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A COMPARISON OF THE HORIZONTAL AND VERTICAL VESTIBULO-OCULAR REFLEXES OF THE RABBIT

By N. H. BARMACK

From the Department of Ophthalmology, Neurological Sciences Institute, Good Samaritan Hospital and Medical Center, Portland, Oregon 97209, U.S.A.

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SUMMARY

1. The gain and phase of the horizontal (H.v.o.r.) and vertical (V.v.o.r.) vestibulo-ocular reflexes were measured in rabbits. The V.v.o.r. was evoked by sinusoidal rolls about the longitudinal axis of the rabbit. This axis was maintained at different orientations with respect to the earth horizontal axis: V.v.o.r. 0°, prone; V.v.o.r. 90°, 'nose-up'; V.v.o.r. 180°, supine; V.v.o.r. 0°L, left side down.

2. In contrast to the H.v.o.r., the V.v.o.r. 0° had a higher gain (eye velocity/head velocity) and a smaller phase lead (eye position $+180°$ with respect to head position) at low frequencies of sinusoidal vestibular stimulation (± 10 deg, 0·005–0·05 Hz). At higher frequencies (0·05–0·8 Hz), the H.v.o.r. and V.v.o.r. 0° were equivalent in both gain and phase.

3. The low-frequency gain of the V.v.o.r. was smallest in the 'nose-up' orientation. The V.v.o.r. 90° was equivalent in both gain and phase to the H.v.o.r. over the entire range of frequencies tested (0·005–0·8 Hz). Threshold angular accelerations for the H.v.o.r. and V.v.o.r. 90° were below 0·04 deg/sec^2.

4. The compensatory eye movements of the H.v.o.r. were frequently interrupted by anticompensatory re-setting eye movements. These anticompensatory re-setting eye movements were present in the V.v.o.r. 90°, but not in the V.v.o.r. 0°.

5. An estimate gain and phase of the otolithic component of the V.v.o.r. 0° was derived by subtraction of the V.v.o.r. 90° (semicircular canal signal) from the V.v.o.r. 0° (semicircular canal signal + otolith signal). This procedure was based on the assumption that the signals from the otolith organs and vertical semicircular canals combine linearly.

6. The V.v.o.r. 180° provided an interesting test of the assumption of linear combination of otolithic and semicircular canal signals by reversing the phase of the modulated otolithic signal. The data indicated that the V.v.o.r. 180° is non-linear.

INTRODUCTION

Vestibulo-ocular reflexes have been examined in a wide variety of vertebrates and invertebrates. These studies have provided useful descriptions of how specific populations of labyrinthine afferents encode vestibular stimulation and how this information is processed by the central nervous system where it interacts with sensory inputs of other modalities. The horizontal vestibulo-ocular reflex (H.v.o.r.) has been studied most extensively. This reflex can be attributed almost exlusively to angular displacement of the cupula-endolymph system of the horizontal semicircular canals evoked by angular acceleration of the head about an earth vertical axis (Baarsma & Collewijn, 1974; Goldberg & Fernandez, 1971; Melville Jones & Milsum, 1971; see Mayne, 1974 for review).

The anatomical origins of the vertical vestibulo-ocular reflex (V.v.o.r.) are less well understood. In the rabbit, rotation about an earth horizontal axis (roll) would be expected to activate the anterior and posterior semicircular canals. However, neither of the vertical canals is co-planar with roll stimulation. In the rabbit the plane of the anterior canal forms an angle of about 50 deg with the sagittal plane, and the plane of the posterior canal forms an angle of about 40 deg with the sagittal plane (Barmack & Pettorossi, 1981). In addition to stimulating the vertical semicircular canals, a roll stimulus changes the orientation of the otolith organs with respect to the linear acceleration of gravity, and thereby modulates the shearing forces acting on the hair cells of the utricular and saccular maculae.

In the rabbit, the plane of the utricular macula lies approximately within the plane of the horizontal semicircular canal, based on anatomical observation (N. H. Barmack & V. E. Pettorossi, in preparation). The saccular macula forms a dorsal-ventral angle of 12 deg and a posterior-anterior angle of 15 deg relative to the sagittal plane. However, the precision of anatomical observation concerning the spatial relationships of the semicircular canal and otolith organs is limited by the fact that, with the exception of the anterior semicircular canal, these structures do not lie along a single plane. There are significant deviations from toroidal geometry for both the horizontal and posterior semicircular canals, particuarly for the utricular segments of these canals. There are regional differences in the thickness of the statoconial membranes of both the saccular and utricular maculae, but there is little evidence concerning possible curvature of the surface of the sensory epithelium *in vivo* (Flock, 1964; Lindeman, 1969).

The morphological polarization of individual hair cells, defined by the spatial relationship of the stereocillia to the kinocilium, has the same general pattern for a variety of species of mammals. Basically, all polarities within a two-dimensional plane are represented. Both the saccular and utricular maculae are divided into two areas, the pars interna and pars externa, with opposite hair cell polarizations in these two regions (Flock, 1964; Lindeman, 1969). For the rabbit, the morphological polarizations of hair cells of the saccular and utricular maculae are illustrated by the small arrows in Fig. 1*B* (Lindeman, 1969).

Functional polarization vectors of utricular and saccular primary afferents have been described using behavioural and electrophysiological techniques. In the squirrel monkey, 75 per cent of the primary afferents originating from the utricular macula are excited by ipsilateral tilt of the head, indicating a *functional* preponderance of medio-laterally polarized utricular hair cells. The functional polarization vectors of primary afferents which originate from the sacculus have a preferred vertical orientation with a slight preponderance of saccular afferents with ventrodorsal polarizations. This subpopulation of saccular afferents also has a higher mean resting discharge rate than afferents with dorso-ventral polarizations (Fernandez & Goldberg, 1976*a*). The *functional* polarizations of the utricular and saccular maculae, based on

