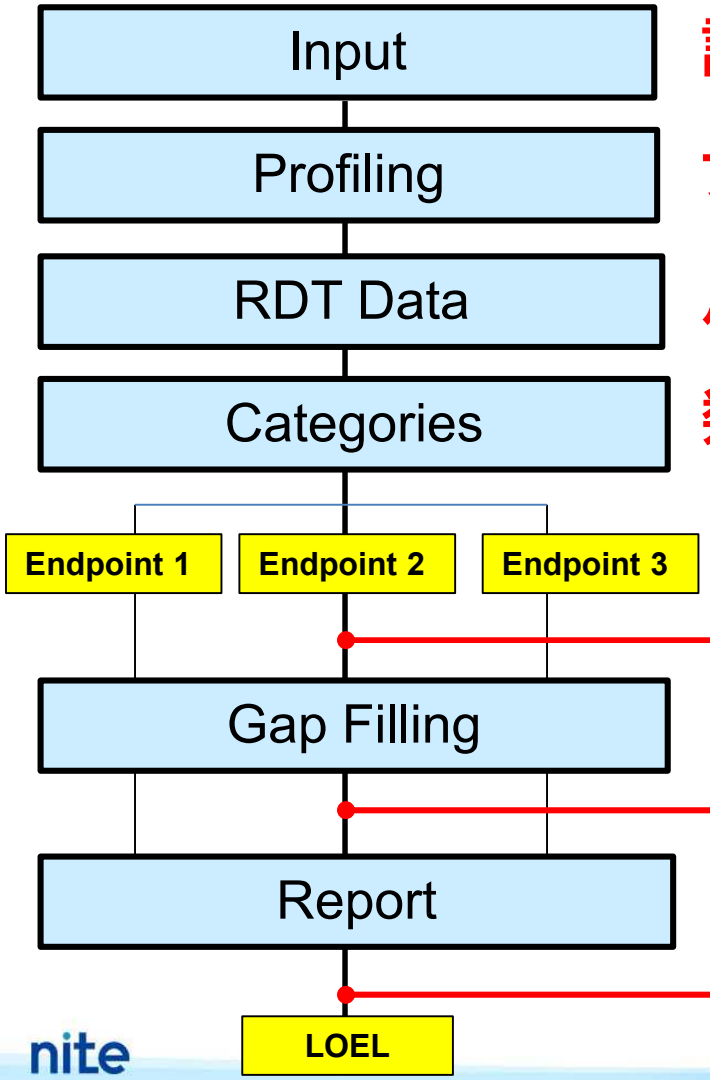
HESSの操作説明

Read-acrossによる反復投与毒性の予測

（独）製品評価技術基盤機構
化学物質管理センター
安全審査課

HESSによる反復投与毒性のデータギャップ補完のワークフロー（OECD Toolboxに準拠）



評価対象物質の入力

プロファイリング（該当カテゴリー等）の抽出

反復投与毒性試験データの抽出

類似物質の抽出

HESS DB等による類似物質のエビデンスの確認（専門家の知見活用）
→必要に応じてサブカテゴリー化

各エンドポイントに対するNOEL/LOELの推定値を求める

レポートを自動作成します

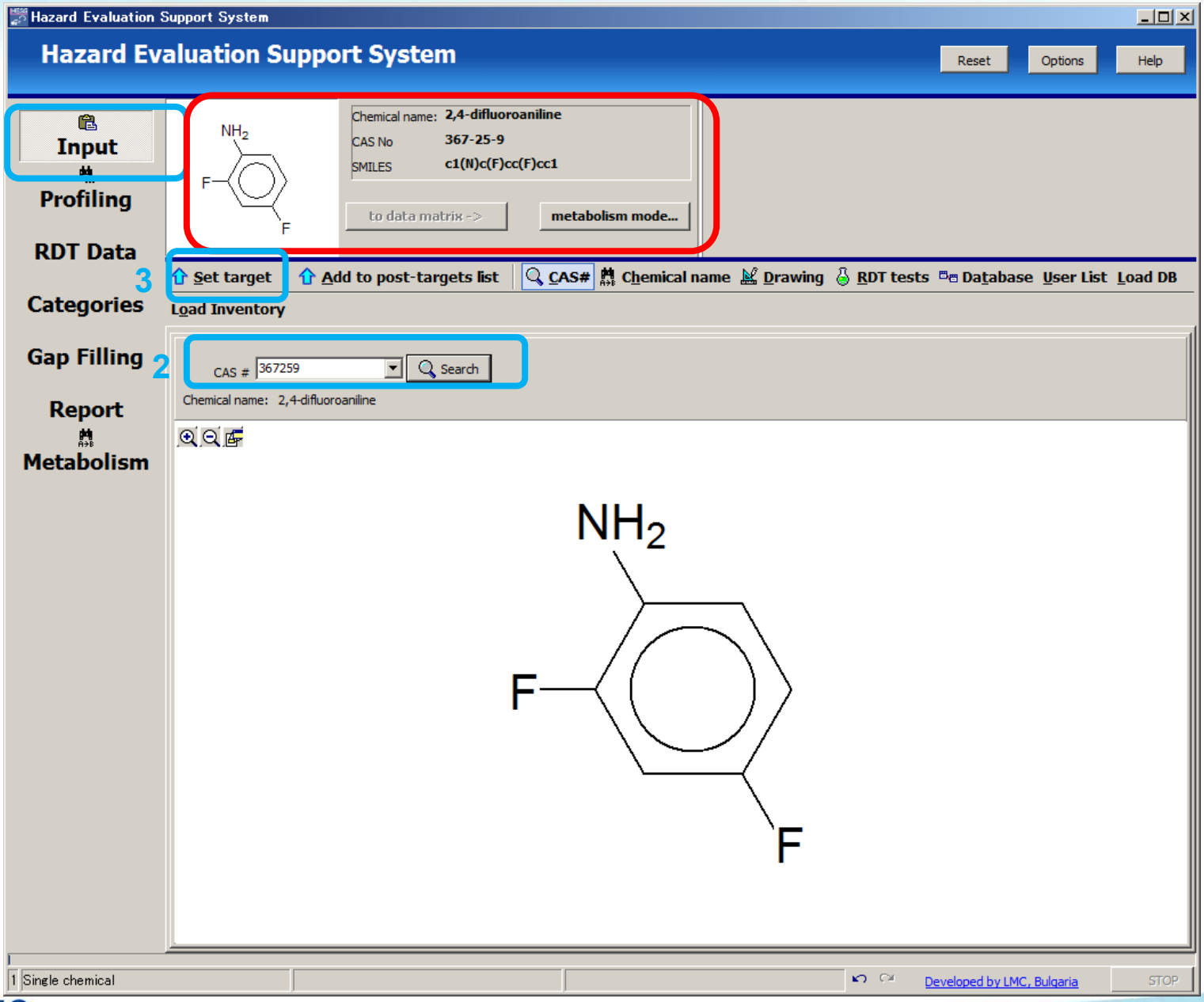
nite

Case study 1:

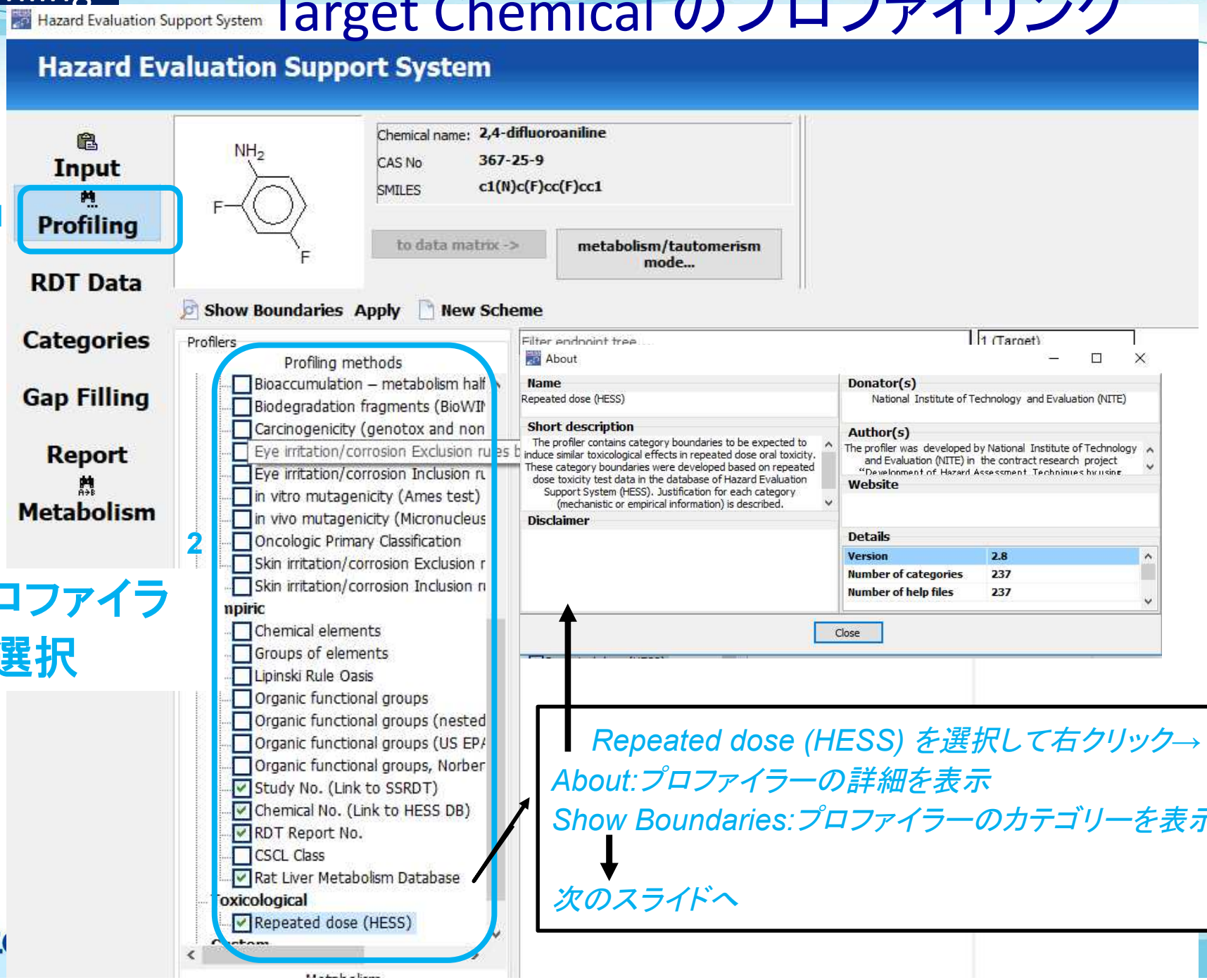
Anemia for 2,4-difluoroaniline

(CAS RN: 367-25-9)

