

7 SEPTEMBER 2001

Acrylamide

AMD

Data Set

| | |
|--------------------------|---|
| Existing Chemical | : Acrylamide |
| CAS No. | : 79-06-1 |
| Molecular Formula | : C ₃ H ₅ NO |
| Molecular Weight | : 71.079 |
| Printing Date | : 7 September 2001 Originally prepared in IUCLID 3.1 |

1.2 SYNONYMS

2-PROPEENAMIDE

2-propenamid

2-PROPENAMIDE

2-Propenamide

2-propenamide

2-Propenamide; Acrylic Amide; Ethylene Carboxamide; Propenamide

Acrylamide Monomer

ACRYLAMIDE

Acrylamide

acrylamide

Acrylamide, Akryyliamidi

Acrylic acid amide

Acrylic amide

acrylic amide

Acrylseureamid

Acrylsäureamid

Ethylencarboxamid

Ethylencarboxamide

ethylenecarboxamide

PROPEENAMIDE

Propenamid

propenamide

Propensaureamid

Vinyl amide

vinyl amide

VINYLAMIDE

2.1 MELTING POINT

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 84 ° C
Decomposition :
Sublimation :
Result :
Conclusion :
Reliability : (2) Reliable with restriction, primary source not in English.
Reference : (Van der Burg, 1922)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 84.5 ± 0.3 ° C
Decomposition :
Sublimation :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance : Purified Acrylamide
Method :
GLP : Pre-GLP
Year :
Test conditions : Constant-temperature-differential melting point apparatus utilizing platinum resistance thermometers as the sensing elements.
Value : = 84.5 ± 0.3° C
Decomposition :
Sublimation :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Carpenter and Davis, 1957)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 84.5 ° C
Decomposition :
Sublimation :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (The Merck Index, 12th Edition)

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Test substance : Acrylamide
Method : other: Estimated by the MPBPWIN Program (v.1.40), using the Adapted Joback Method value
GLP : No data
Year : 2001
Test conditions :
Value : = 83.75 ° C
Decomposition :
Sublimation :
Result :
Conclusion : 1.00 deg C (Gold and Ogle Method)
21.7 deg C (Weighted value)
Mean Melt Pt 42.38 deg C (Joback; Gold, Ogle Methods)
Reliability : (2) reliable with restriction. Data obtained by modeling.
Reference : (Klimisch et al., 1997)(Syracuse Research Corporation 1998)

2.2 BOILING POINT

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 87 ° C @ 0.27 kPa (2.03 mmHg)
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 103 ° C @ 0.67 kPa (5.03 mmHg)
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 116 ° C @ 0.67 kPa (10.5 mmHg)
Pressure :
Pressure unit :
Decomposition :
Result :

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 136 ° C @ 3.3 kPa (24.8 mmHg)
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 87 ° C @ 2 mmHg
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (The Merck Index, 12th Edition)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 103 ° C @ 5 mmHg
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (The Merck Index, 12th Edition)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 125 ° C @ 25 mmHg
Pressure :
Pressure unit :
Decomposition :
Result :
Conclusion :

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Reliability : (1) reliable without restriction
Reference : (The Merck Index, 12th Edition)

2.4 VAPOUR PRESSURE

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : 0.007 mmHg
Temperature : 25 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : 0.033 mmHg
Temperature : 40 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : 0.07 mmHg
Temperature : 50 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Kirk-Othmer, 1991)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : Ca. 15 mmHg
Temperature : 20 ° C
Decomposition :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (DOW, 1989)

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 18.8 mmHg
Temperature : 25 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (MacWilliams, 1978)

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 0.03 mmHg
Temperature : 40 ° C
Decomposition :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Thomas, 1964)

Test substance : Purified Acrylamide
Method : Flow method
GLP : Pre-GLP
Year : 1957
Test conditions : 10⁰ – 150⁰ C
Value : = 0.033 mmHg
Temperature : 40 ° C
Decomposition : No decomposition within the range tested
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Carpenter and Davis, 1957)

Test substance : Purified Acrylamide
Method : Flow-method
GLP : Pre-GLP
Year : 1957
Test conditions :
Value : = 0.08 mmHg
Temperature : 50 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Carpenter and Davis, 1957)

Test substance :
Method :
GLP : No data
Year :

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Test conditions :
Value : = 1.6 mmHg
Temperature : 84.5 ° C
Decomposition :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Lewis, 2000)

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 2.0 mmHg
Temperature : 87 ° C
Decomposition :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Thomas, 1964)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : ca. 2.0 mmHg
Temperature : 87 ° C
Decomposition :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (DOW, 1989)

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 25 mmHg
Temperature : 125 ° C
Decomposition :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Thomas, 1964)

2.5 PARTITION COEFFICIENT

Test substance :
Method : Other: calculated
GLP : Pre-GLP
Year :
Test conditions :

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Log Pow : = -1.65
Temperature : ° C
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (US EPA, 1980)

Test substance :
Method : Other: calculated
GLP : Pre-GLP
Year : 1981
Test conditions : Calculated from HPLC with a retention time of 3.1 minutes
Log Pow : = -1.24
Temperature :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Fujisawa and Masuhara, 1981)

Test substance :
Method : Other: calculated
GLP : Pre-GLP
Year :
Test conditions :
Log Pow : = -1.04
Temperature : ° C
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Hermens and Leeuwangh, 1982)

