3D analysis of the Fv structure of the IgE SPE-7 in complex with acenaphthenequinone

Introduction: Due to their critical function in the immune system, the interactions between antibodies and their antigens have been extensively studied. To fully comprehend the action of these antibody-antigen complexes, it is imperative to unravel their three-dimensional (3D) structure. Recent developments in the field of molecular and structural biology have made it possible to use software programs like PyMOL to study these interactions. In this report, the Fv structure of the IgE SPE-7 in complex with acenaphthenequinone is studied to gain insights into the mechanism of antibody-antigen recognition.

Methods: The Fv structure of the IgE SPE-7 in complex with acenaphthenequinone ($C_{12}H_6O_2$) (Identifier 1OAX) was analyzed in the Protein Data Bank (PDB) using the website: https://www.rcsb.org/structure/1OAX. The PDB (1oax.pdb) and FASTA (rcsb_pdb_1OAX.fasta) files for this complex were downloaded from the website. The PDB file was imported into PyMOL-2.5.5 for further analysis. In PyMOL-2.5.5, water molecules were removed prior to analysis. The structure of the complex was visualized in this software. The FASTA file was imported into the prediction tool Benchling to identify the complementarity-determining regions (CDRs) in the Fv based on the amino acid sequence. The antibody-antigen interactions were analyzed in PyMOL by selecting the acenaphthenequinone ligand and displaying its interactions with the IgE SPE-7 Fv. Polar contacts 'to others excluding solvent' and all pi interactions were considered. These interactions were compared to the interactions in PDB.

Results: A preliminary analysis of the structure was performed in PDB. The structure consists of the Immunoglobulin E (Figure 1A), in complex with the acenaphtenequinone ligand (Figure 1B). H_2O molecules are present in the environment of the complex (Figure 1C).

