SYNAPTIC RIBBON PLASTICITY IN UTRICULAR AND SACULAR MACULAE: NEW CLUES TO FUNCTIONS?

*Muriel D. Ross and **Joseph Varelas, *The University of New Mexico Health Sciences Center, Albuquerque, N.M. ** Lockheed-Martin and NASA Ames Center for Bioinformatics, Moffett Field, CA

INTRODUCTION

Research into the effects of weightlessness on rat vestibular maculae has consistently shown that ribbon synapses in hair cells of the utricular maculae exhibit statistically significant changes in number, kind, and distribution when rats are exposed to space flight (Ross, 1993, 1994, 2000). Synaptic plasticity was most evident when all type II hair cells of these maculae were considered and was confined to type II cells in just the complete hair cells. Analysis of the co-variance of the multiple variables (number, rod or sphere, pairs and groups) by the MANOVA feature of SuperANOVA software demonstrated further that day and weightlessness both had statistically significant effects on type II hair cells (Ross, 2000). These results were obtained from the posterior portion of the utricular macula and did not include the striola. For Neurolab, the striola and parasotriolar area internal to the striola (pars interna) were studied. The Neurolab striolar data in general support the findings in hair cells of the utricular maculae, but effects in the saccular maculae differed. In saccular maculae, ribbon synapses in type I cells fluctuated while synapses in type II hair cells remained relatively stable throughout the flight and up to postflight day 2. Only differences between flight days 2 and 14 in ribbon synapses of type I hair cells were statistically significant. It cannot be argued that the lack of significant differences in type II hair cells was due to inner ears utilized since utricular and saccular samples were matched for the same rats in two cases.

The results in the saccular maculae raise the interesting question whether the anatomical findings signify very different functions for the two maculae, even though both are subject to stimulation by gravitoinertial forces. This report provides the data for the utricular and saccular maculae of the Neurolab experiment and compares the findings with those previously obtained on SLS-1 and SLS-2. It also discusses the findings in light of previous physiological data that indicated different functions for the two maculae (Fernandez et al., 1976a,b).

CURRENT STATUS OF RESEARCH

Methods
The Fisher strain of rats was used for this experiment. Rats were euthanized by decapitation on the ground on day two of flight (Basal), in flight on flight days 2 (FD2) and 14 (FD14), and postflight on days 2 (PF2) and 14 (PF14). Labyrinths were quickly removed from dissected temporal bones, fixed by immersion, and prepared for electron microscopy as described previously (Ross, 2000). Unfortunately, only one of the four FD2 utricular maculae made available to us proved useful for electron microscopic study. Study of the utricular maculae was limited, therefore, to one Basal, one FD2, and one FD 14 sample. Attention then turned to the saccular macula. Study of the saccular maculae is incomplete at this time. Two maculae from Basal, FD2, FD14 and PF2 will be analyzed. This Abstract is based on two samples from FD2 and FD14 and one from a Basal and another from a PF2 rat which have been studied thus far in 50 serial sections cut at 150 nm. The striolar area is identified in the rat by the presence of myelination to the calyx in some afferents. In the case of a FD2 utricular and a FD2 saccular sample, an additional set of 50 sections that crossed from the striola into pars interna was studied, to learn whether differences in synaptic counts and other properties would be evident. Analysis of statistical significance was accomplished using the ANOVA features of SuperANOVA software. All procedures used in this experiment were reviewed by the Animal Use Committee established at NASA Ames Research Center and are in compliance with the Guide for the Use of Laboratory Animals and the Animal Welfare Act.

