P38.03

An evolutionary-driven approach to detect critical coding and non-coding regions in deafness

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Genome-wide studies identifying critical innovations in coding and non-coding DNA are continually growing, but the evidence showing how these genetic changes may simultaneously impact phenotype is scarce. In the inner ear, no studies have previously addressed the relevance of coding and non-coding DNA for its impact on evolution, development or disease. Classical and recent studies on non-syndromic hearing loss, a condition that affects approximately 1 in 1000 human newborns, have identified possible causal genes. We hypothesize that these hearing loss genes must play a critical role in the development and physiology of the mammalian inner ear and have thus been a critical target during evolution. Using a stringent bioinformatic pipeline, we identified signatures of selection at the coding and non-coding level for a compiled dataset including classic non-syndromic deafness genes and novel hearing loss genes candidates recently identified by the Mouse Phenotype Consortium. Using this dual approach, we detected genes showing signatures of accelerated evolution in non-coding conserved regions and in protein-coding regions. We functionally tested several of these regions in transgenic reporter essays, expanding the number of functional genomic territories associated with inner ear physiology and in close association with its development. We predicted the implications of ancestral coding variants in the functional properties of hearing loss genes. Moreover, we suggest that certain deafness genes could have played an important role in the appearance of the functional novelties that characterize the mammalian inner ear.

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P38.04

Peripheral ablation of type 3 adenylyl cyclase contributes to hyperalgesia in mice

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Mammalian adenylyl cyclase (AC) is an important enzyme family catalyzing the conversion of ATP into the second messenger cAMP. The type 3 adenylyl cyclase (AC3) was reported to be predominant in primary cilia throughout the central nervous system (CNS) leading to anosmia, depressive disorder, defects in learning and memory. Here we further showed that AC3 was expressed in dorsal root ganglion (DRG) and spinal cord dorsal horn which was crucial to peripheral sensitization in mice. Double immunofluorescence showed that AC3 immunoreactivity (IR) was expressed in cytoplasm of small to medium sized DRG neurons mainly colocalized with CGRP (a peptidergic nociceptor marker). Furthermore, AC3 IR existed in central or peripheral terminals of CGRP neurons in the superficial dorsal horn and glabrous skin. Mechanical and thermal hyperalgesia was induced in mice of AC3 conditional knockout (AC3 CKO) by intra-DRG injection of adeno-associated virus (AAV) expressing Cre recombinase in *AC3-floxed* mice. Licking and flinching responses were increased in AC3 CKO mice after intraplanta (i.pl.) application of formalin or capsaicin. Meanwhile, whole-cell patch-clamp recordings from DRG neurons showed that the excitability of neurons and transient potential vanilloid 1 (TRPV1)-currents were enhanced significantly and the current density of voltage-gated potassium channels (K_V) was reduced in AC3 CKO neurons. Intrathecal injection of κ opioid receptor (KOR) agonist nalfurafine did not relieve mechanical and thermal hyperalgesia in AC3 CKO mice. Also, the current density of K_V was not reversed by KOR agonist dynorphin A in AC3 CKO mice. These findings suggest that peripheral AC3 may contribute to antinociception in mice by regulating K_V and TRPV1 functions via endogenous KOR signaling.

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P38.05

Lesion of cerebral cortex affects tactile sensitivity and affective motivation behaviors associated with sustained pain

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Pain is an unpleasant sensory and affective experience. Cortical areas associated with the sensory-discriminative properties and emotional–motivational aspects are different. Previous studies have implied important roles of specific cortical areas such as ACC and PFC in the affective processing and descending modulation of pain. However, the roles of cortical structure in modulating complex behaviors and the effects of cerebral cortex lesion remain poorly understood. Here we investigate the effects of cerebral cortex lesion on the behavioral responses to noxious and inocuous stimuli in Tra2b conditional knockout (cKO) "cortexless" mice that was generated by breeding Tra2b flox/flox homozygous mice (provide by Prof. Brunhilde Wirth as a gift) with Emx1-cre homozygous mice (provided by Prof. Jiayi Zhang as a gift). Our results showed that the reflexive defensive responses to noxious stimuli remain undamaged, while the affective motivational behaviors, such as licking, are selectively impaired in cortexless mice. Moreover, cortical lesion enhances tactile sensitivity in both behavioral testing and spinal dorsal horn electrophysiological recording in vivo. These data demonstrated that cerebral cortex plays an important role in the processing of affective motivation responses to sustained pain, and tactile sensory processing.

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P38.06

Changes in the ratio of chondroitin sulfate A and C of perineuronal net components on spinal motoneurons during postnatal development

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Lumbar spinal segments responsible for hind limb movements develop during the first 3 weeks of postnatal development. In rats, descending systems, such as the corticospinal tracts, reach the lumbar spinal cord at postnatal day 9 (P9), and mature from P14 to P21. Primary afferent fibers, including proprioceptive afferents, that project to the spinal cord also mature around P14 and play