Issues in Compound Storage in DMSO

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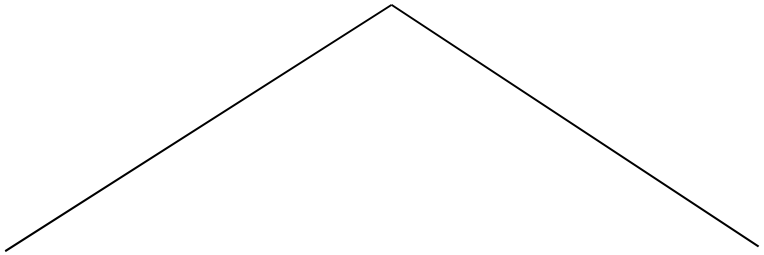
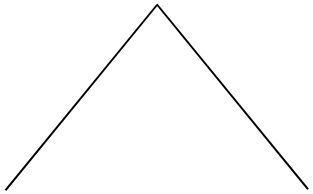
Sample storage in DMSO - overview



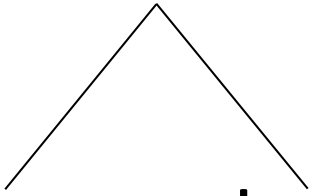
Centralized storage
Business rules enforced
Long storage times



solubility

chemical stability

End user handling / storage
No business rules / chaotic
Short storage times



solubility

chemical stability

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Sample lifetime. What is the key issue?

- **Compound disappears from DMSO solution**
- **What is the explanation?**
- **Chemical integrity**
  - **Keep cold and frozen**
  - **Avoid oxygen**
  - **Keep dry**
- **Compound solubility**
  - **Cold and / or frozen is the worst choice possible**
  - **Avoid freeze thaw cycles**

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Chemical integrity / solubility balance

- **Chris Lipinski's personal opinion**
  - **Reasonable people might disagree**
- **Solubility / precipitation is a bigger problem than chemical integrity**
- **Solubility is the major problem for end users**
  - **Short term handling / storage**
- **Major positive impact is possible with end users**
  - **People issue not a technology issue**

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Where do compounds come from?

- **Legacy compounds are less chemically stable**
  - **Traditional medicinal chemistry**
    - **Good chemistry control of conditions**
    - **Poorer stability compounds can be isolated**
    - **E.g. beta lactams**
- **Combinatorial chemistry**
  - **Poorer chemistry control of conditions**
    - **Stable compounds tend to be isolated**
- **Caveat – combinatorial chemistry**
  - **Acidic hydrolysis due to HPLC additives**
    - **Trifluoroacetic acid, formic acid**

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Compound Solubility in DMSO

- **Compound solubility is determined by 2 factors:**
  - **Solvation energy and crystal disruption**
- **Continuum in the importance of each**
- **Low melting point lipophilic compound**
  - **Solvation most important**
- **High melting point hydrophilic compound**
  - **Crystal disruption most important**

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DMSO Really Helps Solvation

- **Compound has to make a "hole" in DMSO to dissolve**
- **Easier to do this in DMSO than in water**
  - **No H-bond donor / acceptor networks to disrupt**
- **DMSO has a high dielectric constant**
- **Solvates compound dipoles**
  - **Almost all drugs have dipoles**
- **DMSO doesn't solvate hydrocarbons**
  - **E.g. hexane is immiscible in DMSO**