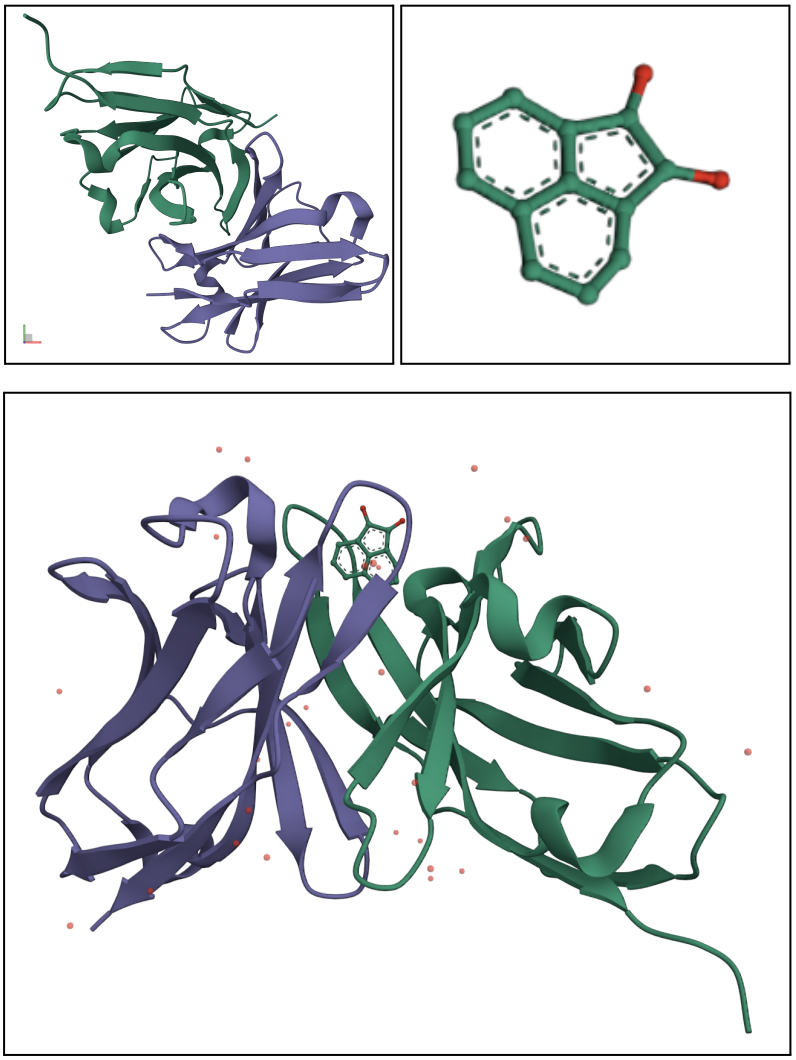

Figure 1: 3D display of one IgE SPE-7 Fv in complex with acenaphthenequinone. Image (A) shows the unbound IgE SPE-7 Fv polymer consisting of a variable heavy chain (green) and a variable light chain (purple). Image (B) shows the unbound acenaphthenequinone ligand consisting of a three-ringed structure with two extruding oxygen atoms (red). Image (C) shows the IgE SPE-7 Fv in complex with acenaphthenequinone, surrounded by H_2O molecules. These images were retrieved from PDB.

The structure was then further analyzed using the PyMol software. Every chain was assigned a different color and labeled with an appropriate name. The secondary structures of both chains are visible, displaying a beta-sheet sandwich structure, stabilized by loops of variable length (Figure 2).

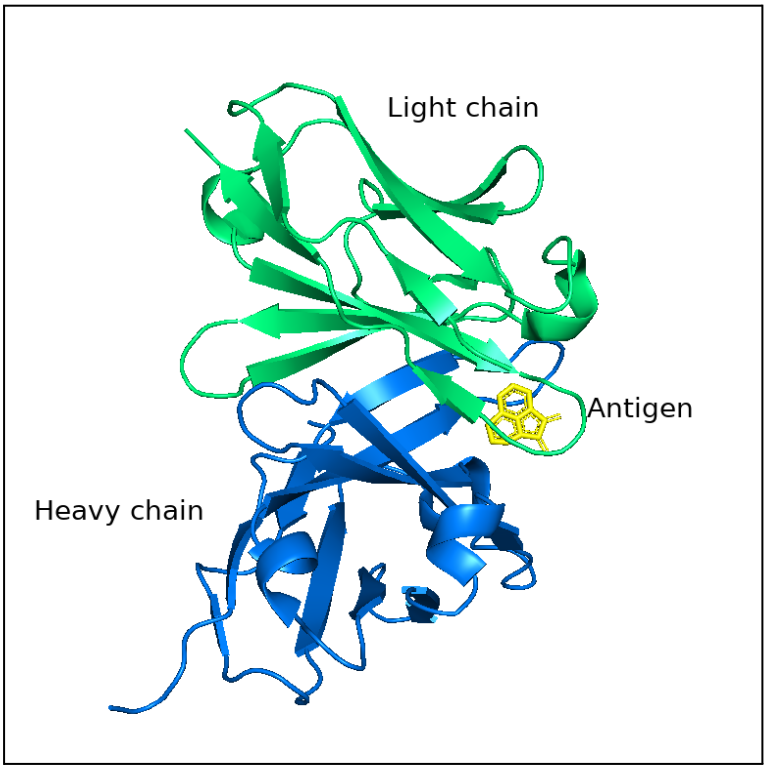

Figure 2: 3D display of one IgE SPE-7 Fv fragment in complex with acenaphthenequinone in which each chain is displayed in a different color and all components are labeled. The light chain is colored limegreen, the heavy chain marine blue, and the antigen, acenaphthenequinone, is shown in yellow. The labels are named accordingly. This image was generated in PyMol.

Using the Benchling prediction tool, the CDRs were identified in both light and heavy chains. These were colored in different gradients of green and blue, respectively (Figure 3). Table 1 provides an overview of the location and length of the identified CDRs for each chain.

Table 1: Overview of the identified CDRs for the light and heavy chains in Benchling. The position of the amino acids (AA) in the chain is provided, as well as the total amount of amino acids in each CDR.

