The Planes of the Utricular and Saccular Maculae of the Guinea Pig

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Abstract: To establish a link between otolith anatomy and function it is necessary to know the regions of the utricular and saccular maculae, which are stimulated by any arbitrary linear acceleration stimulus. That requires accurate information about the location and orientation of the spatially extended maculae in head-fixed coordinates and referred to head-fixed landmarks (such as Reid's line). New data showing the location of the otolithic maculae in the guinea pig with respect to head-fixed stereotaxic coordinates are presented. Guinea pigs were perfused with Karnovsky's fixative and the maculae were exposed while the head was held in a guinea pig stereotaxic device. An electrolytically sharpened fine wire held in a calibrated micromanipulator was touched to points all over the surface of each macula under visual observation with the aid of a high-power operating microscope. The *x, y, z* coordinates of these points were plotted using a three-dimensional plotting program. Both maculae have pronounced curvature so that dorsoventral shear forces will stimulate regions of both the utricular and saccular maculae.

INTRODUCTION

In the literature there are excellent photographs and illustrations of the gross anatomical characteristics of the utricular and saccular maculae (e.g., reference 1), but there is little information about the relationship of the otolith receptor surface to skull landmarks, which is the information needed for interpreting behavioral studies of otolith function. Recent investigators have approximated the otolithic maculae as being flat surfaces, perpendicular to each other and roughly in the cardinal planes of the head, and this simplification is frequently used to model the response of receptors on these complex structures (e.g., references 2 and 3). However even early anatomical investigations showed that such a simplification was not correct.[4-6] The significant error of the more recent approximations is shown by the fact that the most common schematic representation of the human otolithic maculae[3] depicts the long axis of the human saccular macula to be oriented vertically with respect to head, whereas in fact it is oriented almost horizontally with respect to head (i.e., almost in the human horizontal stereotaxic plane).[4,5]

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One reason the orientation of these structures with respect to head is not readily available is that in order to carry out the difficult dissection needed to reveal the maculae, the whole temporal bone is usually removed from the skull, thus losing the very skull landmarks that are necessary for orienting the head in experimental studies. The surgical exposure is difficult because the maculae are delicate membranous structures located in the dense petrous temporal bone. In the case of the utricular macula most receptors are located on a thin membrane that is stretched across the fluid-filled vestibule. The saccular macula adheres to the bony inner wall of the saccule of the inner ear.

There have been attempts to obtain the position of these structures by reconstructions, but these attempts have not been successful as shown by the very large discrepancies between different investigators.[5–7] There are very substantial disagreements in the literature about the orientation and configuration of the maculae; for example de Burlet's data[6] is very different from that of Corvera *et al.*[7] Spoendlin has published quasi-quantitative representations of the human utricular and saccular macula topography[8] that appear to be visual estimates of the contours of the structures rather than being obtained by quantitative measurement.

Because of the importance of the anatomical data for understanding otolithic function we sought to obtain quantitative specification of the location of the otolithic maculae in the guinea pig with respect to head landmarks. The guinea pig was studied because the gross and fine anatomy of its vestibular labyrinth is better understood than any other species. Multiple specimens were studied to demonstrate the extent of variation between animals.

METHODS

Guinea pigs were deeply anesthetized with nembutal and perfused with 400 mL of saline containing heparin followed by 500 mL of phosphate-buffered Karnovsky's fixative (4% paraformaldehyde and 5% glutaraldehyde).[9] The head was mounted in a guinea pig stereotaxic apparatus (Kopf) and the inner ear of one labyrinth was dissected to show the maculae. The otoconia and otoconial membrane were very gently removed by a fine jet of saline. Saline was removed from the surface of the utricular macula so that errors due to refraction would not confound the measurements. The macula surface was not allowed to dry—a thin layer of saline always bathed the macula. Methylene blue was used to show the macula surface, staining individual receptor hair cells that could be just discerned at the maximum power of the operating microscope used here. In freshly fixed specimens the hair cells at the striola (the type I hair cells)[1,10,11] stained more darkly than the surrounding hair cells, allowing measurement of the spatial location of the striola.

The x, y, and z coordinates of up to 300 data points per macula were obtained from the surface of the utricular and saccular macula using a Kopf precision micromanipulator that was calibrated in 50-micron steps and was specially modified to be free of backlash. Under very high power, the tip of a fine probe (electrolytically sharpened 40-micron stainless-steel wire) was touched to points on the surface of the macula. In some animals it was possible to obtain data from the utricular macula, the saccular macula, and the horizontal semicircular canal. The latter served as a reference since the orientation of the plane of the horizontal semicircular canal is very well established,[12] and so the plane of the horizontal canal may possibly be used to relate the macula orientation to head position.