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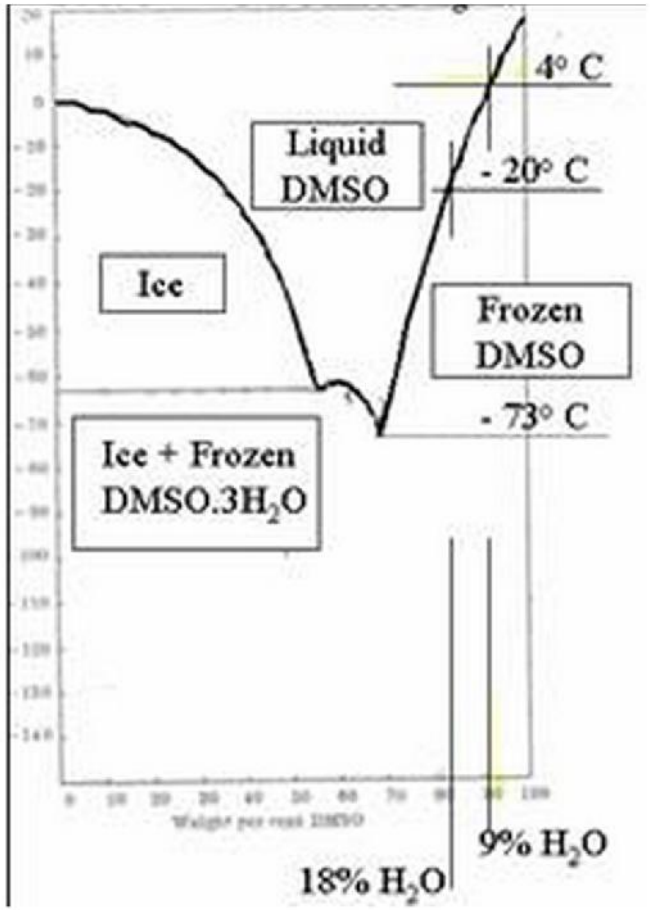
DMSO – water phase diagram



DMSO containing 9% water is unfrozen in the typical lab refrigerator

Very bad practice. Typical biology procedure of storing samples in DMSO in the non-freezer part of a lab refrigerator

Nature (1969), 220, 1315-1317

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Compounds Differ in DMSO Solubility Based on Crystalline Form

- **Oswald's "Rule of Stages"**
- **Sequence of compound batch isolation proceeds towards thermodynamically most stable form**
  1. **Amorphous – highest energy solid form**
  2. **Highest energy crystalline polymorph**
  3. **Lowest energy crystalline polymorph**
- **Amorphous is the highest energy form**
  - **Most soluble in DMSO**
  - **Lowest melting point**

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Timing Factor in Compound DMSO Solubility

- **Once a compound crystallizes from DNSO it will not easily re-dissolve**
  - **Crystallized compound is in a lower energy less DMSO soluble form**
- **Narrow working window (time window) for keeping most compounds dissolved in DMSO**
  - **1 to 2 days at room temperature**
  - **Explains why compounds are active when freshly made but not when stored**
- **Freeze thaw cycles increase the probability of crystallization**

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Precipitation from 60 mM Compound in DMSO

Vials of 60 mM compound in MDSO at room temperature are visually checked the morning after the aqueous solubility assay is completed.

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Compound Insolubility at 60 mM in DMSO

- **Non combinatorial compounds**
- **Traditional medchem synthesis**
- **Solubility test using 60 mM DMSO stock solution**
- **6% insoluble at 60 mM in DMSO**
  - **Cpds dissolved at 60 mM in DMSO**
  - **Vials checked for pptn at rt 24 hr post dissolving**
  - **2381 / 40000 were insoluble**
- **Underestimates DMSO solubility problem**
  - **Because least aqueous soluble were excluded**

