INTERACTION OF RETINOID X NUCLEAR RECEPTOR WITH DIFFERENTS DNA HORMONE RESPONSIVE ELEMENTS

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Retinoid X nuclear receptor (RXR) is a ligand-activated transcription factor class that act on target genes in higher eukaryotes. This receptor is activated by the vitamin A metabolite 9-cis-retinoic acid (9-cRA), whose pleiotropic effects are crucial for the metabolism, development and homeostasis. The transcriptional action of this receptor is mediated by your interaction (like homo- or heterodimers) with small DNA sequences – the hormone responsive elements (HREs). These HREs are derivate from the archetypic sequence AGGTCA. Several ancient studies indicate that RXR, acting as homodimer, recognizes preferentially HREs composed by direct repeats of AGGTCA spaced by one base pair (DR1). However, only a few studies have been carried out to estimate both the affinity constants for the interaction of RXR with HREs and the effect of ligand binding on these constants. In this study, we have investigated the binding isotherms of human RXRΔAB (hRXR ΔAB - a mutant without the amino-therminal domain) with different HREs – the half site AGGTCA, direct repets no spaced and spaced by one, four and five base pairs (DR0,1,4 and 5, respectively), palindromic non spaced sequence (PAL) and everted palindrome spaced by two base pair (F2)- in the absence and presence of 9-cRA. These studies were carried through the fluorescence anisotropy technique. We observed a promiscuous behavior for the interaction of hRXRΔAB with HREs, with no signifcant differences in the ΔG_{diss} values for the hRXRΔAB complex with the majority of the sequences, with and without ligand. However, was observed a significant cooperative behavior to DR-1 and DR-4, indicating protein-protein interaction energetically linked to the interaction of hRXRΔAB with these HREs. The adding of ligand caused a significant depletion on F2 recognition. In the experiments leaded, it was not observed protein-PAL interaction. Take all together, these results showed a preference of holo hRXRΔAB to the direct repeats (here used), and a stronger protein-protein interface (sugesting a more stable complex) in the association with DR1 and DR4.