QSAR TOOLBOX

The OECD QSAR Toolbox for Grouping Chemicals into Categories

OECD QSAR Toolbox v.3.0

Step-by-step example of how to evaluate an ad-hoc category of aliphatic amines and to predict an ecotoxicological endpoint

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Background

- This is a step-by-step presentation designed to take you through the workflow of the Toolbox for evaluating of an ad-hoc category.
- You will learn several new functionalities which we be repeated to assure a consistent category is defined.
- It is assumed that you now have some experience in using the Toolbox so there will be multiple key strokes between screen shots.

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Outlook

- Background
- **Category evaluation - overview**

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Category Evaluation

Two phases of evaluation process

Phase I. **Evaluation of Category & Data Gap Filling:**

- Investigating the structural consistency of an ad hoc category (e.g., a category submitted by an industry consortium to a regulatory assessment program).
- Implementation of an ad hoc category building and data gap filling.

Phase II. **Extension of Category & Data Gap Filling:**

- Search for other analogues which are consistent with the submitted category.
- Data gap filling using new data matrix.

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Outlook

- Background
- Category evaluation – overview
  - **Phase I. Evaluation of Category & Data Gap Filling**
    - *Step 1: Investigating the structural consistency of an ad hoc category.*
      - ***Case study***

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Case Study

- The submission consists of a category with 19 aliphatic amines.
- The predicted ecotoxicological endpoint EC50, 48h, *D.magna* of 2-Butanamine(CAS 13952-84-6) will be reviewed.

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- Background
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  - **Phase I. Evaluation of Category & Data Gap Filling**
    - *Step 1: Investigating the structural consistency of an ad hoc category.*
      - *Case study*
      - ***Workflow***

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow

The following input workflow is used:

- Input the file for submission as an user list.(In this case input file Aliphatic amines.smi*)
- Evaluate the category applying the following profiling schemes:
  - USEPA categories
  - ECOSAR categorization
  - Aquatic toxicity MOA of action
  - Organic Functional Groups (nested)

Aliphatic amines.smi* - file is packed in the zip file with all power point tutorials

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    - *Step 1: Investigating the structural consistency of an ad hoc category.*
      - *Case study*
      - ***Workflow***
        - ***Input***

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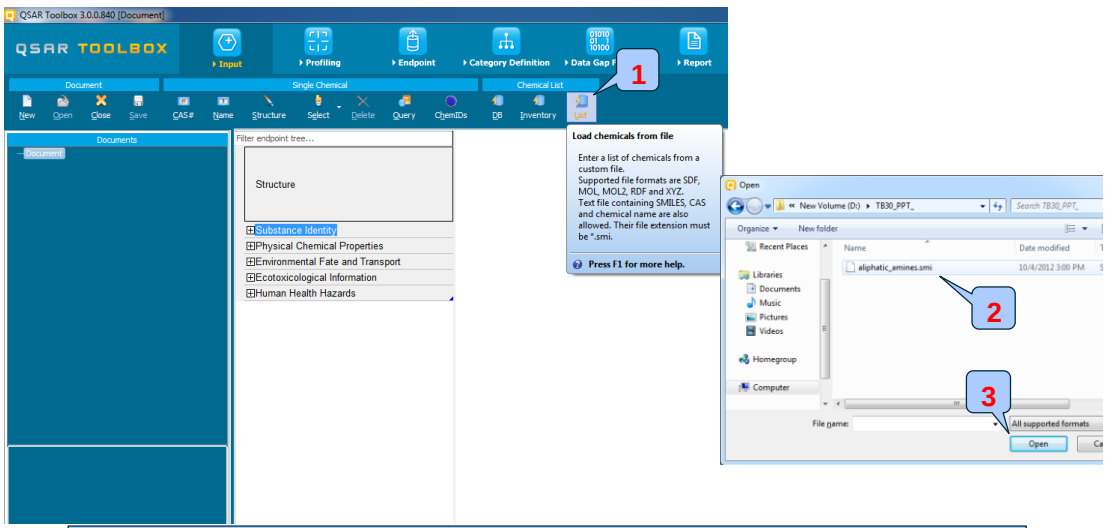
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Input



1. **Click** the List; 2. **Browse** and find the file for input 3. **Open** the file.

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Input

- You have now inserted your chemical list into the system.
- Click on the box next to "Substance Identity"; this displays the chemical identification information (see next screen shot).

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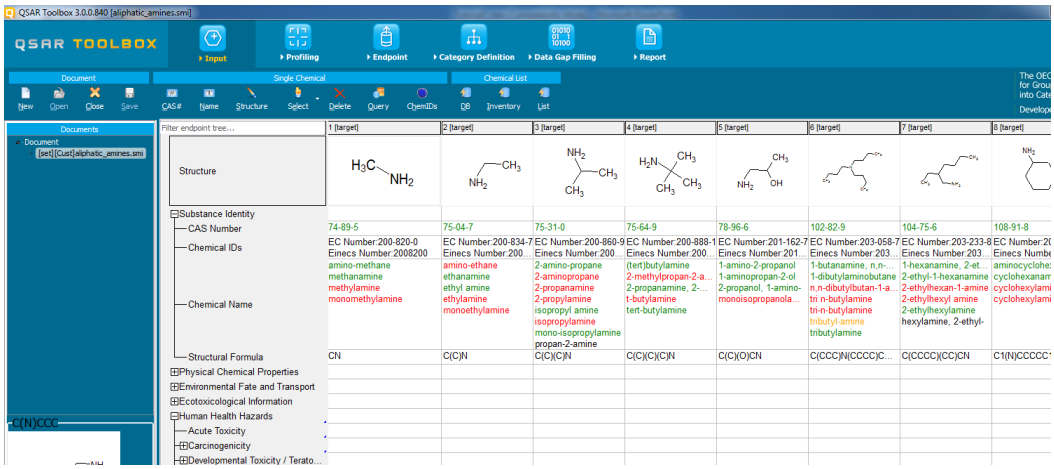
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Input



1. **Click** on "Substance identity"

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Outlook

- Background
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    - *Step 1: Investigating the structural consistency of an ad hoc category.*
      - *Case study*
      - ***Workflow***
        - *Input*
        - ***Profiling***

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling

- The first step of the category evaluation according to Phase I is the consistency check of the category which starts with evaluation of the robustness with respect to structural functionalities. The following schemes could be used for this purpose:
  - US EPA categories
  - MOA of action
  - ECOSAR categorization
  - Organic Functional Groups (nested)

- Select "profiling methods" by clicking on the boxes before the names of the profilers and Click "Apply". Before selecting the profiling methods unselect all (see next screen shot).

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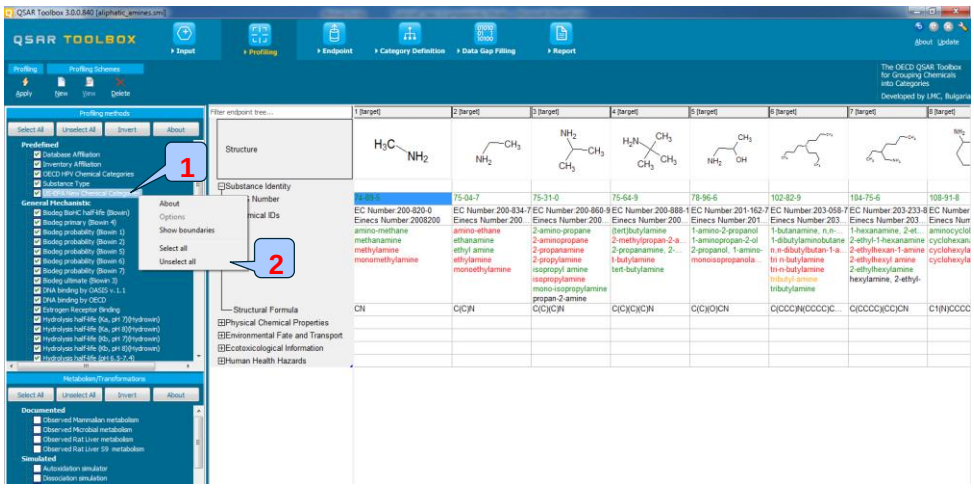
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling



1. **Right click** in space above the profilers; 2. **Select** Unselect All.

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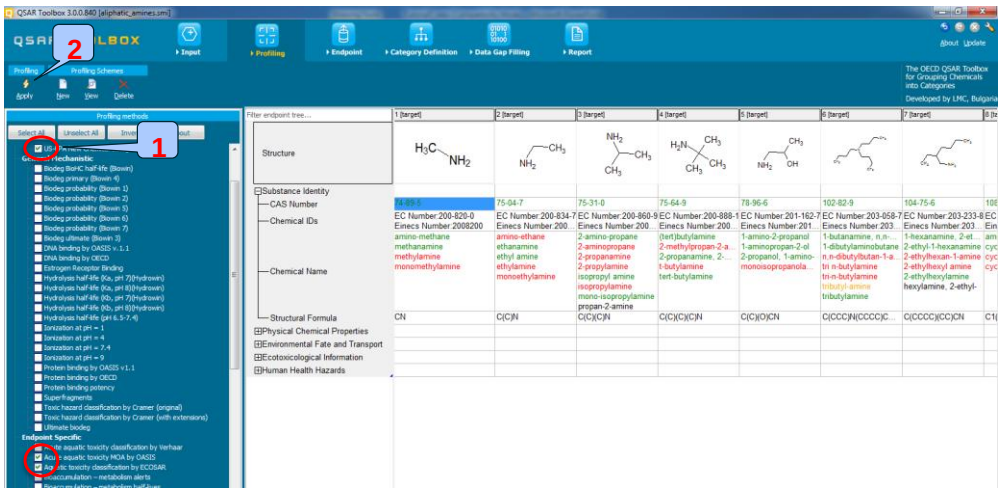
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling



1. **Check** the USEPA, MOA of action, ECOSAR and OFG(nested) profilers; 2. **Click** Apply.

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling

- The actual profiling will take several seconds depending on the number and type of selected profilers.
- The results of profiling automatically appeared as a dropdown box under the target chemical.
- The result from profile statistics, can be seen by right clicking in the space above the profiler in the endpoint tree and select Profile statistics from the dropdown menu (see next screen shot).

