

Relationship of Semicircular Canal Size to Vestibular-Nerve Afferent Sensitivity in Mammals

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Yang A, Hullar TE. Relationship of semicircular canal size to vestibular-nerve afferent sensitivity in mammals. *J Neurophysiol* 98: 3197–3205, 2007. First published October 3, 2007; doi:10.1152/jn.00798.2007. The relationship between semicircular canal radius of curvature and afferent sensitivity has not been experimentally determined. We characterized mouse semicircular canal afferent responses to sinusoidal head rotations to facilitate interspecies and intraspecies comparisons of canal size to sensitivity. The interspecies experiment compared the horizontal canal afferent responses among animals ranging in size from mouse to rhesus monkey. The intraspecies experiment compared afferent responses from the larger anterior canal to those from the smaller horizontal canal of mice. The responses of mouse vestibular-nerve afferents showed a low- and high-frequency phase lead and high-frequency gain enhancement. Regular horizontal-canal afferents showed a sensitivity to 0.5-Hz sinusoidal rotations of 0.10 ± 0.03 (SD) spike \cdot s⁻¹/deg \cdot s⁻¹ and high-gain irregular afferents showed a sensitivity of 0.25 ± 0.11 spike \cdot s⁻¹/deg \cdot s⁻¹. The interspecies comparison showed that the sensitivity of regular afferents was related to the radius of curvature R according to the formula $G_r = 0.23R - 0.09$ ($r^2 = 0.86$) and the sensitivity of irregular afferents was related to radius according to the formula $G_i = 0.32R + 0.01$ ($r^2 = 0.67$). The intraspecies comparison showed that regularly firing anterior canal afferents were significantly more sensitive than those from the relatively smaller horizontal canal, with $G_r = 0.25R$. This suggests that canal radius of curvature is closely related to afferent sensitivity both among and within species. If the relationship in humans is similar to that demonstrated here, the sensitivity of their regular vestibular-nerve afferents to 0.5-Hz rotations is likely to be about 0.67 spike \cdot s⁻¹/deg \cdot s⁻¹ and of their high-gain irregular afferents about 1.06 spikes \cdot s⁻¹/deg \cdot s⁻¹.

INTRODUCTION

An animal's typical head movements have been assumed to be related to the geometry of its semicircular canals (Gray 1907–1908; Howland and Masci 1973a,b; Jones and Spells 1963). Recently, this possible relationship has even been used in an attempt to infer the behaviors of extinct animals from the fossil record (Alonso et al. 2004; Clarke 2005; Rogers 1998, 2005; Spoor et al. 1994, 2002). The conclusions of these studies rely on the controversial assumptions that semicircular canal size determines vestibular afferent sensitivity, and that this sensitivity is matched closely to an animal's behavior to maximize its ability to encode head rotations (Graf and Vidal 1996). Although biophysical models of the semicircular canals suggest that the mechanical sensitivity of the canal may indeed be related to the radius of curvature of the centerline of the

membranous duct (Oman et al. 1987; Rabbitt et al. 2004; Squires 2004), such a relationship has not been demonstrated experimentally.

Early attempts to correlate canal radius of curvature and afferent responses found contradictory results. Ten Kate (1970) studied the relationship of canal size to sensitivity in fish, whose canals continue to grow with body size into adulthood. Contrary to the predictions of a standard model of canal behavior, he found that the threshold amount of rotation required to stimulate the vestibular system did not change despite growth of the labyrinth. He hypothesized that changes in cupular mechanics, efferent tone, hair cell sensitivity, central connections, or extraocular muscle responsiveness compensated for larger canal radius to keep sensitivity constant. Curthoys (1982a) analyzed the relationship between size and sensitivity using a similar developmental paradigm but found different results. He measured the responses of vestibular-nerve afferents in newborn rats as their labyrinths grew to adult dimensions over the first week of life. He found that the sensitivity of the afferents grew with the size of the semicircular canals, although the sensitivity increased more than would be expected based on changes in canal anatomy alone. He hypothesized that the increased sensitivity beyond that explained by canal size might be due to changes in cupular mechanical characteristics or changes in the physiology of the neuroepithelium.

Studies comparing the sensitivity to the size of the semicircular canals of growing animals suffer from the disadvantage that developmental changes affecting the endolymph composition, biomechanics of the cupula, structure of the neuroepithelium, and inherent dynamics of afferents themselves might confound the results. An alternative approach relies on comparing the sensitivity of canals of different sizes within adult animals of the same species. Differences in size among the anterior, horizontal, and posterior semicircular canals have been used to make such comparisons, although these studies have not reported consistent results. In the cat, both the smaller posterior (Anderson et al. 1978) and the larger anterior (Blanks et al. 1975) canals were reported to be the most sensitive; in the squirrel monkey the larger anterior canal was reported to be more sensitive than both of the other canals (Goldberg and Fernandez 1971); and in pigeons, the sensitivities of the larger anterior and smaller horizontal canals were found to be similar (Anastasio et al. 1985). Interpreting the results of these studies is difficult because both Blanks et al. (1975) and Goldberg and

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Fernandez (1971) relied on step stimuli, which are subject to afferent adaptation that may affect measurements of their sensitivity (Boyle and Highstein 1990; Dickman and Correia 1989a; Fernandez and Goldberg 1971, 1976a; Ledoux 1961; Lowenstein 1955; Precht et al. 1971) and measurements of canal sensitivity in pigeon may have been complicated by the fact that in this species the anterior canal is dihedral and its plane of maximum sensitivity is difficult to determine anatomically (Dickman 1996). A third approach, relying on interspecies comparisons of the relationship between canal radius of curvature and sensitivity, has been attempted using biophysical models of the canals but has not been tested experimentally (Oman 1981; Oman et al. 1987; Rabbitt et al. 2004).

Given the importance of understanding the effect of biophysical parameters such as the radius of curvature on the responses of the canals, and noting that existing studies did not show a consistent relationship between canal size and sensitivity, we used two distinct techniques to pursue the question further. We first performed an interspecies comparison between the radius of curvature and the sensitivity of horizontal canal vestibular-nerve afferents. We then performed an intraspecies comparison between the sensitivities of mouse afferents innervating the larger anterior canal and those innervating the smaller horizontal canal. We chose to use mice for this study because they allowed us to compare canal radius of curvature to sensitivity over a wider range of canal sizes than otherwise possible and because the size and orientation of their canals is well defined (Calabrese and Hullar 2006). Furthermore, analyzing the afferent responses of mice is especially important given the increasingly significant role they play in studying the vestibular system (Bagnall et al. 2007; Hoebeek et al. 2005; Jones et al. 2005; Puyal et al. 2006). The responses of mouse afferents to sinusoidal rotations in an *in vitro* preparation have been described (Lee et al. 2005) but the data presented here and in similar experiments (Han et al. 2006; Jones et al. 2007) are expected to serve as a baseline of comparison for future *in vivo* studies of the mouse peripheral vestibular system.

METHODS

All experiments were conducted in accordance with a protocol approved by the Animal Studies Committee of the Washington University School of Medicine. In all, 56 mice of either sex of strain C57/BL6 (Jackson Labs) and 8–12 wk in age were anesthetized with a mixture of ketamine (87 mg/kg) and xylazine (13 mg/kg) and the scalp was opened sagittally. The external auditory canals and maxilla were fixed into a custom-built stereotaxic frame so that the horizontal semicircular canals were parallel to earth-horizontal as determined by skull landmarks (Calabrese and Hullar 2006). The stereotaxic frame was secured onto a custom-built motion delivery device (Shot, Greenville, IN) capable of providing sinusoidal rotation in any canal plane. A craniotomy was performed on the animal's left side and the paraflocculus was suctioned from the subarcuate fossa to expose the porus of the internal auditory canal. A borosilicate pipette (model 30-31-1; FHC, Bowdoinham, ME) filled with 3 M NaCl was oriented over the nerve using a three-dimensional micromanipulator (model US-3F; Narishige International USA, East Meadow, NY) and advanced into the vestibular nerve using a one-dimensional hydraulic drive (model MO-22; Narishige International USA).