Target Chemicalの選択



Target Chemical のプロファイリング



プロファイラの選択

Repeated dose (HESS) を選択して右クリック→
About:プロファイラーの詳細を表示
Show Boundaries:プロファイラーのカテゴリーを表示

次のスライドへ

プロファイル表示(1)

Repeated dose (HESS) (Toxicological) – Profiling Scheme Browser

Advanced

Repeated dose (HESS) - Category definitions

- 4,4'-Diethylaminoethoxyhexestrol (Hepatotox
- 4,4'-Methylenedianilines/benzidines (Hepatobi
- 4-Aminopyrazolopyrimidine (Hepatotoxicity) A
- Acetaminophen (Hepatotoxicity) Alert
- Acrylamides (Neurotoxicity) Rank C
- Aflatoxin B1 (Hepatotoxicity) Alert
- Ajmaline (Hepatotoxicity) Alert
- Aliphatic amines (Mucous membrane irritation)
- Aliphatic nitriles (Hepatotoxicity) Rank B
- Aliphatic/Alicyclic hydrocarbons (Alpha 2u-glob
- Allopurinol (Hepatotoxicity) Alert
- Allyl esters (Hepatotoxicity) Rank A
- Alpha olefin (Less susceptible) No Rank
- Alpha-Amanitin (Amatoxin) (Hepatotoxicity) A
- Alpha-Naphthyl-isothiocyanate (Hepatotoxicit
- Amine oxides (Less susceptible) No Rank
- Amineptine (Hepatotoxicity) Alert
- Amiodarone (Hepatotoxicity) Alert
- Anilines (Hemolytic anemia with methemoglobi
- Anilines (Hepatotoxicity) Rank C
- Aromatic hydrocarbons (Liver enzyme inductic
- Azithromycin (Hepatotoxicity) Alert
- Azobenzenes (Hemolytic anemia with methem
- Benzene/ Naphthalene sulfonic acids (Less sus
- Benzenesulfonamides (Toxicity to urinary syst
- Beta-Naphthylisothiocyanate (Hepatotoxicity)
- Bosentan (Hepatotoxicity) Alert
- Bromfenac (Hepatotoxicity) Alert
- Carbamazepine (Hepatotoxicity) Alert
- Carbon Disulfide (Hepatotoxicity) Alert
- Carboxylic acids (Hepatotoxicity) No rank
- Chloramphenicol (Hepatotoxicity) Alert
- Chloroquine (Hepatotoxicity) Alert
- Chlorphentermine (Hepatotoxicity) Alert
- Chlorpromazine (Hepatotoxicity) Alert
- Cisplatin (Hepatotoxicity) Alert
- Clindamycin (Hepatotoxicity) Alert
- Clofibrate (Hepatotoxicity) Alert
- Coumarin (Hepatotoxicity) Alert
- Cuprizone (Hepatotoxicity) Alert
- Cycloheximide (Hepatotoxicity) Alert
- Cyclophosphamide (Hepatotoxicity) Alert
- Cyclosporin A (Hepatotoxicity) Alert
- Cyproterone Acetate (Hepatotoxicity) Alert
- Danazol (Hepatotoxicity) Alert
- Dantrolene (Hepatotoxicity) Alert

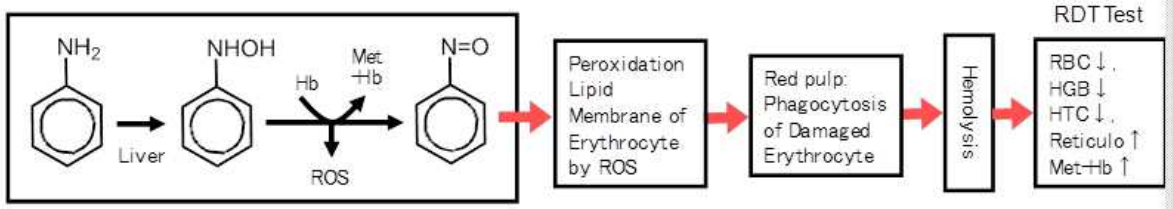
Profile Description

Anilines (Hemolytic anemia with methemoglobinemia) Rank A

1. Toxicity Information

The toxicant of methemoglobinemia induced by anilines is considered to be N-hydroxyl anilines that are metabolites of anilines in the liver[1,2]. The hemolytic anemia induced by anilines is considered to be related to the oxidation of erythrocytes by N-hydroxyl anilines[3, 4].

1) Anilines are metabolized in hepatocytes by oxidases such as P450 to N-hydroxyl anilines.
2) N-hydroxyl anilines react with hemoglobin (Hgb) in erythrocytes to produce nitrosoaniline and methemoglobin (Met-Hgb). The resulting increase in the concentration of Met-Hgb is observed in hematological examination.
3) Erythrocytes are degenerated (peroxidation of lipid membrane etc.) by reactive oxygen species (ROS) produced in the above reaction[3].
4) Phagocytosis of degenerate erythrocytes, mainly in the spleen, results in hemolysis[4].
5) The result is: decrease in red blood cells (RBC), decrease in Hgb, decreased hematocrit (Hct) and increase in reticulocytes (Ret) observed upon hematological examination in RDT test. In addition, pigmentation of hemosiderin and congestion are observed in the spleen on histopathological examination[5].
6) As a compensatory response to anemia, extramedullary hematopoiesis (mainly in the spleen) is observed on histopathological examination[4].



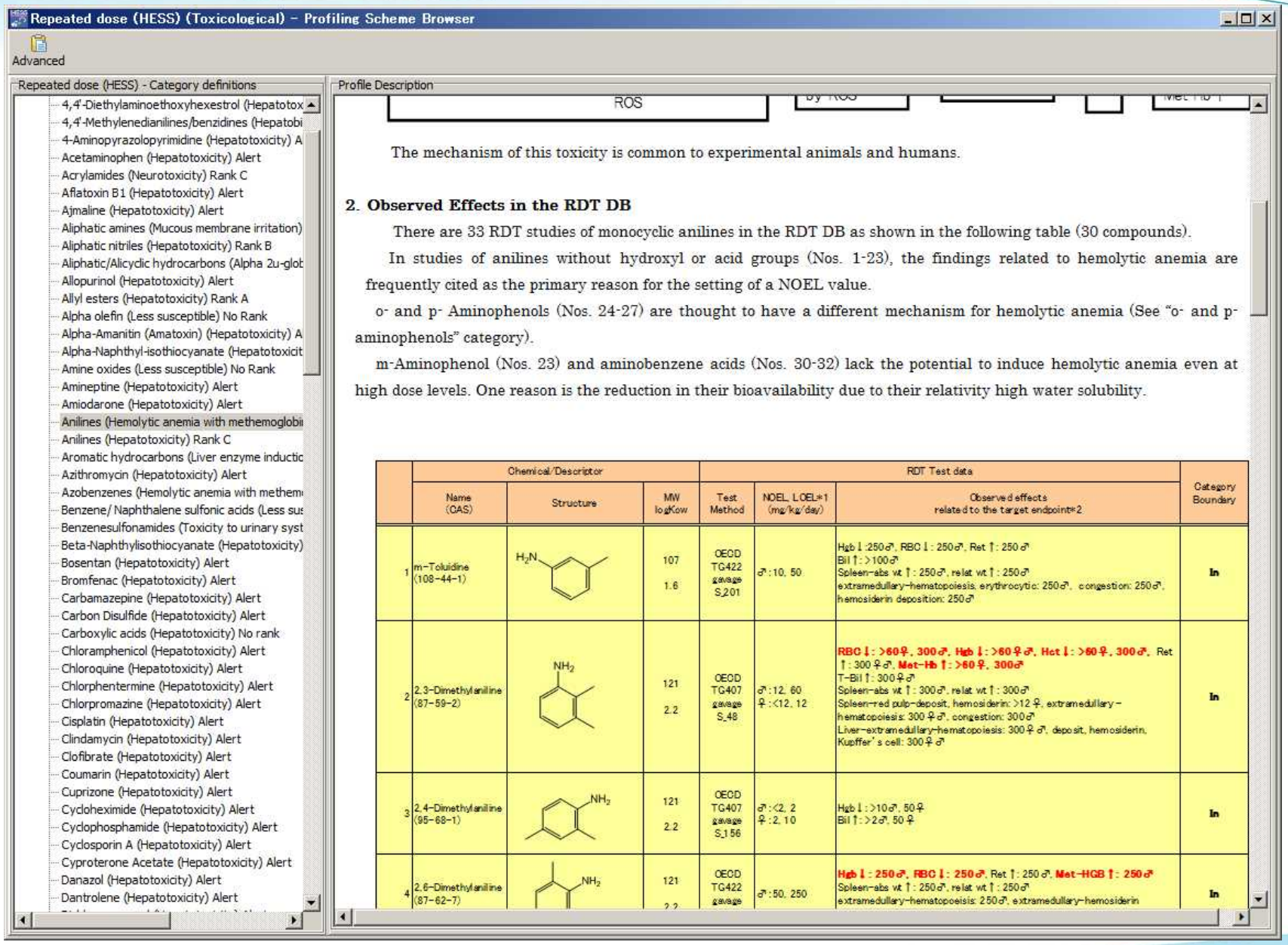
The mechanism of this toxicity is common to experimental animals and humans.

2. Observed Effects in the RDT DB

There are 33 RDT studies of monocyclic anilines in the RDT DB as shown in the following table (30 compounds).

In studies of anilines without hydroxyl or acid groups (Nos. 1-23), the findings related to hemolytic anemia are frequently cited as the primary reason for the setting of a NOEL value.