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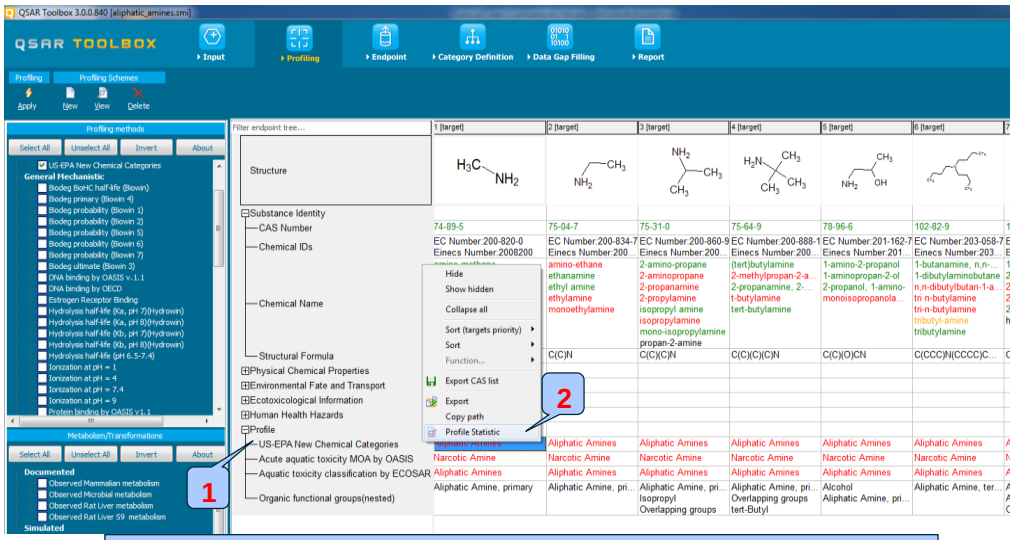
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling



1. **Right click** above US-EPA profiler; 2. **Select** Profile Statistics.

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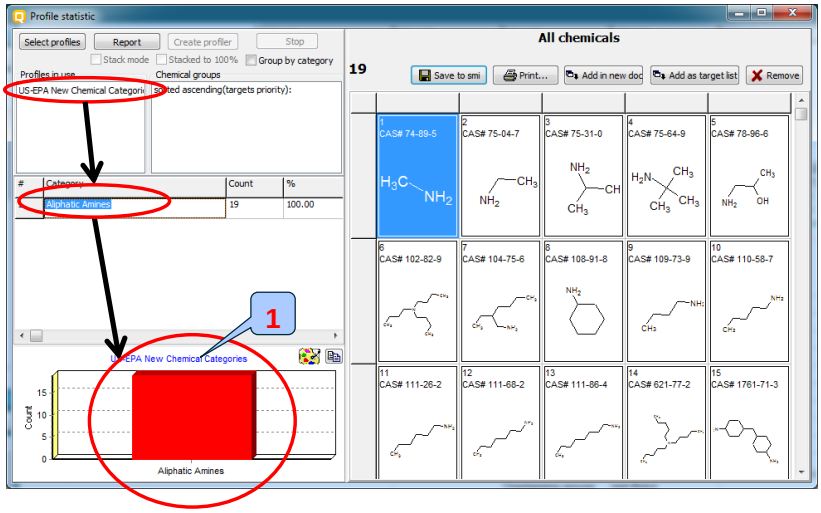
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling/Statistics according to US-EPA



1.In this case all 19 chemicals are Aliphatic amines according to US-EPA profiler.

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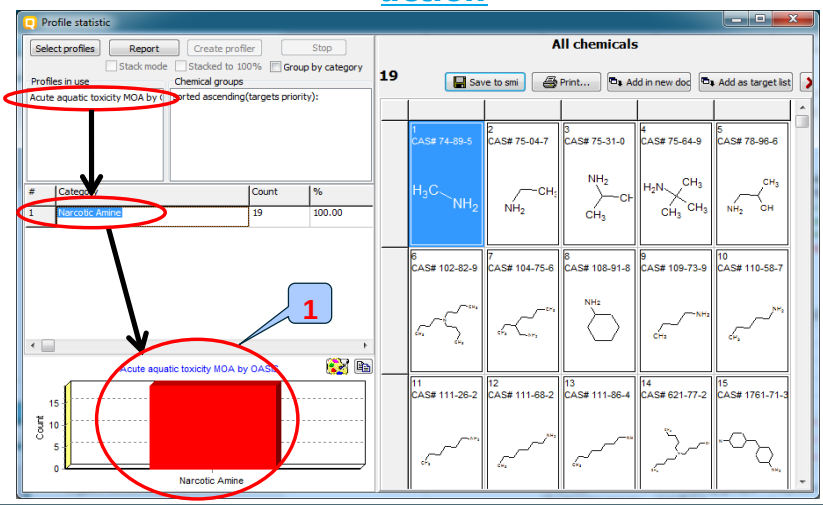
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling/Statistics according to OASIS MOA of action



1.In this case all 19 chemicals are Narcotic amines according to MOA of action profiling scheme

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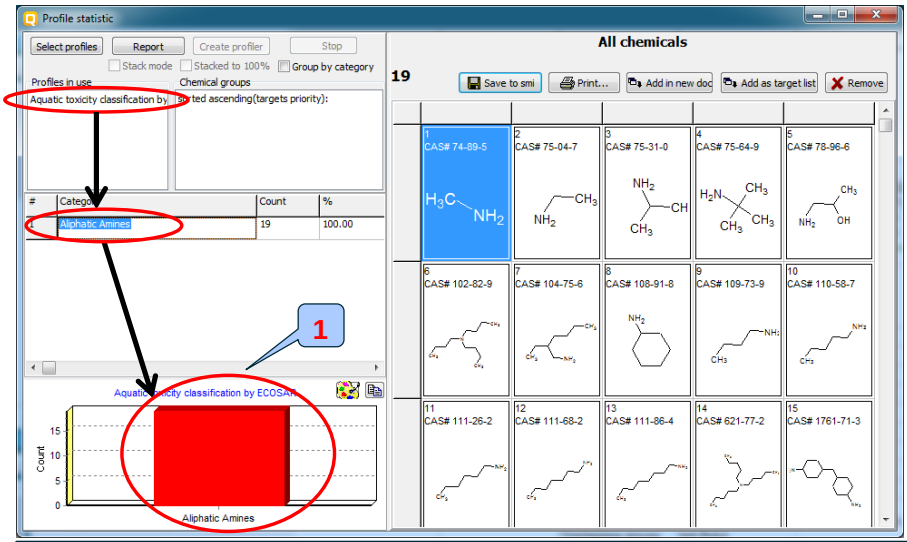
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling/Statistics according to ECOSAR



1.In this case all 19 chemicals are Aliphatic amines according to ECOSAR scheme

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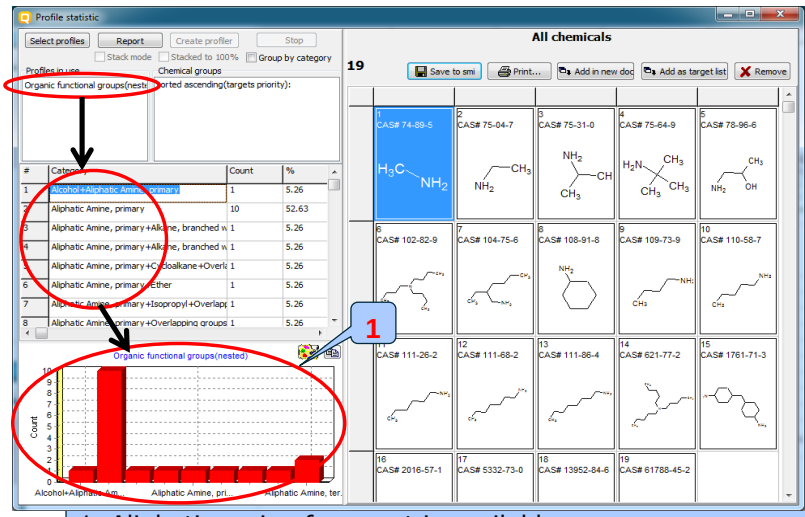
Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Workflow/Profiling/Statistics according to OFG(nested)



1. Aliphatic amine fragment is available.

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Phase I: Evaluation of Category & Data Gap Filling

Step 1. Investigating the structural consistency of an ad hoc category

Recap

- **Chemicals are defined as:**
  - Aliphatic amines (broader category than primary amines) – US EPA categories, ECOSAR classification
  - The statistics of organic functional groups provide detailed alert description of all 19 structures. However, all 19 chemicals have aliphatic amines fragment.
- **It could be concluded that the category is consistent with respect to structural functionalities (chemicals are empirically similar).**

QSAR TOOLBOX

Outlook

- Background
- Category evaluation – overview
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    - *Step 1: Investigating the structural consistency of an ad hoc category.*
    - *Step 2: Investigating the a*pplicability domain of an ad hoc category.

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Phase I: Evaluation of Category & Data Gap Filling

Step 2. Investigating the applicability domain of an ad hoc category

- Boundaries of structural functionalities
  - Aliphatic amines
- Parametric boundaries
  - log Kow (from 0.64 to 7.71)
  - Molecular weight (from 31 to 269 Da)
  - Water solubility (from 0.48×10^{-1} to 1×10^{6} mg/l)

You are now ready to extract the 2D and/or 3D parameters (see next screen shot).

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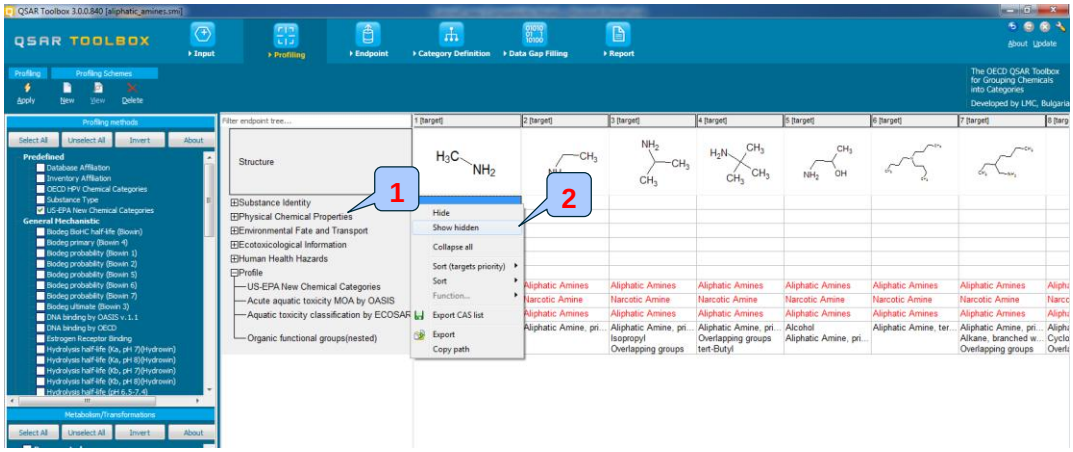
Phase I: Evaluation of Category & Data Gap Filling

