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	Human multidomain p53in complex with non specific DNA	MX-1597
ID23-2	28th of November 2013	
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Experimental Report

Aim of the experiment and specific background :

We aim to solve the co-crystal structure of the human transcription factor containing both the p53 DNA-binding and oligomerization domains in complex with non-specific DNA. We have already determined the structure of the co-crystal structure of p53 in complex with specific consensus DNA as well as in complex with one of its natural response element (*CDKN1A* (*p21*)) (Figure 1) (1-3). Our structure shows two p53 dimers, having in total four DNA binding domains, bound to double stranded DNA. We observed that the loop L1 that interacts with DNA has two different conformations, recessed and extended. We demonstrated that the p53-DNA binding occures via an induced fit mechanism with a switch in the conformation that involves the loop L1 (2). We solved the structure of the loop mutant p53 in its apo form and in complex with DNA (4,5,6).

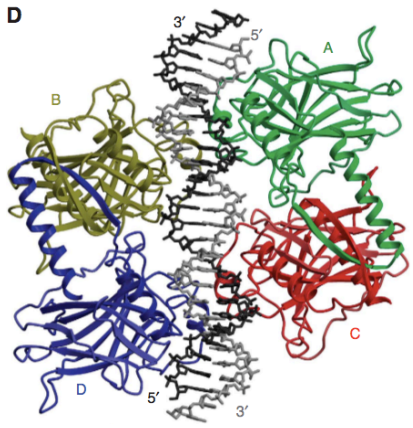

Figure 1. Structure of p53 in complex with DNA

Experimental method :

We purified a stable p53 containing both the DNA-binding and oligomerization domains. We proceeded then to screen crystallographic conditions in order to obtain crystals. All experiments, from purification through crystal screening, were performed at 4 degrees Celsius to reduce precipitation and increase the chances of obtaining crystals.
We obtained p53 crystals with different sizes in a range of 30 to 50 microns (Figure 2).

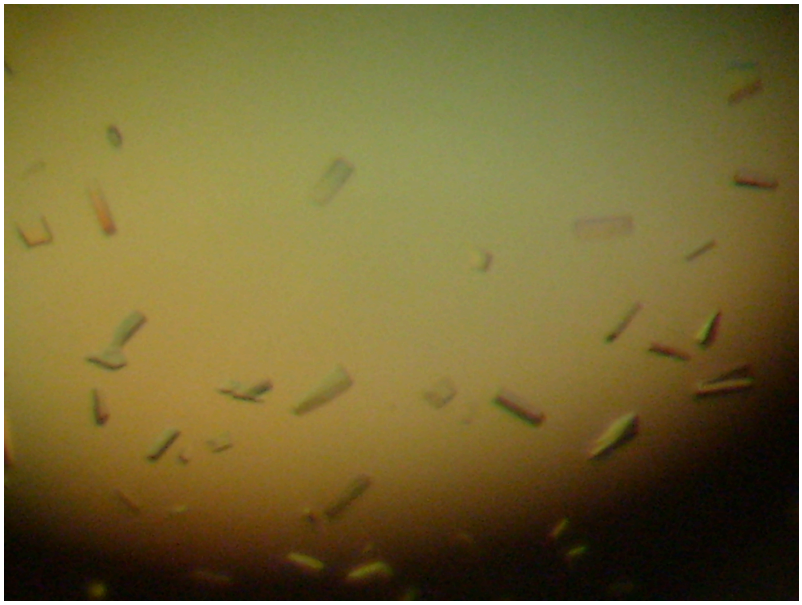

Figure 2. Crystals of p53 with non-specific DNA

Results:

We were able to collect diffraction data from the four complex of p53 with non specific we had. We obtained a resolution average of about 3.5-4 Angström. However, after processing the data we were not able to determine a high resolution structure. We plan to optimize the conditions of the crystal growth in order to have better crystals.

References :

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   Crystal structure of a multidomain human p53 tetramer bound to the natural CDKN1A (p21) p53-response element. Mol Cancer Res. 2011 Nov; 9(11): 1493-9

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