	CDR1	**CDR2**	**CDR3**
Light chain	23-36 (14 AA)	52-58 (7 AA)	91-99 (9 AA)
Heavy chain	26-32 (7 AA)	52-57 (6 AA)	99-109 (11 AA)

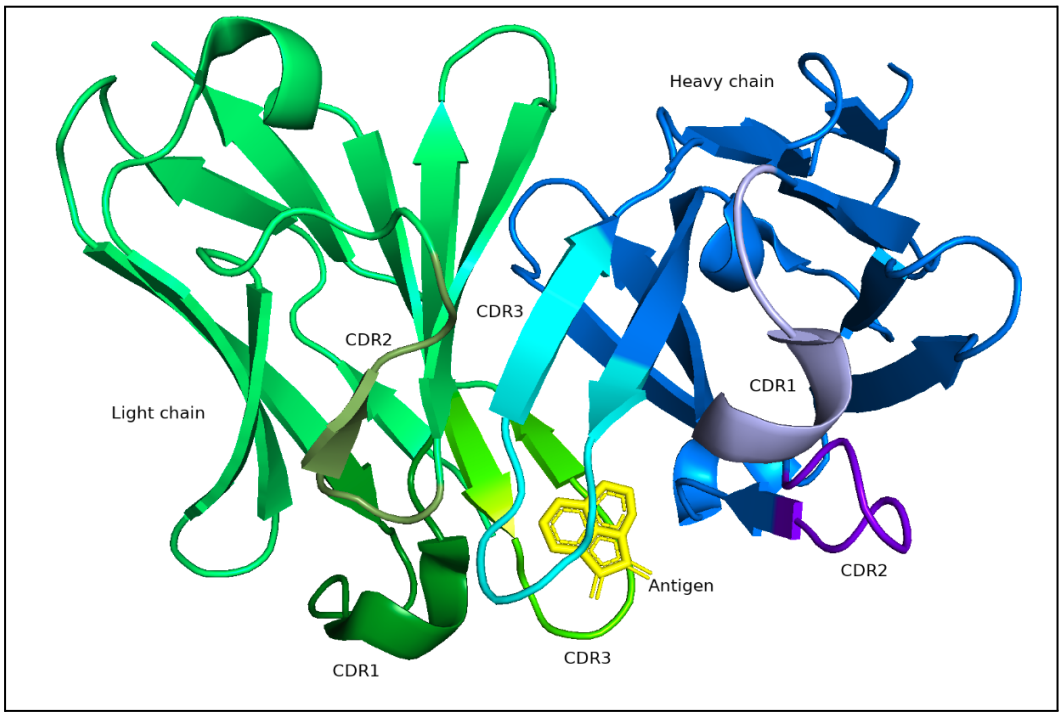

Figure 3: 3D display of one IgE SPE-7 Fv fragment in complex with acenaphthenequinone in which each CDR is displayed in different shades of green or blue for the light and heavy chains, respectively. All components are labeled. The framework regions of the light chain are colored limegreen, the CDR1 in forest green, the CDR2 in smudge green, and the CDR3 in chartreuse green. The framework regions of the heavy chain are colored marine blue, the CDR1 in lightblue, the CDR2 in purple blue, and the CDR3 in cyan. The antigen, acenaphthenequinone, is shown in yellow. The labels are named accordingly. This image was generated in PyMol.

Interactions between the antibody and antigen were analyzed in PyMOL. Two polar interactions were identified, these are hydrogen bonds with an average distance of 3.4 Angstroms. The residues participating in these interactions are arginine 50 and tyrosine 105, both in the heavy chain (Figure 4). Tyrosine 105 is part of the CDR3, whereas arginine 50 is not part of a CDR. No interactions were identified with the light chain. PyMOL did not identify pi-stacking interactions. The two polar interactions were also present in the PDB (Figure A.1). Additionally, in the PDB, seven pi-stacking interactions were identified. These interactions between the antigen and light chain residue tryptophan 93 and heavy chain residue tyrosine 105 further stabilize the antibody-antigen complex.