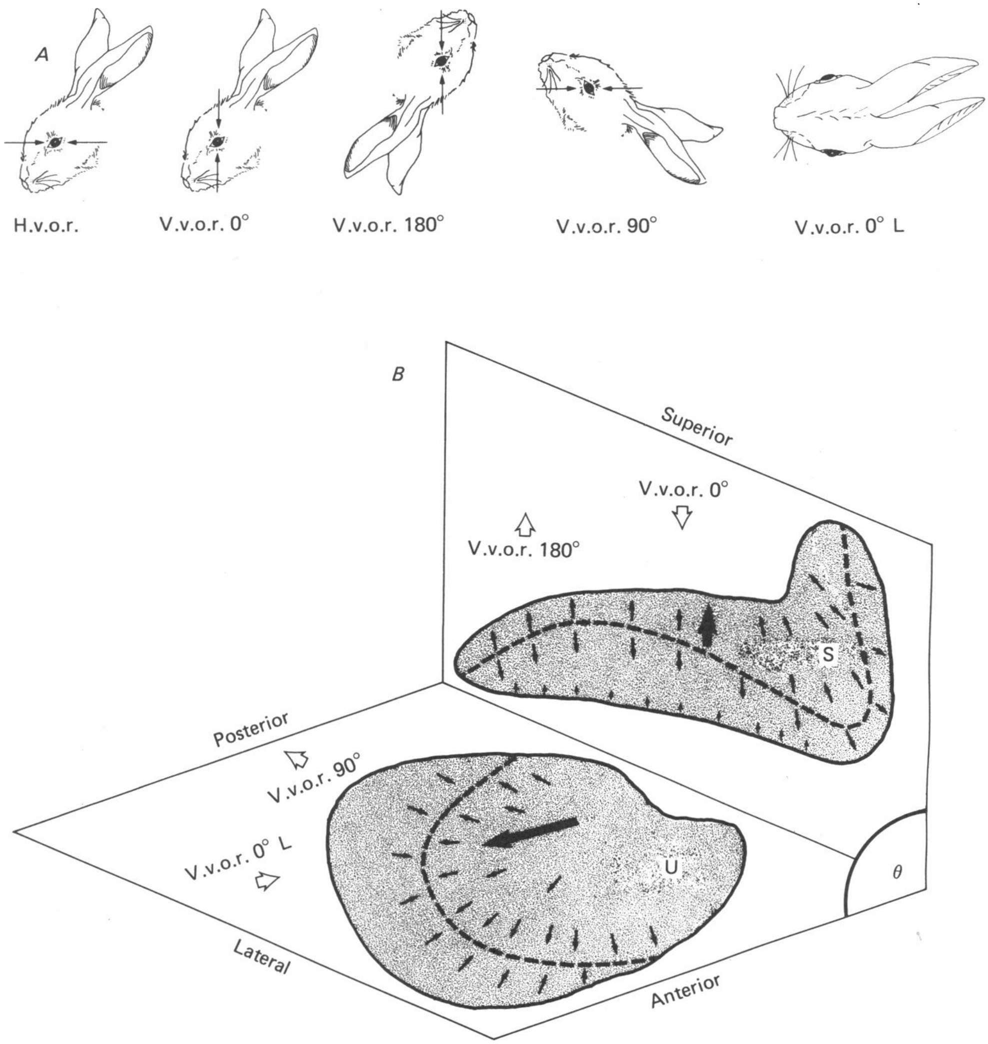

Fig. 1. Orientation of the rabbit's head during different types of vestibular stimulation, and the influence of head orientation on the relationship of the utricular and saccular maculae to the linear acceleration of gravity. *A*, sinusoidal stimulation about the earth vertical axis evoked the horizontal reflex H.v.o.r. Sinusoidal roll stimuli about the earth horizontal axis evoked the vertical reflex V.v.o.r. 0° in the prone orientation, the V.v.o.r. 90° in the 'nose-up' orientation, the V.v.o.r. 180° in the supine orientation, and the V.v.o.r. 0°L in the 'left side down' orientation. *B*, the ***morphological polarizations*** of otolithic hair cells of the rabbit are illustrated with small arrows (see Lindeman, 1969). The ***functional polarizations*** of two populations of primary afferents recorded from the superior and inferior divisions of the vestibular nerve of squirrel monkeys (Fernandez & Goldberg, 1976*a*) are illustrated by two large arrows. The gravitational vectors relative to the otoliths are illustrated for different head orientations.

electrophysiological recordings from the squirrel monkey, are drawn with large arrows in Fig. 1*B*. To summarize, although the morphological polarization of hair cells of both the utricular and saccular maculae suggests that these structures encode shearing forces pan-directionally within a plane, electrophysiological recordings indicate that utricular afferents are maximally excited by linear acceleration directed mediolaterally and that saccular afferents are excited to a lesser extent by linear acceleration directed ventrodorsally. Therefore, the contributions of the utricular and saccular maculae to the vertical vestibulo-ocular reflex would depend on the orientation of the head with respect to gravitational acceleration. The magnitude of the gravitational vector, which is co-linear with the functional polarization vector of an otolith macula is trigonometrically related to the cosine of the angle of gravitational acceleration and the functional polarization vector. The change in the gravitational vector acting co-linearly with the functional polarization vector of an otolith macula is related to the sine of their angle. For example, if a rabbit's head is maintained in a prone orientation about the earth horizontal axis, the gravitational vector would form an angle of 90° with respect to the mediolaterally polarized utricular maculae. The population of utricular hair cells would be minimally activated ($\cos 90° = 0$) but maximally sensitive ($\sin 90° = 1$) to angular roll deviations from this orientation. The activity of the ventrodorsally polarized saccular afferents would be maximally decreased in the prone orientation ($\cos 180° = -1$) and minimally sensitive to deviations from this orientation ($\sin 180° = 0$). For $\pm 10°$ sinusoidal roll deviations from the prone orientation, the modulation of the gravitational vector acting co-linearly with the functional polarization vector of the utricular maculae is eleven times greater than the modulation of the gravitational vector acting co-linearly with the functional polarization vector of the saccular maculae. If the rabbit was oriented 'nose-up' (V.v.o.r. 90°), then sinusoidal oscillation of the head about the longitudinal axis of the rabbit would not change the orientation of the otolith organs relative to gravitational acceleration. If the rabbit lay on its left side, (V.v.o.r. 0°L), then symmetrical rolls about this orientation would cause maximal changes in the gravitational vector acting co-linearly with the functional polarization vectors of the saccular maculae. If the rabbit was supine, (V.v.o.r. 180°), then again the utricular maculae would be maximally sensitive to roll stimulation, but because of the inverted head orientation, the utricular signal would be phase shifted by 180 deg with respect to the signals originating from the vertical semicircular canals, which are primarily sensitive to angular acceleration.

The influences of these different vestibular stimuli on the vertical vestibulo-ocular reflex of rabbits are examined in the present experiment. It will be shown that the phase and gain of the V.v.o.r. 0° are different from the phase and gain of the H.v.o.r., reflecting a contribtion from the otolith organs to the V.v.o.r. 0° at low frequencies of sinusoidal roll. In addition to the differences in phase and gain, the dynamics of eye movements evoked during stimulation of the H.v.o.r. and V.v.o.r. 0° are different. The compensatory eye movements of the H.v.o.r. are frequently interrupted by anticompensatory or 'return eye movements' (Benson & Bodin, 1966). However, these anticompensatory eye movements are absent from the V.v.o.r. 0°. These differences between the phase, gain and dynamics of eye movements evoked during stimulation of the H.v.o.r. and V.v.o.r. 0° are eliminated when the rabbit is orientated 'nose-up'. The phase and gain of eye movements evoked during stimulation of the H.v.o.r. and V.v.o.r. 90° are identical. The compensatory eye movements of the V.v.o.r. 90°, like those of the H.v.o.r., are interrupted by anticompensatory re-setting eye movements. Thus, it appears that the otolith organs account both for the low-frequency gain and the absence of anticompensatory eye movements during stimulation of the V.v.o.r. 0°. These findings have been communicated previously in preliminary form (Barmack, 1977).

METHODS

Surgical procedures. In preparatory operations, twenty-eight albino and pigmented rabbits were anaesthetized with ketamine hydrochloride (50 mg/kg intramuscularly) and halothane. Each rabbit's head was aligned in a stereotaxic apparatus so that the lambda suture was 1·5 mm below the bregma suture. Two stainless-steel screws (10–32) were anchored to the calvarium with four smaller peripherally placed stainless steel screws (2–56) and dental cement. The two larger screws mated with a steel rod later used to restrain the head of the rabbit and to support the eye position detection device. The rod was tilted in the posterior-anterior direction at an angle of 12 deg in order to align the plane of the horizontal semicircular canals with the horizontal plane of the rate table.

Vestibular stimulation. The horizontal and vertical vestibulo-ocular reflexes (H.v.o.r. and V.v.o.r.) were tested in unanaesthetized rabbits. The head was fixed in the centre of rotation of a biaxial rate table and the body was firmly encased in 'egg carton' foam rubber. Rotation of the rate table about the earth vertical and earth horizontal axes was servo-controlled. The position output signal of the rate table was of constant amplitude for sinusoidal input signals equal to or less than 0·8 Hz, ±10 deg. The V.v.o.r. was evoked by sinusoidal rolls about the longitudinal axis of the rabbit which was oriented parallel to the earth horizontal axis (prone, V.v.o.r. 0°), vertically ('nose-up', V.v.o.r. 90°), horizontally but inverted ('supine', V.v.o.r. 180°), or horizontally with the left side down (V.v.o.r. 0°L). These different modes of vestibular stimulation are illustrated in Fig. 1*A*. Vestibular stimulation was always performed with both eyes of the rabbit completely occluded by three layers of black cloth.

Eye position recording. Eye position was measured with an infra-red light projection technique (Barmack, 1976). The eye was anaesthetized topically with proparacaine hydrochloride. A small suction cup bearing a light-emitting diode was attached to the anaesthetized eye; this diode projected a narrow beam of infra-red light onto a photosensitive X–Y position detector (United Detector Technology, SC-50) which was fixed relative to the head and located 3–5 mm from the tip of the diode (G.E. SSL-315). The photosensitive surface gave a continuous X–Y indication of the position of the incident centroid of infra-red light. The system was calibrated by moving the eye on which the diode was mounted through known angular displacements. This system had a sensitivity of 60 mV/deg and was linear to within 5 per cent for deviations of the eye of ±15 deg. Eye velocity was measured by electronic differentiation of the eye position signal.