Step 2. Investigating the applicability domain of an ad hoc category

Extracting 2D and 3D parametrs



1. Right click above endpoint tree; 2. **Select** Show hidden.

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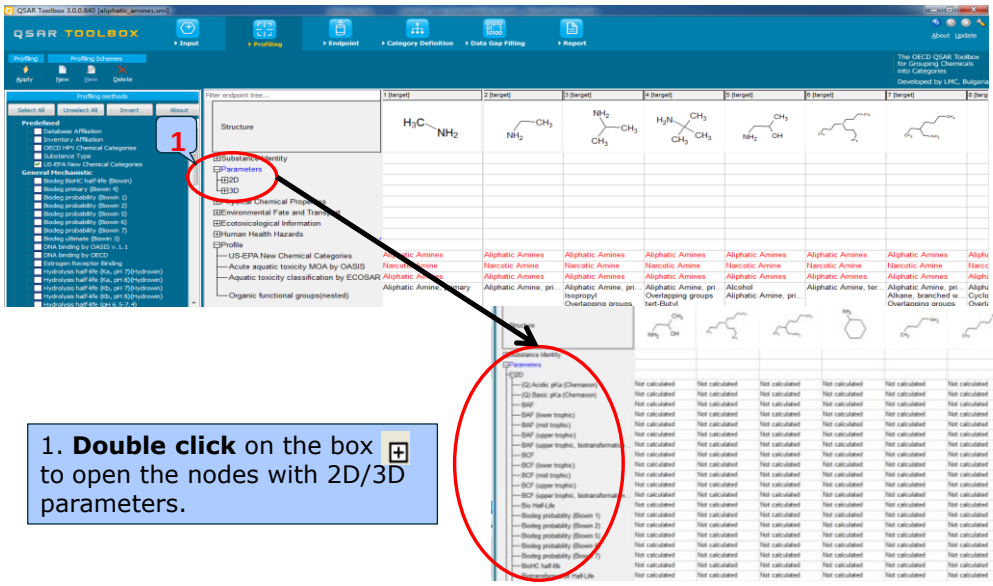
Phase I: Evaluation of Category & Data Gap Filling

Step 2. Investigating the applicability domain of an ad hoc category

Extracting 2D and 3D parametrs



1. **Double click** on the box ⊞ to open the nodes with 2D/3D parameters.

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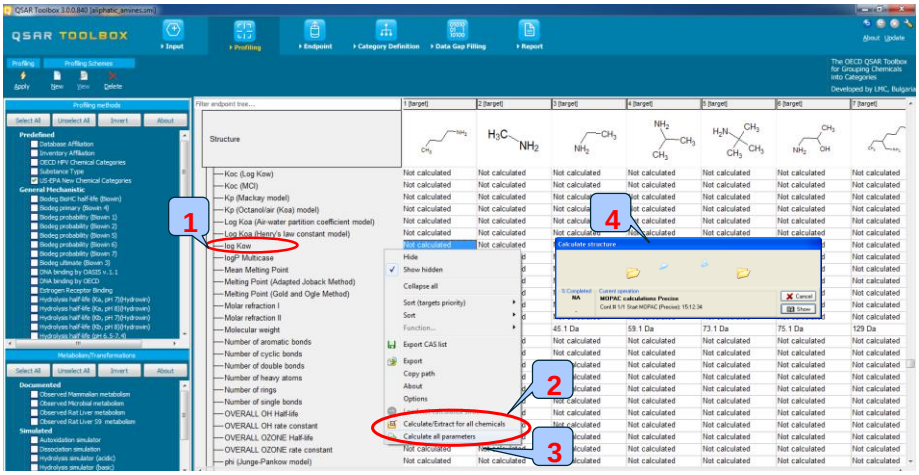
Phase I: Evaluation of Category & Data Gap Filling

Step 2. Investigating the applicability domain of an ad hoc category

Extracting 2D and 3D parametrs



1. **Right click** above the parameter in the endpoint tree; 2. The user can calculate all parameters or extract current parameter for all chemicals in the current row. 3. In this exercise calculate all (2D) parameters.

If some of the parameters (in case of new chemicals) are not calculated then the message (4) appears If you want to stop the calculation process, click the **Cancel** button.

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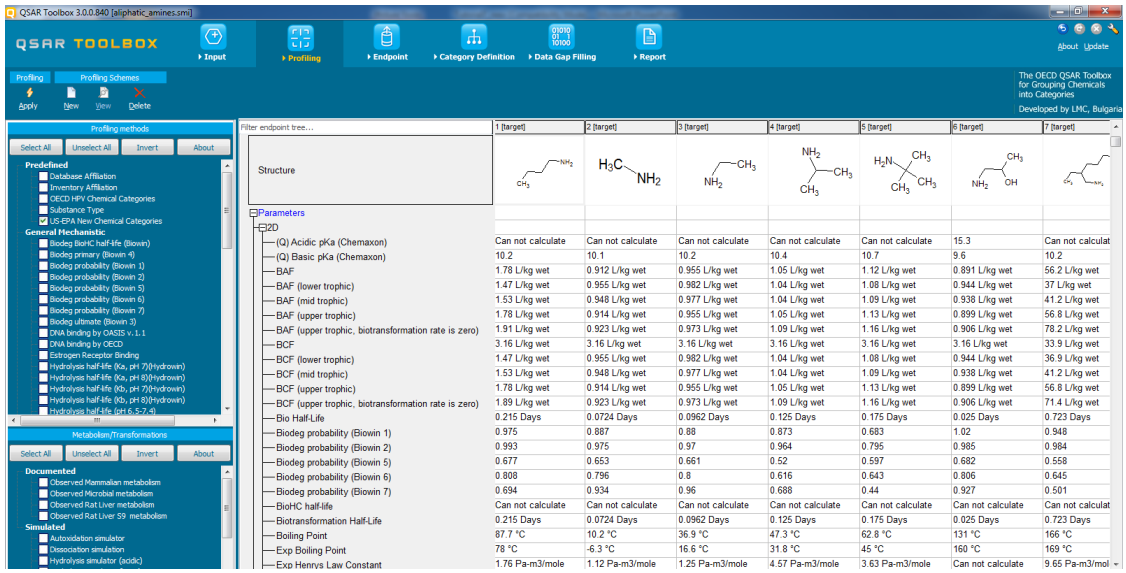
Phase I: Evaluation of Category & Data Gap Filling

Step 2. Investigating the applicability domain of an ad hoc category

Extracting 2D and 3D parametrs - results



QSAR TOOLBOX

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    - *Step 3: Reading data for the analogues*

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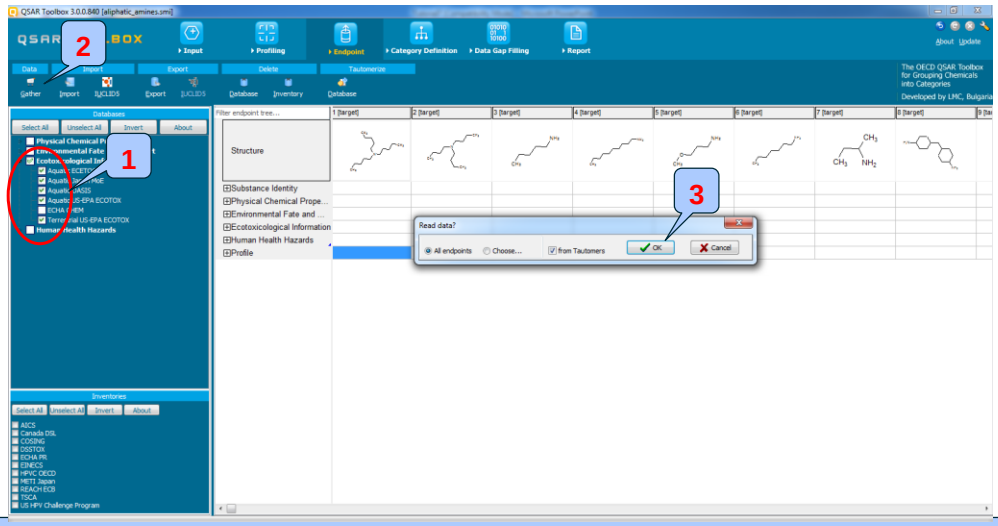
Phase I: Evaluation of Category & Data Gap Filling

Step 3. Reading data

- Next, the Data matrix is constructed by extracting available experimental results for all 19 members of the category.
- Based on preceding category evaluation, no outliers have been identified violating the structural and mechanistic consistency of the category.

Phase I: Evaluation of Category & Data Gap Filling

Step 3. Reading data



1. **Select** databases related to aquatic toxicity; 2. **Click** Gather; 3. **Click** OK.

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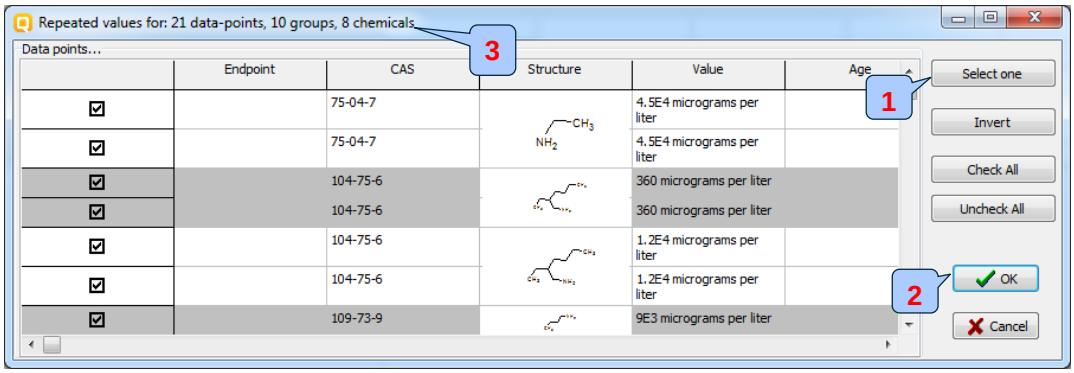
Phase I: Evaluation of Category & Data Gap Filling

Step 3. Reading data

Due to the overlap between the Toolbox databases same data for intersecting chemicals could be found simultaneously in more than one database. The data redundancy is identified and the user has the opportunity to select either a single data value or all data values.