The data points were entered into a statistical program, S-Plus,[13] running under Unix, and a program called xgobi[14] allowed the data points to be rotated *en bloc* to show more clearly the projection of the maculae into the major skull planes and to demonstrate the curvature of the macula surfaces.

RESULTS

FIGURES 1 to 4 show views of the guinea pig otolithic maculae in stereotaxic coordinates. The data from a number of animals are included (FIG. 3) to demonstrate the similarity of these data across different animals. FIGURES 1 to 3 show that the utricular macula is a curved structure, with the anterior portion being upturned like the front of a toboggan. To the extent that it is meaningful to assign a "plane" to such a curved structure, then one could describe "the plane" of the utricular macula as being pitched nose down by about 20 deg with respect to the plane of the horizontal semicircular canal. Such a coarse description does not do justice to the curved surface topography of the utricular macula (see FIGS. 1–3).

The pitch angle of the horizontal canal appears to be very large—around 45 deg (FIG. 1), but this is merely a consequence of the rather unusual definition of the stereotaxic horizontal plane in the guinea pig.[12] In fact the natural head position of the guinea pig is pitched down by about 30 deg relative to the stereotaxic horizontal so the horizontal canal is held close to being earth horizontal.[12]

The data from all the specimens are similar (FIG. 3) and that figure shows that there are systematic curves in the planes of both the utricular and saccular maculae. The saccular macula is attached tightly to the curving bony wall of the vestibule and has very close connections to Scarpa's ganglion through the thin cribriform bony plate on which the saccular macula is located. The saccular macula exhibits a complex twisted surface with clear

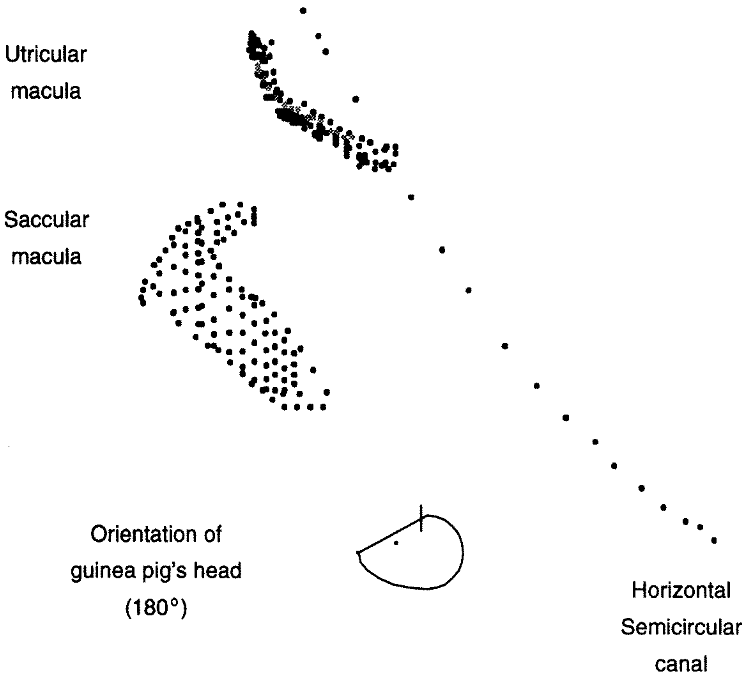

FIGURE 1. View from a directly side-on point of view of points from the surface of the guinea pig utricular macula, saccular macula, and horizontal canal from a left labyrinth in stereotaxic space. The edges of the figure correspond to the horizontal (X) and dorsoventral (Z) guinea pig stereotaxic axes.