Voltages were measured between a silver wire introduced into the open end of the micropipette and a ground electrode secured in the cervical musculature. Afferent spike trains were amplified and re-

corded in the standard way for later off-line analysis using programs custom-written for the Matlab working environment (The MathWorks, Natick, MA). A fiber's responses to rotations and static tilts allowed it to be categorized as an otolith or canal afferent. The responses of canal afferents to hand rotations in each canal plane were observed and the fiber was assigned to the canal whose plane matched the most responsive direction. Only otolith fibers and canal fibers originating from the horizontal or anterior (superior) semicircular canals were used in this study. A total of 15–20 s of spontaneous discharge of each afferent were recorded before sinusoidal stimulation in the plane of the canal began. All afferents were recorded at a peak velocity of 40 deg/s, affording a good signal-to-noise ratio over the frequency range tested. Horizontal canal afferents were stimulated over a maximal frequency range of 0.01–4 Hz, whereas anterior canal data presented in this study were collected at 0.5 Hz only. Over the frequencies and velocities tested here, no afferents demonstrated cutoff, an effect observed previously in mammalian species including gerbil (Schneider and Anderson 1976), chinchilla (Hullar et al. 2005), and rhesus macaque monkeys (Ramachandran and Lisberger 2005); and nonmammalian species such as bullfrog (Segal and Outerbridge 1982), goldfish (Hartmann and Klinke 1980), and pigeon (Dickman and Correia 1989b). Each animal breathed spontaneously throughout the experimental procedure and its body temperature was held constant at 35.5–36.5°C using a servocontrolled heating blanket (model 40-90-8B, FHC).

Measurements of canal dimensions were drawn from the literature for the following animals: mouse (Calabrese and Hullar 2006); rat and guinea pig (Curthoys and Oman 1986); cat (Curthoys et al. 1977a); squirrel monkey (Ramprashad et al. 1984); chinchilla (Ramprashad et al. 1984); and rhesus macaque (Jones and Spells 1963). The published sensitivities of vestibular-nerve afferents were compiled for the following animals: rat and guinea pig (Curthoys 1982b); cat (Tomko et al. 1981); squirrel monkey (Goldberg et al. 1982); chinchilla (Baird et al. 1988); and rhesus macaque (Haque et al. 2003). The radius of curvature of most of the animals was measured using either histologic sections (Curthoys et al. 1977a; Ramprashad et al. 1984) or photographs of the intact membranous duct (Curthoys and Oman 1986; Jones and Spells 1963). In the mouse, the radius was measured from micro-CT scans of the bony canal (Calabrese and Hullar 2006). Comparisons of afferent sensitivity were made at 0.5 Hz, a frequency that lies comfortably above the lower corner frequency in all the animals tested here. Several different peak velocities of sinusoidal stimulation were used in the various animal studies cited here, but this is not expected to influence the results because the linearity of sinusoidal responses of primary vestibular-nerve afferents to moderate rotations has been demonstrated in several separate studies (Baird et al. 1988; Hullar et al. 2005; Sadeghi et al. 2007).

RESULTS

Three major types of afferents have been described in mammals: regular, high-gain irregular, and low-gain irregular. Recent results describing the responses of low-gain irregular fibers have shown that at frequencies >10 Hz they actually have a sensitivity approaching that of high-gain fibers but maintain a consistent phase lead over other fiber types at all frequencies tested, giving rise to the suggestion that they might be more accurately called "phase-led" fibers (Hullar et al. 2005). Categorization of afferent types depends on their normalized coefficient of variation, or CV* (Goldberg 1984). A standardized relationship between coefficient of variation and firing rate was determined by measuring the interspike interval (ISI) and coefficient of variation (CV) of 25 otolith afferents from 11 animals during static tilts. The CV* of each afferent was defined as its coefficient of variation when firing with an

TABLE 1. Constants relating CV and interspike intervals (ISIs) to CV^* in the mouse

| ISI | <i>a</i> | <i>b</i> |
|-------|----------|----------|
| 0.010 | 0.72 | 0.92 |
| 0.013 | 0.88 | 0.98 |
| 0.018 | 1.05 | 0.97 |
| 0.020 | 1.12 | 0.95 |
| 0.023 | 1.17 | 0.95 |
| 0.025 | 1.23 | 0.92 |
| 0.028 | 1.22 | 0.85 |
| 0.030 | 1.27 | 0.84 |
| 0.035 | 1.33 | 0.81 |
| 0.040 | 1.29 | 0.72 |
| 0.045 | 1.26 | 0.69 |
| 0.050 | 1.18 | 0.61 |
| 0.060 | 1.05 | 0.46 |
| 0.070 | 0.92 | 0.30 |
| 0.080 | 0.89 | 0.20 |
| 0.010 | 0.72 | 0.92 |

Formula is $CV(ISI) = a(ISI)(CV^*)^{b(ISI)}$, where ISI is the mean interspike interval (in seconds). Data collected from mouse otolith afferents during static tilts.

ISI of 15 ms. The CV^* for each afferent was described according to the relationship $CV(ISI) = a(ISI)(CV^*)^{b(ISI)}$. Values of the coefficients *a* and *b* relating ISI, CV, and CV^* are shown in Table 1.

Recordings from 45 mice yielded usable data from 87 horizontal canal afferents and 25 anterior canal afferents. The mean and CV of ISIs during spontaneous discharge of the horizontal semicircular afferents are shown in Fig. 1. The superimposed curves, determined using the coefficients shown in Table 1, separate the population of fibers into regular afferents with a $CV^* < 0.1$ and irregular afferents with a $CV^* > 0.1$. This level for distinguishing regular from irregular fibers is identical to that used previously in chinchillas (Baird et al. 1988; Hullar et al. 2005) but slightly lower than the threshold

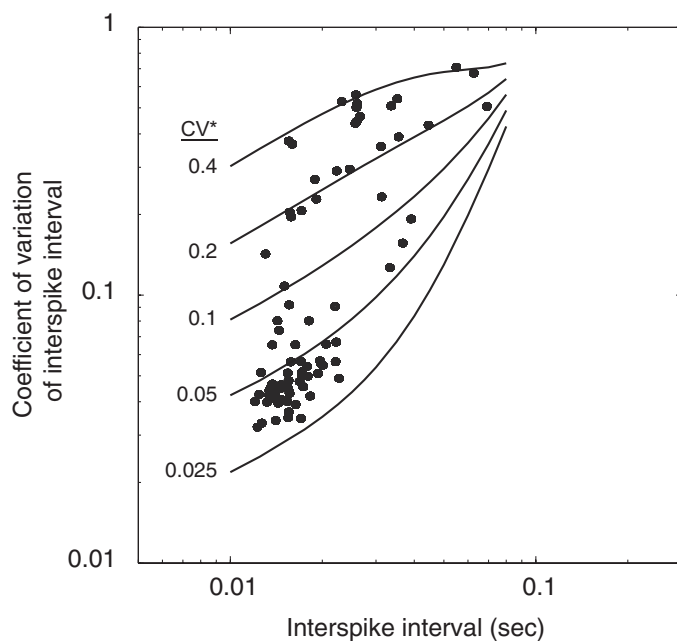


FIG. 1. Relationship of coefficient of variation (CV) and interspike interval for the population of horizontal semicircular canal afferents. Solid lines separate the population of afferents according to their normalized CV (CV^*).

of $CV^* = 0.15$ used in rhesus monkeys (Sadeghi et al. 2007). The relationship between CV and ISI among the population of spontaneously firing afferents shown here is similar to that previously reported for chinchilla (Baird et al. 1988; Hullar et al. 2005) and squirrel monkey (Goldberg et al. 1984). The gain and phase with respect to head velocity of each afferent at 0.5 Hz, 40 deg/s, plotted as a function of CV^* , are shown in Fig. 2. Low-gain (high phase lead) irregular afferents were defined as those fibers with a sensitivity < 0.2 spikes/s per deg/s at 0.5 Hz and a $CV^* > 0.3$, distinguishing a population of fibers similar to that initially described in chinchillas (Baird et al. 1988). The resting firing rate, CV^* , and sensitivity of each fiber type is shown in Table 2.