1. Click Select one; 2. **Click** OK
3. The indication for repeating values

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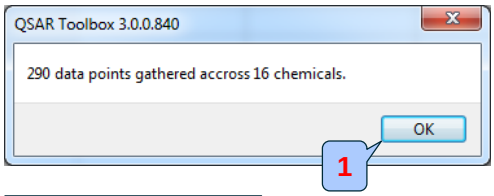
Phase I: Evaluation of Category & Data Gap Filling

Step 3. Reading data

The system automatically gives indication for the number of gather experimental data points



1. **Click** OK

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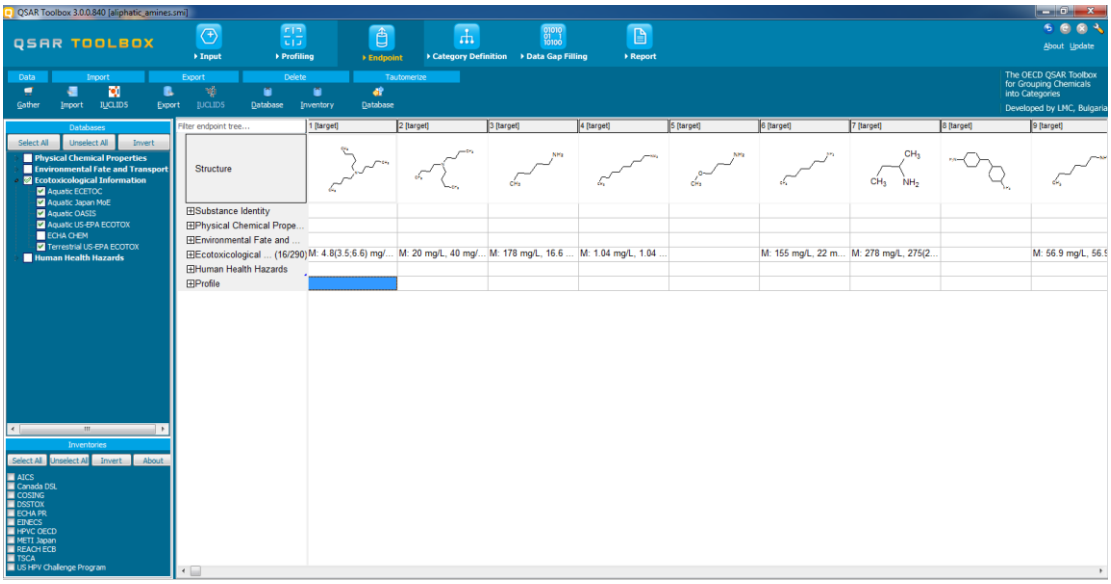
Phase I: Evaluation of Category & Data Gap Filling

Step 3. Reading data.

Inserting data into the data matrix



QSAR TOOLBOX

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    - *Step 3: Reading data for the analogues*
    - *Step 4: Data gap filling for 2-Butanamine*

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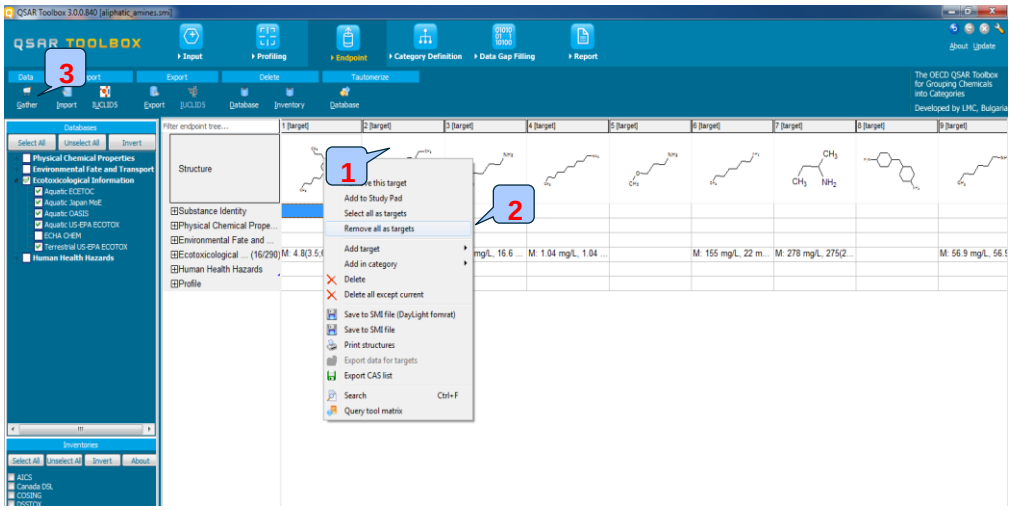
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine

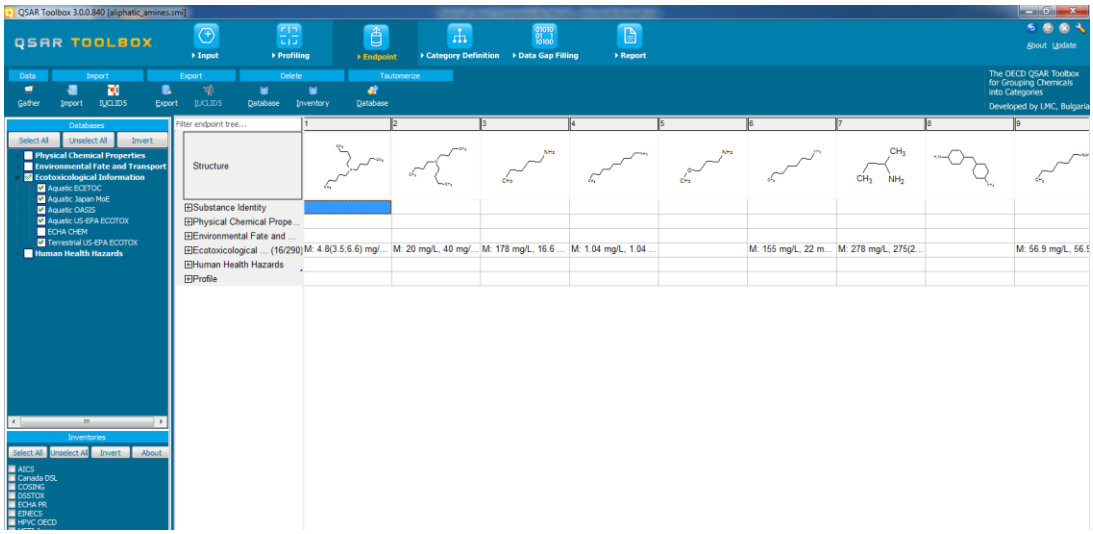


1. **Right click** above the target chemicals. 2. **Select** Remove all as targets.3. **Go** to Data Gap Filling

QSAR TOOLBOX

Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine



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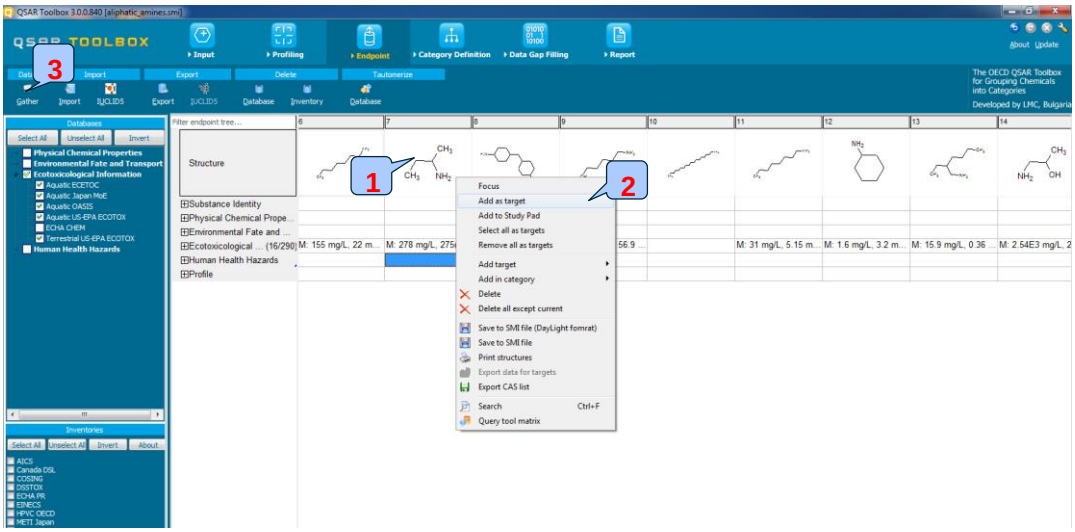
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine



1. **Right click** above the target chemical (2-Butanamine); 2. **Select** Add as target.3. **Go** to Data Gap Filling

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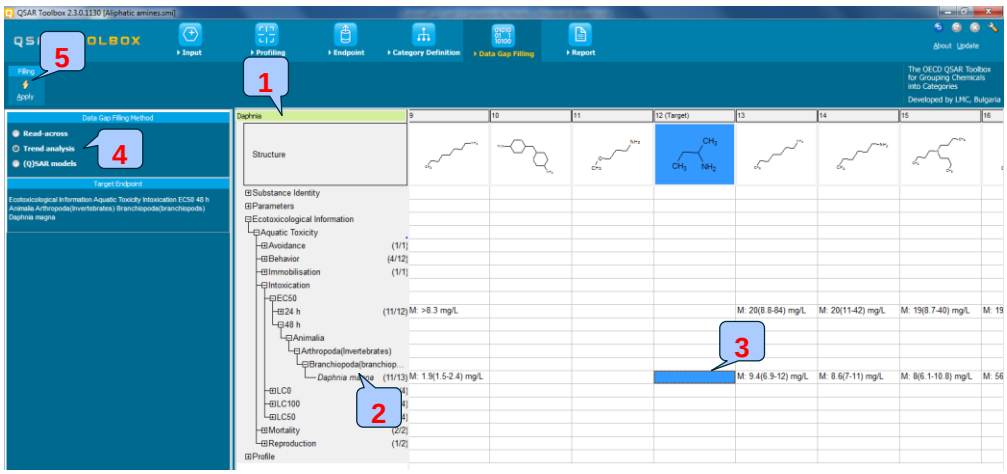
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine

Navigate to the target endpoint: Intoxication, D.magna,EC50, 48h



1. **Type** Daphnia in the filter field; 2. Navigate to target endpoint by opening the nodes and 3. **Highlight** the gap which will be filled in; 4. **Select** Trend analysis; 5. **Click** Apply.