プロファイル表示(2)


Repeated dose (HESS) (Toxicological) – Profiling Scheme Browser
Advanced
Repeated dose (HESS) - Category definitions
4,4'-Diethylaminoethoxyhexestrol (Hepatotox
4,4'-Methylenedianilines/benzidines (Hepatobi
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Carboxylic acids (Hepatotoxicity) No rank
Chloramphenicol (Hepatotoxicity) Alert
Chloroquine (Hepatotoxicity) Alert
Chlorphentermine (Hepatotoxicity) Alert
Chlorpromazine (Hepatotoxicity) Alert
Cisplatin (Hepatotoxicity) Alert
Clindamycin (Hepatotoxicity) Alert
Clofibrate (Hepatotoxicity) Alert
Coumarin (Hepatotoxicity) Alert
Cuprizone (Hepatotoxicity) Alert
Cycloheximide (Hepatotoxicity) Alert
Cyclophosphamide (Hepatotoxicity) Alert
Cyclosporin A (Hepatotoxicity) Alert
Cyproterone Acetate (Hepatotoxicity) Alert
Danazol (Hepatotoxicity) Alert
Dantrolene (Hepatotoxicity) Alert
Profile Description
ROS
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There are 33 RDT studies of monocyclic anilines in the RDT DB as shown in the following table (30 compounds).
In studies of anilines without hydroxyl or acid groups (Nos. 1-23), the findings related to hemolytic anemia are frequently cited as the primary reason for the setting of a NOEL value.
o- and p- Aminophenols (Nos. 24-27) are thought to have a different mechanism for hemolytic anemia (See "o- and p-aminophenols" category).
m-Aminophenol (Nos. 23) and aminobenzene acids (Nos. 30-32) lack the potential to induce hemolytic anemia even at high dose levels. One reason is the reduction in their bioavailability due to their relativity high water solubility.
Chemical/Descriptor
RDT Test data
Category Boundary
Name (CAS)
Structure
MW logKow
Test Method
NOEL, LOEL*1 (mg/kg/day)
Observed effects related to the target endpoint*2
1 m-Toluidine (108-44-1) 107 1.6 OECD TG422 gavage S_201 ♂:10, 50 Hgb↓:250♂, RBC↓:250♂, Ret↑:250♂ Bil↑:>100♂ Spleen-abs wt↑:250♂, relat wt↑:250♂ extramedullary-hematopoiesis, erythrocytic: 250♂, congestion: 250♂, hemosiderin deposition: 250♂ In
2 2,3-Dimethylaniline (87-59-2) 121 2.2 OECD TG407 gavage S_48 ♂:12, 60 ♀:<12, 12 RBC↓:>60♀, 300♂, Hgb↓:>60♀♂, Het↓:>60♀, 300♂, Ret↑:300♀♂, Met-Hb↑:>60♀, 300♂ T-Bil↑:300♀♂ Spleen-abs wt↑:300♂, relat wt↑:300♂ Spleen-red pulp-deposit, hemosiderin: >12♀, extramedullary-hematopoiesis: 300♀♂, congestion: 300♂ Liver-extramedullary-hematopoiesis: 300♀♂, deposit, hemosiderin, Kupffer's cell: 300♀♂ In
3 2,4-Dimethylaniline (95-68-1) 121 2.2 OECD TG407 gavage S_156 ♂:<2, 2 ♀:2, 10 Hgb↓:>10♂, 50♀ Bil↑:>2♂, 50♀ In
4 2,6-Dimethylaniline (87-62-7) 121 OECD TG422 gavage ♂:50, 250 Hgb↓:250♂, RBC↓:250♂, Ret↑:250♂, Met-HGB↑:250♂ Spleen-abs wt↑:250♂, relat wt↑:250♂ extramedullary-hematopoiesis: 250♂, extramedullary-hemosiderin In

プロファイル表示(3)

Repeated dose (HESS) (Toxicological) – Profiling Scheme Browser

Advanced

Repeated dose (HESS) - Category definitions

- Repeated dose (HESS)
  - 2-Acetylaminofluorene (Hepatotoxicity) Alert
  - 3-Methylcholantrene (Hepatotoxicity) Alert
  - 4,4'-Diethylaminoethoxyhexestrol (Hepatotox
  - 4,4'-Methylenedianilines/benzidines (Hepatobi
  - 4-Aminopyrazolopyrimidine (Hepatotoxicity) A
  - Acetaminophen (Hepatotoxicity) Alert
  - Acrylamides (Neurotoxicity) Rank C
  - Aflatoxin B1 (Hepatotoxicity) Alert
  - Ajmaline (Hepatotoxicity) Alert
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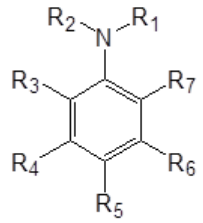
  - Chlorpromazine (Hepatotoxicity) Alert
  - Cisplatin (Hepatotoxicity) Alert
  - Clindamycin (Hepatotoxicity) Alert
  - Clofibrate (Hepatotoxicity) Alert
  - Coumarin (Hepatotoxicity) Alert
  - Cuprizone (Hepatotoxicity) Alert
  - Cycloheximide (Hepatotoxicity) Alert

Profile Description

3. Category Boundary in the Profiler

The following structure is defined as the structural boundary of the category.



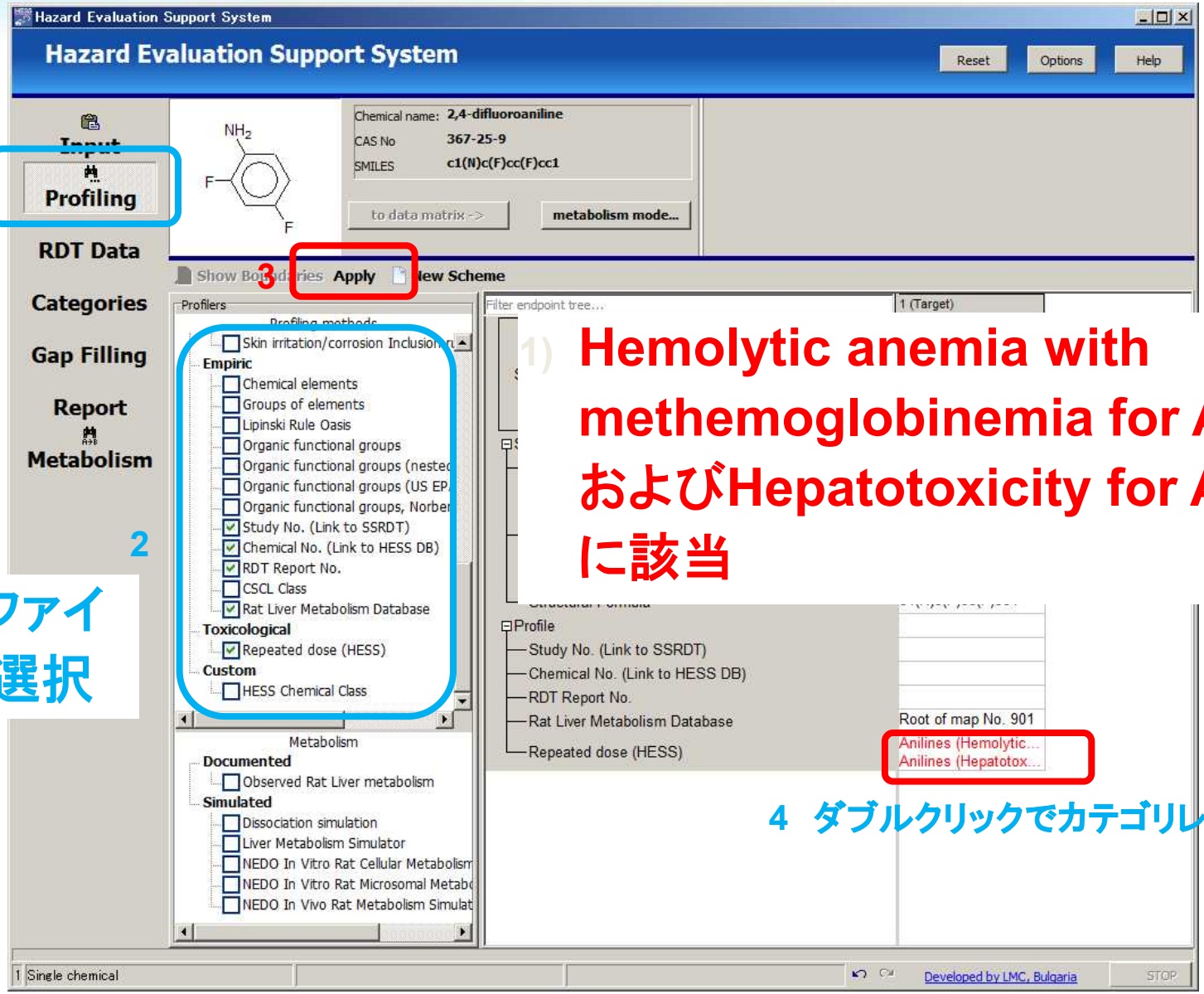
R_1, R_2 = H, methyl or ethyl. R_3~R_7 = H, alkyl, halo, alkoxy, NO_2, NH_2.

4. Rank of reliability of the categories in the profiler

*Rank of reliability	A	B	C
Toxicity Mechanism	Well known	Well known	Not well known
Category Boundaries	Validated by the RDT test data for enough number of compounds.	Not validated by the RDT test data. (RDT data for enough number of compound doses not available). But, estimated by the test data other than RDT test such as SDT and in vitro.	Experientially defined by using the RDT test data.