The responses of each mouse afferent over the range of frequencies studied here are shown in Fig. 3. Consistent with previous studies in other mammalian species, most afferents in each subset showed an increase in sensitivity from low to high frequencies and a phase lead re head velocity at both low and high frequencies (Baird et al. 1988; Fernandez and Goldberg 1971). The average responses of each group are shown in Fig. 4. In chinchillas, Baird et al. (1988) described the behavior of chinchilla afferent responses using a transfer function of the

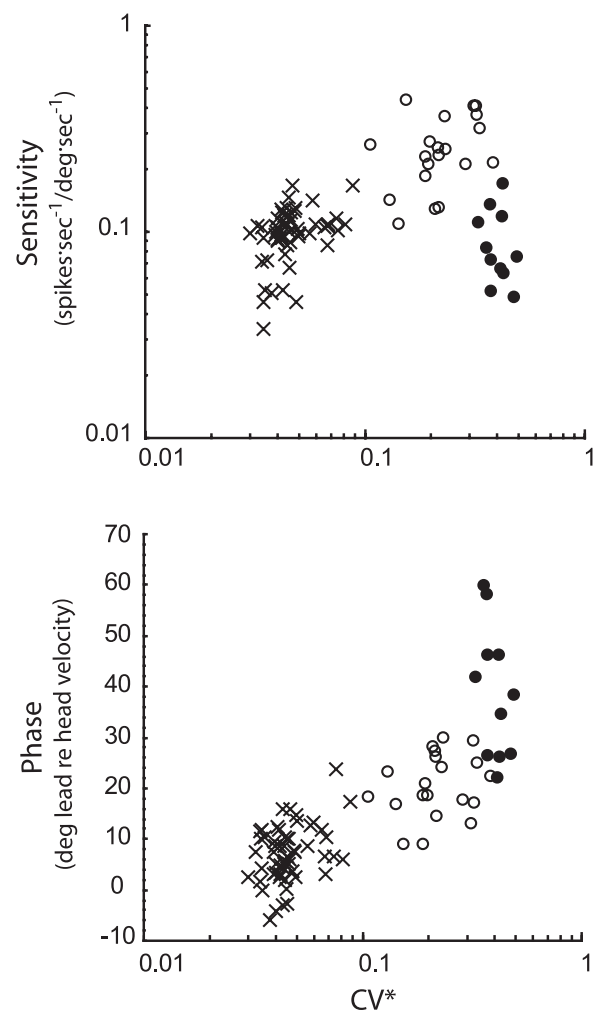


FIG. 2. *Top*: sensitivity (spikes $s^{-1}/deg s^{-1}$) of horizontal semicircular canal afferents. *Bottom*: phase re head velocity of the same fibers. x, regular fibers; open circles, high gain irregular fibers; filled circles, low-gain (phase-lead) irregular fibers.

TABLE 2. Characteristics of mouse afferent responses

| | <i>n</i> | Resting Rate | CV* | Sensitivity |
|--------------------------------------|----------|--------------|-------------|-------------|
| Horizontal canal afferents | 83 | 55.1 ± 17.0 | 0.13 ± 0.13 | 0.14 ± 0.08 |
| Regular horizontal canal afferents | 56 | 62.4 ± 13.0 | 0.05 ± 0.01 | 0.10 ± 0.03 |
| High-gain horizontal canal afferents | 20 | 43.7 ± 16.2 | 0.23 ± 0.08 | 0.25 ± 0.11 |
| Low-gain horizontal canal afferents | 11 | 39.4 ± 15.2 | 0.41 ± 0.05 | 0.07 ± 0.03 |
| Regular anterior canal afferents | 22 | 68.6 ± 18.2 | 0.05 ± 0.01 | 0.17 ± 0.04 |

Values are means ± SD. Resting firing rate is given in spikes/s; CV* represents normalized coefficient of variation of interspike intervals; sensitivity is given in units of spike s⁻¹/deg s⁻¹ measured with respect to 0.5-Hz sinusoidal rotations with a peak head velocity of 40 deg/s.

form $H = G \times (\tau_L s / [(\tau_L s + 1)(\tau_S s + 1)])(\tau_F s + 1)^k$, where G is the midband gain of the system, τ_L is the long time constant of the torsion pendulum equation, τ_S is its short time constant, τ_F is a high-frequency lead term time constant, and k is a fractional operator. The same form was chosen to fit the data presented here because of the subjective similarity of the mouse and chinchilla afferent Bode plots, the same range of frequencies tested, and the close phylogenetic relationship of the animals. As in Baird et al.'s equations, the short time constant was set to 0.007 and the high-frequency lead term time constant was set to 0.2. For mouse regular afferents, the

resulting transfer function took the form $0.09\{(3.0s)/(3.0s + 1)(0.007s + 1)\}(0.2s + 1)^{0.03}$; for high-gain irregular afferents it took the form $0.27\{(2.5)/(2.5 + 1)(0.007s + 1)\}(0.2s + 1)^{0.4}$; and for low-gain irregular afferents it took the form $0.10\{(2.6s)/(2.6s + 1)(0.007s + 1)\}(0.2s + 1)^{0.5}$.

By including the results obtained from mouse vestibular afferents reported here, a wide range of canal sizes and afferent responses could be compared. The canal radius of curvature ranged in size from that of the mouse (0.725 mm) to that of the rhesus macaque (2.55 mm). The sensitivities of horizontal semicircular canal afferents to rotations at 0.5 Hz are shown as a function of radius of curvature of the canal in Fig. 5. Both regular and high-gain irregular afferents show a correlation with canal size, although the relationship is much more robust for regular fibers. Low-gain irregulars could not be included here because their responses have not been reported for many species. The sensitivity of regular afferents G_r , in spikes · s⁻¹/deg · s⁻¹, is related to the radius of curvature R according to the formula $G_r = 0.23R - 0.09$ ($r^2 = 0.86$) and the sensitivity of irregular afferents G_i is related to the radius of curvature according to the formula $G_i = 0.32R + 0.01$ ($r^2 = 0.67$).

An intraspecies comparison of canal size to afferent sensitivity was made by comparing the sensitivities of a population of 25 regularly firing anterior canal afferents rotated at 0.5 Hz, 40 deg/s and the population of regularly firing horizontal canal afferents already described. Irregular afferent responses were