Behavioural testing. The *gain* (G) of the vestibulo-ocular reflex was determined from measurements of the peak eye velocities attained during each half-cycle of sinusoidal rotation; $(V_R + V_L)/2V_T$, where V_R = peak compensatory eye velocity to the right; V_L = peak compensatory eye velocity to the left; V_T = peak table velocity. The *phase* of the v.o.r. (eye position + 180° with respect to head position) was measured at each half-cycle of rotation. At stimulation frequencies below 0·02 Hz, two cycles of stimulation were usually measured. At frequencies above 0·04 Hz, three or more cycles were usually measured. In preliminary tests of the v.o.r. it was observed that random switching between high and low frequencies seemed to cause a disruption in the behavioural state of the rabbits, requiring examination of the v.o.r. at each frequency of stimulation for a longer duration. Consequently, a method of ascending order of stimulus presentation was adopted, which reduced the time needed to collect data and appeared to maintain the rabbits in a more uniform behavioural state. All data were recorded with an FM tape recorder and subsequently measured and photographed from a storage cathode ray oscilloscope.

RESULTS

Comparison of H.v.o.r. and V.v.o.r. 0°. The eye movements evoked by sinusoidal oscillation about the earth vertical axis (H.v.o.r.) and about the earth horizontal axis (V.v.o.r. 0°) had different characteristics over the entire range of frequencies tested (0·005–0·08 Hz). At low frequencies of stimulation (0·005–0·02 Hz) the H.v.o.r. had a gain of less than 0·25, and a phase lead larger than 90 deg (Fig. 2). As the frequency of stimulation increased, the gain increased and the phase lead was reduced. At frequencies of stimulation above 0·02 Hz the compensatory eye movements of the H.v.o.r. were repeatedly interrupted by anticompensatory re-setting eye movements (Fig. 2). By contrast, the V.v.o.r. 0° had a gain greater than 0·5 and a phase lead of less than 5 deg at frequencies of stimulation below 0·02 Hz. At frequencies of stimulation in the 0·02–0·06 Hz range, both the gain and the phase lead of the V.v.o.r. 0° increased. At frequencies of stimulation above 0·06 Hz, the gain of the H.v.o.r. 0° continued to increase but the phase lead began to decrease (Figs. 2 and 3). The compensatory eye movements evoked by sinusoidal roll stimulation were never interrupted by the anticompensatory re-setting eye movements which characterized the V.v.o.r. at higher frequencies of stimulation. The marked difference in gain and phase between the H.v.o.r. and the V.v.o.r. 0° at low frequencies of stimulation confirmed the expectation that the V.v.o.r. 0° receives a contribution from the otolith organs as well as from the vertical semicircular canals (Fig. 3). This otolithic contribution appears to account for the low frequency gain of the V.v.o.r. 0° *and* to

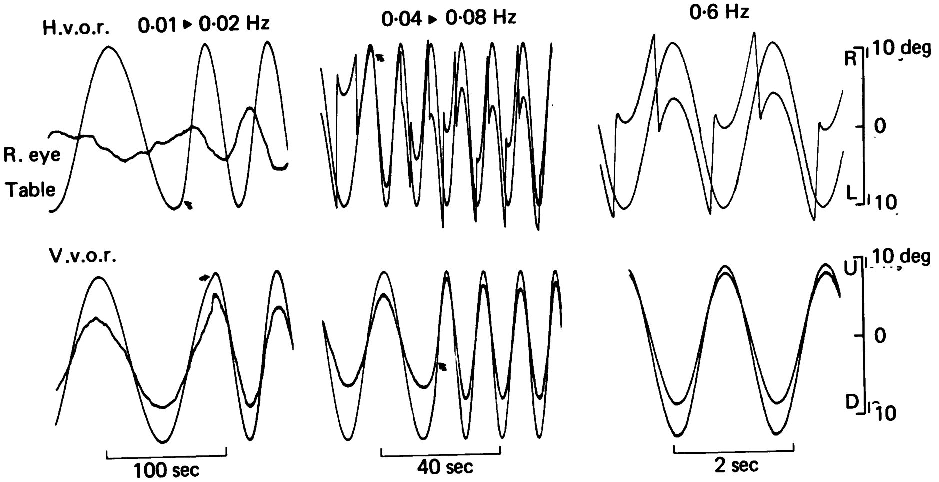

Fig. 2. Eye movements evoked by sinusoidal rotation about the earth vertical and earth horizontal axes. The horizontal vestibulo-ocular reflex (H.v.o.r.) has a low gain and a large phase lead at low frequencies of stimulation. The vertical vestibulo-ocular reflex (V.v.o.r. 0°) has a larger gain and smaller phase lead at low frequencies (0·01–0·04 Hz). The arrows indicate transitions between different frequencies of stimulation. The calibrations indicate the position of the right eye. The table position signal is inverted relative to the right eye to facilitate phase comparisons.

interact with information which originates from the semicircular canals in a way which prevents the occurrence of anticompensatory re-setting eye movements.

The occurrence of anticompensatory re-setting eye movements during rotation about the earth vertical axis seemed to be dependent on both eye position and head velocity. These re-setting eye movements were more likely to be initiated from less extreme eye deviations during vestibular stimulation at higher frequencies. During vestibular stimulation with step rotations of 20 deg about the earth vertical axis, compensatory eye movements were invariably followed by anticompensatory re-setting eye movements (Fig. 4*A*). However, the same rotational steps about the earth horizontal axis (prone orientation) evoked only compensatory eye movements (Fig. 4*B*). The head velocity during the vertical steps could be increased to approximately three times the head velocity attained during horizontal steps, without triggering anticompensatory re-setting eye movements.

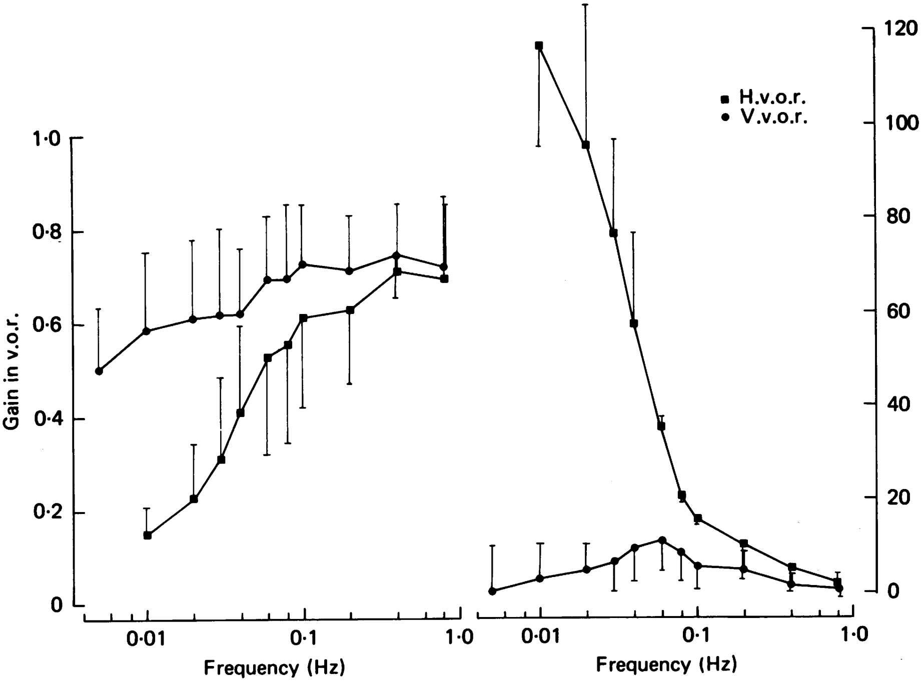

Fig. 3. Comparison of the H.v.o.r. and V.v.o.r. 0°. The polar mean phase and gain for the H.v.o.r. (squares) and V.v.o.r. 0° (circles) are illustrated for twenty-one rabbits. One standard deviation (+ or −) is indicated for each of the data points.