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    - *Step 3: Reading data for the analogues*
    - *Step 4: Data gap filling for 2-Butanamine*
      - *Step 4.1*: How to reorder the endpoint tree*

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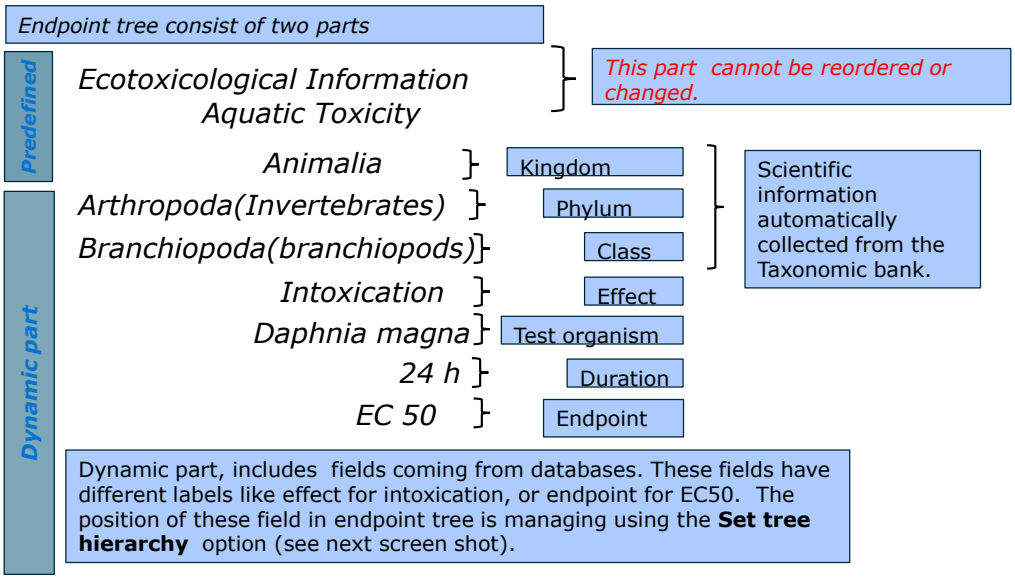
Phase I: Evaluation of Category & Data Gap Filling

Step 4.1*. How to reorder the endpoint tree

* Detailed information describing how to reproduce the order of endpoint tree shown in the current presentation



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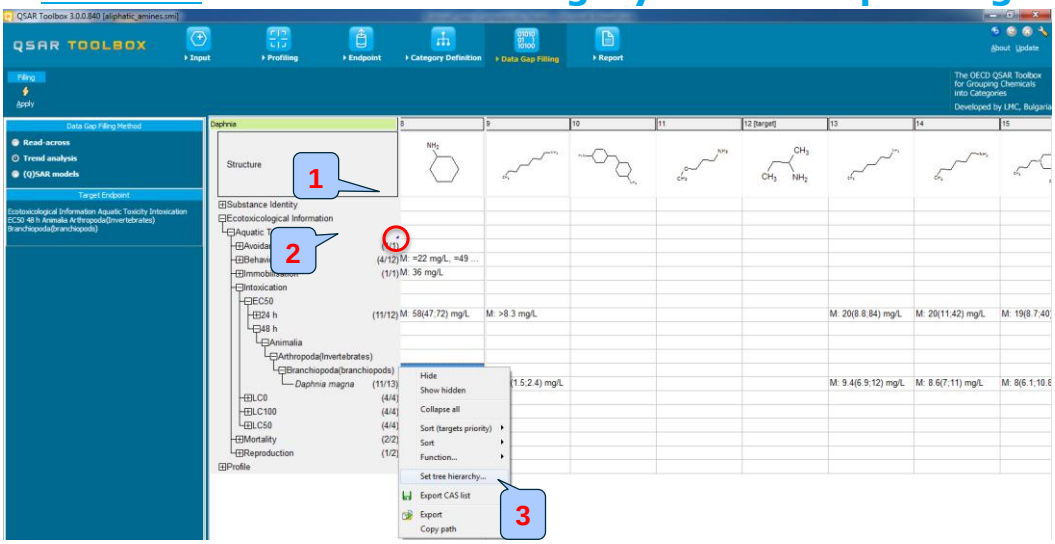
Phase I: Evaluation of Category & Data Gap Filling



1. The little blue triangle is indication that the endpoint tree below is ordered. To see order of nodes of the tree, 2. **Right click** above endpoint tree near this triangle; 3. Select Set tree hierarchy.

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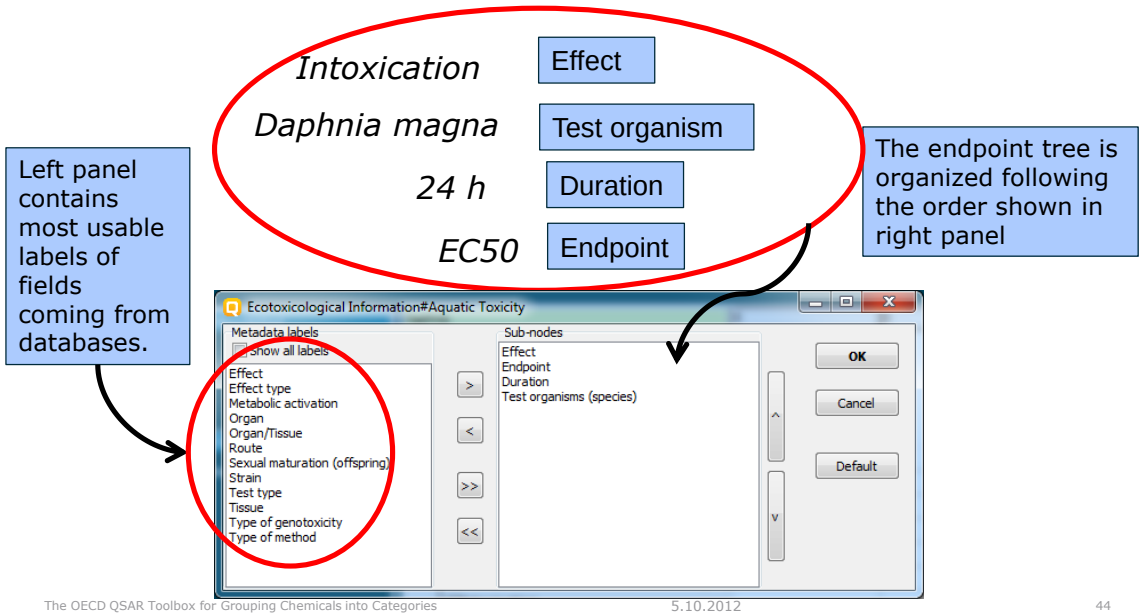
Phase I: Evaluation of Category & Data Gap Filling

Step 4.1. How to reorder the endpoint tree



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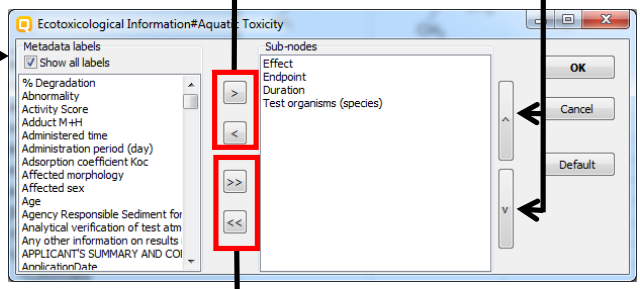
Phase I: Evaluation of Category & Data Gap Filling

Step 4.1. How to reorder the endpoint tree

All available fields coming from databases could be shown when the box **Show all labels** is selected.

To remove or change some of the fields which are visualizing on the tree, highlight the field from left panel and move it to the right panel using the **Right** or **Left** button.

Reordering of the fields is possible using the **Up** and **Down** buttons.



To move all fields form one panel to another use **Right** and **Left** button.

QSAR TOOLBOX

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    - *Step 3: Reading data for the analogues*
    - *Step 4: Data gap filling for 2-Butanamine*
      - *Back to Trend analysis*

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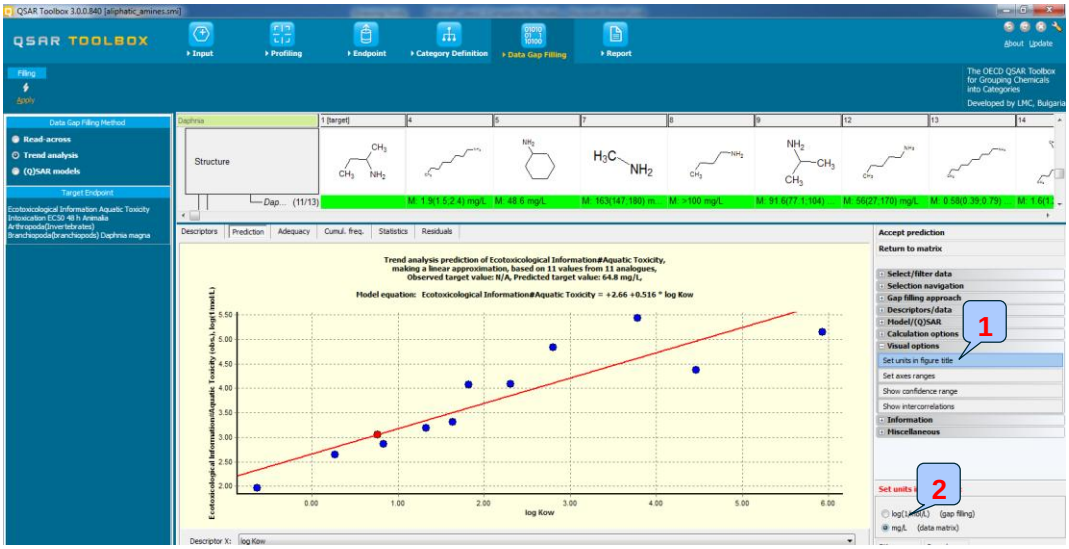
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine



1. To see predicted value in log (1 mol/L) go to **Visual options** and select **Set unit in figure title**; 2. **Select mg/L.**

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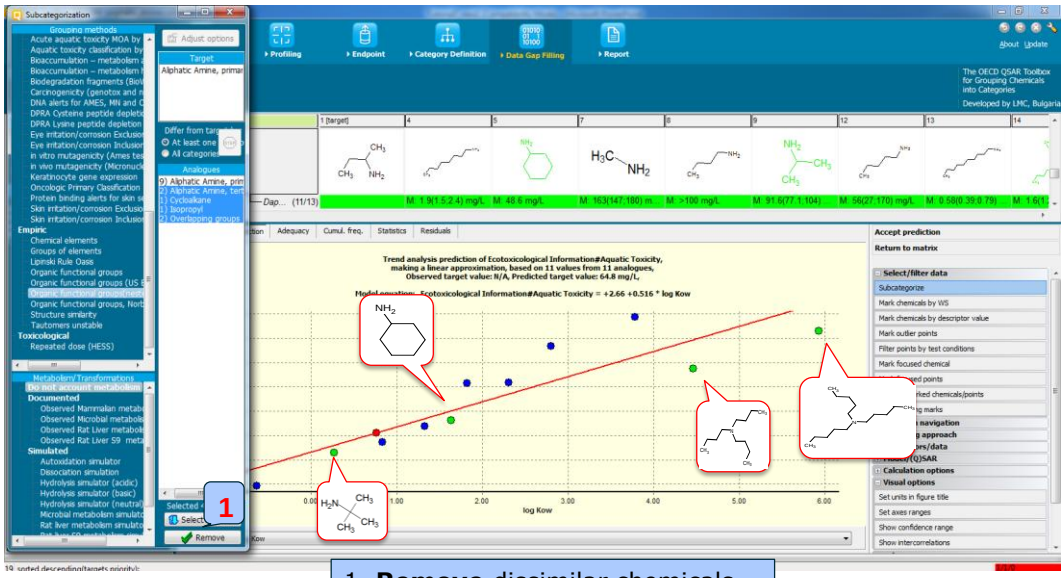
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine/*Subcategorize by OFG(nested)*