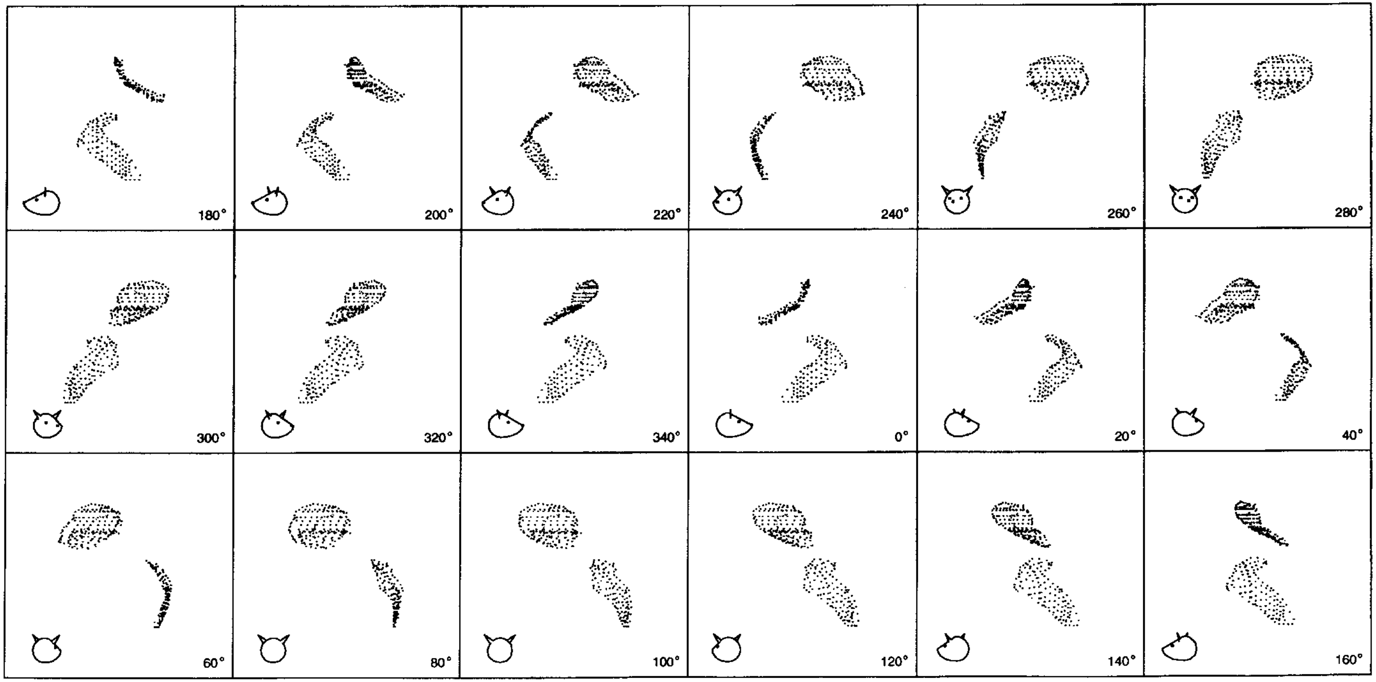

FIGURE 2. Single frames—"still photos"—from the rotation sequence of the guinea pig utricular and saccular macula rotated in 20-degree steps around a *Z*-axis. The view is from a directly lateral point of view, and the small schematic guinea pig head at the bottom of each inset shows head orientation.

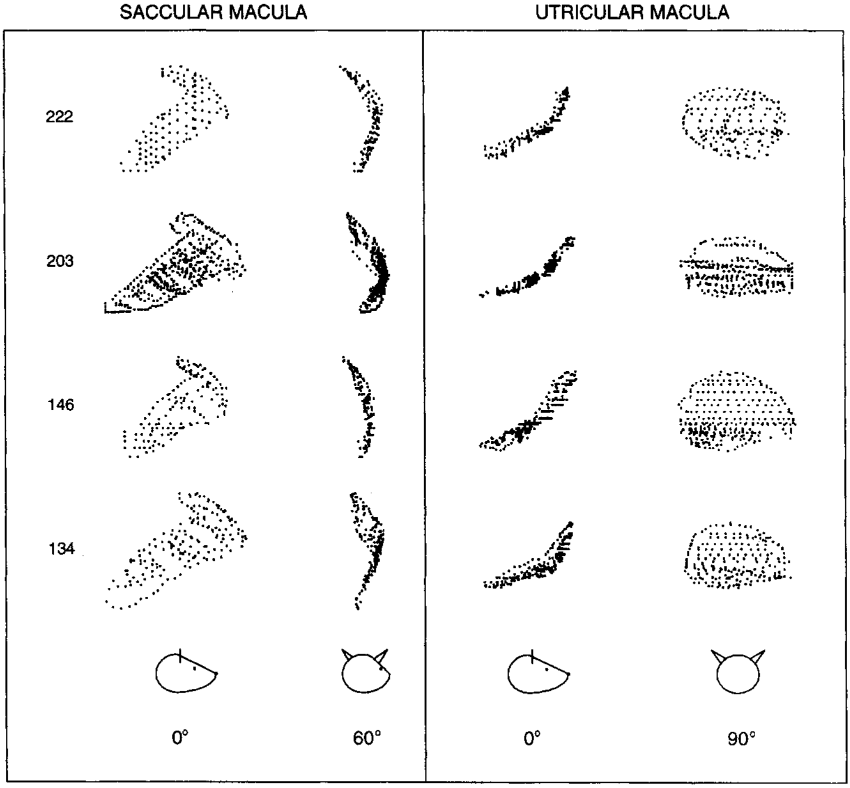

FIGURE 3. Views of the saccular and utricular maculae from 4 animals to show the similarity between animals. The maculae have been rotated to give an "edge-on" view that shows the curvature so clearly. The maculae have been arbitrarily shifted in the dorsoventral dimension to allow comparison between animals.

