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AFM and BSEM: Novel approaches to the Basilar Membrane

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For over a half century, researchers have probed cochlear biophysics with increasingly sophisticated technologies. Recently, we brought two new approaches, atomic force microscopy (AFM) and back scatter electron microscopy (BSEM), to bear on the question of how basilar membrane structural variations affect mechanical responses. Hemi-cochleae and basilar membrane segments were obtained from rat, guinea pig, and gerbil ears by temporal bone microdissection. Hemi-cochleae in PBS or formalin were imaged in a hydrated, uncoated state (VP wet mode) in Petri dishes mounted on boutons using an Hitachi S3400 N-1 SEM. Membrane segments were excised, trimmed of laminae and ligament remnants, and mounted on polylysine/albumin-coated glass for AFM imaging and force measurements. Both BSEM and AFM data indicate two distinct regions consistent with softer ground substance separating relatively stiff,ordered fiber bundles oriented radially in pectinate regions. Fibers and bundles ranged 0.3 to 1 micron in diameter with elasticity values, based on a Hertzian contact model, in the hundreds of kPa range, consistent with mixed elastin and collagen. Greatest variations occurred in matrix spacing, suggesting differences are attributable largely to packing density. Middle turn ground substance spacing averaged 1.5 microns, consistent with previously reported values (Naidu and Mountain, 2007; Fung, 1993).