*Rank of reliability for each category is marked in the name of the category

Target Chemical のプロファイリング



1

2

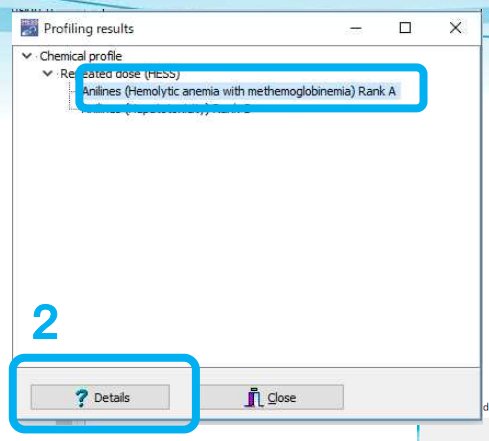
3

プロファイルの選択

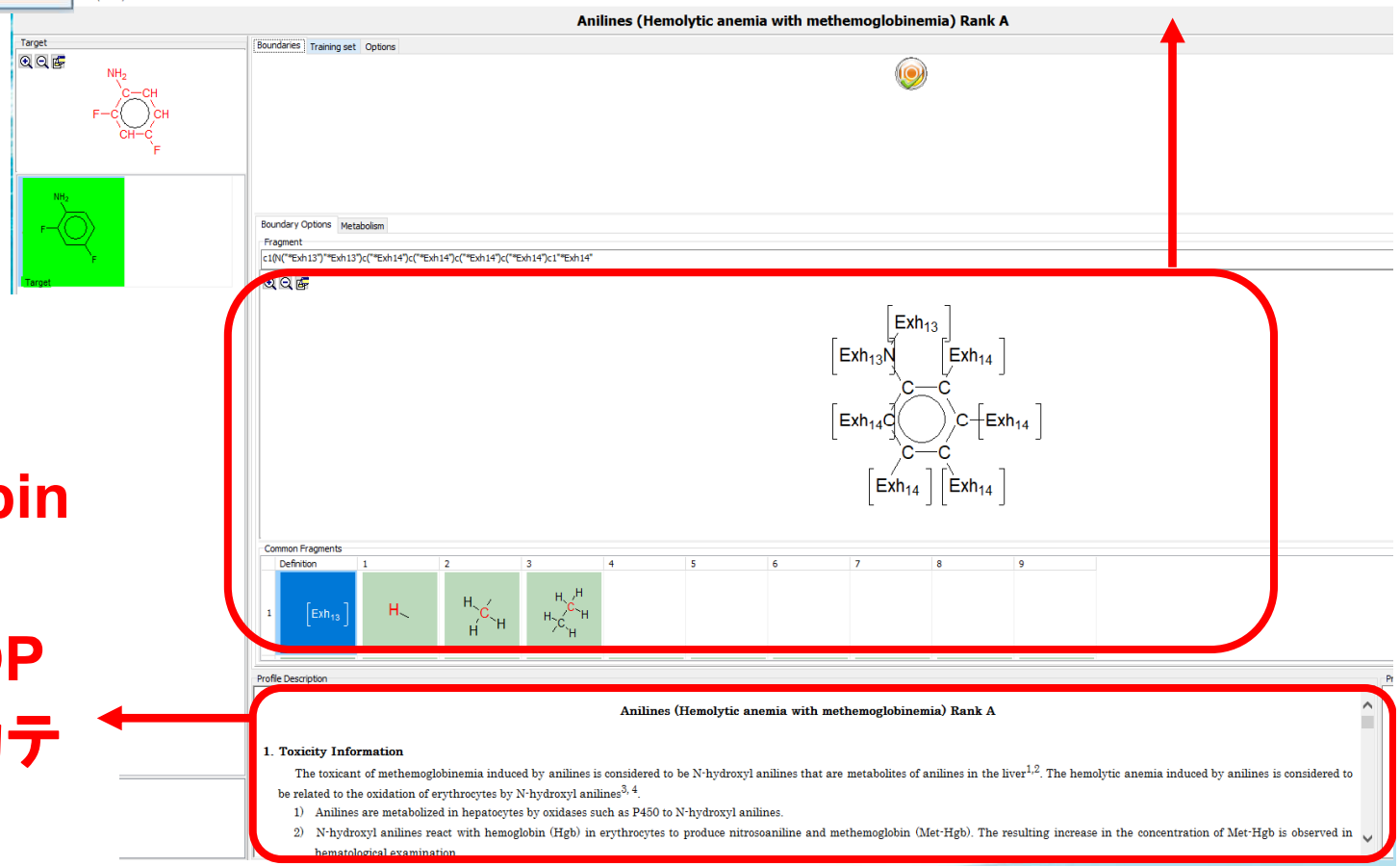
1) **Hemolytic anemia with methemoglobinemia for Anilines およびHepatotoxicity for Anilines に該当**

