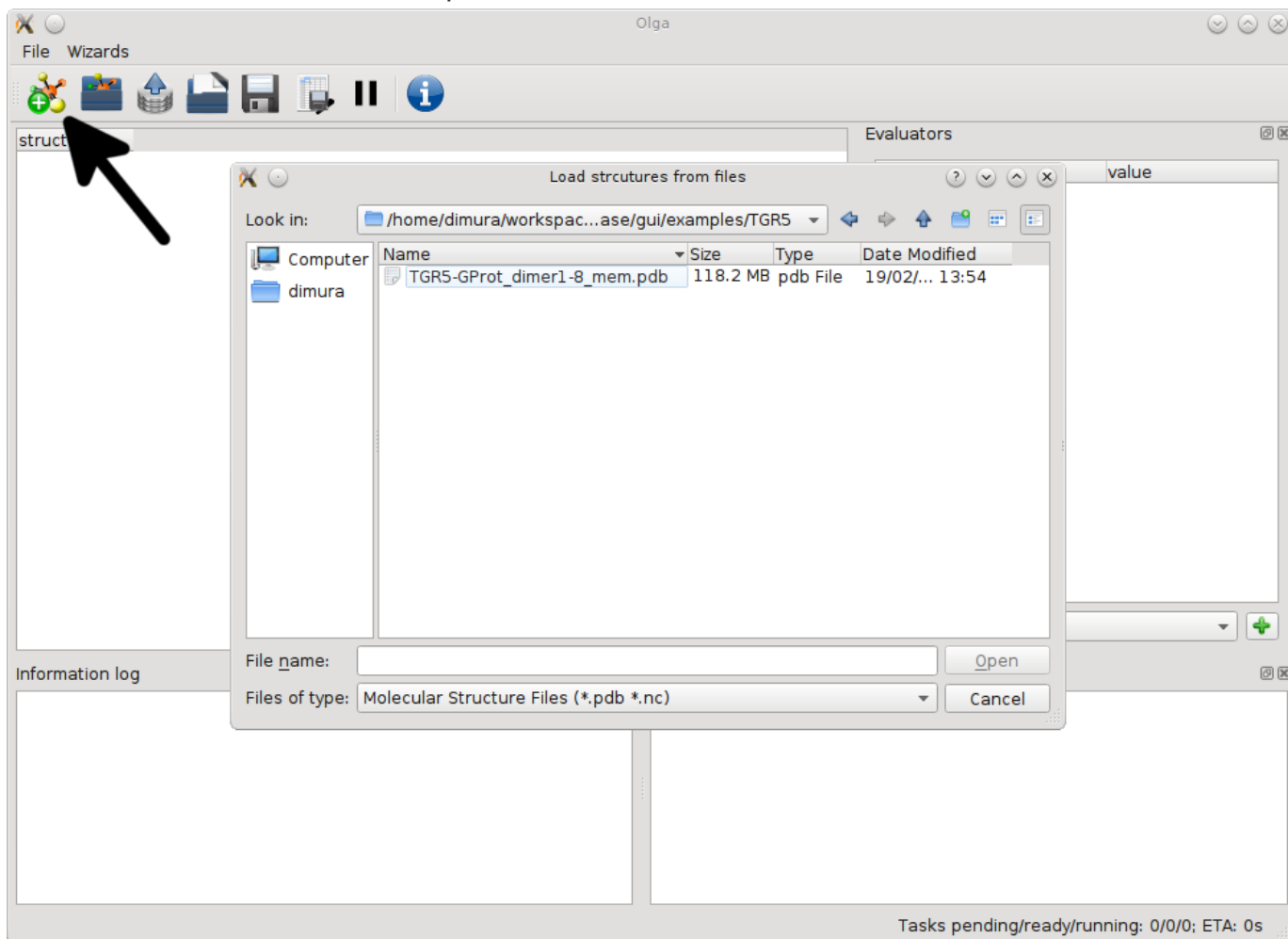


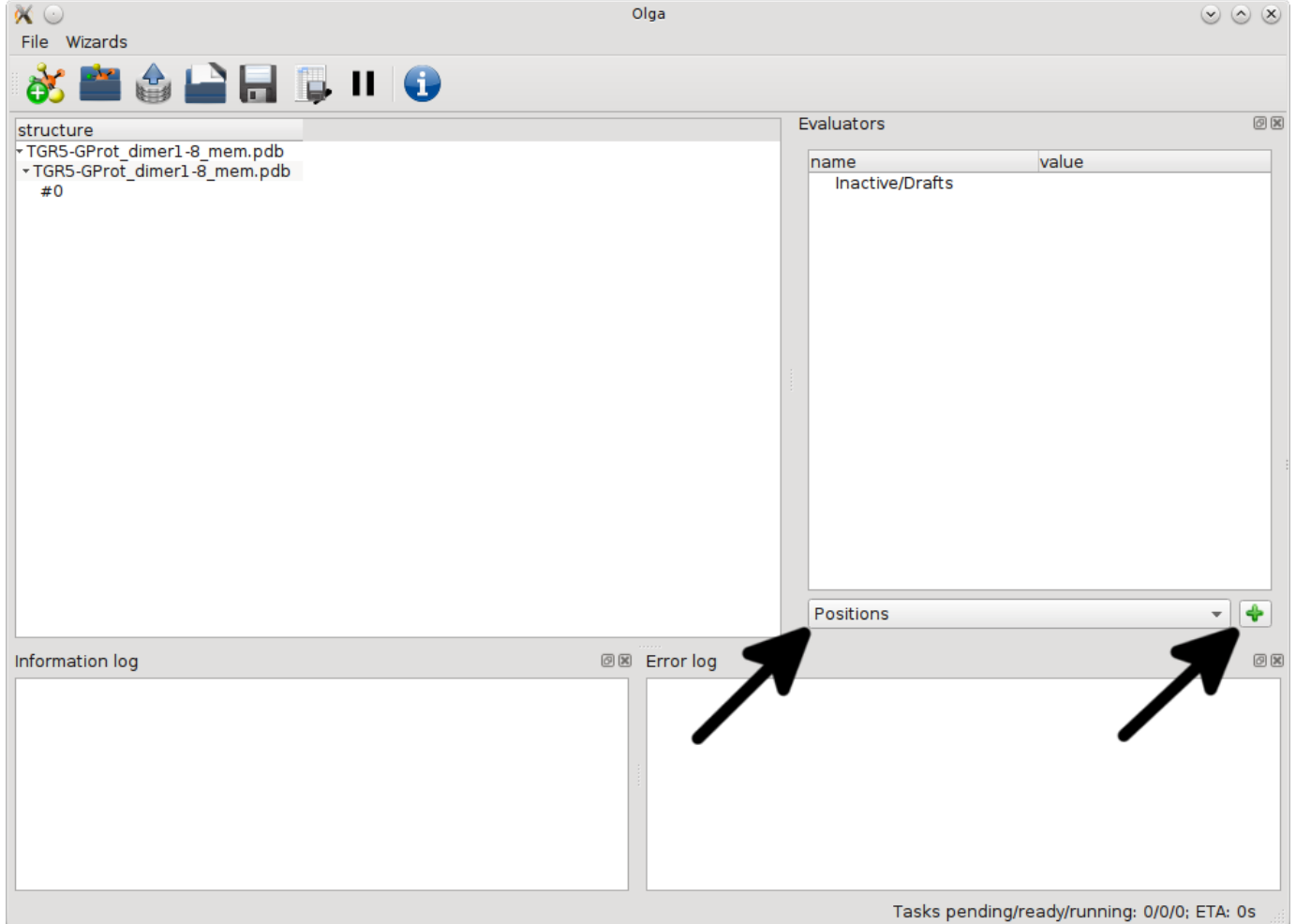
# Tutorial: screening of structural models and generation of accessible volumes