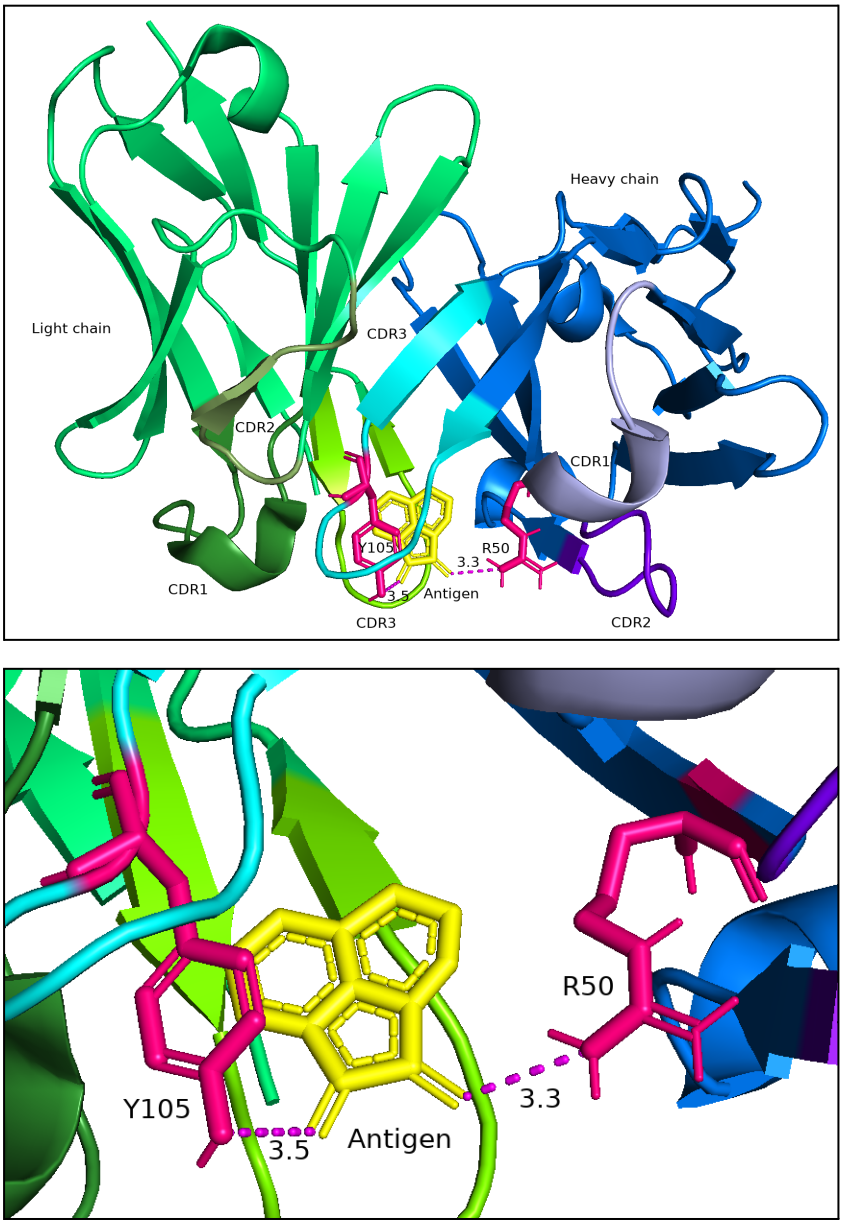

Figure 4: 3D display of one IgE SPE-7 Fv fragment in complex with acenaphthenequinone with the identified polar interactions. These interactions are crucial for antigen recognition by the antibody. Image (A) shows the entire Fv fragment. Image (B) shows a zoom on the interactions between the antibody and the antigen. The interaction partners of the antigen are tyrosine 105 (Y105) and arginine 50 (R50) which are marked in magenta. The polar interactions are marked by dashed lines. The distance of each interaction is provided in Angstroms. The framework regions of the light chain are colored limegreen, the CDR1 in forest green, the CDR2 in smudge green, and the CDR3 in chartreuse green. The framework regions of the heavy chain are colored marine blue, the CDR1 in light blue, the CDR2 in purple blue, and the CDR3 in cyan. The antigen, acenaphthenequinone, is shown in yellow. The labels are named accordingly. These images were generated in PyMol.