Comparison of the H.v.o.r. and V.v.o.r. 90°. The possible otolithic contribution to the V.v.o.r. 0° was evaluated by using roll stimulation with the head of the rabbit oriented 'nose-up' (V.v.o.r. 90°). In this orientation, rotation of the rabbit about its longitudinal axis does not change the orientation of the otoliths with respect to gravity (Fig. 1). Therefore, the V.v.o.r. 90° should reflect stimulation of only the vertical semicircular canals. As expected, vestibular stimulation in this 'nose-up' orientation reduced the gain and caused an increased phase lead at the lower frequencies. The V.v.o.r. 90° was equivalent in both gain and phase to the H.v.o.r. (Figs. 5 and 6). Furthermore, the compensatory eye movements of the V.v.o.r. 90°, like those of the H.v.o.r., were interrupted by anticompensatory re-setting eye movements at frequencies of stimulation above 0·1 Hz (Fig. 5*B*).

At a stimulation frequency of 0·01 Hz, which produced peak angular accelerations of 0·04 deg/sec^2, the H.v.o.r. and the V.v.o.r. 90° had gains of 0·13 and 0·09 respectively. In one rabbit there was a clear reflex eye movement during V.v.o.r. 90° stimulation at a frequency of 0·005 Hz, which caused peak angular accelerations of 0·01 deg/sec^2. A threshold angular acceleration for evoking the H.v.o.r. and V.v.o.r. 90° in rabbits is probably closer to the lower value of 0·01 deg/sec^2. Although this value is lower than that reported previously, based on photographic measurements of eye position (Ter Braak, 1936), the difference can probably be attributed to a greater sensitivity of the present photoelectric technique of measuring eye position.

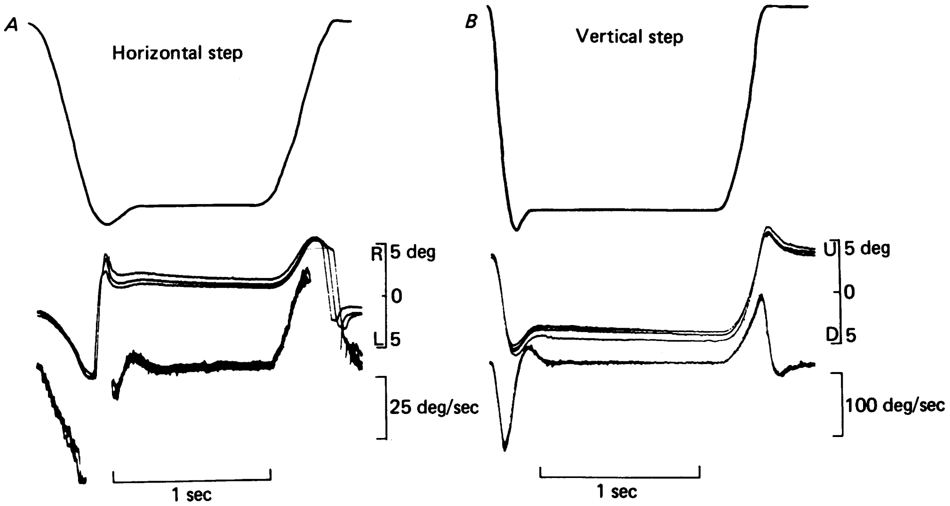

Fig. 4. Eye movements evoked by horizontal and roll steps. *A*, step stimulation of 20 deg amplitude about the earth vertical axis evokes horizontal compensatory eye movements followed by anticompensatory re-setting eye movements. *B*, step roll stimulation about the earth horizontal axis (prone orientation) evokes vertical compensatory eye movements, without any anticompensatory re-setting eye movements. The traces, top to bottom are: rate table (head) position (same calibration, but inverted with respect to the right eye), right eye position, right eye velocity. Note that the calibration for vertical eye velocity is four times greater than for horizontal eye velocity.

If one assumes that signals from the otolith organs and the vertical semicircular canals combine vectorially to produce the V.v.o.r. 0°, then it should be possible to obtain an estimate of the otolithic component of this reflex by subtracting vectorially the V.v.o.r. 90° (semicircular canal signal) from the V.v.o.r. 0° (semicircular canal signal + otolith signal). The results of such a vector subtraction are illustrated in Fig. 7. These data suggest a relatively high gain (0·4–0·5) for the otolithic component of the V.v.o.r. 0° at low frequencies of stimulation (0·005–0·01 Hz). As the frequency increases the gain decreases and the phase *lag* increases.

V.v.o.r. 180°. Under normal circumstances, during roll deviations of the head from the prone orientation, the vertical semicircular canals and the utricular maculae act synergistically. Rolls from the prone orientation onto the left side (i.e. clockwise rolls, viewed from in front of the rabbit) would cause ampullofugal flow of endolymph in the left anterior and posterior semicircular canals which in turn would cause excitation in the hair cells in the ampullae of these vertical canals (Estes, Blanks & Markham, 1975). Rolls from the prone orientation onto the left side would also increase the gravitational vector acting on mediolaterally polarized hair cells of the left utricular maculae. A clockwise roll onto the left side from an initial prone orientation would cause ampullopetal endolymph flow in the right anterior and posterior semicircular canals and reduce the primary afferent activity originating from the ampullae of these canals. Similarly the signal originating from the right utricular macula would be reduced, since the gravitational vector would be directed oppositely to its functional polarization vector. However, if a rabbit were placed in

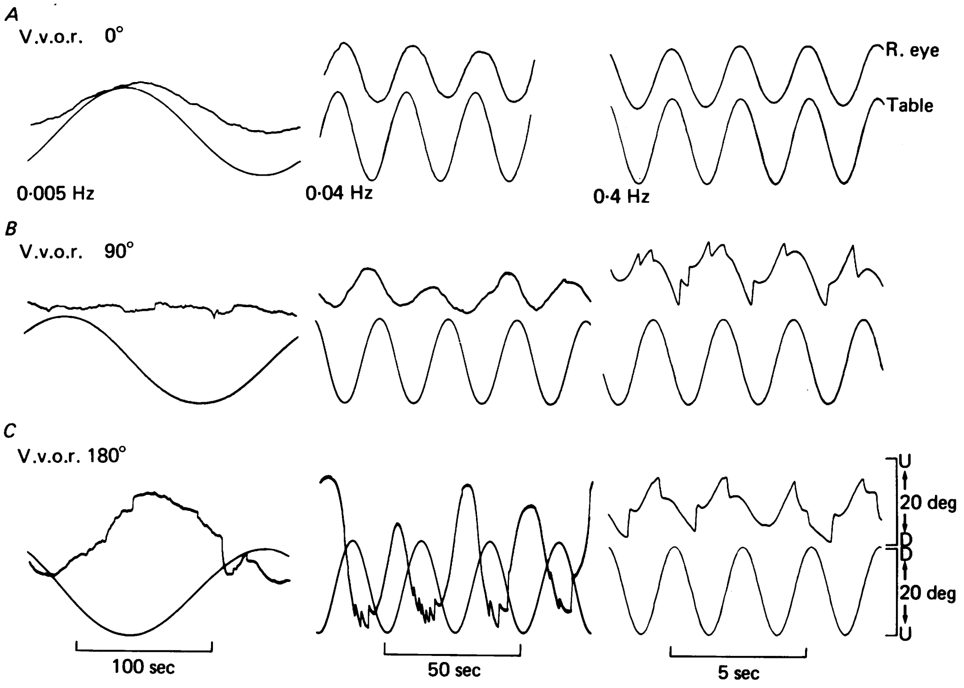

Fig. 5. Vertical eye movements evoked during sinusoidal roll stimulation with the rabbit's longitudinal axis maintained at three different orientations: V.v.o.r. 0° (prone), V.v.o.r. 90° ('nose-up'), V.v.o.r. 180° (supine). Note the appearance of anticompensatory re-setting eye movements at 0·4 Hz during the V.v.o.r. 90°. Note also that the gain of the V.v.o.r. 180° at 0·04 Hz is greater than one.

a supine orientation, then the phase of the signals from the utricular maculae would be shifted by 180° with respect to the signals from the vertical semicircular canals. A clockwise roll of the rabbit (viewed from in front), from an initial supine orientation, would excite primary afferents originating from the ampullae of the *left* vertical semicircular canals and it would also excite primary afferents originating in the mediolaterally polarized hair cells of the *right* utricular macula. This reversal in phase of the utricular signals is due to the inversion of the utricular maculae relative to gravitational acceleration. The signals originating from the saccular maculae would also be modified. In the prone orientation the gravitational vector would be directed oppositely to the weak ventrodorsal polarization vectors of both saccular maculae. In the supine orientation, however, the gravitational vector would have the same direction as the polarization vectors of the saccular maculae, increasing the level of excitation of *both* saccular maculae.

