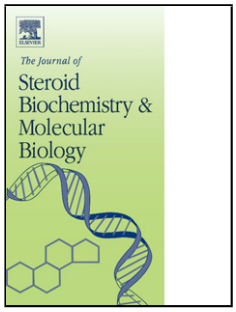

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<AT>The impact of VDR expression and regulation in vivo

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<ABS-HEAD>Highlights► Expression of the VDR is regulated in a tissue-specific manner. ► Tissue-specific regulatory regions in *Vdr* gene locus are identified. ► Hormonal regulatory mechanisms of *Vdr* expression in bone and kidney are revealed. ► Low amounts of the VDR are systemically functional in vivo despite reduced activity. ► Mutation in S208 of human VDR has no effect on its activity and phenotypes in vivo.

<ABS-HEAD>Abstract

<ABS-P>The vitamin D receptor (VDR) mediates the pleiotropic biological actions of 1,25-dihydroxyvitamin D_3 ($1,25(OH)_2D_3$). These actions include orchestration of mineral homeostasis which is coordinated by the kidney, intestine, bone and parathyroid gland wherein the VDR transcriptionally regulates expression of the genes involved in this complex process. Mutations in human VDR (hVDR) cause hereditary vitamin D resistant rickets, a genetic syndrome characterized by hypocalcemia, hyperparathyroidism and rickets resulting from dysregulation of mineral homeostasis. Expression of the VDR is regulated by external stimuli in a tissue-specific manner. However, the mechanisms of this tissue-specificity remain unclear. Studies also suggest that phosphorylation of hVDR at serine 208 impacts the receptor's transcriptional activity. These experiments were conducted in vitro, however, and therefore limited in their conclusions. In this report, we summarize (1) our most recently updated ChIP-seq data from mouse tissues to identify regulatory regions responsible for the tissues-specific regulation of the VDR and (2) our studies to understand the mechanism of hormonal regulation of *Vdr* expression in bone and kidney in vivo using transgenic mouse strains generated by mouse mini-genes that contain comprehensive genetic information capable of recapitulating endogenous *Vdr* gene regulation and expression. We also defined the functional human *VDR* gene locus in vivo by using a human mini-gene comparable to that in the mouse to generate a humanized VDR mouse strain in which the receptor is expressed at normal levels (normal expressor). The present report also shows that a humanized mouse model in which the VDR is expressed at levels about 10-fold lower than the normal expressor mouse rescued the VDR-null phenotype despite its reduced transcriptional activity relative to wildtype expression. We also generated an additional humanized mouse model expressing hVDR bearing a mutation converting serine 208 to alanine (hVDR-S208A). In spite of the mutation, target gene expression induced by the ligand was unchanged relative to a mouse strain expressing comparable levels of wildtype hVDR. Further characterization also showed that serum calcium and parathyroid hormone levels were normal and alopecia was not observed in this hVDR-S208A mouse strain as well. Taken together, our in vivo studies using ChIP-seq analyses and the mini-gene transgenic mice improve our understanding of the tissue-