## Screening

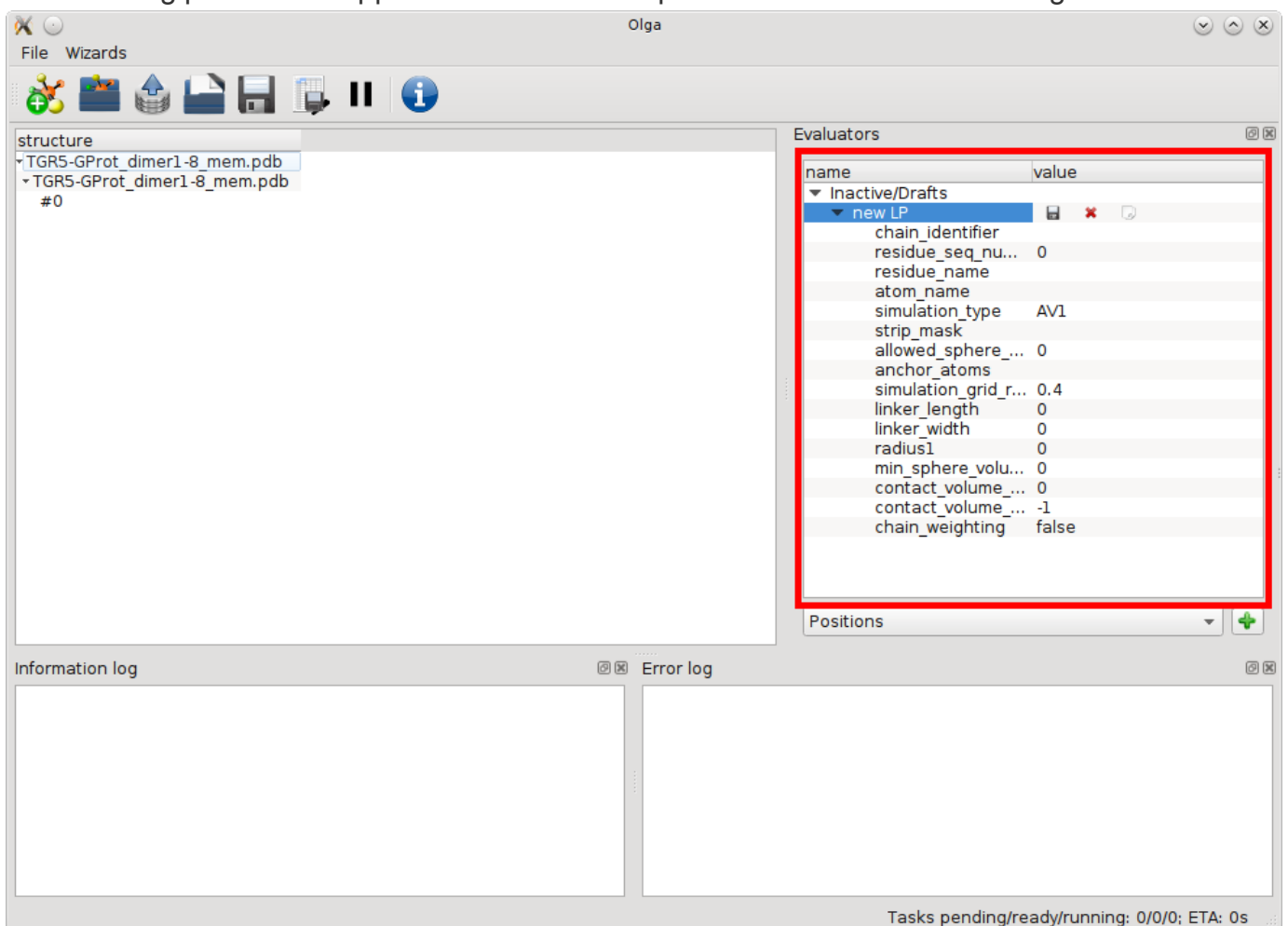
1. Start Olga software
2. Press "Import structures" button to load a set of PDB files. Select the files that you need to screen, use shift/ctrl buttons to select multiple files.



3. Create a labelling position evaluator. To do this, you need to select "Position" from the dropdown menu and then press "+" button.



New labelling position will appear in the evaluators panel. Unfold it to see its settings.