1. **Remove** dissimilar chemicals

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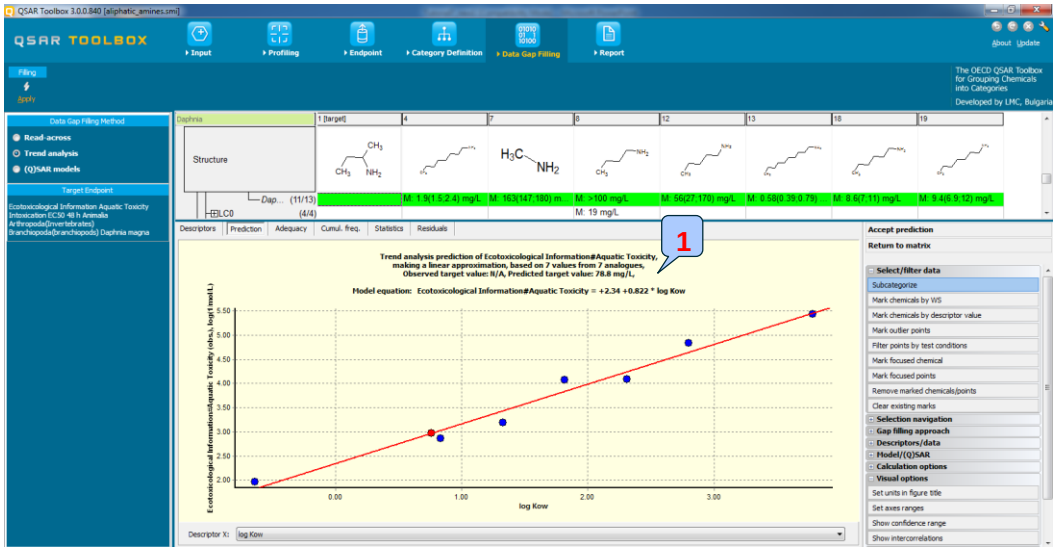
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine/*Predicted result*



1. Predicted result is 78.8 mg/l

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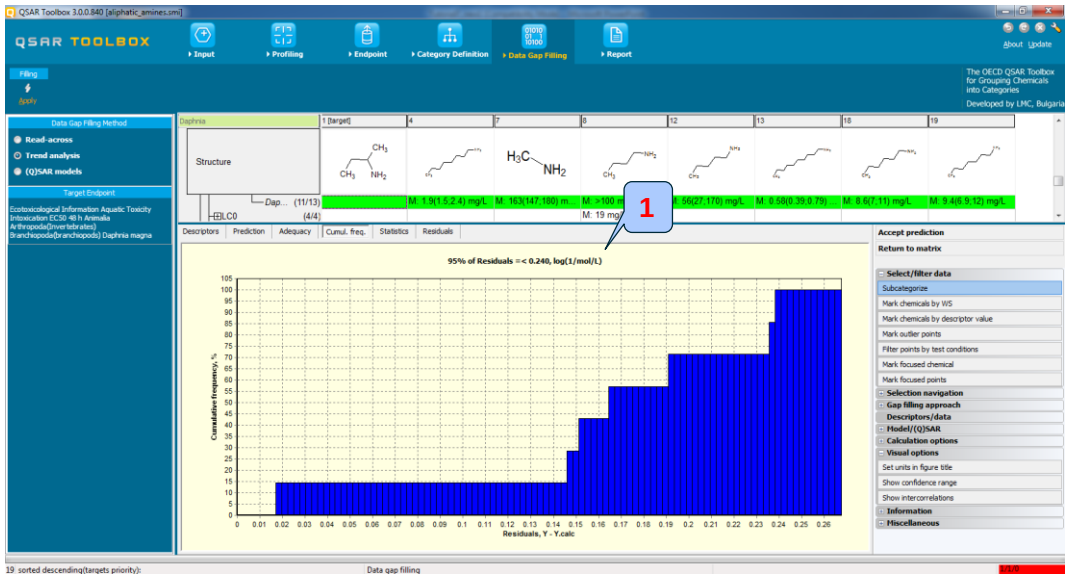
Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine/*Cumulative frequency*

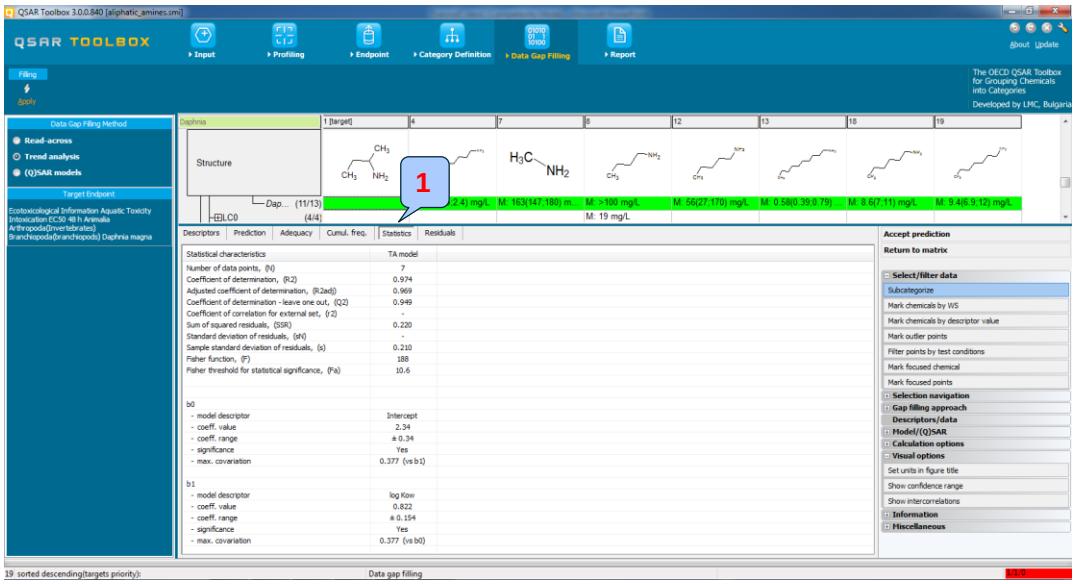


1. Cumulative frequency is less then 0.25 log units

QSAR TOOLBOX

Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine/*Statistics*



QSAR TOOLBOX

Phase I: Evaluation of Category & Data Gap Filling

Step 4. Data Gap Filling for 2-Butanamine/*Interpretation of the result*

- The structurally similar analogs across category of aliphatic amines is used for data gap filling
- Subcategorization by OFG(nested) is applied
- The prediction based on the defined category is acceptable.
- The predicted value based on predefined category of aliphatic amines is 78.8 mg/l

QSAR TOOLBOX

Outlook

- Background
- Category evaluation – overview
  - Phase I. Evaluation of Category & Data Gap Filling
  - **Phase II. Extension of the Category& Data gap Filling**

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

- The extension of the category is performed by using Phase II of the category evaluation process (*Extension of Category & Data Gap Filling*). Other analogues are searched in the Toolbox, which are structurally and mechanistically consistent with the predefined category.
- The structural analogues could be defined using ECOSAR grouping

QSAR TOOLBOX

Outlook

- Background
- Category evaluation – overview
  - Phase I. Evaluation of Category & Data Gap Filling
  - **Phase II. Extension of the Category& Data gap Filling**
    - Step 1: Category definition

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition

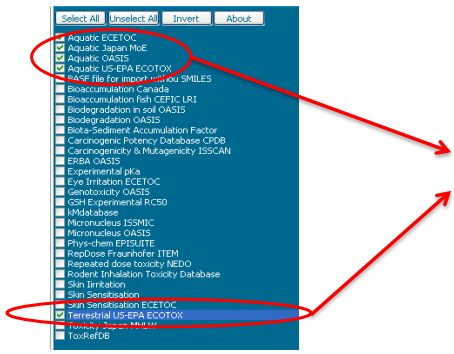
- The ECOSAR strict category is used to define an broader category used in further analysis.
- According to ECOSAR categorization **a list of 318 aliphatic amines have been selected** for which data are available in the Toolbox (identified in the Toolbox databases).
- The same endpoint: EC 50 48h *D.magna* will be predicted as with the predefined category

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition

- Define ECOSAR category
- Before defining the category, the following databases related to the predicted endpoint are selected:

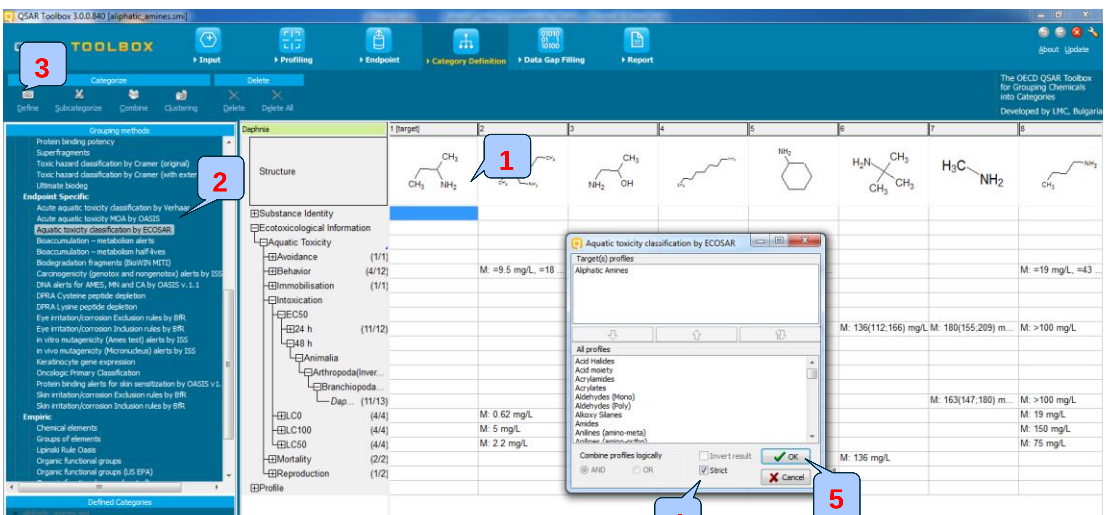


Aquatic ECETOC
Aquatic Japan MoE
Aquatic OASIS
Aquatic US-EPA ECOTOX
Terrestrial US-EPA ECOTOX

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition/*Defining ECOSAR (strict)*

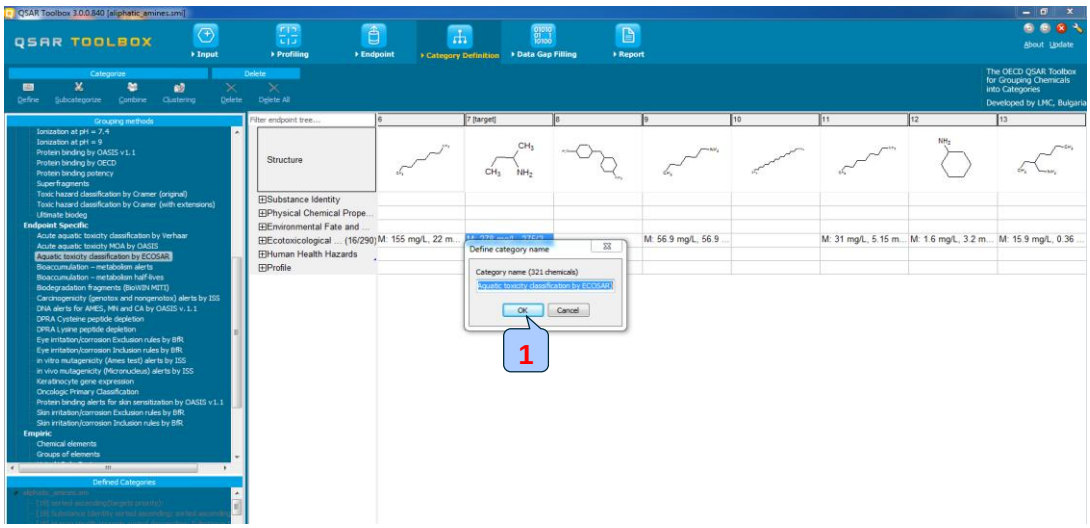


1. **Select** the target chemical 2. **Highlight** "Aquatic toxicity classification by ECOSAR"; 3. **Click** Define; 4. **Select** Strict. 5. **Click** OK

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition/*Defining ECOSAR (strict)*



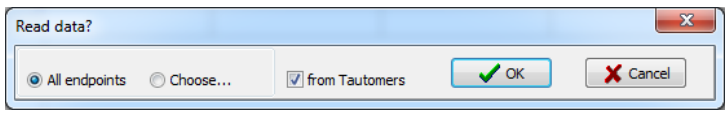
1. **Click** OK to confirm the name of the category.

