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ACCEPTED MANUSCRIPT

Connexins, E-Cadherin, Claudin-7 and β-catenin transiently form junctional nexuses during the post-natal mammary gland development

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ABSTRACT

Gap junctions are intercellular channels made of connexins (Cxs) that allow direct communication between adjacent cells. Modulation of Cxs has been associated with abnormal development and function of the mammary gland and breast cancer. However, the mechanisms underlying their expression during normal mammary gland are not yet known. Cxs interact with components of tight and adherens junctions. Thus, we hypothesized that the expression levels of Cxs vary during mammary gland development and are regulated through stage-dependent interactions with members of the tight and adherens junctions. Our specific objectives were to: 1) determine the expression of Cxs and tight and adherens junction proteins throughout development and 2) characterize Cxs interactions with components of tight and adherens junctions. Murine mammary glands were sampled at various developmental stages (prepubescent to post-weaning). RT-qPCR and western-blot analyses demonstrated differential expression patterns for all gap (Cx43, Cx32, Cx26, Cx30), tight (Claudin-1, -3, -4, -7) and adherens (β-catenin, E- and P-cadherins) junctions throughout development. Interestingly, coimmunoprecipitation demonstrated interactions between these different types of junctions. Cx30 interacted with Cx26 just at the late pregnancy stage. While Cx43 showed a persistent interaction with β-catenin from virginity to post-weaning, its interactions with E-cadherin and Claudin-7 were transient. Cx32 interacted with Cx26, E-cadherin and β-catenin during lactation. Immunofluorescence results confirmed the existence of a junctional nexus that remodeled during mammary gland development. Together, our results confirm that the expression levels of Cxs vary concomitantly and that Cxs form junctional nexuses with tight and adherens junctions, suggesting the existence of common regulatory pathways.

KEYWORDS

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