- Now Labelling Position properties can be filled out. First one has to choose the simulation type (here AV1 is selected). In this example Donor label is shown, GFP is used as a dye (radius1 = 20 Angstrom). It is attached by linker consisting of 62 aminoacids, therefore linker length at maximum

extension is defined as  $\text{linker\_length} = 62 * 3.7 + 20 = 249.4$ , where 3.7 is the length of a single amino acid and 20 is the effective radius of the GFP. Here linker width of 7 Angstrom is used, digitization step is set to 2 Angstrom ( $\text{simulation\_grid\_resolution} = 2$ ). You will need to specify chain id, residue id, residue name and atom name of the atom, to which fluorophore linker is attached. User can specify `allowed_sphere_radius` option, which tells algorithm to ignore obstacles in the given radius in AV simulation. Since in this example the dye (GFP) is attached to the protein by a peptide linker, `chain_weighting` option is activated to account for the polypeptide chain dynamics. This option reweights the probability density distribution within the AV as described by ... The name of the labelling position can be changed by double-clicking on the corresponding field (here "A:ALA1088").

The screenshot shows the Olga software interface. The main window is titled "Olga". It features a menu bar with "File" and "Wizards". Below the menu bar is a toolbar with icons for file operations and simulation controls. The main workspace is divided into several panels:

- Structure Table:** A table with columns "structure", "A", "B", and "A:ARG1101-B:". It contains two rows for "TGR5-GPr..." and one row for "#0" with values 1, 1, and 56.091231.
- Evaluators Panel:** A panel on the right showing a list of parameters and their values. The "Inactive/Drafts" section is expanded, showing "A:ALA1088" selected. The parameters and values are:
 

| name                            | value |
|---------------------------------|-------|
| chain_identifier                | A     |
| residue_seq_number              | 1088  |
| residue_name                    | ALA   |
| atom_name                       | CA    |
| simulation_type                 | AV1   |
| strip_mask                      |       |
| allowed_sphere_radius           | 15    |
| anchor_atoms                    |       |
| simulation_grid_resolution      | 2     |
| linker_length                   | 249.4 |
| linker_width                    | 7     |
| radius1                         | 20    |
| min_sphere_volume_fraction      | 0     |
| contact_volume_thickness        | 0     |
| contact_volume_trapped_fraction | -1    |
| chain_weighting                 | true  |
- Positions Panel:** A dropdown menu labeled "Positions" with a green plus icon.
- Information log and Error log:** Two empty text areas at the bottom for logging.
- Task Status:** A status bar at the bottom right showing "Tasks pending/ready/running: 0/3/0; ETA: 0s".

5. Copy the Donor labelling position and modify the copy to represent the Acceptor.

The screenshot shows the Olga software interface. The 'structure' panel on the left displays a table with columns A, B, and A:ARG1101-B:A. The 'Evaluators' panel on the right shows a list of evaluators. An arrow labeled 'copy LP' points to the 'B:ALA1088' entry. The 'Positions' dropdown is set to 'Positions'. The status bar at the bottom indicates 'Tasks pending/ready/running: 3/3/4; ETA: 0s'.

| name                            | value |
|---------------------------------|-------|
| ▶ A:ALA1088                     | ✗     |
| ▼ B:ALA1088                     | ✗     |
| chain_identifier                | B     |
| residue_seq_number              | 1088  |
| residue_name                    | ALA   |
| atom_name                       | CA    |
| simulation_type                 | AV1   |
| strip_mask                      |       |
| allowed_sphere_radius           | 15    |
| anchor_atoms                    |       |
| simulation_grid_resolution      | 2     |
| linker_length                   | 249.4 |
| linker_width                    | 7     |
| radius1                         | 20    |
| min_sphere_volume_fraction      | 0     |
| contact_volume_thickness        | 0     |
| contact_volume_trapped_fraction | -1    |
| chain_weighting                 | true  |

6. Activate Donor and Acceptor labelling position drafts by pressing "save" button.

The screenshot shows the Olga software interface. The 'structure' panel on the left is empty. The 'Evaluators' panel on the right shows a list of evaluators. The 'Inactive/Drafts' section is selected. Arrows point to the 'B:ALA1088' entry and its 'chain\_identifier' field. The 'Positions' dropdown is set to 'Positions'. The status bar at the bottom indicates 'Tasks pending/ready/running: 0/0/0; ETA: 0s'.