4 ダブルクリックでカテゴリレポートへリンク

Target Chemical のプロファイリング

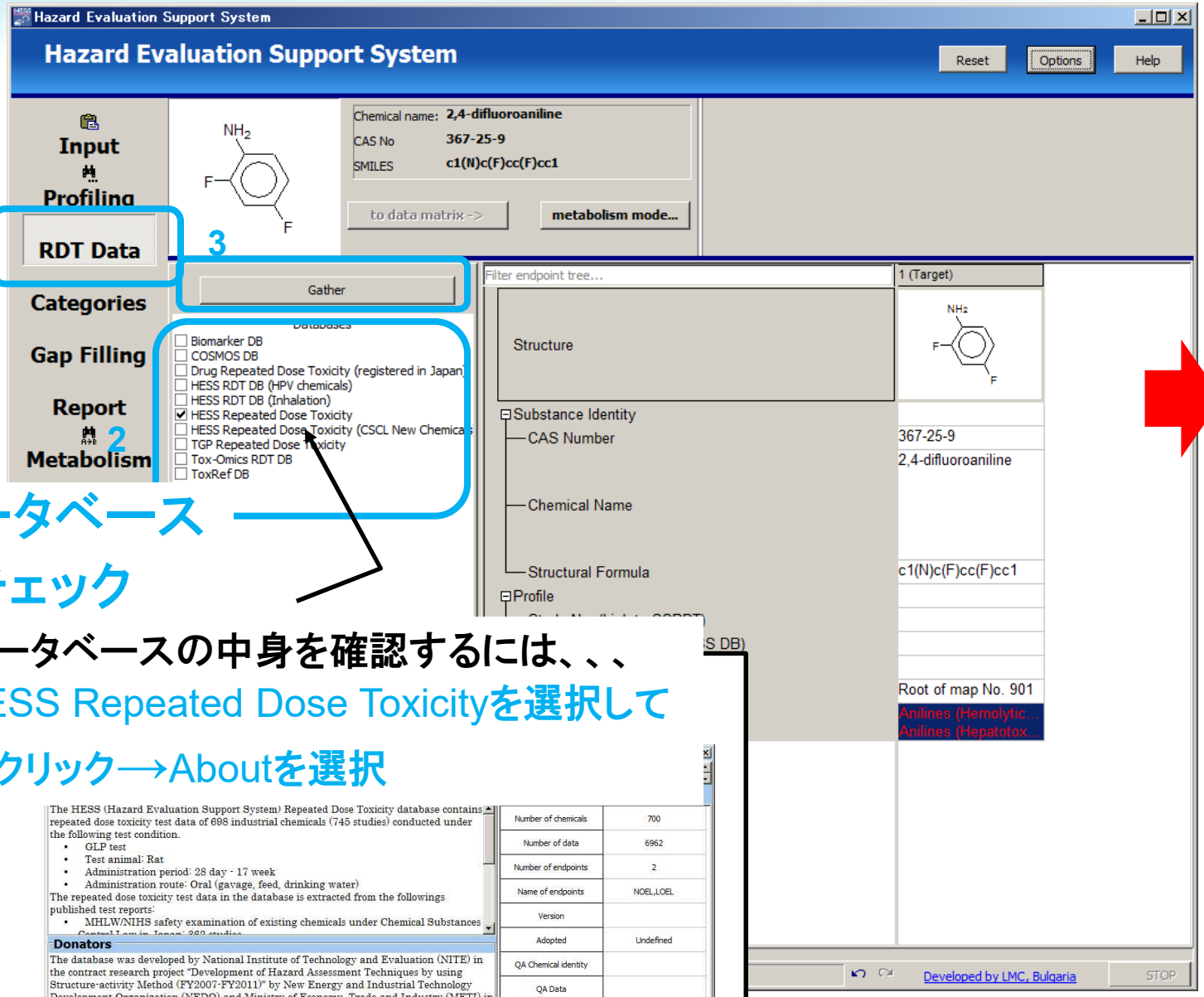


1) **Hemolytic anemia with methemoglobinemia for Anilines の構造領域**



1) **Hemolytic anemia with methemoglobinemia for AnilinesのAOPに基づく毒性カテゴリー情報**

Target Chemical のRDT dataの収集



データベースにチェック

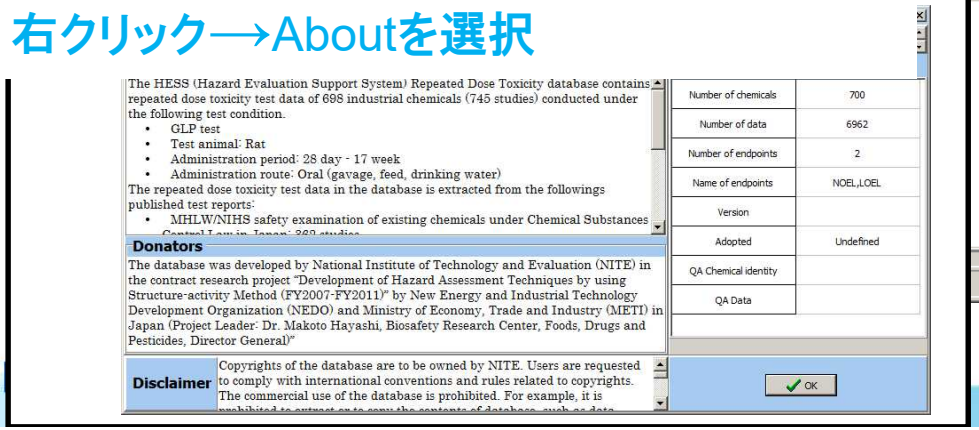
4

選択したDBからはデータはみつからなかった

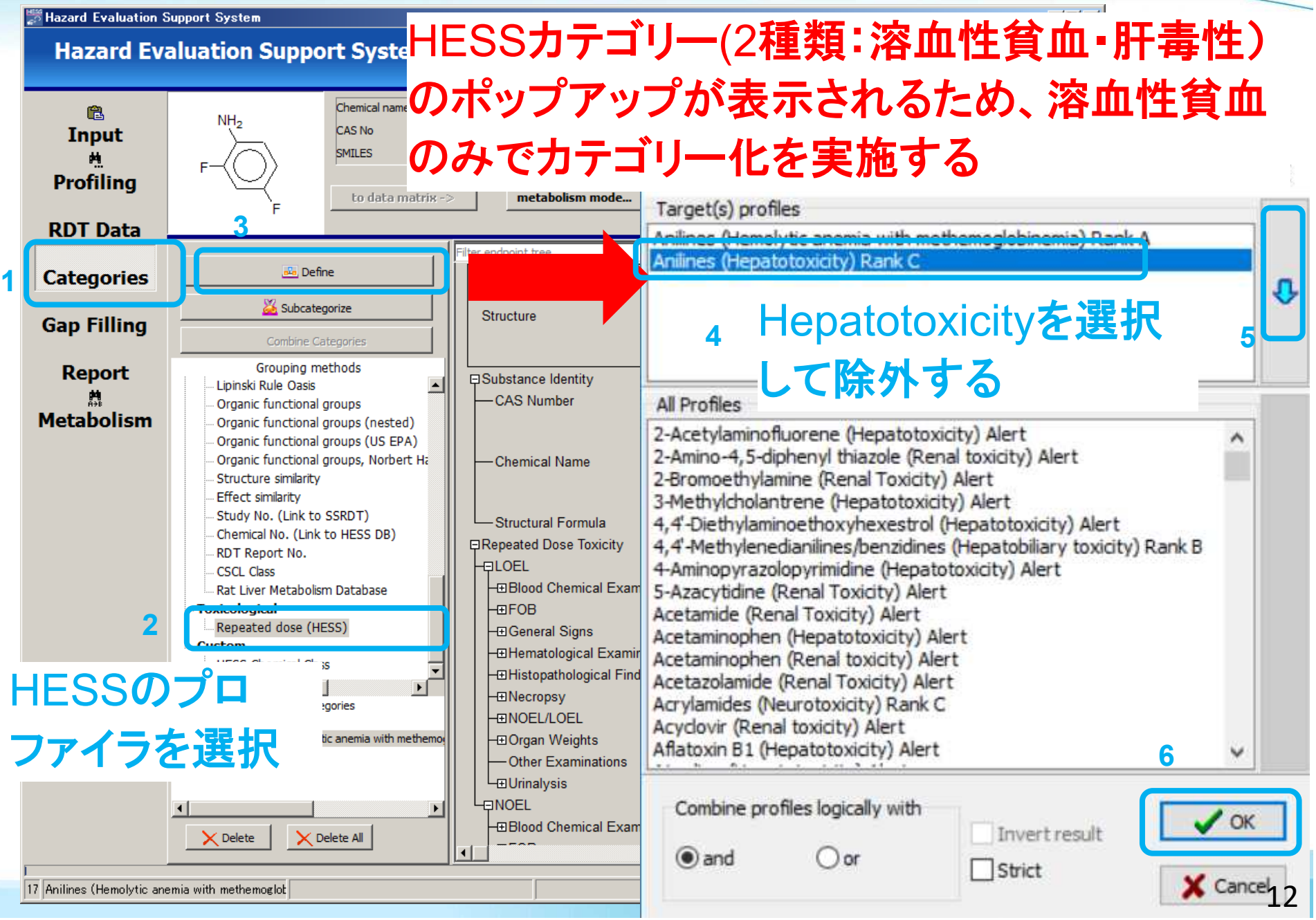
データベースの中身を確認するには、、、

HESS Repeated Dose Toxicityを選択して

右クリック→Aboutを選択



Target Chemicalの類似物質検索 (1)



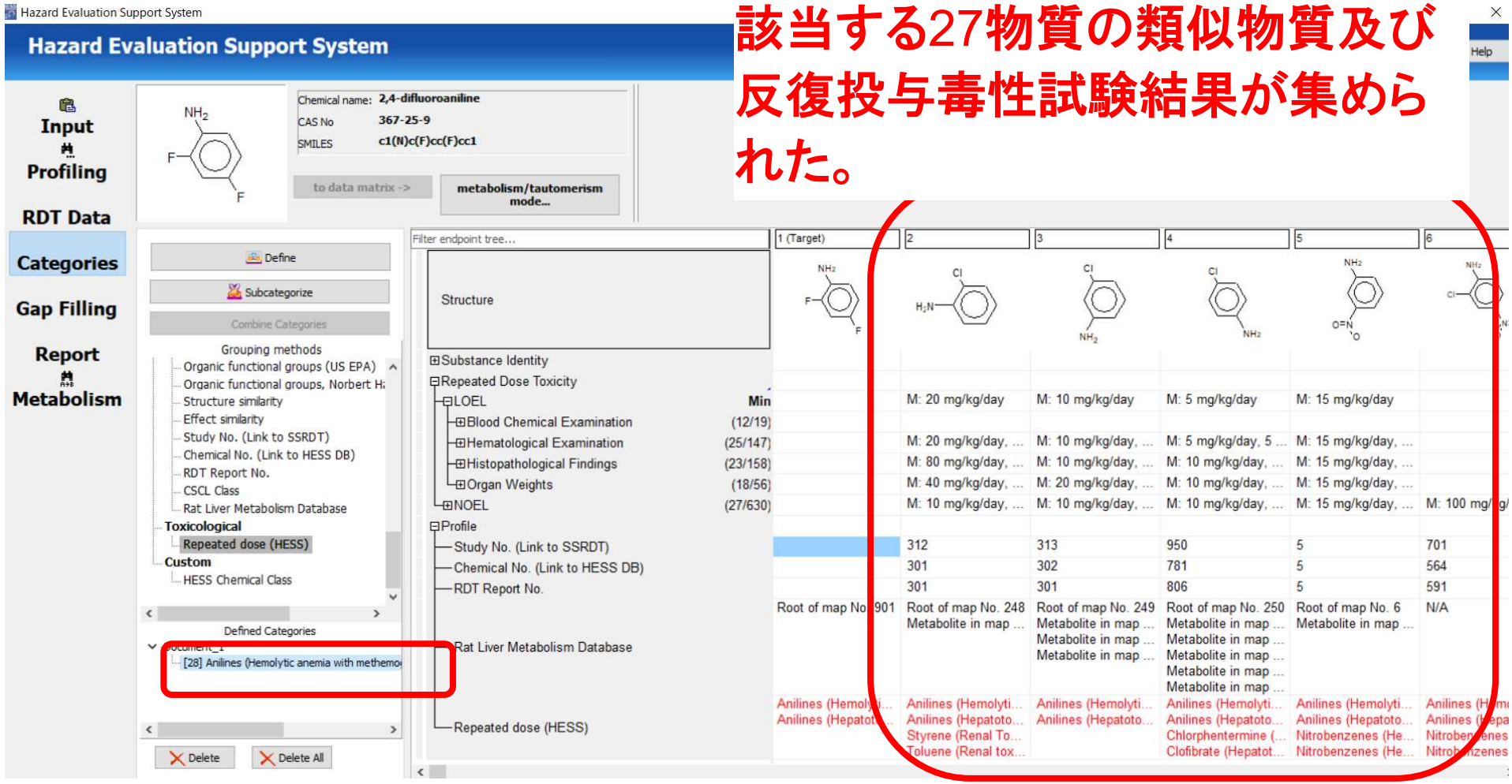
HESSカテゴリー(2種類：溶血性貧血・肝毒性）のポップアップが表示されるため、溶血性貧血のみでカテゴリー化を実施する

Hepatotoxicityを選択して除外する

HESSのプロファイラを選択

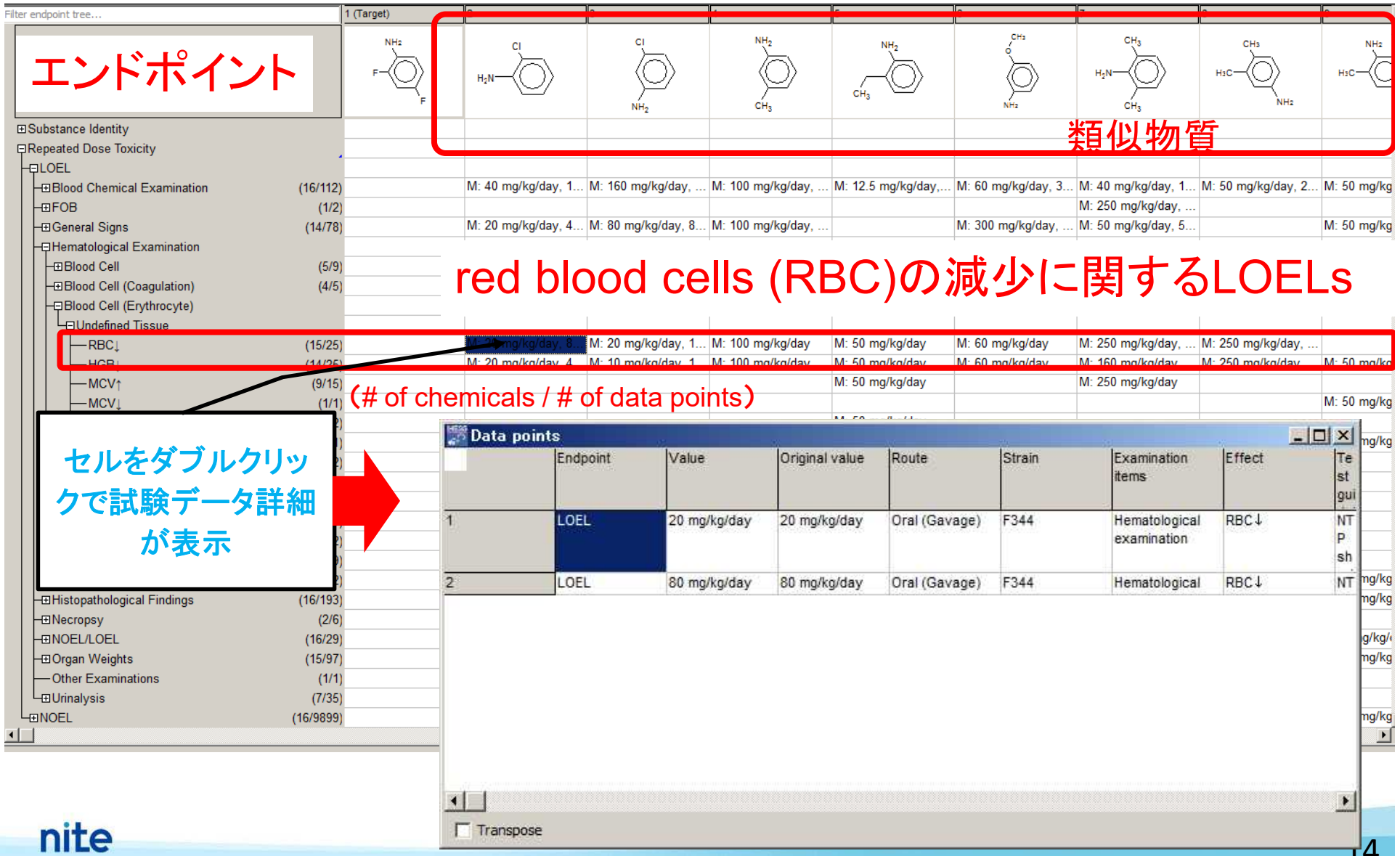
Target Chemicalの類似物質検索(2)