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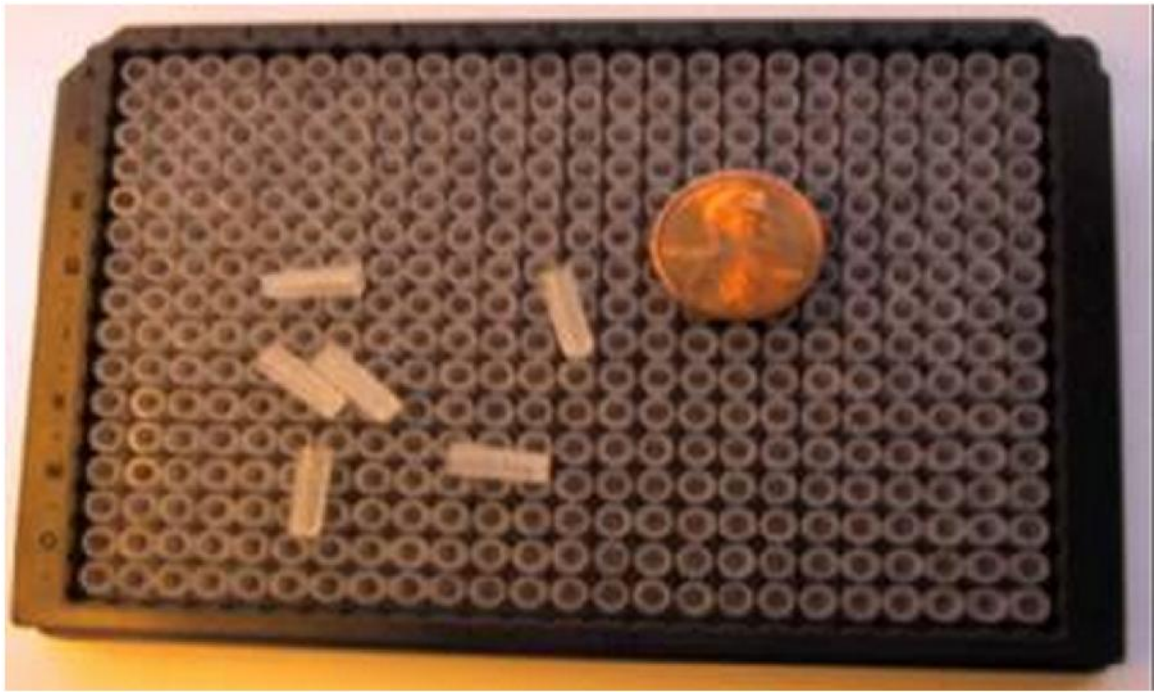
384 Well Format Plate



Visual precipitation detection is impossible for compounds in DMSO distributed in this format

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DMSO Solubility Data Source

www.gaylordchem.com

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Data View in Gaylord Web Page

SOLUBILITIES OF ACTIVE PHARMACEUTICALS COMPOUNDS IN DIMETHYL SULFOXIDE

Pharmaceutical Nomenclature	CAS Number	solubility g/100ml DMSO@25°C	solubility g/100g DMSO@25°C	solubility g/100g solution@25°C
Acetominophen	103-90-2	84.3	77.0	43.5
Acetyl-D-Glucosamine	10036-64-3	4.9	4.5	4.3
Acyclovir USP	59277-89-3	15.4	14.0	12.3
Albuterol Sulfate		0.6	0.5	0.5
Aloe Vera Powder Freeze Dried 200:1	N/A	20.4	18.6	15.7
Aloe Vera Powder Freeze Dried 200:1		20.4	18.6	15.7
Amitriptyline Hydrochloride USP	549-18-8	25.4	23.2	18.8
Amphotericin B USP (Oral Grade)	1397-89-3	0.0	0.0	0.0
Androstendione	63-05-8	5.5	5.0	4.8
Baclofen	1134-47-0	0.0	0.0	0.0
Beclomethasone Dipropionate USP	1134-47-0	38.5	35.1	26.0
Beta Glucan (1,3) FG	N/A	0.0	0.0	0.0
Betamethasone Dipropionate USP	5593-20-4	89.6	81.8	45.0
Biotin (D) USP (Vitamin H)	58-85-5	23.4	21.4	17.6

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Capturing Gaylord Solubility Data

- **Save web pates as Adobe PDF file**
- **Open PDF file with Adobe Acrobat**
- **Select table with Acrobat text table tool**
- **Copy table to clipboard**
- **Paste to Accord for Excel**
- **Merge in structures from SciFinder, Isisbase**

"*Single-Mode Compound Retrieval for QSAR, QSPR Data Sets, and Batch Mode Exact Structure Searching*" - Christopher A. Lipinski, Journal of Pharmaceutical Sciences, (2002), 91 (12), in press.

