Topic: 3. Heart Failure and Cardiomyopathy**A Transcriptional Co-activator, p300 is Involved in the Epigenetic Gene Activation on Hypertrophic Response Gene Promoters in Heart Failure**

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Introduction and Objectives

An intrinsic histone acetyltransferase (HAT), p300, is required for acetylation and the transcriptional activity of GATA4, as well as for pathological left ventricular hypertrophy (LVH) and the development of heart failure (HF) *in vivo*. Many studies of histone modification have been performed within the flexible tails of these histones, such as on the lysines H3K9 and H3K14. Although most previously studied histone modifications have been carried out within these flexible tails, it has been reported recently that the lysine H3K122 is a novel site of the histone globular domain acetylated by p300, and that its acetylation activates gene transcriptions by destabilizing histone-DNA binding and increasing the accessibility of transcriptional factors to DNA. However, little is known about the extent to which histone modifications directly affect LVH and HF. We hypothesized that p300 induces epigenetic changes through the acetylation of both the globular domain and the tail domain of histone during the LVH and HF.

Materials and Methods

First, to investigate whether the acetylation of H3K122 in the globular domain and H3K9 and H3K14 in the tail domain increased in cardiomyocyte hypertrophy, western blotting and chromatin-immunoprecipitation (ChIP) assays were performed using neonatal rat cultured cardiomyocytes with phenylephrine (PE). Second, neonatal rat cultured cardiomyocytes were treated by using siRNA of p300 or curcumin, a p300-specific HAT inhibitor. Third, to investigate the role of p300 HAT activity in histone acetylation *in vivo*, we utilized mice overexpressing p300 in the heart, which induces LVH.

Finally, to investigate whether these acetylation changes occurred during the development of LVH and HF, an *in vivo* ChIP assay was performed using the hypertensive heart disease model of Dahl salt-sensitive rats.

Results

Western blotting indicated that treatment with PE increased the acetylation of H3K122, H3K9, and H3K14 in cardiomyocyte hypertrophy. The ChIP assay demonstrated that PE increased the recruitment of acetylated H3K122 and H3K9 onto ANF and BNP promoters containing the GATA element. Time course analyses of the acetylated levels by western blotting indicated that acetylated H3K9 increased after PE stimulation but acetylated H3K122 increased slowly after PE stimulation compared with acetylated H3K9. These acetylations were significantly inhibited by both p300 knockdown using siRNA and treatment with curcumin. Conversely, *in vivo* ChIP assays in mice overexpressing p300 indicated that p300 overexpression increased the recruitment of acetylated H3K122 and H3K9 onto ANF and BNP promoters containing the GATA element. Next, in the hypertensive heart disease model of Dahl salt-sensitive rats, *in vivo* ChIP assays revealed that acetylation of H3K9 was increased around the ANF and BNP promoters containing the GATA element at the LVH stage, but that of H3K122 was increased at the HF stage.

Conclusions

These results indicate that the acetylation of H3K122 in the globular domain of histones by p300 is a key event of the transition from LVH to HF. ■