HESSのカテゴリ(溶血性貧血)に該当する27物質の類似物質及び反復投与毒性試験結果が集められた。



類似物質のLOELs/NOELs

類似物質のRDT data

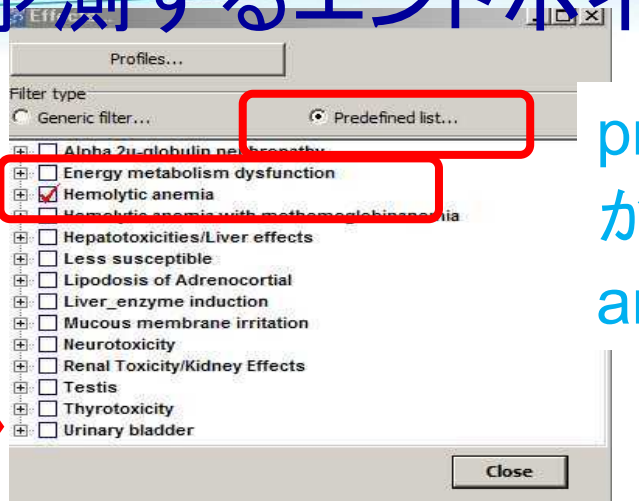


Categories

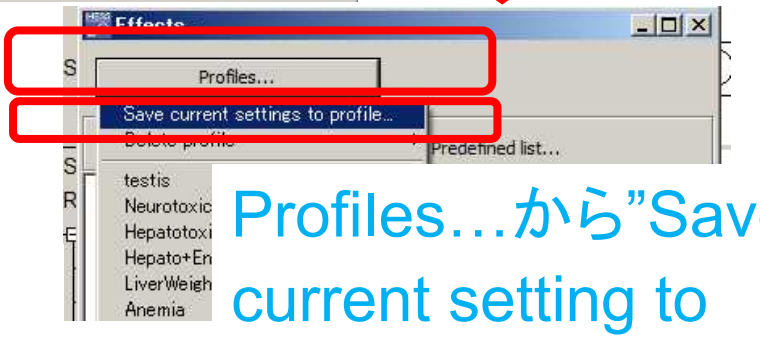
LOELs/NOELsを予測するエンドポイントの選択(1)

エンドポイントツリー

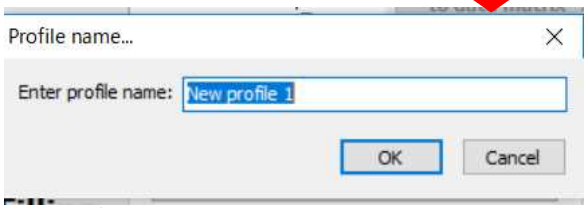
青丸あたりで右クリック　Filter effects -> edit filters



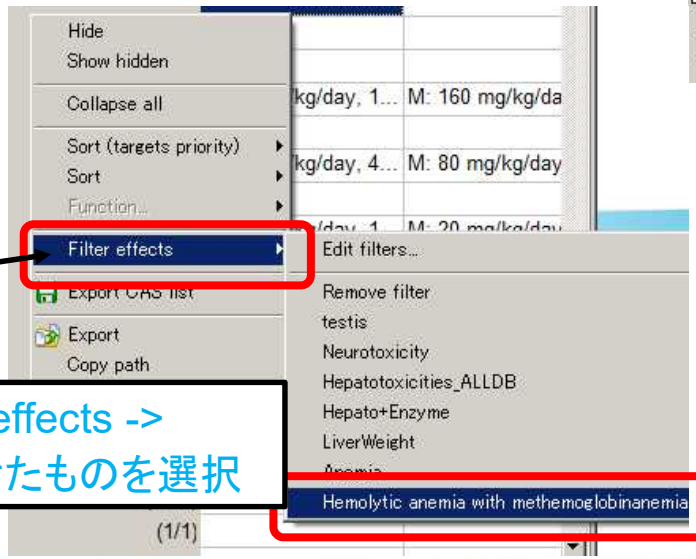
predefined list から“Hemolytic anemia”を選択



Profiles…から”Save current setting to profiles …”を選択



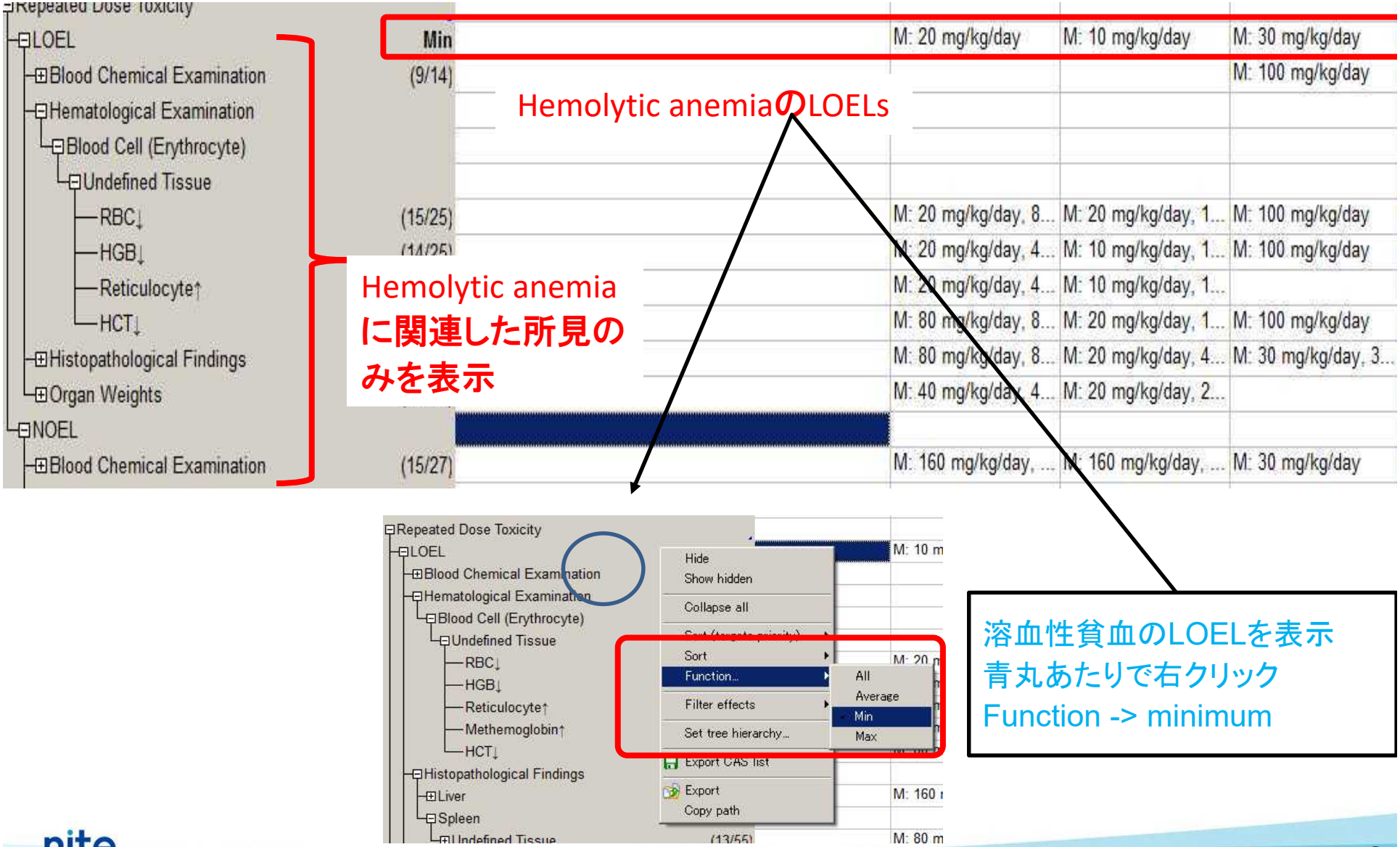
名前をつけたら、OKを押してClose



右クリック　Filter effects -> 自分で名前をつけたものを選択

Categories

LOELs/NOELsを予測するエンドポイントの選択 (2)