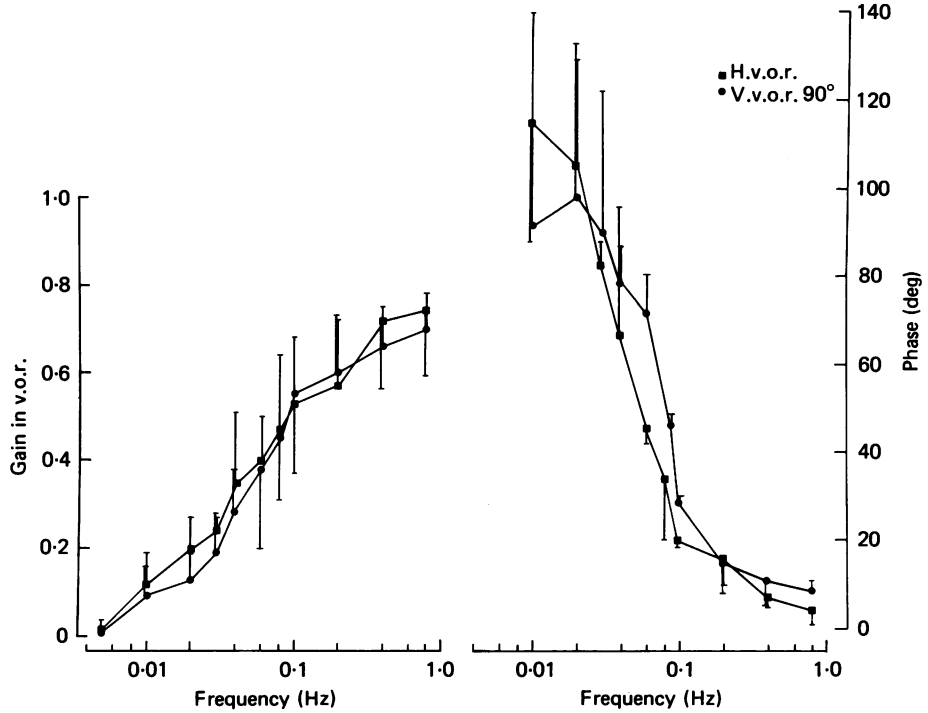

Fig. 6. Comparison of the H.v.o.r. and V.v.o.r. 90°. The mean polar phase and gain were computed for nine rabbits. Only one rabbit had a non-zero gain of the V.v.o.r. 90° and 0·005 Hz. One standard deviation (+ or −) is indicated for each of the data points.

If the signals originating from the semicircular canals and the otolith organs added vectorially to produce the V.v.o.r. 0° it would be possible to obtain a prediction of the V.v.o.r. 180° by subtracting the otolith signal (V.v.o.r. 0°–V.v.o.r. 90°) from the semicircular canal signal (V.v.o.r. 90°). This prediction of the V.v.o.r. 180°, derived from independent measurements of the V.v.o.r. 0° and the V.v.o.r. 90° was not confirmed by measurement of the V.v.o.r. 180°. The discrepancy between the expected and obtained results was maximal over the frequency range of 0·02–0·08 Hz (Fig. 8). The V.v.o.r. 180° in this frequency range had a much higher gain and greater phase lead than would be predicted by the vector subtraction of the otolith signal from the semicircular canal signal (Fig. 8). In some rabbits which were tested in the supine orientation, eye movements appeared as a nystagmus with a reversal in the direction of the beats roughly in phase with head velocity in the frequency range of 0·02–0·08 Hz. Interestingly, it is in this frequency range that there is an overlap between semicircular canal and otolithic signals. At frequencies below 0·02 Hz, the vertical semicircular canals make a negligible contribution to the V.v.o.r. 0° ($G < 0{\cdot}1$) and at frequencies above 0·08 Hz the contribution of the otolith organs to the V.v.o.r. 0° is greatly attenuated.

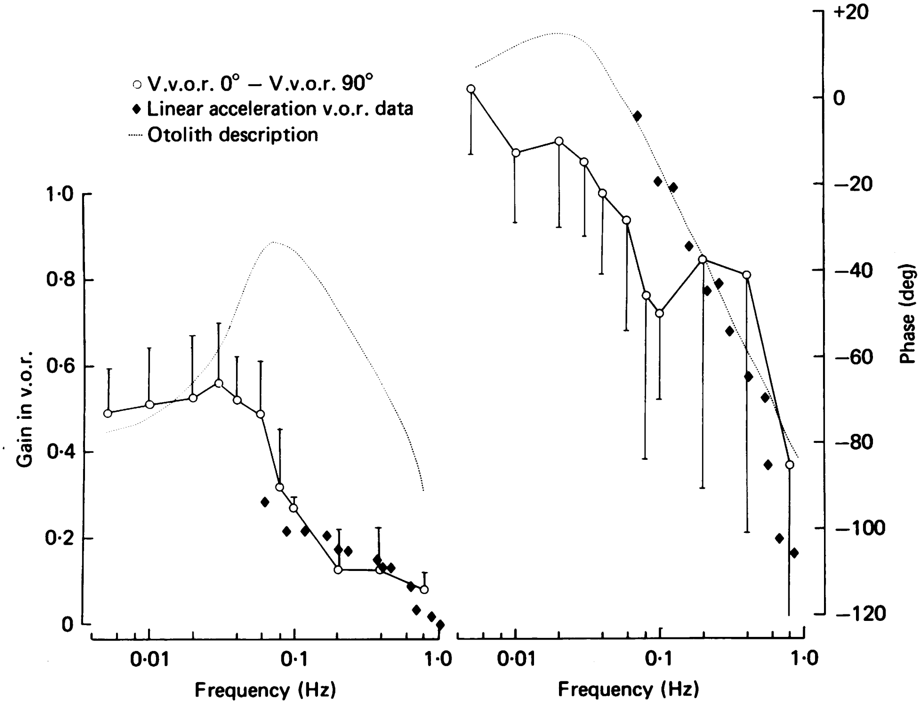

Fig. 7. Derived prediction of the otolithic contribution to the V.v.o.r. 0°. The filled circles indicate the predicted frequency response of the otoliths obtained by polar subtraction of the mean V.v.o.r. 90° from the V.v.o.r. 0° ($n = 9$). The dotted line indicates the frequency response of the otoliths obtained from a descriptive model (Young & Meiry, 1968). The filled diamonds indicate the frequency response obtained from stimulation of the v.o.r. with linear accelerations (Baarsma & Collewijn, 1975). One standard deviation (+ or −) is indicated for each data point.