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Capturing Gaylord Solubility Data

	A	B	C	D	E	F
1	CHEMISTRY	NAME	CAS	Pharmaceutical_Nomenclature	ORGANIC	SALT
2	10191-41-0	Vitamin_E	10191-41-0	Vitamin_E	1	0
3	79-81-2	Vitamin_A_Palmitate	79-81-2	Vitamin_A_Palmitate_(18,000_U/mL)	1	0
4	2777-48-4	Capsaicin	2777-48-4	Capsaicin_Synthetic	1	0
5	137-58-6	Lidocaine	137-58-6	Lidocaine_USP	1	0
6	15687-27-1	Ibuprofen	15687-27-1	Ibuprofen_USP	1	0
7	93-14-1	Guaifenesin	93-14-1	Guaifenesin_USP	1	0
8	67-97-0	Vitamin_D3	67-97-0	Vitamin_D3_(2400_U/mL)	1	0
9	22071-15-4	Ketoprofen	22071-15-4	Ketoprofen_USP	1	0
10	552-22-7	Theymollodide	552-22-7	Theymol_Iodide_Purified	1	0
11	6493-05-5	Pentoxifylline	6493-05-5	Pentoxifylline	1	0
12	5534-09-8	Beclomethasone_Dipropionate	5534-09-8	Beclomethasone_Dipropionate_USP	1	0
13	630-56-8	Hydroxyprogesterone_Caproate	630-56-8	Hydroxyprogesterone_Caproate_USP	1	0
14	57-13-6	Progesterone	57-13-6	Progesterone_USP	1	0
15	98-92-0	Niacinamide	98-92-0	Niacinamide_USP	1	0
16	57-13-6	Urea	57-13-6	Urea_USP	1	0
17	114-07-8	Erythromycin	114-07-8	Erythromycin_USP	1	0
18	154-17-6	2-Deoxy-D-Glucose	154-17-6	2-Deoxy-D-Glucose_(2)	1	0
19	64-86-8	Colchicine	64-86-8	Colchicine_USP	1	0

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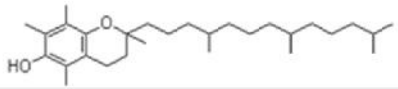
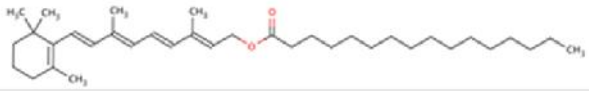
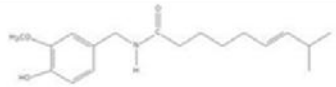
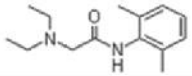
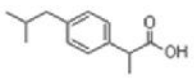
DMSO Melting Point Data in Accord for Excel

	A	B	C	D	E	F	G
1	CHEMISTRY		NAME	CAS	Pharmaceutical_Nomenclature	ORGANIC	SALT
2	10191-41-0		Vitamin_E	10191-41-0	Vitamin_E	1	0
3	79-81-2		Vitamin_A_Palmitate	79-81-2	Vitamin_A_Palmitate_(18,000_U/mL)	1	0
4	2444-48-4		Capsaicin	2444-48-4	Capsaicin_Synthetic	1	0
5	137-58-6		Lidocaine	137-58-6	Lidocaine_USP	1	0
6	15687-27-1		Ibuprofen	15687-27-1	Ibuprofen_USP	1	0

Live chemistry structures can be used in calculations

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Parsing Compounds with DMSO solubility

- **107 compounds with DMSO solubility**
  - **Remove salts**
  - **Remove organo-metallics**
  - **Remove cpds with no structure**
  - **Remove cpds with zero DMSO solubility**
  - **Remove cpds without melting points**
- **57 compounds for analysis**