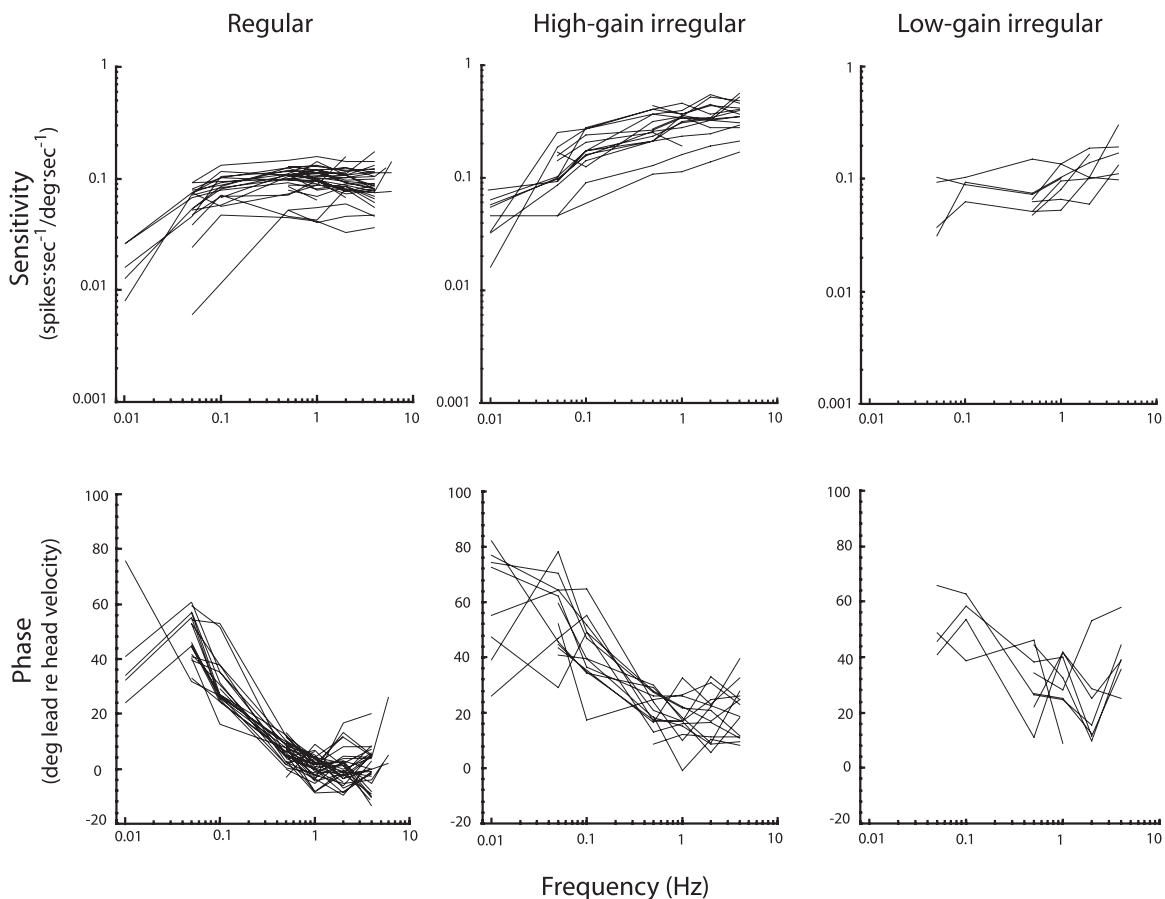


FIG. 3. Responses of all horizontal canal afferents included in this study. *Left*: regular fibers. *Middle*: high-gain irregular fibers. *Right*: low-gain (phase-led) irregular fibers.

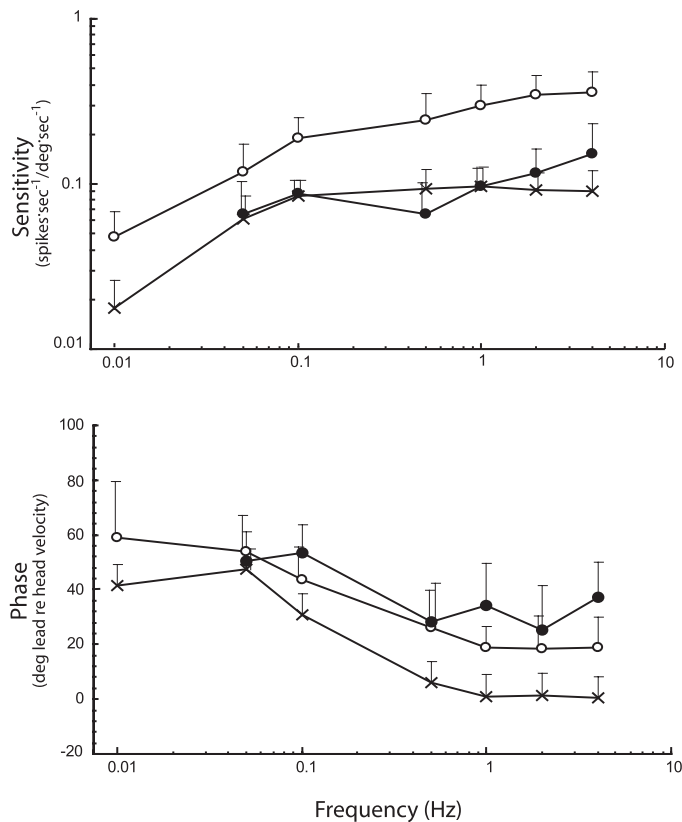


FIG. 4. Average sensitivities of all afferents. *Top*: sensitivity ($\text{spikes} \cdot \text{s}^{-1} / \text{deg} \cdot \text{s}^{-1}$). *Bottom*: phase re head velocity. \times , regular fibers; open circles, high-gain irregulars; filled circles, low-gain (phase-led) irregulars. Bars = SD.

not used for this comparison because their greater range of sensitivities would be likely to obscure any difference in sensitivity between the two canals. The average radius of curvature of the horizontal canals of the C57/BL6 strain of mouse used here is 0.725 mm and of the anterior canals is

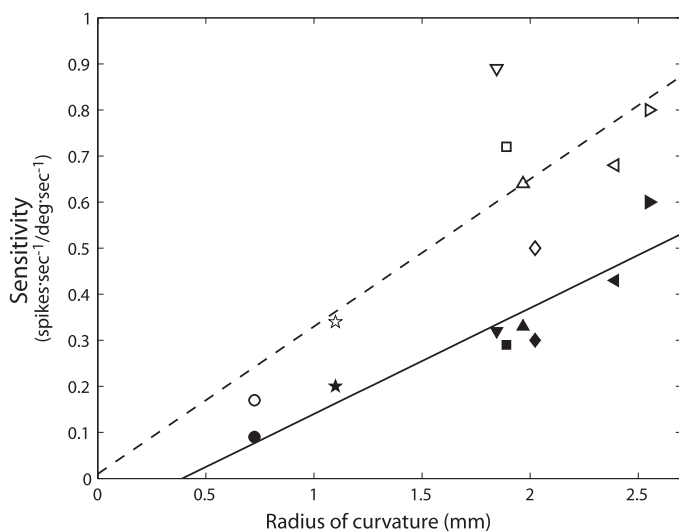


FIG. 5. Relationship of sensitivity to canal dimension in several mammals. Regular fibers, closed symbols with solid line representing best least-squares linear fit. High gain irregular fibers, open symbols with dashed line representing best least-squares linear fit. Mouse: circles; rat: pentagrams; guinea pig: squares; cat: down-pointing triangles; squirrel monkey, up-pointing triangles; chinchilla, diamonds; gerbil: left-pointing triangles; rhesus monkey, right-pointing triangles.

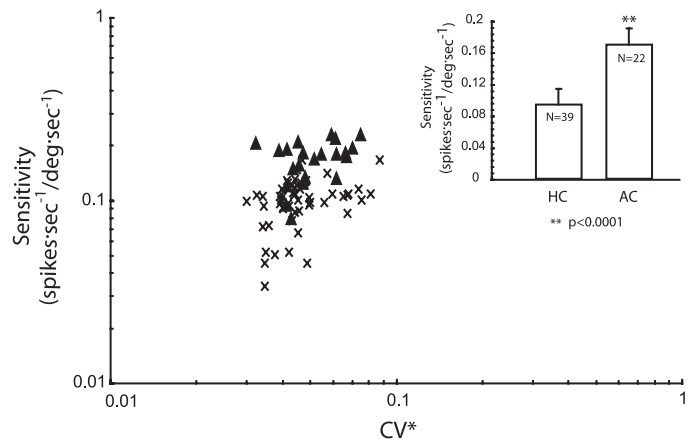


FIG. 6. Sensitivity ($\text{spikes} \cdot \text{s}^{-1} / \text{deg} \cdot \text{s}^{-1}$) as a function of CV^* of the population all regularly firing afferents used in this study. \times , horizontal canal fibers; closed triangles, anterior canal fibers. *Inset*: average sensitivities of populations of fibers. HC, horizontal canal fibers; AC, anterior canal fibers. Bars = SD. ** $p < 0.0001$.