Vertical step responses at different head orientations. The presence of anticompensatory re-setting eye movements in the V.v.o.r. 90° and their absence from the V.v.o.r. 0° suggested that these eye movements might be prevented by otolithic signals. This was tested by examining the vertical eye movements evoked during roll step stimulation when the rabbit was oriented ‘nose-up’. In the prone orientation, step roll stimulation evoked purely compensatory vertical eye movements (Fig. 9*A*). In the ‘nose-up’orientation, the same amplitude step stimulation evoked compensatory eye movements which were interrupted by anticompensatory re-setting eye movements (Fig. 9*B*). In the supine orientation, the re-setting anticompensatory eye movements appeared to sum with the now antiphasic input from the otolith organs (Fig. 9*C*). The dotted curve in Fig. 9*A* depicts the graphic subtraction of the step response in the ‘nose-up’ orientation from the step response in the prone orientation. It reveals that the onset of the presumed otolithic component of the response is about 50 msec slower than the presumed semicircular canal component.

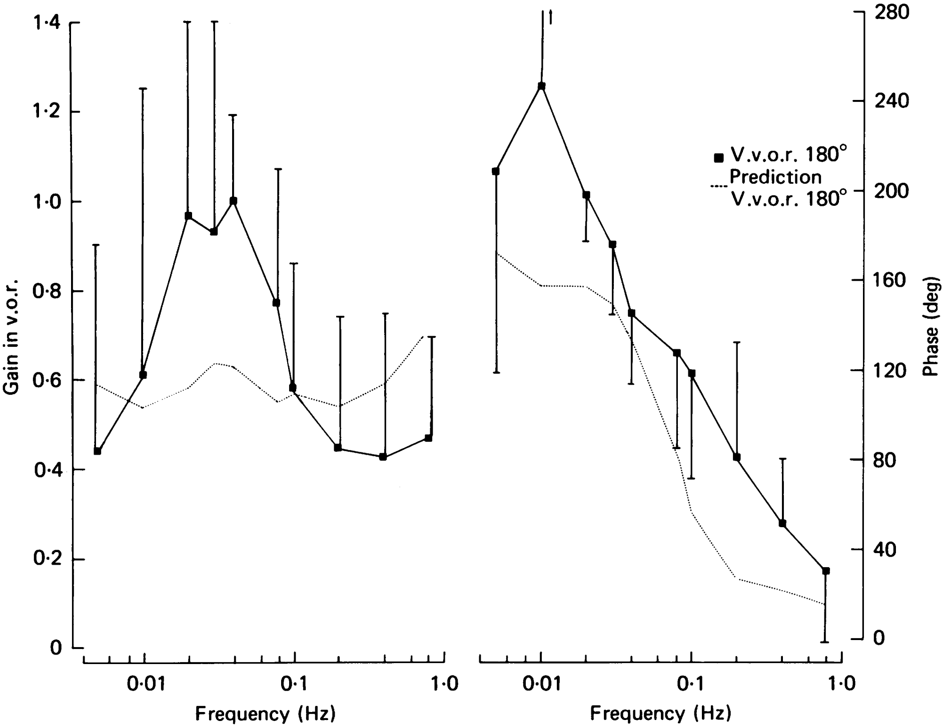

Fig. 8. Comparison of the predicted and obtained V.v.o.r. 180°. The dashed line indicates the predicted V.v.o.r. 180°, assuming a linear combination of signals originating from the semicircular canals and otoliths. The derived otolith signal (V.v.o.r. 0° − V.v.o.r. 90°) was phase-shifted by 180 deg and summed vectorially with the semicircular canal signal (V.v.o.r. 90°). One standard deviation (+ or −) is indicated for each data point.

V.v.o.r. 0°L. Sinsoidal rolls about the left-side-down orientation (V.v.o.r. 0°L) would be expected to *modulate* maximally the gravitational shearing forces acting co-linearly with the functional polarization vector of the saccular maculae (Fig. 1). In this orientation the efficacy of a ± 10 deg sinusoidal stimulus in modulating the gravitational shearing forces acting co-linearly with the functional polarization vector

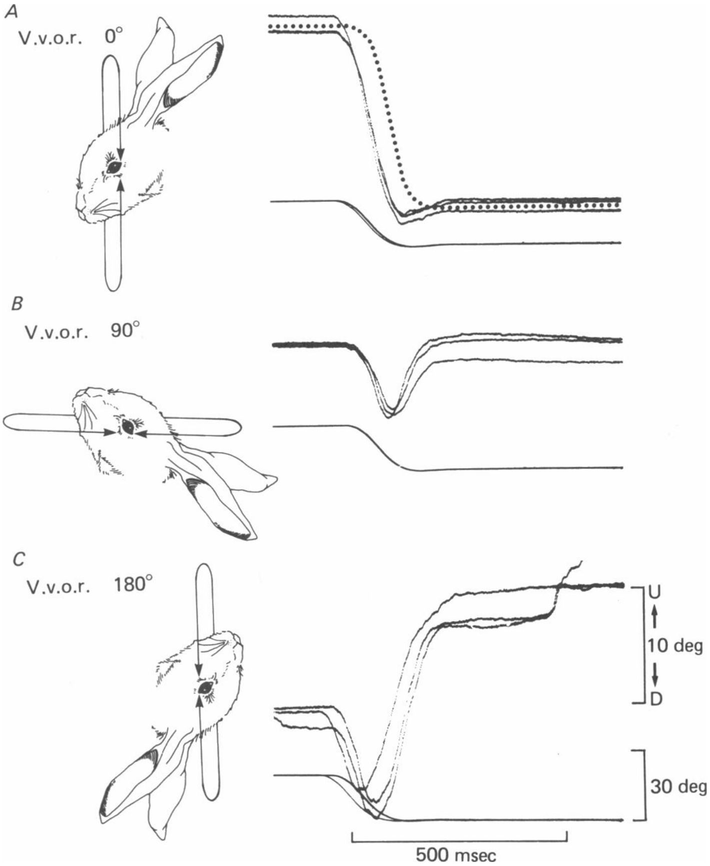

Fig. 9. Vertical eye movements evoked by roll steps with the longitudinal axis of the rabbit maintained at three different orientations: *A*, V.v.o.r. 0° (prone); *B*, V.v.o.r. 90° ('nose-up'); and *C*, V.v.o.r. 180° (supine). The dotted curve in *A* indicates the 'predicted' otolith response, obtained by subtraction of the V.v.o.r. 90° from the V.v.o.r. 0°. Each set of multiple traces indicates the position of the left eye (upper traces). The table position signal is inverted (lower traces).

of the utricular maculae would be reduced relative to the same amplitude stimulation in the prone orientation by a factor of 11. The V.v.o.r. 0°L had a greatly attenuated low frequency gain with a large phase lead (Fig. 10). On the basis of these data it appears that saccular maculae make only a nominal direct contribution to the V.v.o.r. 0°. Nevertheless, it is possible that the saccular maculae could *indirectly* influence the V.v.o.r. 0° or V.v.o.r. 180° by altering the 'steady-state' excitability of secondary vestibular neurones which receive convergent stimulus-modulated signals from the vertical semicircular canals.

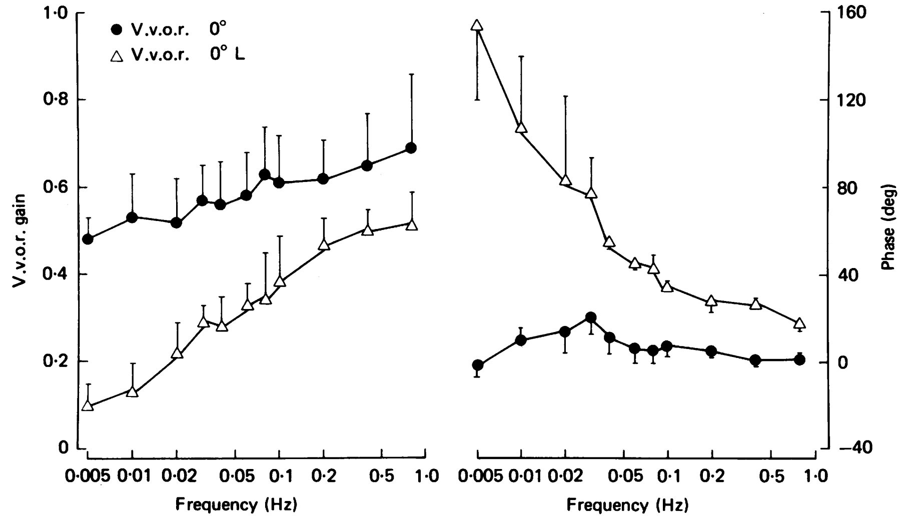

Fig. 10. Comparison of the V.v.o.r. 0° with the V.v.o.r. 0°L. The mean polar phase and gain for the V.v.o.r. 0° (filled circles) and V.v.o.r. 0°L (open triangles) were computed for four rabbits. One standard deviation (+ or −) is illustrated for each data point.