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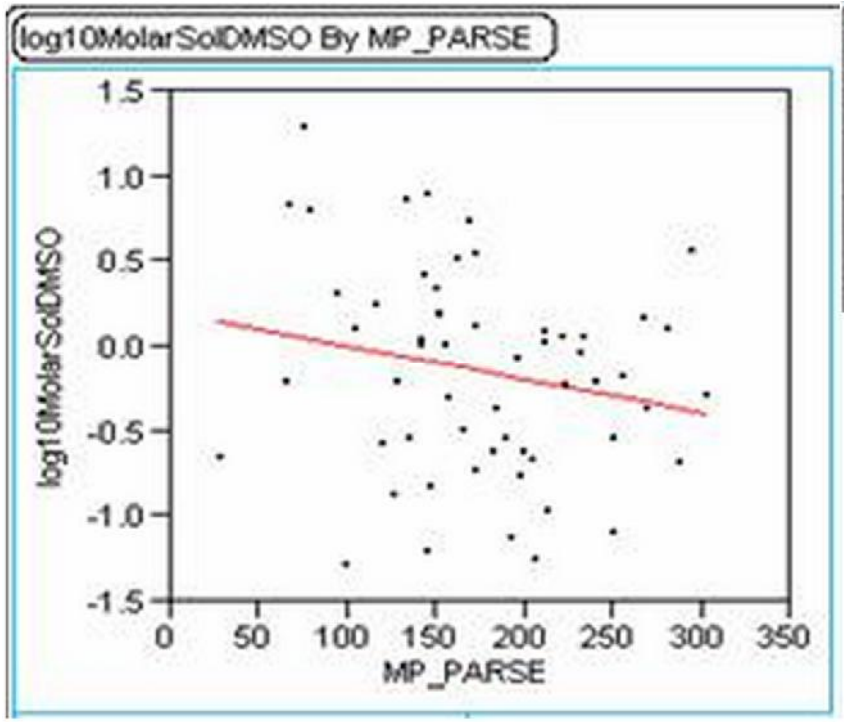
Parsing Compounds with DMSO solubility



Linear Fit

log10MolarSolDMSO = 0.2009 – 0.00196 MP_PARSE

Summary of Fit

RSquare	0.042016
RSquare Adj	0.024598
Root Mean Square Error	0.591044
Mean of Response	-0.14443
Observations (or Sum Wgts)	57

Melting point from Merck Index. Highest polymorph value used. Lower value in range used if a range was reported.

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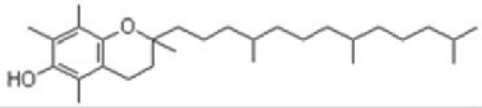
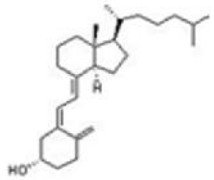
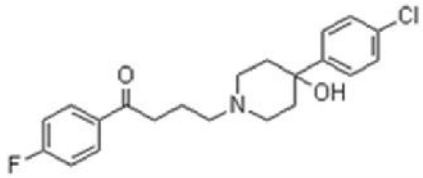
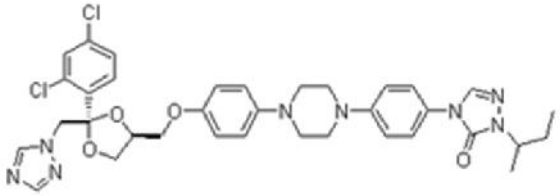
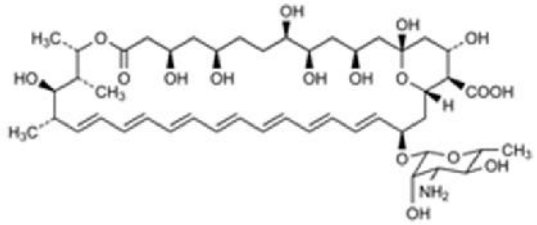
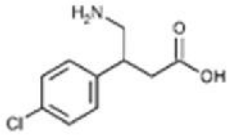
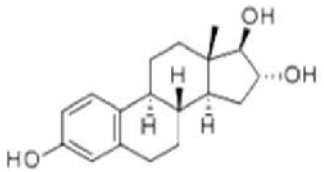
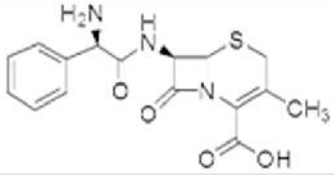
Some compounds with zero DMSO solubility

10191-41-0	
67-97-0	
52-86-8	
84625-61-6	

1397-89-3	
1134-47-0	
50-27-1	
15686-71-2	

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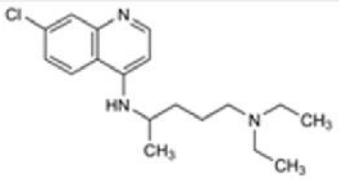
More compounds with zero DMSO solubility