Appendix

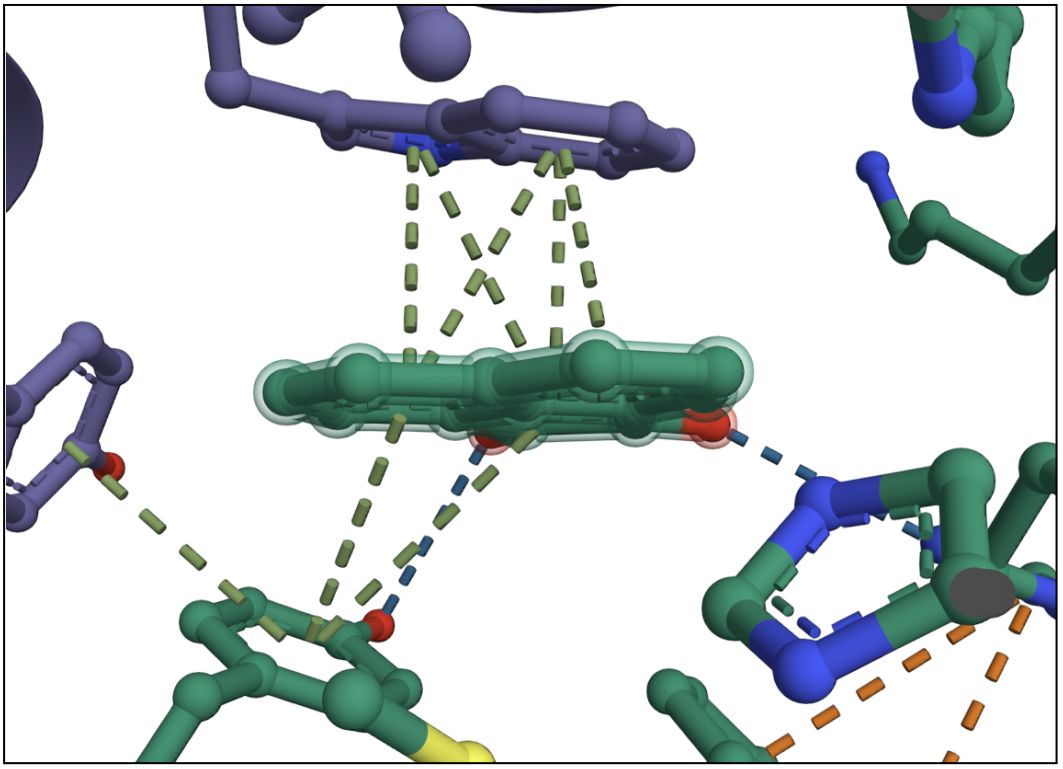

Figure A.1: 3D display of the interaction between the IgE SPE-7 Fv fragment and its antigen, acenaphthenequinone. The antigen is represented by the central ring structure. It interacts with heavy chain residues tyrosine 105 and arginine 50 through hydrogen bonds (indicated in blue dashed lines). The tyrosine residue is situated above the antigen in this image. The arginine residue is at the bottom right. Additionally, the antigen interacts with light chain residue tryptophan 93 through five pi stacking interactions (indicated in green dashed lines) and two pi stacking interactions with heavy chain residue tyrosine 105. The tryptophan residue is situated at the bottom left. These pi-stacking interactions contribute to the antibody-antigen binding but were not identified in PyMOL. These images were retrieved from PDB.

Figure A.2: Object Control Panel in PyMol. Labels in the PyMOL figures are similar to the names in this Object Control Panel.