory neurones in the medulla of the cat. *J. Physiol.* **234**, 87–89*P*.

MENDELL, L. M. & HENNEMAN, E. (1971). Terminals of single Ia fibres: location, density and distribution within a pool of 300 homonymous motoneurones. *J. Neurophysiol.* **34**, 171–187.

RALL, W., BURKE, R. E., SMITH, T. G., NELSON, P. G. & FRANK, K. (1967). Dendritic location of synapses and possible mechanisms for the monosynaptic EPSP in motoneurones. *J. Neurophysiol.* **30**, 1169–1193.

RAPOPORT, S., SUSSWEIN, A., UCHINO, Y. & WILSON, V. J. (1977). Properties of vestibular neurones projecting to neck segments of the cat spinal cord. *J. Physiol.* **268**, 493–510.

REIGHARD, J. & JENNINGS, H. S. (1935). *Anatomy of the Cat*, 3rd edn. New York, N.Y.: Holt.

RICHMOND, F. J. R. & ABRAHAMS, V. C. (1975). Morphology and enzyme histochemistry of dorsal muscles of the cat neck. *J. Neurophysiol.* **38**, 1312–1321.

SCOTT, J. G. & MENDELL, L. M. (1976). Individual EPSPs produced by single triceps surae Ia afferent fibres in homonymous and heteronymous motoneurones. *J. Neurophysiol.* **39**, 679–692.

SHIMAZU, H. (1967). Mutual interactions between bilateral vestibular nuclei and their significance in motor regulation. In *Neurophysiological Basis of Normal and Abnormal Motor Activities*, ed. M. D. Yahr and D. P. Purpura, pp. 155–176. New York: Raven.

SHIMAZU, H. & PRECHT, W. (1966). Inhibition of central vestibular neurones from the contralateral labyrinth and its mediating pathway. *J. Neurophysiol.* **29**, 467–492.

SMITH, T. G., WUERKER, R. B. & FRANK, K. (1967). Membrane impedance changes during synaptic transmission in cat spinal motoneurones. *J. Neurophysiol.* **30**, 1072–1096.

THOMAS, R. C. & WILSON, V. J. (1965). Precise localization of Renshaw cells with a new marking technique. *Nature, Lond.* **206**, 211–213.

WATT, D. G. D., STAUFFER, E. K., TAYLOR, A., REINKING, R. M. & STUART, D. G. (1976). Analysis of muscle receptor connections by spike-triggered averaging. 1. Spindle primary receptors and tendon organ afferents. *J. Neurophysiol.* **39**, 1375–1392.

WILSON, V. J. & MAEDA, M. (1974). Connections between semicircular canals and neck motoneurones in the cat. *J. Neurophysiol.* **37**, 346–357.

WILSON, V. J., UCHINO, Y., SUSSWEIN, A. & RAPOPORT, S. (1976). Properties of vestibular neurones projecting to the neck segments of the cat spinal cord. *Neurosci. Abs.* **2**, 1522.

WILSON, V. J., WYLIE, R. M. & MARCO, L. A. (1968). Synaptic inputs to cells in the medial vestibular nucleus. *J. Neurophysiol.* **31**, 176–186.

WILSON, V. J. & YOSHIDA, M. (1969*a*). Comparison of effects of stimulation of Deiters' nucleus and medial longitudinal fasciculus on neck, forelimb and hindlimb motoneurones. *J. Neurophysiol.* **32**, 743–758.

WILSON, V. J. & YOSHIDA, M. (1969*b*). Monosynaptic inhibition of neck motoneurones by the medial vestibular nucleus. *Expl. Brain Res.* **9**, 365–380.