50-63-5



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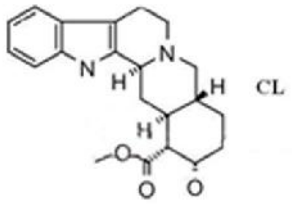

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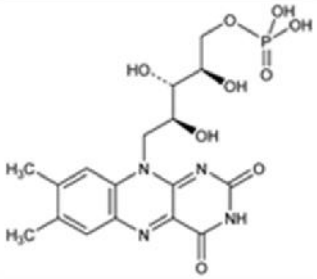

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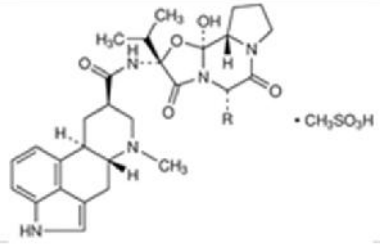

Will be glad to forward entire solubility data set in machine readable format upon request

Christopher_A_Lipinski@Groton.Pfizer.Com

DMSO solubility for 68 cpds v.s. 24 calculations

Variable	logS_DMSO_(MVL)	Variable	logS_DMSO_(MVL)
logS_H2O_(MVL)	0.3695	CX	-0.4685
HBD	-0.0997	MW	-0.4542
ClogP	-0.1089	CMR	-0.4229
ClogP_TS	-0.1089	Ro5_Violations	-0.375
Polar_Area_TS	-0.1139	Ro5_Alert	-0.3458
POL	-0.1143	AndrewBind	-0.3278
HBDNR	-0.1193	UB	-0.3212
PRX	-0.1205	NOCount	-0.2687
INTHB	-0.1319	HBA	-0.2676
RotBonds	-0.157	HBA_TS	-0.2676
MlogP	-0.1968	NO	-0.2671
HBC	-0.2331	tpsa	-0.2504

Nothing useful observed in this data set

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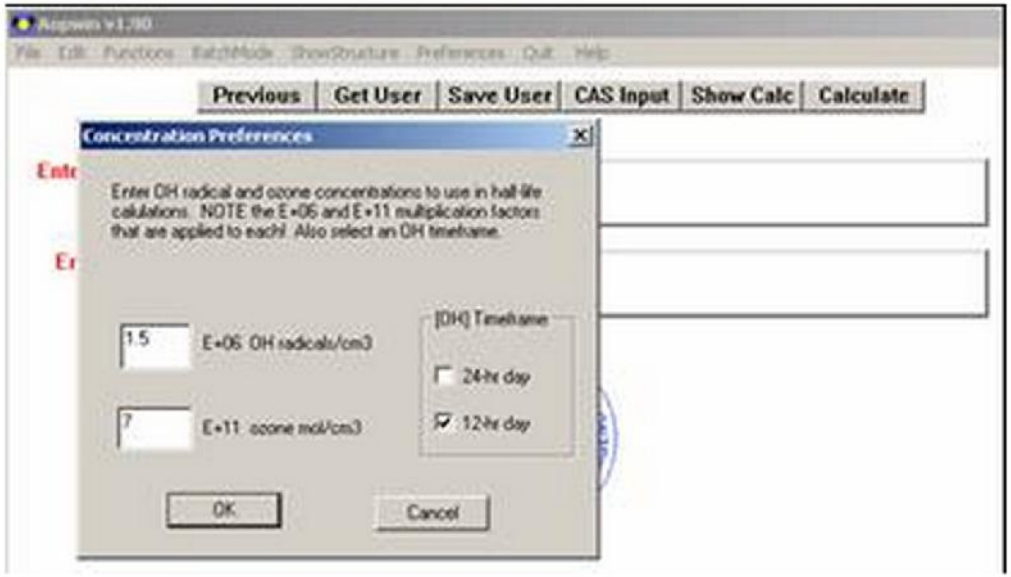
Calculating Oxidative Chemical Stability

Syracuse Research Corporation Aopwin software, free download from:

http://www.epa.gov/oppt/exposure/docs/episuitedl.htm



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