1.032 mm (Calabrese and Hullar 2006). The sensitivity of horizontal and anterior canal fibers recorded at 0.5 Hz is shown in Fig. 6. There was a highly significant difference ($P < 0.0001$) between the mean sensitivity of the horizontal and anterior canal afferent populations. The relationship of canal sensitivity to radius of curvature between the horizontal and anterior canals is $0.25 \text{ spike} \cdot \text{s}^{-1} / \text{deg} \cdot \text{s}^{-1}$ per mm, a figure in remarkable agreement with the value of $0.23 \text{ spike} \cdot \text{s}^{-1} / \text{deg} \cdot \text{s}^{-1}$ per mm calculated in the interspecies comparison.

The sensitivity of a canal to rotations can be represented as the length of a vector drawn through its center of curvature and in the direction of its axis of maximum response (Rabbitt 1999). The combined sensitivity of the six-canal system to a particular rotation depends on the orientation of the canals with respect to each other and to the axis of rotation (Brichta et al. 1988; Calabrese and Hullar 2006; Day and Fitzpatrick 2005; Hullar and Williams 2006; Mazza and Winterson 1984). A perfectly orthogonal set of three canals, each with a unit sensitivity, would have a response to a rotation about any axis equal to one, whereas two orthogonal sets representing the labyrinths of both ears would have a uniform response equal to two. The mouse semicircular canals on each side are neither orthogonal to one another nor parallel to their contralateral synergistic partners, suggesting that the system's overall response to a head rotation depends on the direction of the axis of rotation. Shown in Fig. 7A is the overall sensitivity of the six semicircular canals of the mouse to head rotations in any direction, assuming that all canals have sensitivities equal to one. Figure 7B shows the same data, after adjustment for the relative sensitivities of each of the canals as determined by their relative radii and normalizing so that the average sensitivity remains equal to one. The figure indicates instead that in both cases, the system's response to rotations about the naso-occipital axis is enhanced relative to other directions.

DISCUSSION

The relationship of semicircular canal radius of curvature to afferent sensitivity was determined. Mouse afferent responses were recorded to facilitate both interspecies and intraspecies comparisons. Both techniques showed a close relationship

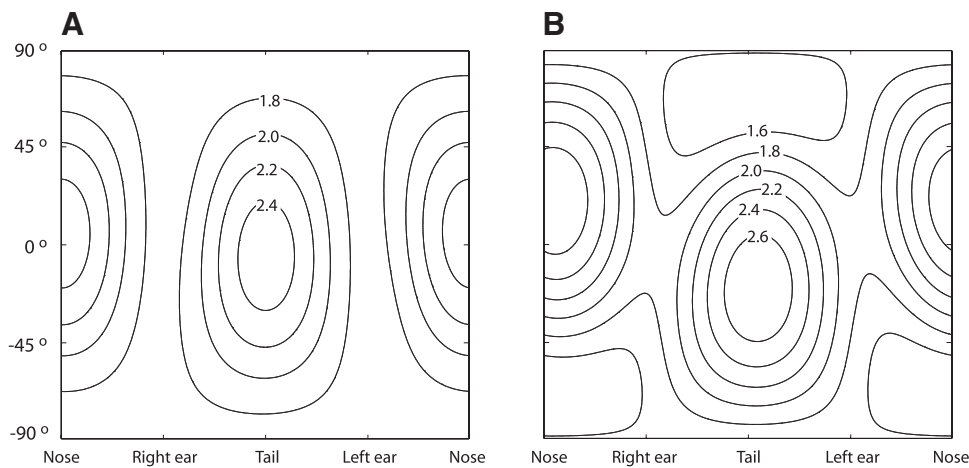


FIG. 7. Overall response of semicircular canals to rotations in any direction. *Left*: all canal sensitivities weighted equally. *Right*: canal sensitivities weighted by radius of curvature of the canals. Size of canals in both cases normalized so average size equals one. *x*-axis, azimuth of rotation vector centered on origin of mouse head coordinate system. *y*-axis, elevation of rotation vector, in degrees, with +90 defined as vertical in the dorsal direction and -90 defined as vertical in the ventral direction.

between canal size and afferent responses. These findings extend our understanding of information processing in the vestibular system, its comparative anatomy and evolution, vestibular responses in humans, and the fundamental biomechanics of the semicircular canals.

The mean resting rates of regular and irregular mouse afferents are lower than those previously recorded in some other mammals including rhesus monkey (Haque et al. 2003), squirrel monkey (Lysakowski et al. 1995), and chinchilla (Baird et al. 1988) but equivalent to or higher than rates described in cat (Tomko et al. 1981), rat, and guinea pig (Curthoys 1982b). Regular fibers make up 76% of the afferent population in rhesus monkeys (Haque et al. 2003), 58% in guinea pigs (Curthoys 1982b), 43% in rats (Curthoys 1982b), and 35% in gerbils (Schneider and Anderson 1976). These values are difficult to compare directly because they rely on varying criteria to classify afferents into groups, but squirrel monkey, mouse, and chinchilla afferents have been divided using a uniform method based on the normalized coefficient of variation (CV^*) of their interspike intervals at rest, so that regular afferents have a $CV^* < 0.1$ and irregular afferents have a $CV^* > 0.1$ (Goldberg et al. 1984). The normalized coefficient of variation in these three species ranges from < 0.05 to > 0.4 , with a concentration of afferents with a $CV^* < 0.05$. The proportion of regular afferents is 42% in squirrel monkeys (Lysakowski et al. 1995), 67% in chinchillas (Baird et al. 1988), and 67% in mice. The irregular afferent populations in several mammals have been shown to include both high-gain and low-gain (phase-led) groups. Low-gain irregular afferents represented 29% of the afferent population in squirrel monkeys, 17% of the afferent population in chinchillas, and 10% of the afferent population in mice. These results indicate that there is not a clear relationship between animal size and either resting firing rate or regularity of discharge of the overall population of afferents.

Responses of mouse afferents over the range 0.05–4 Hz follow the same pattern of low- and high-frequency phase lead with low-frequency gain attenuation and high-frequency gain enhancement as previously shown in several other mammalian species (Baird et al. 1988; Curthoys 1982b; Fernandez and Goldberg 1971; Haque et al. 2003; Schneider and Anderson 1976; Tomko et al. 1981). Between 2 and 4 Hz, the sensitivity of low-gain (phase-led) mouse irregular afferents surpasses that of regular afferents and their phase lead re head velocity is

greater than that of both regular and high-gain irregular afferents. The shape of the Bode plots characterizing these responses matches the results of recent studies of high-frequency afferent dynamics in chinchillas (Hullar et al. 2005), although further recordings of mouse afferents responding to stimuli > 4 Hz are necessary to explore this similarity further.

The most significant difference between the dynamic responses of mouse afferents and those of other animals is the extraordinarily low sensitivity of mouse fibers. Signal detection theory indicates that low-sensitivity afferents would require a more regular rate of discharge to carry information as reliably as the higher-sensitivity fibers shown here to be typical of larger species (Green and Swets 1966). Because neither the discharge regularity of individual afferents nor the proportion of all afferents that discharge regularly appears to be greater in mice than in animals with larger, more sensitive canals, their lower sensitivity is likely to decrease their ability to accurately encode small changes in head velocity. Complicating this problem is that the mouse canal's neuroepithelium is significantly smaller than that of larger rodents (Desai et al. 2005) and the number of vestibular-nerve afferents is smaller than that in other mammals (Bäurle and Guldin 1998). To overcome these disadvantages, mice may use their vibrissae, vision, audition, proprioception, or other orientation cues to augment their limited vestibular information (Dräger and Hubel 1975). Alternatively, they may not require a vestibular system able to measure subtle changes in head velocity because relatively few of their rotational head movements may occur at velocities slow enough to require a particularly sensitive system.

