Abstract

The development, maturation, and viability of inner ear neurosensory cells depend on the spatiotemporal expression of multiple transcriptional factors. Based on three mouse models [Tg(*Pax2-Isl1*)], *Sox2CKO*, and *Neurod1CKO,* this thesis investigates the function of three transcriptional factors ISL1, SOX2 and NEUROD1 in the neurosensory development of the inner ear.

The mouse mutant [Tg(*Pax2-Isl1*)] carries transgenic sequence containing *Isl1* gene under *Pax2* regulatory sequence in its genome. ISL1 ectopic expression driven by *Pax2* regulatory sequence resulted in the enlarged cochleovestibular ganglion and accelerated neurite extension in [Tg(*Pax2-Isl1*)] embryos. In adult mutants, we detected an early onset of age-related hearing loss correlating with the worsening function of outer hair cells. These changes were associated with the loss of medial olivocochlear efferent neuron fibers innervating outer hair cells. For the first time, we showed that the age-related hearing loss (presbyacusis) might be caused by efferent innervation defects besides hair cell loss and spiral ganglion degeneration. In addition to presbyacusis, [Tg(*Pax2-Isl1*)] mice suffered from hyperactivity that was diminished by the administration of picrotoxin – channel blocker for GABA receptor chloride channels. This indicates that ISL1 overexpression impacts GABAergic neuron function. Besides changes in GABAergic signalization, [Tg(*Pax2-Isl1*)] mice had the reduced size of inferior colliculus and the aberrant morphology of the cerebellum, corresponding to the abnormal vestibular phenotype of these mutant mice.

For analysis of SOX2 function during inner ear development, we generated a new mouse model with conditional deletion of *Sox2* in ISL1 positive cells using the Cre-loxP system (*Sox2CKO*). In *Sox2CKO*, a limited number of hair cells of neurosensory origin differentiated in the utricle, saccule, and cochlear base, while all cristae and the cochlear apex were devoid of any hair cells derived from sensory precursors. Early developed neurons innervating the vestibular end organs and cochlear base differentiated in *Sox2CKO*. However, all these early differentiated neurons died later by apoptosis due to a lack of neurotrophins that are produced by hair cells. Late forming apical neurons in the cochlea did not differentiate at all in *Sox2CKO*. Our results demonstrate that transcriptional factor SOX2 is necessary for the differentiation of late forming neurons in the

cochlear apex, for the maintenance of vestibular and basal spiral ganglion neurons, and for the differentiation of hair cells derived from sensory progenitors.

We used the Cre-loxP system to conditionally delete *Neurod1* gene in the inner ear but to retain NEUROD1 expression in the auditory nuclei and auditory midbrain (*Neurod1CKO*). Using *Neurod1CKO,* we demonstrated how developmental changes in the periphery affect the function of the central auditory system. The primary consequence of *Neurod1* deletion was shortened cochlea containing reduced number of spiral ganglion neurons with disorganized innervation and missing tonotopy. Abnormalities in the inner ear resulted in the size reduction of the cochlear nucleus and a loss of tonotopic organization of the central auditory pathway. These changes altered tuning properties of inferior colliculus neurons – truncated frequency range, worsened frequency selectivity, and abnormal responses in behavioral tests.