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition/*Reading data*

- The Toolbox will now retrieve those chemicals that have the same protein binding mechanism than the target compound.
- The Toolbox automatically request the user to select the endpoint that should be retrieved.
- The user can either select the specific endpoint or by default choose to retrieve data on all endpoints (see below).

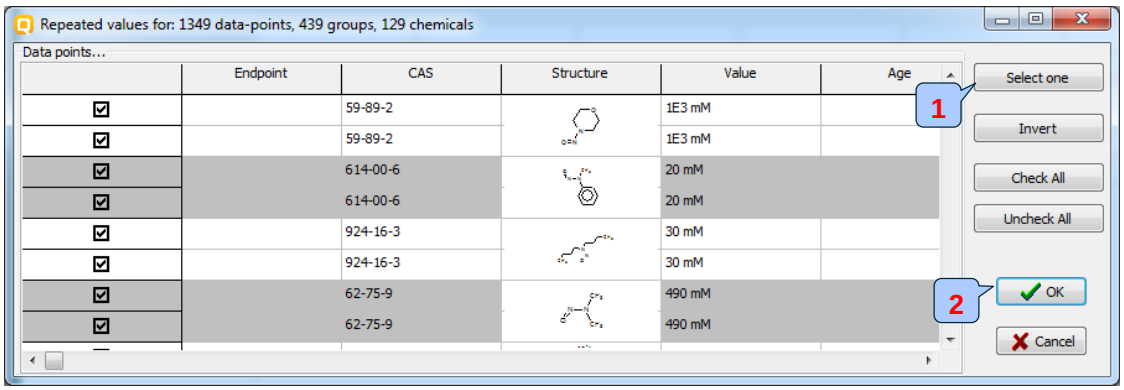


- In this example, as only databases are selected that contain information for aquatic toxicity endpoint, both options give the same results.

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition/*Reading data*



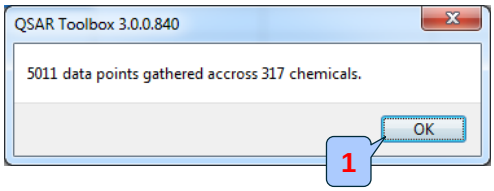
1. **Click** Select one and then; 2. **Click** OK.

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 1: Category definition/*Reading data*

The system automatically gives indication for the number of gather experimental data points



1. **Click** OK

QSAR TOOLBOX

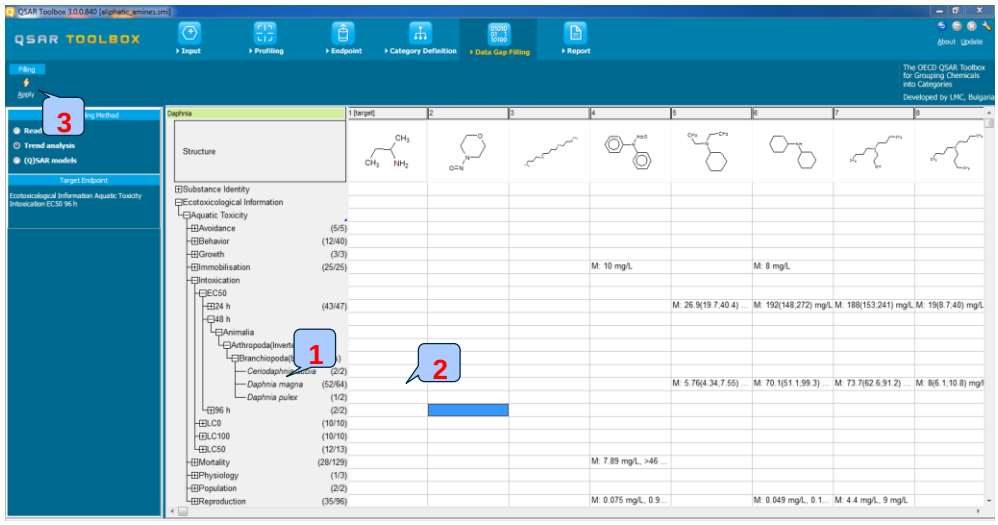
Outlook

- Background
- Category evaluation – overview
  - Phase I. Evaluation of Category & Data Gap Filling
  - **Phase II. Extension of the Category& Data gap Filling**
    - Step 1: Category definition
    - Step 2: Navigate to the target endpoint

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 2: Navigate to the target endpoint/*EC 50, 48h, D.magna*



1. Navigate to target endpoint by **opening the nodes**; 2. **Highlight the gap** which will be fill in. 3. **Move** to Data Gap Filling

QSAR TOOLBOX

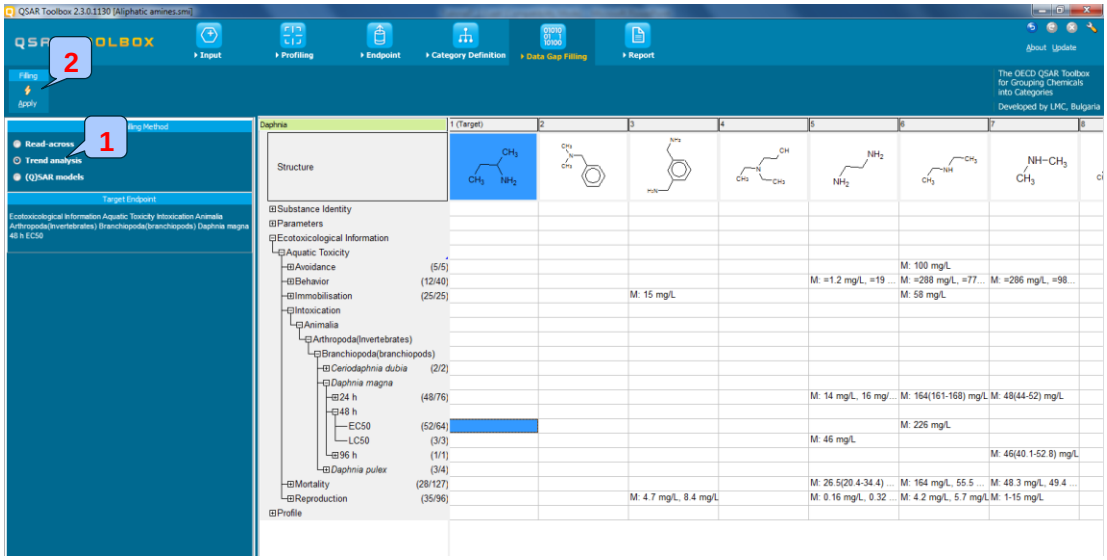
Outlook

- Background
- Category evaluation – overview
  - Phase I. Evaluation of Category & Data Gap Filling
  - **Phase II. Extension of the Category& Data gap Filling**
    - Step 1: Category definition
    - Step 2: Navigate to the target endpoint
    - Step 3: Data Gap Filling

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling

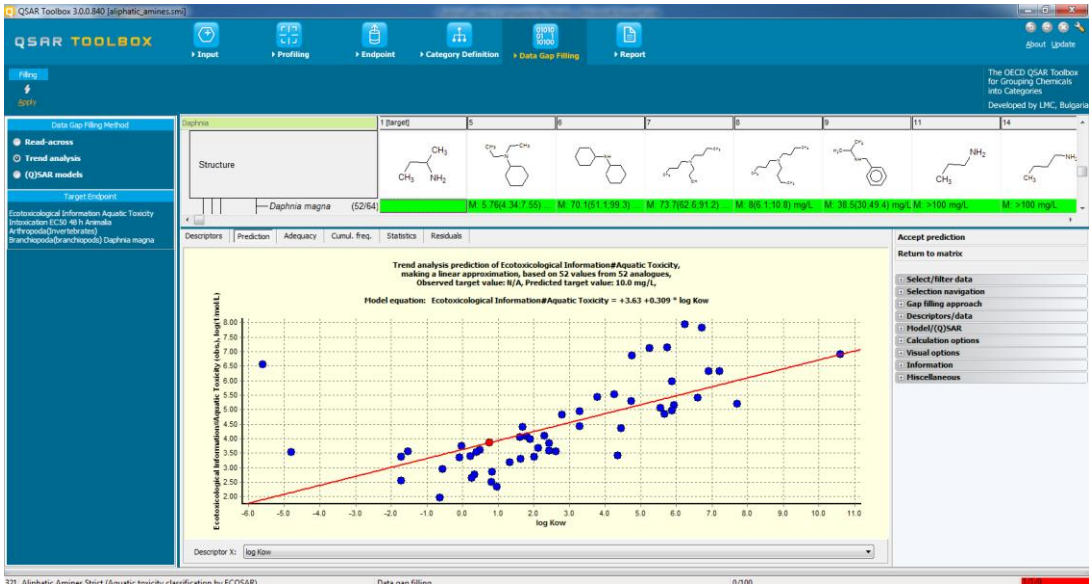


1. **Select** Trend analysis; 2. **Click** Apply

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

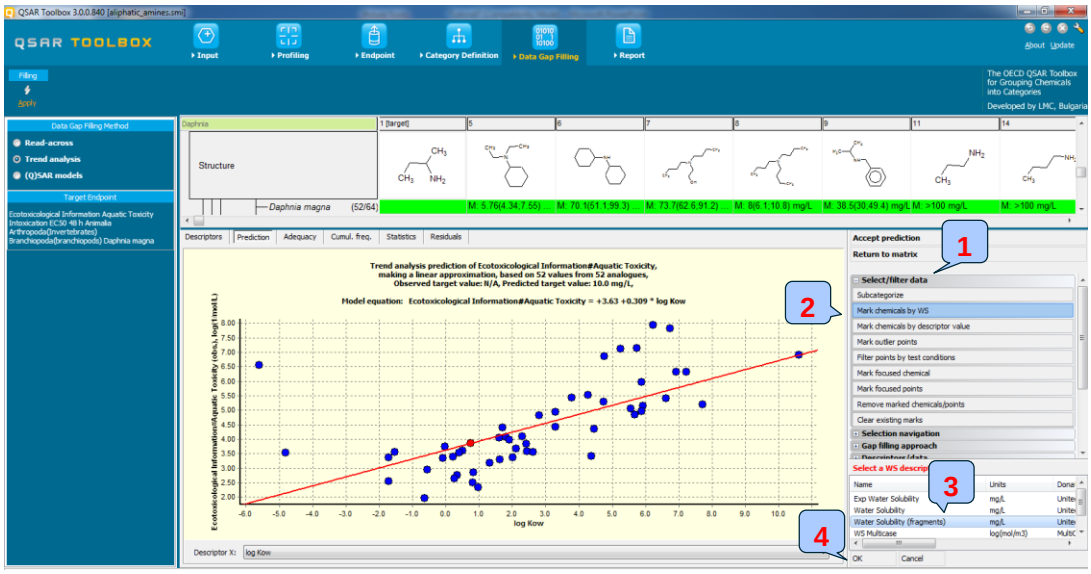
Step 3: Data Gap Filling



QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling/*Subcategorize by Water solubility*