Results
Utricular macula: Mean values for number of ribbon synapses in type I cells of the utricular striolar area were as follows: Basal; 2.034; FD2, 3.512; and FD 14, 2.400. The increment in synaptic mean value in ribbon synapses from the Basal to FD2 was significant (p< 0.0044) as were differences in values for sphere-like ribbons (p< 0.0002). FD2 mean values for total synapses (p< 0.0408) and spheres (p< 0.0379) differed from FD14 values. For the parasotriolar...
area, pars interna, the mean value of synaptic ribbons was 3.543, differing significantly from the basal (p< 0.0013) as did also sphere-like ribbons (p< 0.0014). Set two of FD2 also differed significantly from FD14 in total synapses (p< 0.0209), and groups (p< 0.0447).

For type II hair cells, the Basal mean value for ribbon synapses at the striola was 4.744. For FD2, the mean value was 9.000 and for FD14 it was 6.884. The differences between synaptic means between the Basal and FD2 were significant for total synapses (p< 0.0001), spheres (p< 0.0034), rods (p< 0.0016) and pairs (p< 0.0003). FD14 differed from the Basal in total synapses (p< 0.0306), spheres (p< 0.0093), and pairs (p< 0.0074). For set two FD2, the mean value of total synapses was 7.929. This value differed from the Basal (p< 0.0023) as did sphere-like ribbons (p< 0.0008) and pairs (p< 0.0012). The two series from FD2 were essentially similar.

Saccular macula: In type I cells of the saccular macula, mean values were: Basal, 3.545; FD2, 2.646; FD14, 3.788; PF2, 3.250. FD2 differed significantly from FD14 in total synapses (p< 0.0277) and in spheres (p< 0.0074). In type II cells, the mean values of synaptic number were as follows: Basal: 6.939; FD2, 6.317; FD14, 6.382, PF2, 6.480. None of these or other mean values (pairs, etc.) differed significantly.

Conclusions

Findings in the striolar area of the utricle were essentially similar to those obtained from the posterior part of the macula on SLS-1 and SLS-2. That is, synapses increased in the hair cells, but changes were greater overall in the case of type II cells. Nevertheless, the results in the striolar area of both the utricle and the saccule differ from those obtained previously; i.e., in type II hair cells, synapses had doubled to 11.4±7.2 on day 13 of a 14 day flight (Ross, 2000). This is not surprising since the striola differs morphologically from other portions of the macula in organization of hair cells, afferents and processes. In the saccular samples of this experiment, in contrast to the utricular, ribbon synapses in type I cells had declined by FD2 but fluctuated on FD14 and PF2. In type II hair cells, a slight decline in synaptic mean occurred by FD2 and was maintained through PF2. None of the changes in type II cells was statistically significant. The fact that in two cases saccular and utricular maculae came from the same inner ears precludes a difference in findings due to sampling. The conclusion thus far is that saccular and utricular maculae differ in responses to weightlessness, with utricular type II hair cells showing the greater plasticity in weightlessness. This result may be related to functional differences described by Fernandez and Goldberg (1976a,b). Their experimental findings indicated that utricular afferents were most responsive to static tilt in the X direction (left-right) while saccular afferents were primarily sensitive to Z direction (vertical or dorsoventral) tilt. Saccular maculae had a lower sensitivity to static tilt. Present and previous findings (Ross, 2000) additionally show that weightlessness has differing effects on striolar and posterior portions of the utricular macula. This is likely due to varying morphological features of receptive fields. The resultant of gravitoinertial vectors acting on type I and type II hair cells of a receptive field, plus parallel processing by many afferents, determines the message delivered to central sites. Type II cells may be particularly sensitive to static stimuli and gravity while type I cells may be more sensitive to phasic stimuli and translational accelerations (Ross, 2000). According to this hypothesis, the maculae use two kinds of receptors as comparators to determine the afferent’s signal, with morphological variations from location to location (including macular geometry and otoconial loading) contributing to the outcome.

FUTURE PLANS:

Work is to be completed on postflight (PF2) and Basal samples. All data will be subjected to analysis of variance.

REFERENCES

INDEX TERMS

Ribbon synapses, vestibular system, hair cells, saccule, utricle, macula, synaptic plasticity, Neurolab