projections into the frontal plane as well as its predominant sagittal projection. The saccular macula is not simply a sheet of receptors in the sagittal plane but has a considerable projection into the frontal plane (FIG. 4) so that even interaural linear accelerations will stimulate the receptors on the guinea pig saccular maculae. This is clearly seen in direct views looking down on the saccular maculae that show the curved projection of the macula clearly (FIG. 4).

DISCUSSION

Contrary to the simplifications imposed on the maculae by artists and illustrators, it is clear from our data that curvature and asymmetry are more prominent features of the maculae than the flatness and symmetry that have characterized artistic representations. The results confirm other published data concerning the curvature of the utricular macula: Lindeman's photos show this arrangement in the guinea pig,[1] as does Curthoys.[15]

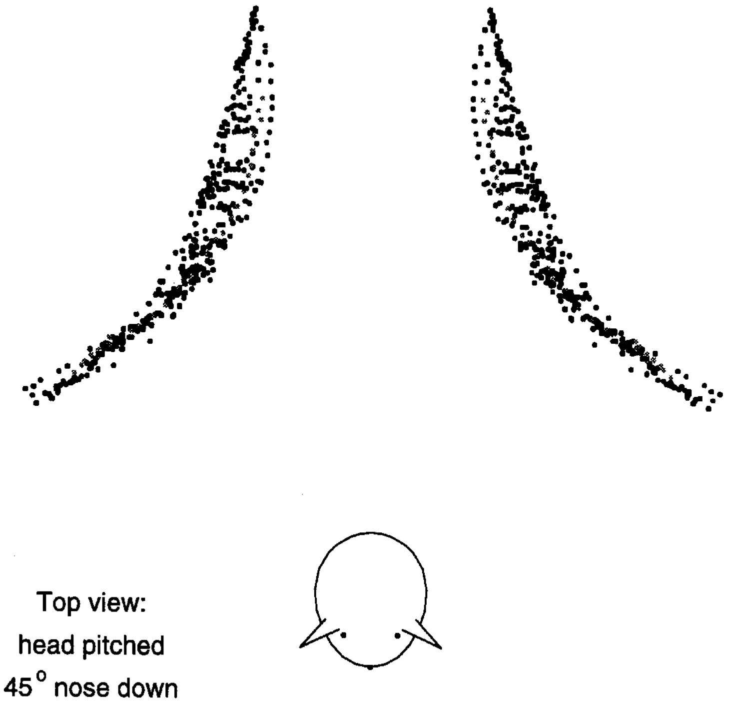

FIGURE 4. To show the curvature of the saccular macula in the guinea pig. This is a directly dorsal view of the data from a single animal with the head pitched nose down by 45 deg (as shown by the inset of the schematic head). The data from a single labyrinth have been reflected around the sagittal plane to represent the maculae in both labyrinths.

Appropriately sectioned temporal bones of the chinchilla also show this curved upturn of the anterior portion of the utricular macula.[10]

The data points were obtained from maculae in animals that had been fixed by perfusion. Could fixation have substantially altered the very characteristics under investigation? Could it have deformed the orientation of the maculae so that the data are invalid? Could the prominent curvature shown in these results be caused by shrinkage or fixation artifact? There are two strong reasons to doubt that fixation affected these results. First, the fixative used (phosphate-buffered Karnovsky's) was specifically chosen because electronmicroscopic evidence has shown that the particular concentration used here caused minimal shrinkage of labyrinth membranes at the electronmicroscopic level.[9] Second, inspection of the maculae in living anesthetized animals confirms the orientation and curvature shown by the quantitative measures. In some anesthetized guinea pigs the inner ear was opened and the utricular and saccular macula were each visualized directly through the endolymph, and they showed similar patterns of curvature to the data presented here.

The following elaborates how this spatial configuration data are important for understanding otolith function. Each macula is covered by receptor hair cells, each with its own preferred direction (polarization). The receptor hair cells are systematically arranged around the surface of the maculae with their polarization directions being exactly opposite on either side of the striola.[1] To relate these structures to function it is necessary to consider the individual hair-cell responses and use that information to determine which hair cells will be stimulated by any linear force stimulus to result in the discharge pattern across fibers in the vestibular nerve.