DISCUSSION

The otolithic contribution to the V.v.o.r. 0°. The principle observation of the present experiment is that the V.v.o.r. 0° has a high gain and a small phase lead at frequencies of stimulation below 0·02 Hz. At these same frequencies of stimulation the H.v.o.r. has a low gain and a large phase lead. The low frequency characteristics of the V.v.o.r. 0° and V.v.o.r. 180° can be attributed to a stimulus-modulated otolithic signal. This signal could be eliminated by orienting the rabbit 'nose-up' so that sinusoidal oscillation of the rabbit about its longitudinal axis did not alter the orientation of the otoliths with respect to gravitational acceleration (V.v.o.r. 90°). It has been demonstrated electrophysiologically that the majority of otolith hair cells respond optimally to shearing forces directed along orthogonal axes (Fernandez & Goldberg, 1976*b*; Loe, Tomko & Werner, 1973). For shearing forces directed orthogonally to the optimal hair cell polarization axis but within the plane of the maculae, the sensitivity of otolith hair cell responses is reduced by 85–90 per cent. Forces which compress or extend the hair cells do not alter their excitability (Fernandez & Goldberg, 1976*b*).

Specificity of otolithic contributions to the V.v.o.r. 0°. There are two reasons for ascribing the otolithic contribution to the V.v.o.r. 0° to the utricular maculae and not the saccular maculae. First, the functional polarization vector of each utricular macula is directed mediolaterally (Fernandez & Goldberg, 1976*a*). This means that most utricular hair cells would be maximally sensitive to small rolls of the head about an initial orientation in which gravitational acceleration acted orthogonally to the plane of utricular maculae (i.e. V.v.o.r. 0°, V.v.o.r. 180°). Conversely, utricular hair cells would be minimally sensitive to rolls about an initial orientation in which gravitational acceleration was co-linear with the functional polarization vector in either direction (V.v.o.r. 0°L). The second reason for attributing the low-frequency characteristics of the V.v.o.r. 0° to the utricular maculae is that in contrast to the saccular maculae, the utricular maculae are reciprocally polarized (Fig. 1). Thus, for roll deviations from an initial prone orientation (V.v.o.r. 0°) the activity which originates from the utricular macula which is ipsilateral to the direction of the roll would increase and the activity originating from the utricular macula contralateral to the direction of the roll would decrease. This modulated utricular activity would be *synergistic* with the vertical canal signals for deviations from the prone orientation, but would be antiphasic to these signals for roll deviations from the supine orientation (V.v.o.r. 180°). The low-frequency gain of the V.v.o.r. 0° was severely attenuated when the rabbit was oriented 'left side down' (V.v.o.r. 0°L). Sinusoidal deviations from this orientation would be expected to evoke a maximal modulation of signals of saccular origin, and a minimal modulation of signals of utricular origin. Since the ventrodorsal functional polarization of the saccular maculae is weaker than the mediolateral functional polarization of the utricular maculae, it might be anticipated that rolls about the longitudinal axis of the rabbit would be less effective in evoking a signal of saccular origin. Furthermore, the functional polarization vectors of the saccular maculae are nearly co-linear and in phase, in contrast to the oppositely polarized utricular maculae. This means that for roll stimulation about the earth horizontal axis, the signals from the saccular maculae would co-vary rather than vary reciprocally. These facts would suggest that only the utricular maculae and the vertical semicircular canals would contribute directly to the conjugately organized V.v.o.r. 0° and V.v.o.r. 180°. The saccular maculae could contribute to torsional eye movements, a possibility not examined in the present experiment, or they might make an indirect contribution to vertical eye movements by modulating the activity of secondary cells in the vestibular nuclei which also receive convergent activity originating from the vertical semicircular canals at the level of the vestibular nuclei.

Comparison of the derived otolith response with other data and descriptive models. An estimate of the gain and phase of the otolithic component of the V.v.o.r. 0° was obtained in the present experiment by vector subtraction of the V.v.o.r. 90° (semicircular canal signal) from the V.v.o.r. 0° (otolith signal + semicircular canal signal). This derived estimate of otolith function was in reasonable agreement with the estimate obtained from measurements of the vestibulo-ocular reflex evoked by sinusoidal linear accelerations of rabbits (Baarsma & Collewijn, 1975). However, due to technical limitations, measurements of the v.o.r. at low frequencies of linear acceleration, where the present results indicate the highest otolith gain, have not been made (see filled diamonds, Fig. 7). The estimate of the otolithic contribution to the V.v.o.r. 0° obtained in the present experiment is at variance with a descriptive model of the otoliths which was based on ocular counter-rolling in human subjects (Young & Meiry, 1968). A time constant of less than 150 msec for the otolithic component of the roll-step response was 'predicted' by graphic subtraction of the 'nose-up' step roll response from the 'prone' response. This time constant is at least an order of magnitude smaller than the time constant obtained from step linear accelerations of rabbits (Baarsma & Collewijn, 1975) and would not be predicted from a second order linear model of the data obtained from sinusoidal roll stimulation. However, electrophysiological recordings from both primary otolithic afferents and second-order cells in the vestibular nuclei have revealed that many otolith-related cells exhibit a gain enhancement at higher frequencies of stimulation (Fernadez & Goldberg, 1976*c*; Schor, 1974; Vidal, Jeannerod, Lifschitz, Levitan, Rosenberg & Segundo, 1971).

Non-linearities of the V.v.o.r. 0°. The estimate of otolith function obtained in the present experiment is based on the assumption that the signals which originate from the vertical semicircular canals and from the otoliths combine linearly, presumably at the level of the vestibular nuclei. The same vector subtraction procedure was used to predict the V.v.o.r. 180°, and assumed that the supine orientation caused a phase shift of the otolith signal of 180° with respect to the semicircular canal signal. However, this assumption of linearity appears to be tenable only as it pertains to signals of utricular origin. Signals of utricular origin should have equivalent gains for sinusoidal deviations from the prone and supine orientations (V.v.o.r. 0°, V.v.o.r. 180°), and the phase of these signals relative to vertical semicircular canal signals should be shifted by 180° by a change from the prone to supine orientation. In agreement with this idea, the gain of the V.v.o.r. 180° at the lowest frequency tested, 0·005 Hz, was equivalent to the gain of the V.v.o.r. 0° at the same frequency, with a phase shift of about 180°. The equivalence in gain of this component at 0·005 Hz in the prone and supine orientations agrees with the observation that the sensitivity of utricular hair cells is not altered by gravitational forces which act orthogonally to the plane of the macula (Fernandez & Golberg, 1976*b*).

However, the signals which originate from the saccular maculae should not be similarly influenced by the prone and supine orientations. In both orientations the saccular signals should be relatively insensitive to sinusoidal deviations. But in the prone orientation the 'steady-state' signals from the weakly ventrodorsally polarized saccular maculae should be at a relative minimum, and in the supine orientation both 'steady-state' signals should be at a relative maximum. If saccular afferents converge centrally with afferents from the vertical semicircular canals, then it is possible that increased 'steady-state' primary afferent activity of saccular origin could enhance the response of secondary vestibular neurones to a stimulus-modulated input originating from the vertical semicircular canals. The departure of the measured from the 'predicted' V.v.o.r. 180° is consistent with this idea (Fig. 8). It is also possible that 'steady-state' primary afferent activity which originates from the semicircular canals could interact centrally with convergent stimulus-modulated primary activity of otolithic origin. Such a convergent interaction might account, in part, for the 'barbecue nystagmus' which is observed in subjects rotated at a constant velocity about an earth horizontal axis (Benson & Bodin, 1966; Young, 1967).