| name                 | value |
|----------------------|-------|
| ▼ Inactive/Drafts    |       |
| ▶ A:ALA1088          | ✗     |
| ▼ B:ALA1088          | ✗     |
| chain_identifier     | B     |
| residue_seq_number   | 1088  |
| residue_name         | ALA   |
| atom_name            | CA    |
| simulation_type      | AV1   |
| strip_mask           |       |
| allowed_sphere...    | 15    |
| anchor_atoms         |       |
| simulation_grid_r... | 2     |
| linker_length        | 249.4 |
| linker_width         | 7     |
| radius1              | 20    |
| min_sphere_volu...   | 0     |
| contact_volume...    | 0     |
| contact_volume...    | -1    |

7. Create a distance evaluator. To do this select "Distance" from dropdown menu and press "+", as in step 3.

8. Select the distance type, donor and acceptor labelling position, specify the Förster radius. If available, you can specify the measured distance and error.

The screenshot shows the Olga software interface. The main window is titled 'Olga'. It has a menu bar with 'File' and 'Wizards'. Below the menu bar is a toolbar with various icons. The interface is divided into several panels:

- structure**: A tree view showing a structure named 'TGR5-GPr...' with a sub-entry '#0'.
- Evaluators**: A table showing the configuration of evaluators. The table has two columns: 'name' and 'value'. The table is expanded to show the configuration for 'A:ARG1101-B:ARG110'. The configuration includes:

| name                 | value     |
|----------------------|-----------|
| ▼ Inactive/Drafts    |           |
| ▼ A:ARG1101-B:ARG110 |           |
| distance_type        | RDAMeanE  |
| position1_name       | A:ALA1088 |
| position2_name       | B:ALA1088 |
| distance             | 0         |
| error_neg            | 1         |
| error_pos            | 1         |
| Forster_radius       | 52        |
| ► Positions          |           |
| AV File              |           |
| Distances            |           |
- Information log**: A panel for displaying information logs.
- Error log**: A panel for displaying error logs. It contains two error messages:

```
Can not set position_1: A:ARG1101-B:ARG1101 copy
Can not set position_2: A:ARG1101-B:ARG1101 copy
```

At the bottom of the window, there is a status bar that reads: 'Tasks pending/ready/running: 0/0/0; ETA: 0s'.

9. Save the distance evaluator to start calculation. Once the distance evaluator is saved, a corresponding column will appear next to the structure list. At first this column will only show "...". Once the corresponding calculation is finished, calculated distance value will show up (or "nan" if the

distance could not be calculated).

The screenshot shows the PyMOL GUI. In the 'structure' panel, the tree is expanded to show 'TGR5-GProt\_dimer1-8\_mem.pdb' and its sub-entry '#0' with a value of 55.095137. A black arrow points to this value. The 'Evaluators' panel on the right shows a table with the following data:

| name                | value     |
|---------------------|-----------|
| Inactive/Drafts     |           |
| Positions           |           |
| Distances           |           |
| A:ARG1101-B:ARG1... | ✖         |
| distance_type       | RDAMeanE  |
| position1_name      | A:ALA1088 |
| position2_name      | B:ALA1088 |
| distance            | 0         |
| error_neg           | 1         |
| error_pos           | 1         |
| Forster_radius      | 52        |

The 'Information log' and 'Error log' panels are empty. The status bar at the bottom indicates 'Tasks pending/ready/running: 0/1/0; ETA: 0s'.