Many evolutionary and comparative anatomic studies have hypothesized that the radius of curvature of the semicircular canals determines vestibular afferent sensitivity, and that this sensitivity is in turn related to head motion (Gray 1907–1908; Howland and Masci 1973a,b; Jones and Spells 1963; Spoor and Zonneveld 1998). The data here represent the first experimental evidence that the sensitivity of the semicircular canals to sinusoidal head rotations shows a close relationship to canal size across a variety of mammalian species. Although the regression is strong, it is important to note that methodologic differences may have influenced the results somewhat. The radius of curvature was determined from the membranous duct for all the animals included in this study except for the mouse, which was determined from the bony canal. It seems unlikely, however, that the difference between these two techniques is

significant. Previously published data indicate that the cross-sectional radius of the mouse bony canal is between 50 and 75 microns and that its radius of curvature is 725 microns (Calabrese and Hullar 2006). The radius of the membranous duct must therefore lie between 650 and 800 microns. Inspection of Fig. 5 indicates that this range of values does not significantly affect the overall relationship between radius of curvature and sensitivity. Furthermore, this range of uncertainty is likely to be a substantial overestimate given the observation that the membranous duct of small rodents fills a large portion of the entire lumen of the bony canal and the radius of curvature of the two must therefore be quite similar (Ramprashad et al. 1984). Another difference is that the radius of some of the animals was measured by fitting a circle to the long and slender portion of the canal (Calabrese and Hullar 2006; Curthoys and Oman 1986), whereas others outlined the entire circuit around the duct and utricle, calculating its maximal diameter, and dividing by two (Curthoys et al. 1977a; Ramprashad et al. 1984). Such methods are likely to give similar results if the circuit around the canal and utricle approximates a circle. Several descriptions of the mammalian labyrinth indicate that this is a valid approximation (Curthoys and Oman 1986, 1987; Lewis et al. 1985). A direct comparison between the two methods performed on the same species by the same author also indicates that they give similar results (Curthoys and Oman 1986; Curthoys et al. 1977b).

Although the data here indicate that the sensitivity of a canal is related to its size, it remains controversial whether the sensitivity of the canals is in turn related to head motion. Several authors have hypothesized that animals with slower head movements typically have more sensitive canals able to accurately measure low velocities, whereas animals with faster head motions benefit from less sensitive canals to avoid excitatory or inhibitory saturation of the system (Alonso et al. 2004; Clarke 2005; Jones and Spells 1963). Others have suggested that larger, more sensitive canals may help to provide fine-grained information to support the complex head and body movements of particularly agile animals (Gray 1907–1908; Hadziselimovic and Savkovic 1964; Matano et al. 1985; Spoor et al. 1994, 2002, 2007). A recent study indicated that more highly maneuverable animals do have somewhat larger canals than slower-moving animals of similar size and hypothesized that particularly agile species would require a robust vestibulocollic response to avoid saturating their sensitive canals (Spoor et al. 2007). Although an argument against this theory is that the inherent dynamics of the vestibulocollic reflex and the limited range of motion of the head on the body may make it impossible for the afferents of agile animals with large canals to avoid becoming saturated, experimental evidence from primates shows that saturation of the canals may not actually constrain an animal's behavior significantly. Given the resting firing rate and sensitivity of squirrel monkey afferents, their typical frequency and velocity of head motions indicate that their afferents may often experience inhibitory saturation (Armand and Minor 2001; Fernandez and Goldberg 1976b) but, despite this, these animals are considered to be particularly agile (Spoor et al. 2007).

The data presented here constitute the first experimental evidence confirming that, within a species, afferent sensitivity varies with canal size. The semicircular canal system is there-

fore particularly sensitive to rotations in certain directions and less sensitive to rotations in others, with the direction of maximum sensitivity determined in part by the relative radii of the canals. This contradicts the hypothesis that the sensitivity of each canal's afferents are normalized in the vestibular periphery so that rotations of the head about any axis are represented equally for interpretation by the brain (Graf and Vidal 1996). An enhanced sensitivity of the vestibular system to rotations in certain directions has been linked theoretically to the typical directions of an animal's movement. Two possible relationships between typical directions of head rotation and directional sensitivity of the system of semicircular canals have been proposed. The direction of greatest sensitivity of the semicircular canal system may coincide with the typical axes of low-velocity head rotations to facilitate their accurate measurement, whereas more vigorous head rotations may occur in less sensitive directions. This model has been used to infer the rate at which dinosaurs moved their heads in the vertical and horizontal planes (Alonso et al. 2004; Clarke 2005). Alternatively, the direction of greatest sensitivity of the canal system may correlate with the direction in which an animal's most vigorous motion occurs because those motions may be best controlled by a system maximally sensitive in the same direction. This theory has been used to relate the increased relative radius of the anterior canals to greater motion in the sagittal plane during the development of bipedalism among primates (Spoor and Zonneveld 1998; Spoor et al. 1994).

The data shown here explore this issue further by examining quantitatively the relative effect of canal orientation and radius on the overall sensitivity of the vestibular system to rotations in various directions. Figure 7A shows that, compared with a six-canal system with a uniform sensitivity to rotations in any direction equal to 2, the response of the mouse semicircular canals to a rotation about the nasooccipital axis is increased by 26% to a value of 2.52 because of the orientation of the canals. Figure 7B shows that variability in the radii of the canals increases their response to rotations about the nasooccipital axis by another 13% to a value of 2.78. This suggests that changes in canal orientation may be more significant than changes in canal radius as determinants of overall sensitivity. If head motion does correlate with directional sensitivity of the system, the orientation of the canals may therefore be a better indicator of typical head motions than differences in the sensitivities of individual canals. A small increase in the radius of the anterior canal has been correlated to the adoption of bipedal locomotion in primates (Spoor and Zonneveld 1998; Spoor et al. 1996). This has been explained by assuming that increased movements in the sagittal plane accompanied the development of an upright gait, and that larger, more sensitive anterior canals allowed better measurement of these movements. Notably, these studies found little change in the orientation of the semicircular canals toward the sagittal plane. The importance of orientation of the canals to overall sensitivity of the system, however, suggests that it is unlikely that only the size, and not the orientation, of the canals would change during a major evolutionary step if in fact changes in vestibular sensitivity were functionally correlated to the evolutionary process. The validity of this theoretical point depends on further experiments to determine the relationship, if any, between the anatomy of an animal's semicircular canal system and its typical movements.

If the ratio in humans is similar to that calculated here between canal radii of curvature, the results presented here may allow the behavior of vestibular-nerve afferents to be estimated in human subjects. The radius of curvature of the human horizontal semicircular canal is 3.2 mm (Curthoys and Oman 1987; Igarashi 1967) that, according to the relationship determined earlier, would correspond to a sensitivity of regularly firing afferents of $0.67 \text{ spike s}^{-1}/\text{deg s}^{-1}$ and of high-gain irregular afferents of $1.06 \text{ spikes s}^{-1}/\text{deg s}^{-1}$ at 0.5 Hz. While running, the maximal velocity of human head motions in the horizontal plane may reach 590 deg/s and in the vertical plane 240 deg/s and are likely to occur at frequencies of ≥ 0.5 Hz (Grossman et al. 1988). If the baseline firing rate of human afferents is approximately equivalent to those reported for macaque monkeys of 102 spikes/s for regular afferents and 68 spikes/s for irregular afferents (Haque et al. 2003), the sensitivities calculated here indicate that these fibers may commonly enter inhibitory saturation even during regular daily activities.