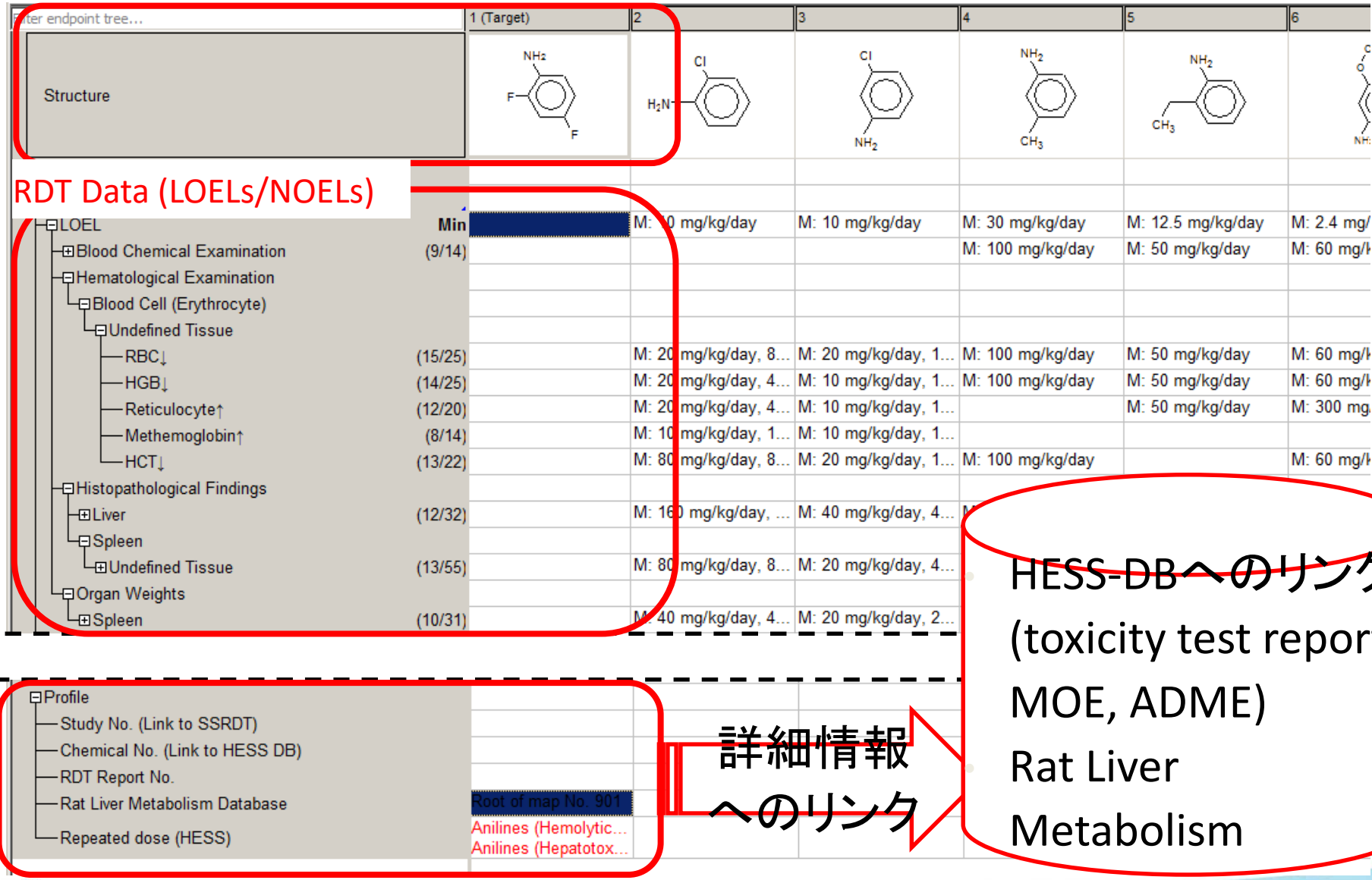


Categories

類似物質の確認

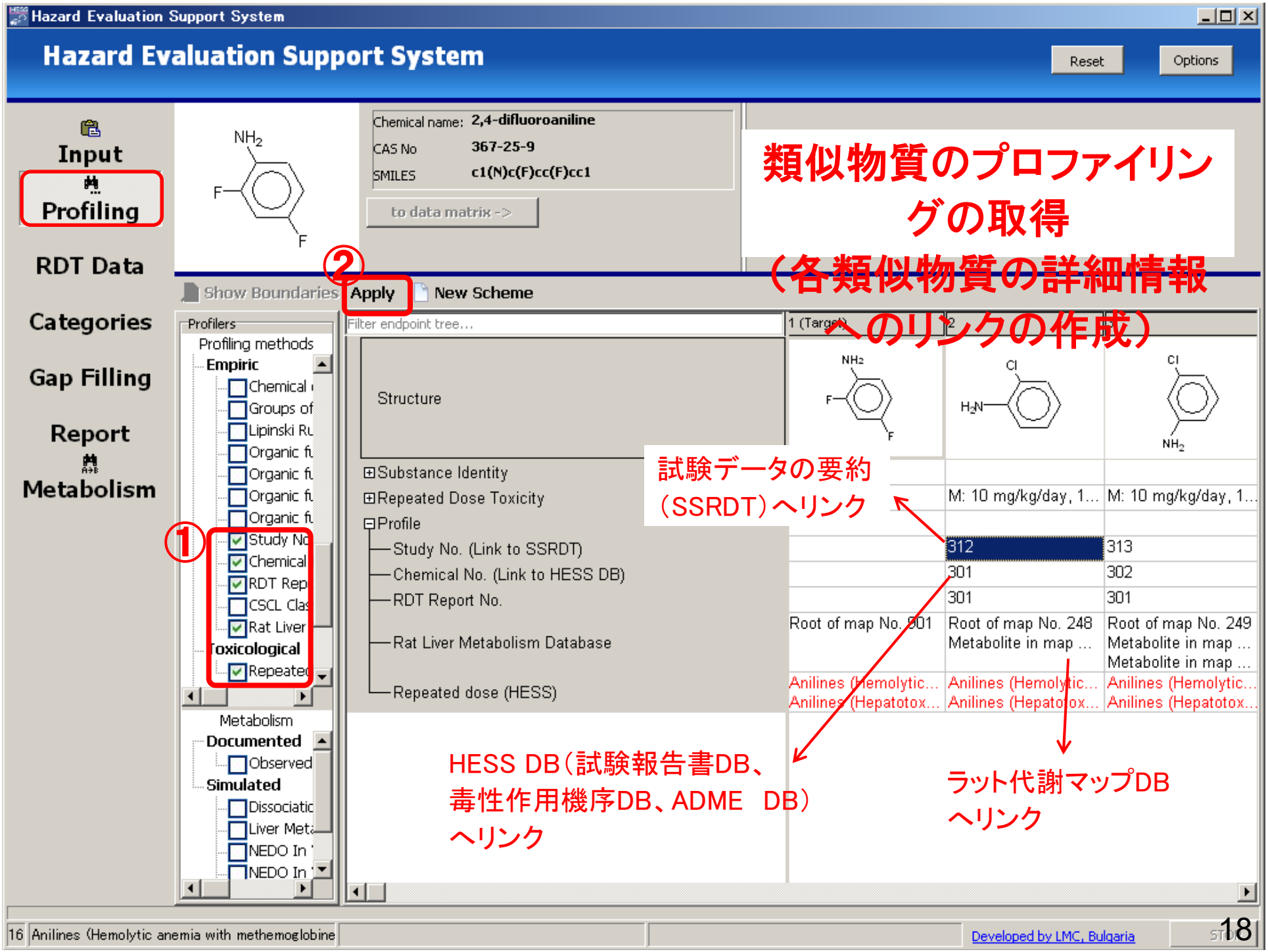
Chemical Structure

RDT Data (LOELs/NOELs)



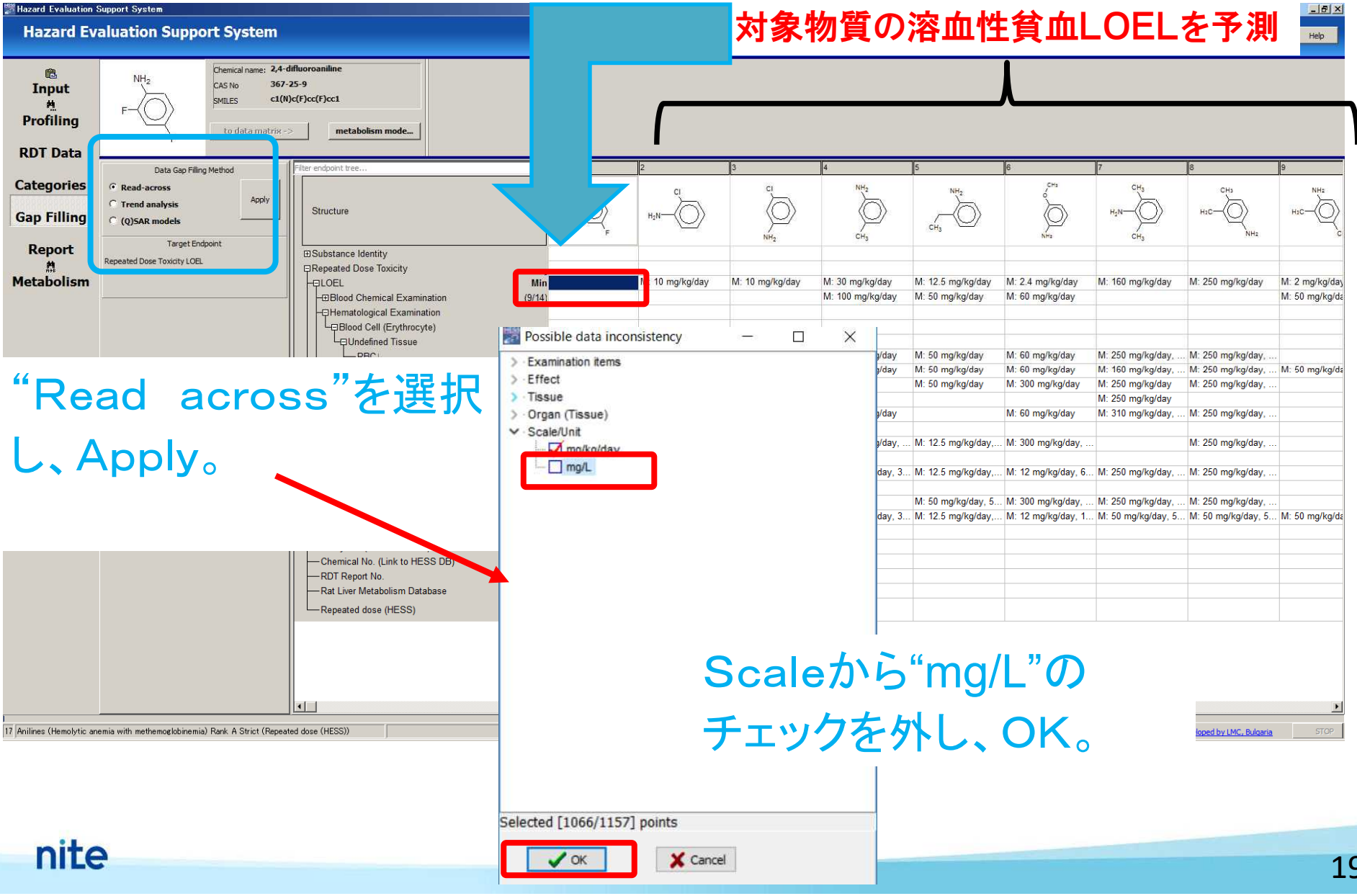
詳細情報へのリンク

HESS-DBへのリンク
(toxicity test report, MOE, ADME)
Rat Liver Metabolism


Hazard Evaluation Support System
Hazard Evaluation Support System
Reset
Options
Input
Profiling
RDT Data
Categories
Gap Filling
Report
Metabolism
Chemical name: 2,4-difluoroaniline
CAS No 367-25-9
SMILES c1(N)c(F)cc(F)cc1
to data matrix ->
②
Show Boundaries
Apply
New Scheme
Profilers
Profiling methods
Empiric
Chemical
Groups of
Lipinski Ru
Organic fu
Organic fu
Organic fu
Organic fu
Study No
Chemical
RDT Rep
CSCL Clas
Rat Liver
Toxicological
Repeated
①
Metabolism
Documented
Observed
Simulated
Dissociatic
Liver Meta
NEDO In
NEDO In
Filter endpoint tree...
Structure
Substance Identity
Repeated Dose Toxicity
Profile
Study No. (Link to SSRDT)
Chemical No. (Link to HESS DB)
RDT Report No.
Rat Liver Metabolism Database
Repeated dose (HESS)
1 (Target)
2
M: 10 mg/kg/day, 1...
M: 10 mg/kg/day, 1...
312
313
301
302
301
301
Root of map No. 501
Root of map No. 248
Metabolite in map ...
Root of map No. 249
Metabolite in map ...
Metabolite in map ...
Anilines (Hemolytic...
Anilines (Hepatotox...
Anilines (Hemolytic...
Anilines (Hepatotox...
Anilines (Hemolytic...
Anilines (Hepatotox...
16 Anilines (Hemolytic anemia with methemoglobine
Developed by LMC, Bulgaria
類似物質のプロファイリングの取得
（各類似物質の詳細情報へのリンクの作成）
試験データの要約（SSRDT）へリンク
HESS DB（試験報告書DB、毒性作用機序DB、ADME　DB）へリンク
ラット代謝マップDB へリンク