10. Results table can be saved by pressing "Export calculated values" button.

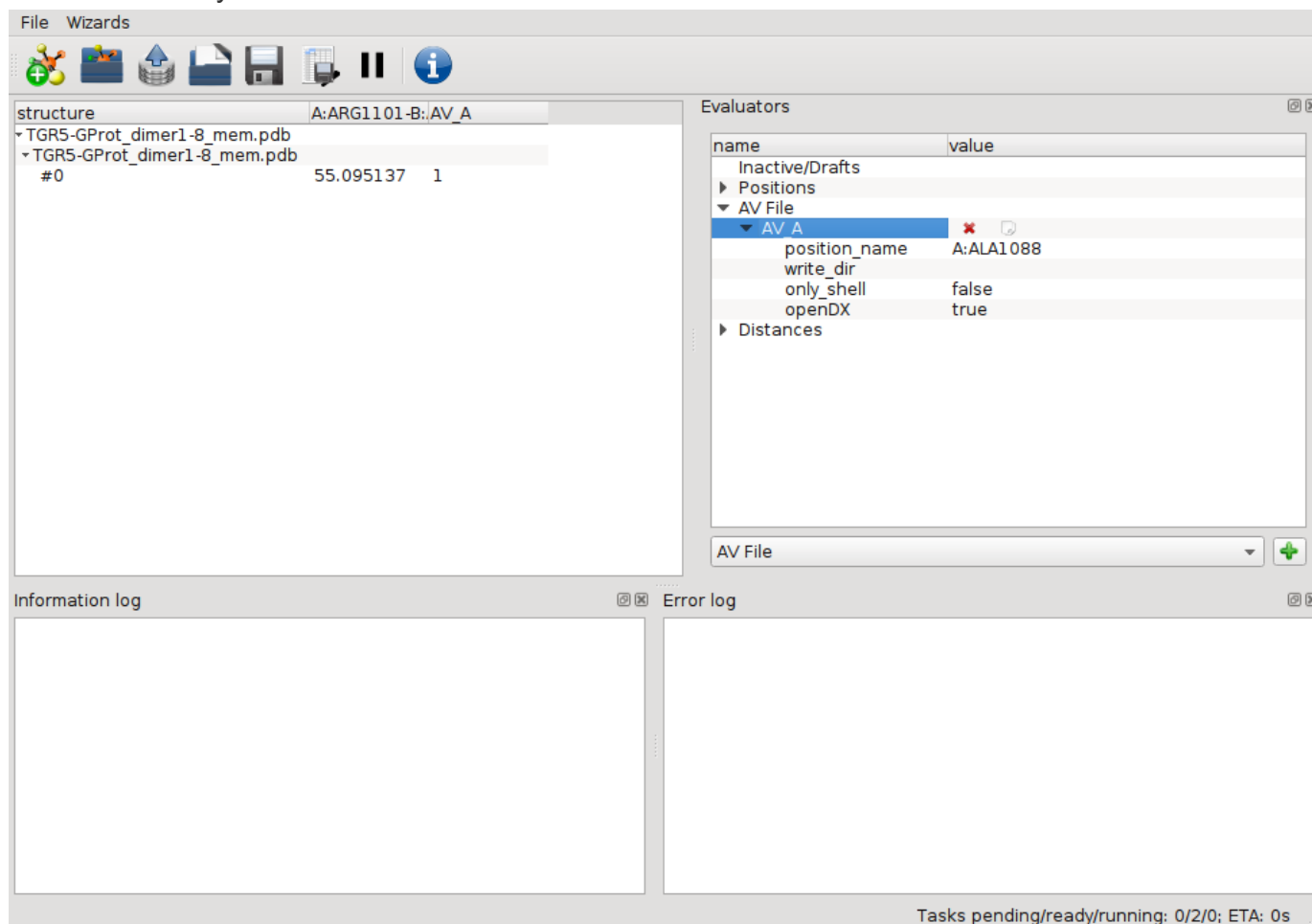
The screenshot shows the PyMOL GUI with the 'Export data' dialog box open. The dialog has a 'Look in:' field showing the path '/home/dimura/workspac...ase/gui/examples/TGR5'. Below this is a list of files and folders:

| Name         | Size     | Type     | Date Modified   |
|--------------|----------|----------|-----------------|
| distance.ha4 | 73 bytes | ha4 File | 20/07/... 14:49 |

The 'File name:' field is set to 'distance.ha4' and the 'Files of type:' dropdown is set to 'Tab-separated values (\*.ha4)'. The 'Save' button is highlighted. The status bar at the bottom indicates 'Tasks pending/ready/running: 0/1/0; ETA: 0s'.

## Saving the Accessible Volume files

1. Create "AV file" evaluator.
2. Select the labelling position, format (openDX or .xyz) and directory to save the files (write\_dir). If write\_dir is empty, directory of the PDB file is used. Once the AV File evaluator is saved, program will automatically simulate AVs and save AV clouds for each of the loaded PDBs.



3. AV cloud files can be now found in the write\_dir. In this example .dx format is used. Here Pymol software is used to view the resulting cloud.