Similar calculations indicate that most, if not all, afferents enter inhibitory cutoff during the head-thrust maneuver. In this test, patients with unilateral vestibular loss experience an increased latency and reduced gain of the vestibuloocular reflex during a rapid rotation of the head (~ 250 deg/s) toward the side of a unilateral vestibular loss (Halmagyi and Curthoys 1988). The relationship of a canal's radius to its sensitivity demonstrated here indicates that, at this velocity, the firing rates of both regular and irregular afferents on the functional side are likely to decrease to zero and prevent the reflex from acting accurately. The functional effect of a lower stimulus amplitude, which may silence irregular afferents without completely silencing regular afferents, is unknown but may be useful in determining the function of each group of fibers.

The finding that afferent sensitivity is closely correlated to canal radius of curvature is surprising given the complexity of biophysical models of the canals (Oman et al. 1987; Rabbitt et al. 2004; Squires 2004). These models show that factors such as the radius of the membranous duct, the dimensions of the utricle, the eccentricity of the canal, the density and viscosity of the endolymph, and the mechanical properties of the cupula itself are all likely to have independent effects on the sensitivity of the canal. Effects of these properties, and those relating cupular sensitivity to afferent responses, remain to be quantified further experimentally.

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REFERENCES

- Alonso PD, Milner AC, Ketcham RA, Cookson MJ, Rowe TB. The avian nature of the brain and inner ear of *Archaeopteryx*. *Nature* 430: 666–669, 2004.
- Anastasio TJ, Correia MJ, Perachio AA. Spontaneous and driven responses of semicircular canal primary afferents in the unanesthetized pigeon. *J Neurophysiol* 54: 335–347, 1985.
- Anderson JH, Blanks RHI, Precht W. Response characteristics of semicircular canal and otolith systems in cat. I. Dynamic responses of primary vestibular nerve fibers. *Exp Brain Res* 32: 491–507, 1978.
- Armand M, Minor LB. Relationship between time- and frequency-domain analyses of angular head movements in the squirrel monkey. *J Comput Neurosci* 11: 217–239, 2001.
- Bagnall MW, Stevens RJ, du Lac S. Transgenic mouse lines subdivide medial vestibular nucleus neurons into discrete, neurochemically distinct populations. *J Neurosci* 27: 2318–2330, 2007.
- Baird RA, Desmadryl G, Fernandez C, Goldberg JM. The vestibular nerve of the chinchilla. II. Relation between afferent response properties and peripheral innervation patterns in the semicircular canals. *J Neurophysiol* 60: 182–203, 1988.
- Bärle J, Guldin WO. Unbiased number of vestibular ganglion neurons in the mouse. *Neurosci Lett* 246: 89–92, 1998.
- Blanks RHI, Estes MS, Markham CH. Physiologic characteristics of vestibular first order canal neurons in the cat. II. Response to constant angular acceleration. *J Neurophysiol* 38: 1250–1268, 1975.
- Boyle R, Hightstein SM. Resting discharge and response dynamics of horizontal semicircular canal afferents of the toadfish, *Opsanus tau*. *J Neurosci* 10: 1557–1569, 1990.
- Brichta AM, Acuna DL, Peterson EH. Planar relations of semicircular canals in awake, resting turtles, *Pseudemys scripta*. *Brain Behav Evol* 32: 236–245, 1988.
- Calabrese DR, Hullar TE. Planar relationships of the semicircular canals in two strains of mice. *J Assoc Res Otolaryngol* 7: 151–159, 2006.
- Clarke AH. On the vestibular labyrinth of *Brachiosaurus brancai*. *J Vestib Res* 15: 65–71, 2005.
- Curthoys IS. Postnatal development changes in the response of rat primary horizontal semicircular canal neurons to sinusoidal angular accelerations. *Exp Brain Res* 47: 295–300, 1982a.
- Curthoys IS. The response of primary horizontal semicircular canal neurons in the rat and guinea pig to angular acceleration. *Exp Brain Res* 47: 294–1982, 1982b.
- Curthoys IS, Blanks RHI, Markham CH. Semicircular canal radii of curvature (R) in cat, guinea pig and man. *J Morphol* 151: 218–225, 1977a.
- Curthoys IS, Markham CH, Curthoys EJ. Semicircular duct and ampulla dimensions in cat, guinea pig and man. *J Morphol* 151: 17–34, 1977b.
- Curthoys IS, Oman CM. Dimensions of the horizontal semicircular duct, ampulla and utricle in rat and guinea pig. *Acta Otolaryngol* 101: 1–10, 1986.
- Curthoys IS, Oman CM. Dimensions of the horizontal semicircular duct, ampulla and utricle in the human. *Acta Otolaryngol* 103: 254–261, 1987.
- Day BL, Fitzpatrick RC. Virtual head rotation reveals a process of route reconstruction from human vestibular signals. *J Physiol* 567: 591–597, 2005.
- Desai SS, Ali H, Lysakowski A. Comparative morphology of rodent vestibular periphery. II. Cristae ampullares. *J Neurophysiol* 93: 267–280, 2005.
- Dickman JD. Spatial orientation of semicircular canals and afferent sensitivity vectors in pigeons. *Exp Brain Res* 111: 8–20, 1996.
- Dickman JD, Correia MJ. Responses of pigeon horizontal semicircular canal afferent fibers. I. Step, trapezoid, and low-frequency sinusoid mechanical and rotational stimulation. *J Neurophysiol* 62: 1090–1101, 1989a.
- Dickman JD, Correia MJ. Responses of pigeon horizontal semicircular canal afferent fibers. II. High-frequency mechanical stimulation. *J Neurophysiol* 62: 1102–1112, 1989b.
- Drager UC, Hubel DH. Responses to visual stimulation and relationship between visual, auditory, and somatosensory inputs in mouse superior colliculus. *J Neurophysiol* 38: 690–713, 1975.
- Fernandez C, Goldberg JM. Physiology of peripheral neurons innervating semicircular canals of the squirrel monkey. II. Response to sinusoidal stimulation and dynamics of peripheral vestibular system. *J Neurophysiol* 34: 661–675, 1971.
- Fernandez C, Goldberg JM. 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, 1976a.
- Fernandez C, Goldberg JM. Physiology of peripheral neurons innervating otolith organs of the squirrel monkey. III. Response dynamics. *J Neurophysiol* 39: 996–1008, 1976b.
- Goldberg JM, Fernandez C. 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, 1971.
- Goldberg JM, Fernandez C, Smith CE. Responses of vestibular-nerve afferents in the squirrel monkey to externally applied galvanic currents. *Brain Res* 252: 156–160, 1982.
- Goldberg JM, Smith CE, Fernandez C. Relation between discharge regularity and responses to externally applied galvanic currents in vestibular nerve afferents of the squirrel monkey. *J Neurophysiol* 51: 1236–1256, 1984.

