

# Transcriptional mechanism of COMP gene expression and chondrogenesis

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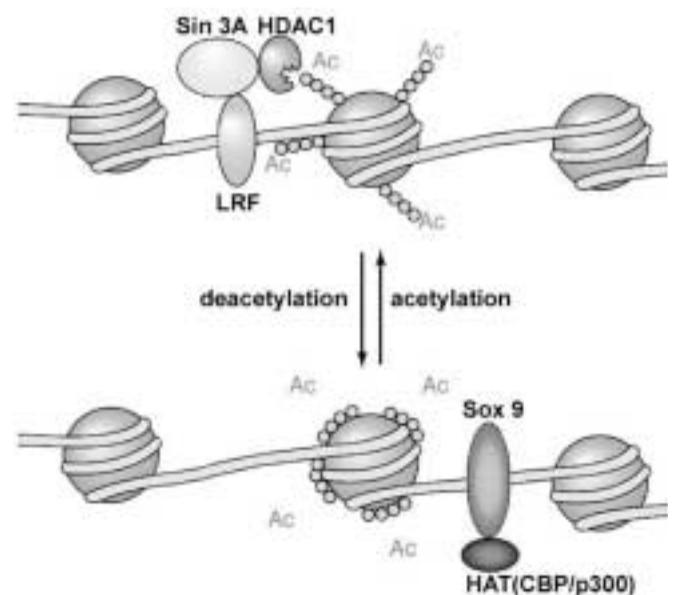
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Gene cis-elements (such as promoters, enhancers and locus control regions) and trans-elements (such as transcription factors, chromatin-remodeling complexes, and histone-modification enzymes) come together to orchestrate a finely tuned sequence of events that results in the complex pattern of tissue specific gene expression that is observed during chondrocyte development. Our laboratory has identified transcriptional regulators of cartilage oligomeric matrix protein (COMP) gene expression. COMP encodes a noncollagenous extracellular matrix protein whose expression marks a variety of musculoskeletal tissues, including chondrocytes, tenocytes, ligament cells, and osteoblasts. To understand the mechanisms controlling this differentiation process, we cloned 1.9 kilobases of the 5' flanking sequence of the promoter region of the murine COMP gene. We then delineated a negative regulatory element (NRE) and a proximal positive region (PPR) in the COMP promoter from a series of reporter gene and gel mobility shift assays<sup>1,2</sup>. A yeast one-hybrid screen identified the leukemia/lymphoma-related factor (LRF) as an NRE-binding transcriptional repressor. LRF showed dose-dependent inhibition of COMP-specific reporter gene activity and overexpression of LRF-repressed BMP-2-induced endogenous expression of COMP and chondrogenesis in high-density micromass cultures of C3H10T1/2 progenitor cells. LRF associates with histone deacetylase-1 (HDAC1), and experiments utilizing the HDAC inhibitor tricostatin A revealed that LRF-mediated repression requires deacetylase activity<sup>3</sup>. Subsequent studies revealed that the association of LRF and HDAC1 is bridged by Sin3A co-repressor, since POZ domain of LRF

directly bound to the PAH2 domain of Sin3A<sup>4</sup> and Sin3A was previously reported to interact with HDAC1<sup>5,6</sup>, in addition, Sin3A enhanced the LRF-mediated inhibition on COMP-specific reporter gene activities and endogenous COMP gene expression. Thus, LRF, Sin3A and HDAC1 form a ternary repressor complex that binds to the NRE of COMP promoter.

Intriguingly, Sox9, a key regulator of chondrogenesis, was shown to directly bind to the positive proximal region (PPR, -125 to -75) of COMP promoter and activate the COMP-specific reporter gene, and this transactivation was enhanced with the co-transfection of histone acetylase (HAT), including CREB-binding protein (CBP) and p300<sup>7-9</sup>. Based on our comprehensive studies and the literature, we proposed a model for explaining the transcriptional control of COMP gene expression (Figure 1). In an undifferentiated state LRF



**Figure 1.** A proposed model explaining the transcriptional control of COMP gene expression.

The author has no conflict of interest.

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binds to the NRE located in the distal region of the COMP promoter and recruits histone deacetylase 1 (HDAC1); this interaction is bridged by the Sin3A corepressor. When the LRF/Sin3A/HDAC1 repressor complex associates with the COMP gene promoter, chromatin is condensed and the COMP gene is silenced. When mesenchymal tissues are triggered by extracellular signals for differentiation, however, the repressor complex disassociates from the COMP promoter, and the SOX9/CBP/p300 activator complex binds to the PPR in the proximal region of the COMP promoter, which results in loosening of the chromatin structure and activation of COMP gene expression.

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