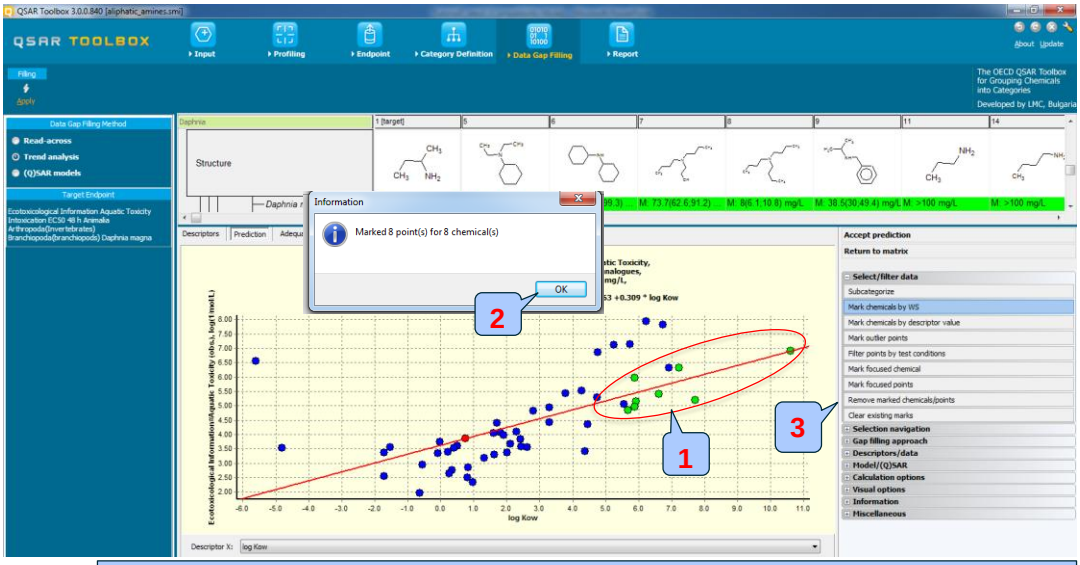


1. **Open** Select/Filter data panel; 2. **Click** Mark chemical by Sw; 3. **Select** Water solubility (fragments); 4. **Click** OK

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling/*Subcategorize by Water solubility*

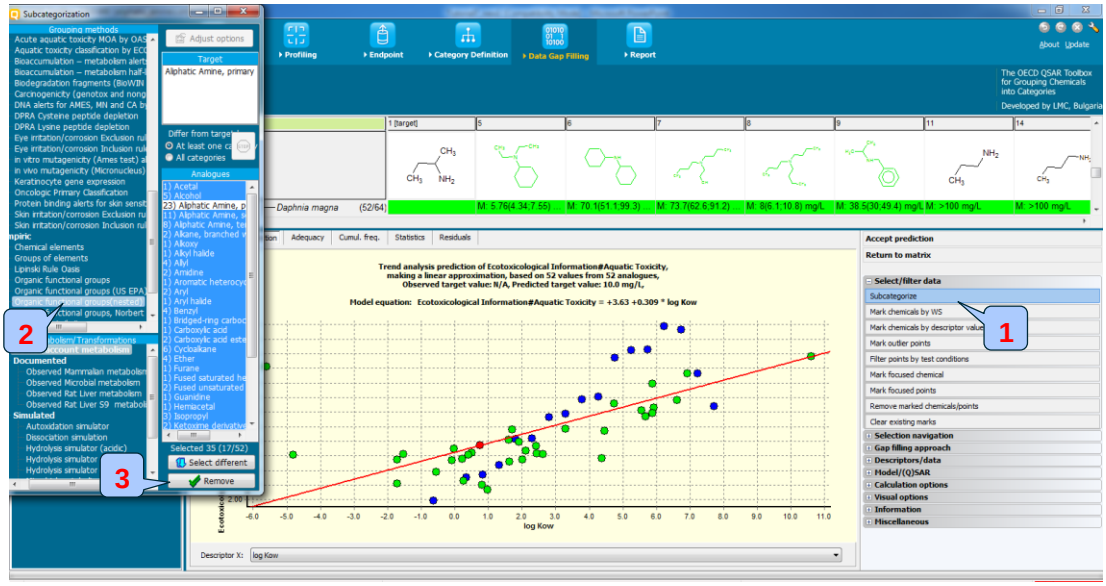


1. Eight chemicals are marked in green. 2.**Click** OK. 3. **Remove** marked chemicals

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling/*Subcategorize by OFG(nested)*



1. **Click** Subcategorize 2. **Select** OFG(nested) 3. **Remove** dissimilar chemicals

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

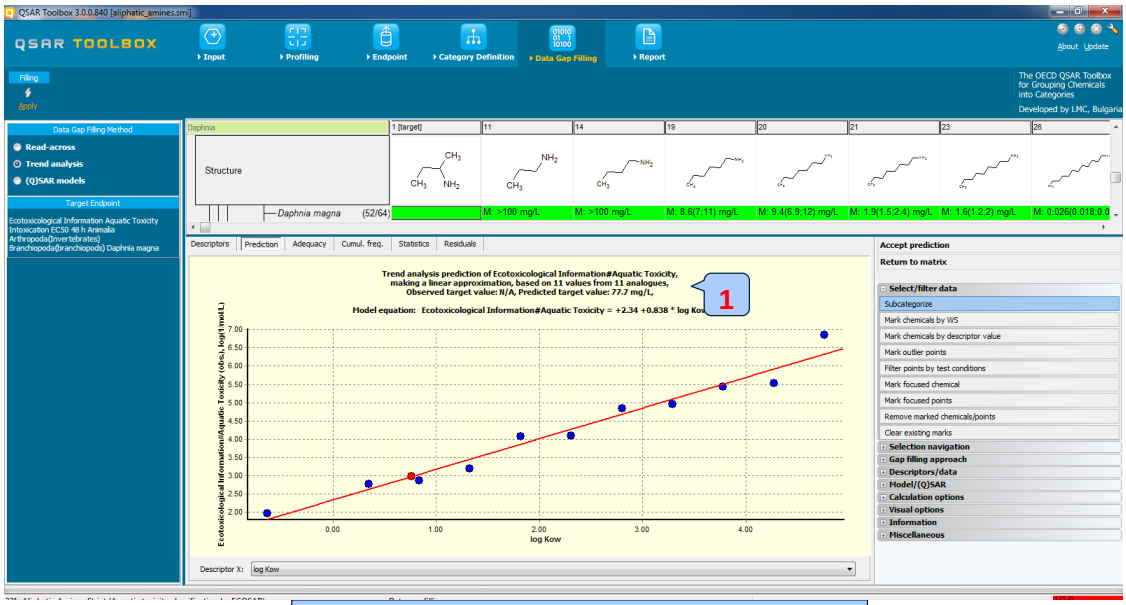
Step 3: Data Gap Filling/*Subcategorize by Lipinski rules*

Chemicals with very long chain could be removed from the category due to their non-bioavailability.
1. **Select** Lipinski rules 2. **Double click** to see "Non-bioavailable" chemicals 3. **Close** the appeared window
4. **Remove** dissimilar chemicals

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling/*Prediction result*

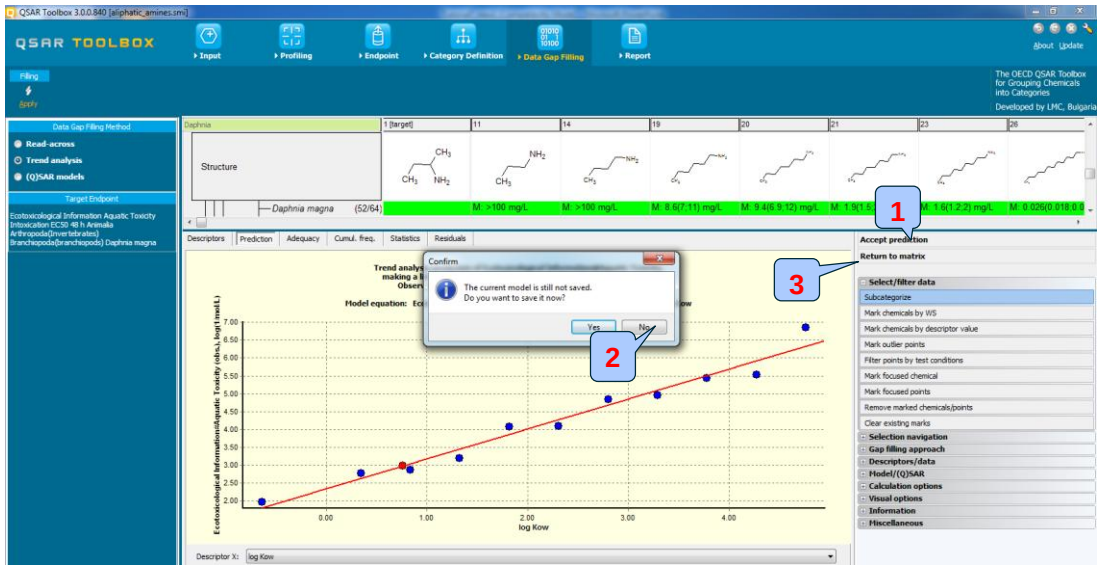


Prediction result is 77.7 mg/l.

QSAR TOOLBOX

Phase II: Extension Category & Data gap filling

Step 3: Data Gap Filling/*Accept the Prediction result*



1. Click on Accept prediction. If you want to save the model click Yes, otherwise click No; 2. **Click** No; 3. **Click** Return to the matrix