- Graf W, Vidal P-P.** Semicircular canal size and upright stance are not interrelated. *J Hum Evol* 30: 175–181, 1996.
- Gray AA.** *The Labyrinth of Animals*. London: Churchill, 1907–1908.
- Green DM, Swets JA.** *Signal Detection Theory and Psychophysics*. New York: Wiley, 1966.
- Grossman GE, Leigh RJ, Abel LA, Lanska DJ, Thurston SE.** Frequency and velocity of rotational head perturbations during locomotion. *Exp Brain Res* 70: 470–476, 1988.
- Hadziselimovic H, Savkovic L.** Appearance of semicircular canals in birds in relation to mode of life. *Acta Anat (Basel)* 57: 306–315, 1964.
- Halmagyi GM, Curthoys IS.** A clinical sign of canal paresis. *Arch Neurol* 45: 737–739, 1988.
- Han G, Lasker D, Park H, Minor L.** Comparison of physiological responses of vestibular-nerve afferents innervating the semicircular canals in the juvenile and adult C5BL/6 mouse. *Assoc Res Otolaryngol Abstr* 1036, 2006.
- Haque A, Angelaki DE, Dickman JD.** Spatial tuning and dynamics of vestibular semicircular canal afferents in rhesus monkeys. *Exp Brain Res* 155: 81–90, 2003.
- Hartmann R, Klinke R.** Discharge properties of afferent fibres of the goldfish semicircular canal with high frequency stimulation. *Pfluegers Arch* 388: 111–121, 1980.
- Hoebek FE, Stahl JS, van Alphen AM, Schonewille M, Luo C, Rutteman M, van den Maagdenberg AM, Molenaar PC, Goossens HH, Frens MA, De Zeeuw CI.** Increased noise level of Purkinje cell activities minimizes impact of their modulation during sensorimotor control. *Neuron* 45: 953–965, 2005.
- Howland H, Masci J.** The functional allometry of semicircular canals, fins, and body dimensions in the juvenile centrarchid fish, *Lepomis gibbosus* (L.). *J Embryol Exp Morphol* 29: 721–743, 1973a.
- Howland HC, Masci J.** The phylogenetic allometry of the semicircular canals of small fishes. *Z Morphol Tiere* 75: 283–296, 1973b.
- Hullar TE, Della Santina CC, Hirvonen TP, Lasker DM, Carey JP, Minor LB.** Responses of irregularly discharging chinchilla semicircular canal vestibular-nerve afferents during high-frequency head rotations. *J Neurophysiol* 93: 2777–2786, 2005.
- Hullar TE, Williams CD.** Geometry of the semicircular canals of the chinchilla (*Chinchilla laniger*). *Hear Res* 213: 17–24, 2006.
- Igarashi M.** Dimensional study of the vestibular apparatus. *Laryngoscope* 77: 1806–1817, 1967.
- Jones GM, Spells KE.** A theoretical and comparative study of the functional dependence of the semicircular canal upon its physical dimensions. *Proc R Soc Lond B Biol Sci* 157: 403–419, 1963.
- Jones SM, Johnson KR, Yu H, Erway LC, Alagramam KN, Pollak N, Jones TA.** A quantitative survey of gravity receptor function in mutant mouse strains. *J Assoc Res Otolaryngol* 6: 1–14, 2005.
- Jones T, Jones S, Hoffman L.** Spontaneous discharge patterns of vestibular neurons in the otoconia-deficient mouse. *Assoc Res Otolaryngol Abstr* 954, 2007.
- Ledoux A.** L'adaptation du système vestibulaire périphérique. *Acta Otolaryngol (Stockh)* 53: 307–316, 1961.
- Lee H-Y, Camp AJ, Callister RJ, Brichta AM.** Vestibular primary afferent activity in an in vitro preparation of the mouse inner ear. *J Neurosci Methods* 145: 73–87, 2005.
- Lewis ER, Leverenz EL, Bialek WS.** *The Vertebrate Inner Ear*. Boca Raton, FL: CRC Press, 1985.
- Lowenstein O.** The effect of galvanic polarization on the impulse discharge from sense endings on the isolated labyrinth of the thorn back ray (*Raja clavata*). *J Physiol* 127: 104–117, 1955.
- Lysakowski A, Minor LB, Fernandez C, Goldberg JM.** Physiological identification of morphologically distinct afferent classes innervating the cristae ampullares of the squirrel monkey. *J Neurophysiol* 73: 1270–1281, 1995.
- Matano S, Kubo T, Niemitz C, Guenther M.** Semicircular canal organ in three primate species and behavioral correlations. *Fortschr Zool* 30: 677–680, 1985.
- Mazza D, Winterson B.** Semicircular canal orientation in the adult resting rabbit. *Acta Otolaryngol* 98: 472–480, 1984.
- Oman C.** The influence of duct and utricular morphology on semicircular canal response. In: *Vestibular System: Function and Morphology*, edited by Gualtierotti T. New York: Springer, 1981, p. 251–274.
- Oman CM, Marcus EN, Curthoys IS.** The influence of semicircular canal morphology on endolymph flow dynamics. *Acta Otolaryngol (Stockh)* 103: 1–13, 1987.
- Precht W, Llinás R, Clarke M.** Physiological responses of frog vestibular fibers to horizontal angular rotation. *Exp Brain Res* 13: 378–407, 1971.
- Puyal J, Martineau M, Mothet JP, Nicolas MT, Raymond J.** Changes in D-serine levels and localization during postnatal development of the rat vestibular nuclei. *J Comp Neurol* 497: 610–621, 2006.
- Rabbitt RD.** Directional coding of three-dimensional movements by the vestibular semicircular canals. *Biol Cybern* 80: 417–431, 1999.
- Rabbitt RD, Damiano ER, Grant JW.** Biomechanics of the semicircular canals and otolith organs. In: *The Vestibular System*, edited by Highstein S, Fay RR, Popper AN. New York: Springer, 2004, p. 153–201.
- Ramachandran R, Lisberger SG.** Normal performance and expression of learning in the vestibulo-ocular reflex (VOR) at high frequencies. *J Neurophysiol* 93: 2028–2038, 2005.
- Ramprasad F, Landolt JP, Money KE, Laufer J.** Dimensional analysis and dynamic response characterization of mammalian peripheral vestibular structures. *Am J Anat* 169: 295–313, 1984.
- Rogers SW.** Exploring dinosaur neuropaleobiology: viewpoint computed tomography scanning and analysis of an *Allosaurus fragilis* endocast. *Neuron* 21: 673–679, 1998.
- Rogers SW.** Reconstructing the behaviors of extinct species: an excursion into comparative paleoneurology. *Am J Med Genet A* 134: 349–356, 2005.
- Sadeghi SG, Chacron MJ, Taylor MC, Cullen KE.** Neural variability, detection thresholds, and information transmission in the vestibular system. *J Neurosci* 27: 771–781, 2007.
- Schneider LW, Anderson DJ.** Transfer characteristics of first and second order lateral canal vestibular neurons in gerbil. *Brain Res* 70: 1–14, 1976.
- Segal BN, Outerbridge JS.** Vestibular (semicircular canal) primary neurons in bullfrog: nonlinearity of individual and population response to rotation. *J Neurophysiol* 47: 545–562, 1982.
- Spoor F, Bajpai S, Hussain ST, Kumar K, Thewissen JG.** Vestibular evidence for the evolution of aquatic behaviour in early cetaceans. *Nature* 417: 163–166, 2002.
- Spoor F, Garland T Jr, Krovitz G, Ryan TM, Silcox MT, Walker A.** The primate semicircular canal system and locomotion. *Proc Natl Acad Sci USA* 104: 10808–10812, 2007.
- Spoor F, Wood B, Zonneveld F.** Implications of early hominid labyrinthine morphology for evolution of human bipedal locomotion. *Nature* 369: 645–648, 1994.
- Spoor F, Wood B, Zonneveld F.** Evidence for a link between human semicircular canal size and bipedal behaviour. *J Hum Evol* 30: 183–187, 1996.
- Spoor F, Zonneveld F.** Comparative review of the human bony labyrinth. *Am J Phys Anthropol Suppl* 27: 211–251, 1998.
- Squires TM.** Optimizing the vertebrate vestibular semicircular canal: could we balance any better? *Phys Rev Lett* 93: 198106, 2004.
- ten Kate JH, van Barneveld HH, Kuiper JW.** The dimensions and sensitivities of semicircular canals. *J Exp Biol* 53: 501–514, 1970.
- Tomko DL, Peterka RJ, Schor RH, O'Leary DP.** Response dynamics of horizontal canal afferents in barbiturate-anesthetized cats. *J Neurophysiol* 45: 376–396, 1981.