Read Acrossによるデータギャップ

類似物質の溶血性貧血LOELから、
対象物質の溶血性貧血LOELを予測



“Read across”を選択し、Apply。

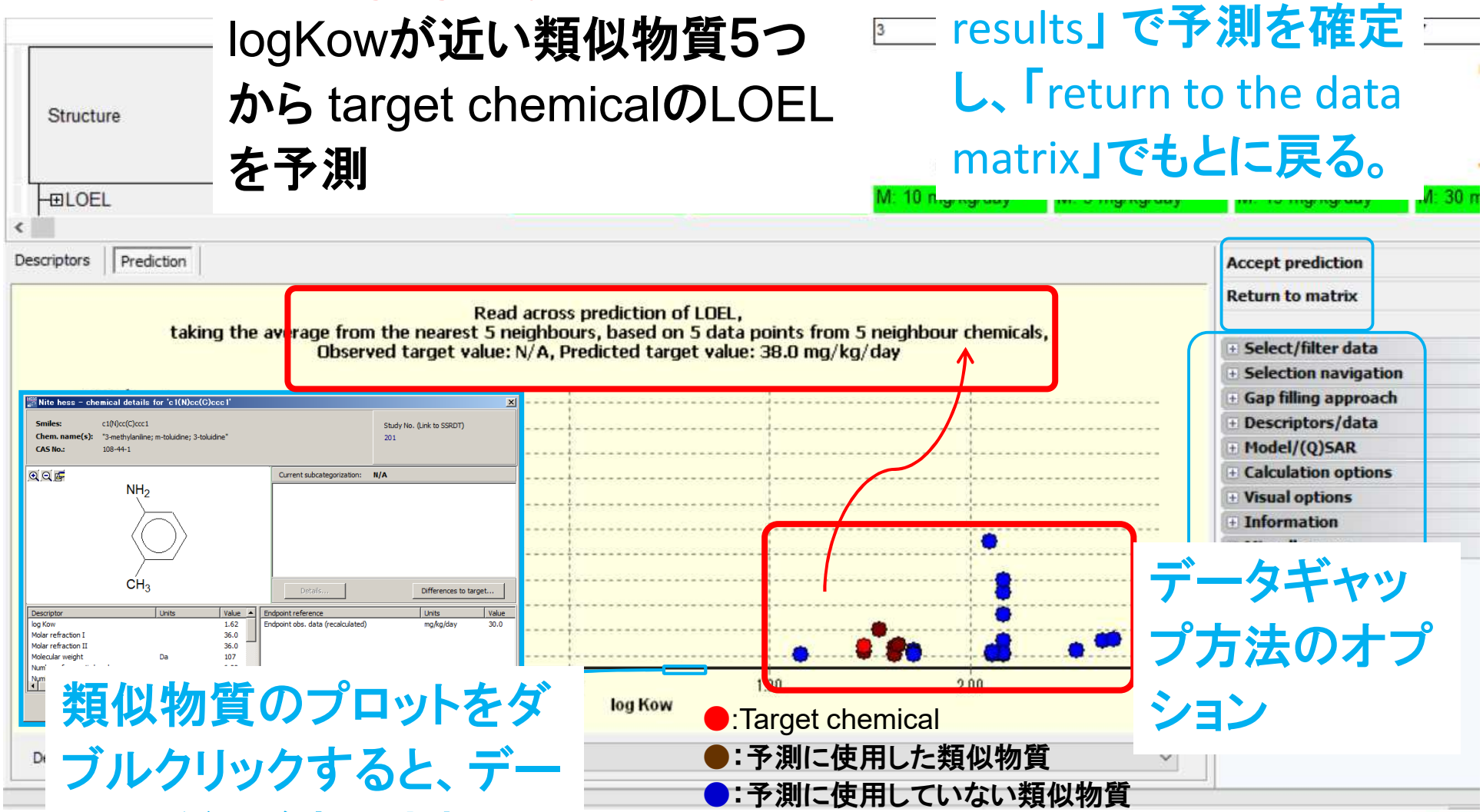
Scaleから“mg/L”のチェックを外し、OK。

LOELの予測

溶血性貧血のLOEL予測値：
38.0 mg/kg/day

logKowが近い類似物質5つからtarget chemicalのLOELを予測

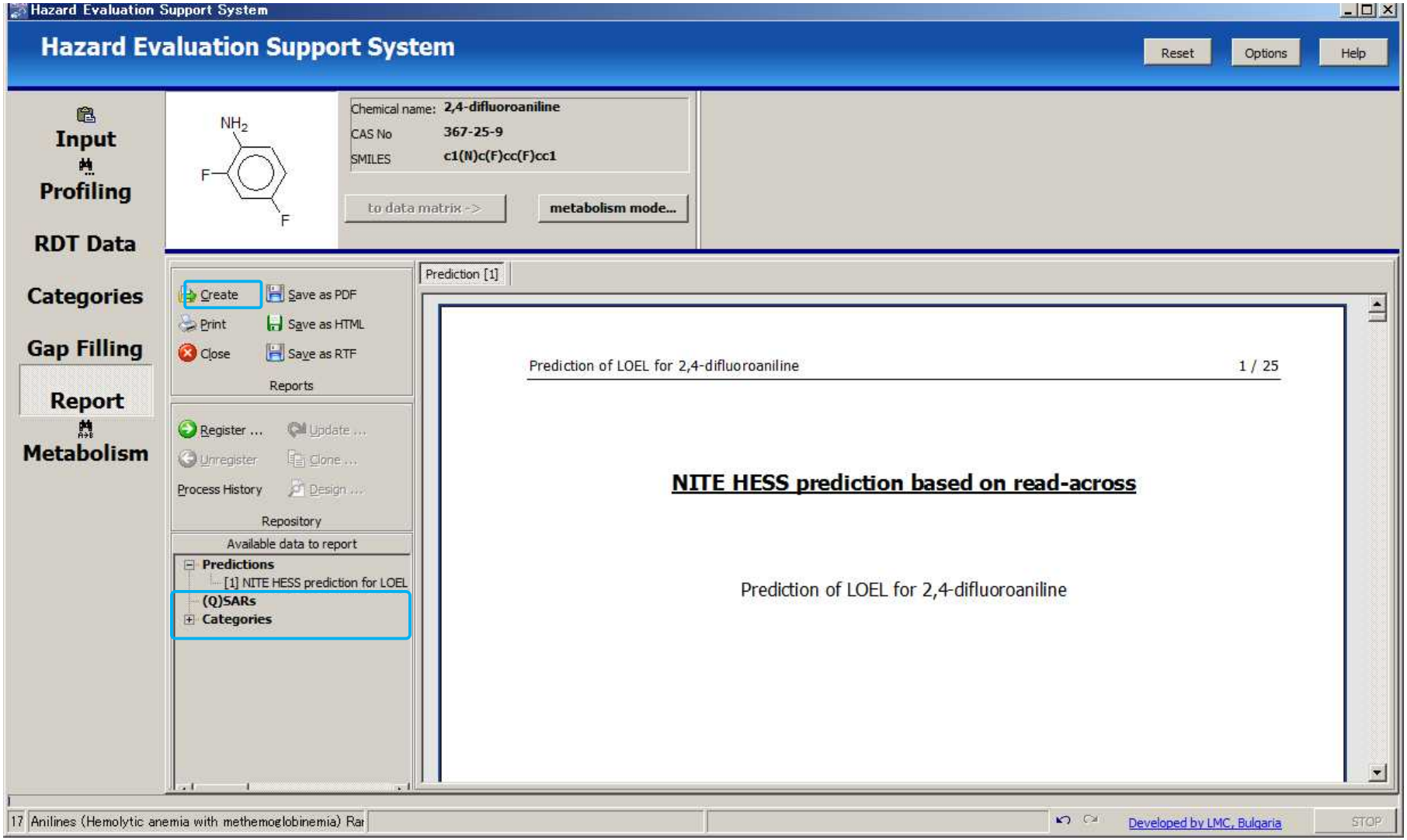
「Accept the prediction results」で予測を確定し、「return to the data matrix」でもとに戻る。



類似物質のプロットをダブルクリックすると、データの詳細が表示される

データギャップ方法のオプション

Report作成


Hazard Evaluation Support System
Hazard Evaluation Support System
Reset
Options
Help
Input
Profiling
RDT Data
Categories
Gap Filling
Report
Metabolism
Chemical name: 2,4-difluoroaniline
CAS No 367-25-9
SMILES c1(N)c(F)cc(F)cc1
to data matrix ->
metabolism mode...
Create
Save as PDF
Print
Save as HTML
Close
Save as RTF
Reports
Register ...
Update ...
Unregister
Clone ...
Process History
Design ...
Repository
Available data to report
Predictions
[1] NITE HESS prediction for LOEL
(Q)SARs
Categories
Prediction [1]
Prediction of LOEL for 2,4-difluoroaniline 1 / 25
NITE HESS prediction based on read-across
Prediction of LOEL for 2,4-difluoroaniline
17 Anilines (Hemolytic anemia with methemoglobinemia) Rat
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STOP