Physiological studies[15] have shown that for an individual hair cell, excitation is maximal when the direction of linear force corresponds to the polarization direction through the stereocilia toward the kinocilium. As the angle between the direction of this imposed linear force and the direction of the morphological polarization changes, so the magnitude of the excitatory neural response declines. Goldberg *et al.* have shown that in the extrastriola region a number of otolithic receptor cells typically synapse on an individual primary afferent[11] and probably as a result, the breadth of tuning of the preferred direction of primary otolithic afferents may be greater than 180 deg.[16] Shear is the effective stimulus for the otoliths—otolithic receptors are not sensitive to compression forces.[17]

Exact information on the orientation and position of the human macular surfaces is not yet available, so linear force stimuli should be specified by their components in the interaural and dorsoventral axes, with the presumption that the human utricular macula will be mainly stimulated by the interaural shear and the saccular macula predominantly by the dorsoventral shear. On the basis of our anatomical evidence from the guinea pig and the available anatomical data in the human,[5–7] this is only a very approximate assignment, and there are regions of the utricular macula that will be stimulated by dorsoventral shear forces.

ACKNOWLEDGEMENTS

We gratefully acknowledge the support of NHMRC of Australia and the partial support from Research Grant 5 R01 DC 02372-02 from the National Institute on Deafness and Other Communication Disorders, National Institutes of Health. The assistance of Warren Davies is gratefully acknowledged.

REFERENCES

1. LINDEMAN, H. H. 1969. Studies on the morphology of the sensory regions of the vestibular apparatus. Adv. Anat. Embryol. Cell Biol. **42**: 1–113.
2. MALCOLM, R. & G. MELVILL JONES. 1974. Erroneous perception of vertical motion by humans seated in the upright position. Acta Otolaryngol. **77**: 274–283.
3. HOWARD, I. P. 1982. Human Visual Orientation. Wiley. New York.
4. ROSENHALL, U. 1972. Vestibular macular mapping in man. Ann. Otol. Rhinol. Laryngol. **81**: 339–351.
5. QUIX, F. H. 1925. The function of the vestibular organ and the clinical examination of the otolithic apparatus. J. Laryngol. Otol. **50**: 425–443.
6. DE BURLET, H. M. 1930. Die Stellung der Maculae acusticae im Schädel des Menschen und einiger Säugetiere. Morphol. Jahrb. **64**: 377–393.
7. CORVERA, J., C. S. HALLPIKE & E. H. J. SCHUSTER. 1958. A new method for the anatomical reconstruction of the human macular planes. Acta Otolaryngol. **49**: 4–16.
8. SPOENDLIN, H. 1965. Strukturelle Eigenschaften der vestibularen Rezeptoren. Schweiz. Arch. Neurol. Neurochir. Psychiatry **96**: 219–230.
9. ANNIKO, M. & P.-G. LUNDQUIST. 1977. The influence of different fixatives and osmolarity on the ultrastructure of the cochlear neuroepithelium. Arch. Otolaryngol. **218**: 67–73.
10. FERNANDEZ, C., R. A. BAIRD & J. M. GOLDBERG. 1988. The vestibular nerve of the Chinchilla. I. Peripheral innervation patterns in the horizontal and superior semicircular canals. J. Neurophysiol. **60**: 167–203.
11. GOLDBERG, J. M., G. DESMADRYL, R. A. BAIRD & C. FERNANDEZ. 1990. The vestibular nerve of the chinchilla. V. Relation between afferent discharge properties and peripheral innervation patterns in the utricular macula. J. Neurophysiol. **63**: 791–804.

12. CURTHOYS, I. S., E. J. CURTHOYS, R. H. I. BLANKS & C. H. MARKHAM. 1975. The orientation of the semicircular canals in the guinea pig. Acta Otolaryngol. **80**: 197–205.
13. BECKER, R. A., J. M. CHAMBERS & A. R. WILKES. 1988. The new S language: a programming environment for data analysis and graphics. Brooks/Cole. Pacific Grove, Calif.
14. SWAYNE, D. F., D. COOK & A. BUJA. 1998. Xgobi: interactive dynamic data visualization in the X-window system. J. Comput. Graph. Stat. **7**: 113–130.
15. CURTHOYS, I. S. 1987. Eye movements produced by utricular and saccular stimulation. Aviat. Space Environ. Med. (Suppl.). **58**: A192–A197.
16. FERNANDEZ, C. & J. M. GOLDBERG. 1976. 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.
17. SHOTWELL, S. L., R. JACOBS & A. J. HUDSPETH. 1981. Directional sensitivity of individual vertebrate hair cells to controlled deflection of their hair bundles. Ann. N.Y. Acad. Sci. **374**: 1–10.