The hypothesized convergence of saccular and vertical semicircular canal signals might resolve a potentially ambiguous situation for an animal which rolls more than 90° from the prone orientation. In this condition the utricular maculae would provide erroneous information about head position, but the vertical semicircular canals, being relatively independent of gravitational acceleration, would always provide correct information about head rotation. An increased 'steady-state' signal of saccular origin when the head is supine would tend to enhance the gain of the 'correct' signal from the vertical semicircular canals relative to the 'incorrect' signal from the utricular maculae, and thereby provide more accurate information concerning head movement. Interestingly, the gain of the V.v.o.r. 180° at the lowest frequency tested, 0·005 Hz, was *not* potentiated relative to the gain of the V.v.o.r. 0° at the same frequency. This finding would imply that there is little convergence between saccular and utricular signals.

Although convergence of vertical semicircular canal signals with otolith signals upon secondary vestibular neurones has been observed (Abend, 1977, 1978; Schor, 1974; N. H. Barmack & V. E. Pettorossi, unpublished observations), there are no electrophysiological data which pertain to the hypothesized lack of convergence of signals which originate from the saccular and utricular maculae.

Generation and suppression of anticompensatory re-setting eye movements. The lack of anticompensatory re-setting vertical eye movements during the V.v.o.r. 0° and their unmasking during sinusoidal or step stimulation in the 'nose-up' orientation suggests the otolithic signals ordinarily suppress these eye movements (Figs. 2, 4, 5 and 9). It is of interest to note that signals of otolithic origin lag the semicircular canal signals by 120–160° over the frequency range of 0·005–0·8 Hz. This phase relationship may indicate a damping influence of the otolithic signals on canal signals. The re-setting eye movements may be triggered by some central set of neurones which detect change in the rate of discharge of secondary vestibular neurones, and the damping influence of the otolith signals may prevent this change in rate from exceeding a threshold. In the 'nose-up' orientation, this damping influence would be eliminated and in the supine orientation, the phase lag of 120–160° would actually become a phase lead of 20–60° and would increase the frequency of anticompensatory re-setting eye movements.

Functional contribution of the otoliths. The frequency response of the otoliths to sinusoidal roll stimulation was derived in the present experiment by vector subtraction of the V.v.o.r. 90° (semicircular canal signal) from the V.v.o.r. 0° (semicircular canal signal + otolith signal). This derived frequency response of the otoliths, in agreement with previous experiments (see Mayne, 1974 for review) suggests that the otoliths would be especially suited for detecting low-frequency changes in head position relative to gravity about the prone orientation (V.v.o.r. 0°). It would be expected that this information, combined with the sensitivity of the semicircular canals to higher frequencies of head movement would be useful in maintaining a relatively fixed spatial reference. In view of the otolithic contributions to vertical eye movements it might be anticipated that non-vestibular sensory information, such as optokinetic and neck proprioceptive inputs would be more important in signalling low-frequency movements of the head in the horizontal plane, since there is no equivalent otolithic signal in this plane. In the present experiment it was not possible to assess the relative contributions of the posterior and anterior canals to the V.v.o.r. 0°, nor was it possible to study directly the otolithic contributions to the V.v.o.r. 0° in the absence of semicircular canal signals. However, in a subsequent experiment the independent contributions of the vertical semicircular canals and otoliths to the V.v.o.r. 0° will be examined by plugging different combinations of vertical semicircular canals.

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REFERENCES

ABEND, W. K. (1977). Functional organization of the superior vestibular nucleus of the squirrel monkey. *Brain Res.* **132**, 65–84.

ABEND, W. K. (1978). Response to constant angular accelerations of neurons in the monkey superior vestibular nucleus. *Exp. Brain Res.* **31**, 459–473.

BAARSMA, E. A. & COLLEWIJN, H. (1974). Vestibulo-ocular and optokinetic reactions to rotation and their interaction in the rabbit. *J. Physiol.* **238**, 603–625.

BAARSMA, E. A. & COLLEWIJN, H. (1975). Eye movements due to linear accelerations in the rabbit. *J. Physiol.* **245**, 227–247.

BARMACK, N. H. (1976). Measurements of stiffness of extraocular muscles of the rabbit. *J. Neurophysiol.* **39**, 1009–1019.

BARMACK, N. H. (1977). A comparison of horizontal and vertical vestibulo-ocular reflexes of the rabbit. *Seventh Annual Meeting, Society for Neuroscience, Anaheim. Abstr.* **3**, 541.

BENSON, A. J. & BODIN, M. A. (1966). Interaction of linear and angular acceleration on vestibular receptors in man. *Aerospace Med.* **36**, 144–158.

ESTES, M. S., BLANKS, R. H. I. & MARKHAM, C. H. (1975). Physiologic characteristics of vestibular first-order canal neurons in the cat. I. Response plane determination and resting discharge characteristics. *J. Neurophysiol.* **38**, 1232–1249.

FERNANDEZ, C. & GOLDBERG, J. M. (1971). Physiology of peripheral neurones innervating semicircular canals of the squirrel monkey. II. Response to sinusoidal stimulation and dynamics of peripheral vestibular system. *J. Neurophysiol.* **34**, 661–675.

FERNANDEZ, C. & GOLDBERG, J. M. (1976*a*). Physiology of peripheral neurons innervating otolith organs of the squirrel monkey. I. Response to static tilts and to long-duration centrifugal force. *J. Neurophysiol.* **39**, 970–984.

FERNANDEZ, C. & GOLDBERG, J. M. (1976*b*). Physiology of peripheral neurons innervating otolith organs of the squirrel monkey. II. Directional selectivity and force–response relations. *J. Neurophysiol.* **39**, 985–995.

FERNANDEZ, C. & GOLDBERG, J. M. (1976*c*). Physiology of peripheral neurons innervating otolith organs of the squirrel monkey. III. Response dynamics. *J. Neurophysiol.* **39**, 996–1008.

FLOCK, Å. (1964). Structure of the macula utriculi with special reference to directional interplay of sensory responses as revealed by morphological polarization. *J. cell Biol.* **22**, 413–431.

GOLDBERG, J. M. & FERNANDEZ, C. (1971). Physiology of peripheral neurons innervating semicircular canals of the squirrel monkey. I. Resting discharge and response to constant angular accelerations. *J. Neurophysiol.* **34**, 635–660.

LINDEMAN, H. H. (1969). Studies on the morphology of the sensory regions of the vestibular apparatus. *Ergbn. Anat. EntwGesch.* **42**, 1–113.

LOE, P. R., TOMKO, D. L. & WERNER, G. (1973). The neural signal of angular head position in primary afferent vestibular nerve axons. *J. Physiol.* **230**, 29–50.

MAYNE, R. (1974). A systems concept of the vestibular organs. In *Handbook of Sensory Physiology. Vestibular system*, vol. 6, ed. KORNHUBER, H. H., pp. 493–580. Berlin: Springer.

MELVILLE JONES, G. & MILSUM, J. H. (1971). Frequency–response analysis of central vestibular unit activity resulting from rotational stimulation of the semicircular canals. *J. Physiol.* **219**, 191–215.

SCHOR, R. H. (1974). Responses of cat vestibular neurons to sinusoidal roll tilt. *Exp. Brain Res.* **20**, 347–362.

TER BRAAK, J. W. G. (1936). Untersuchungen über optokinetischen nystagmus. *Archs néerl. Physiol.* **21**, 309–376.

VIDAL, J., JEANNEROD, M., LIFSCHITZ, W., LEVITAN, H., ROSENBERG, J. & SEGUNDO, J. P. (1971). Static and dynamic properties of gravity-sensitive receptors in the cat vestibular system. *Kybernetik* **9**, 205–215.

YOUNG, L. R. (1967). Effects of linear acceleration on vestibular nystagmus. *Third Symposium on the Role of the Vestibular Organs in Space Exploration, NASA SP-152*, 383–391.

YOUNG, L. R. & MEIRY, J. L. (1968). A revised dynamic otolith model. *Aerospace Med.* **39**, 606–608.