Test substance :
Method : Other: measured
GLP : Pre-GLP
Year : 1980
Test conditions : Calculated using the flask-shaking method
Log Pow : = -0.9
Temperature :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Fujisawa and Masuhara, 1980)

Test substance :
Method : Other: calculated (CLOGP3 computer program based upon the fragment constant methodology)
GLP : No data
Year :
Test conditions :
Log Pow : = - 0.86
Temperature :
Result :
Conclusion :
Reliability : (2) reliable with restriction, values obtained by modeling
Reference : (Lipnick, et al., 1987)

Test substance :

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Method : Other: measured
GLP : Pre-GLP
Year :
Test conditions :
Log Pow : = -.67
Temperature : ° C
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Hansch and Leo, 1979)

2.6.1 WATER SOLUBILITY

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 215.5 g/100 ml at 30 ° C
Solubility :
pH at ° C :
pKa at 25 ° C :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Budavari 1989)

Test substance :
Method :
GLP : No data
Year :
Test conditions :
Value : = 215.5 g/100 ml at 30 ° C
Solubility :
pH at ° C :
pKa at 25 ° C :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (The Merck Index, 12th Edition)

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 2040 g/l at 25 ° C
Solubility :
pH at ° C :
pKa at 25 ° C :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Thomas, 1964)

2. Physico-Chemical Data

Id 79-06-1

Date 7 September 2001

Test substance : Purified Acrylamide
Method :
GLP : Pre-GLP
Year :
Test conditions : Solution saturated at 30 ° C and analysed for Acrylamide; value obtained was then checked by cryoscopic measurements on solutions of known composition.
Value : = 2155 g/l at 30 ° C
Solubility :
pH at ° C :
pKa at 25 ° C :
Result :
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Carpenter and Davis, 1957)

Test substance :
Method :
GLP : Pre-GLP
Year :
Test conditions :
Value : = 4260 g/l at 50 ° C
Solubility :
pH at ° C :
pKa at 25 ° C :
Result :
Conclusion :
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Thomas, 1964)

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Please note, Section 3 is still under review and revision.

3.1.1 PHOTODEGRADATION

Type :
Light source :
Light spect. : Nm
Rel. intensity : Based on Intensity of Sunlight
Deg. Product :
Method :
Year :
GLP : No data
Test substance :
Results : Atmospheric T $\frac{1}{2}$ = 6.6 hours
Reliability : (1) reliable without restriction
Reference : (GEMS, 1986)

Type : Water
Reactant : Hydroxyl radical
Deg. Product : Oxidation products
Method : Kinetic studies at pH 10.7
Year :
GLP : No data
Test substance : Acrylamide
Result : Rate Constant = 3.83×10^{-12} cu. Cm/molecule/sec.
Reliability : (1) Reliable without reservation
Reference : (Anbar and Neta 1967)(Matthews and Sangster 1965)

Deg. Product :
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide
Remark : The photodegradation rate is estimated by the EPIWIN/AOPWIN model which estimates rate constants and half-lives of atmospheric reactions of organic compounds with hydroxyl radicals and ozone in the atmosphere.
Result : Half-Life: 0.955 days
Half-Life: 11.455 hours
Overall OH Rate Constant = 11.2050×10^{-12} cm³/molecule-sec
Reliability : (2) valid with restrictions. Data were obtained by modeling.
Reference : (Klimisch et al. 1997)(Syracuse Research Corporation 1998)

3.1.2 STABILITY IN WATER

Type : Abiotic
Results : Degradation = 100%
Remark : Rapid biodegradation of AMD (100-700h) occurs in natural and polluted environmental water in the range of pH 4-10. Biodegradation occurred all non-sterized conditions (i.e. aerobic and anerobic and light and dark). AMD breakdown occurred due to microbial degradation not chemical reactions.
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980)

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Type : Abiotic
Test substance : Acrylamide
Results :
Remarks : In an aqueous environment, acrylamide hydrolyzes to acrylic acid and ammonia with hydroxy and hydrogen ions acting as catalysts.
Reliability : (2) Reliable with restriction, primary source not in English.
Reference : (Jung et al., 1980)

Type : Abiotic
T1/2 pH4 : at degree C
T1/2 pH7 : at degree C
T1/2 pH9 : at degree C
Deg. Product : Acrylic acid and ammonia
Method : Hydrolysis studies at various pH's and temperatures
Year : 1957
GLP : Pre-GLP
Test substance : Acrylamide
Results : Rate Constants
Alkaline pH -1.47 x 10⁻⁴/mole/sec (55° C)
-3.70 x 10⁻⁴/mole/sec (65° C)
6.66 x 10⁻⁴/mole/sec (75° C)
13.8 x 10⁻⁴/mole/sec (85° C)
Acid ph -1.48 x 10⁻⁴/mole/sec (80° C)
5.5 x 10⁻⁴/mole/sec (95° C)
16.6 x 10⁻⁴/mole/sec (110° C)

Reliability : (1) reliable without restriction
Reference : (Moens and Smets, 1957)

Type : Abiotic
Method : other (calculated): Estimated by the EPIWIN/HYDROWIN Program (v1.67).
Year : 2001
GLP : No
Test substance : Acrylamide
Remark : This program was not able to estimate a hydrolysis rate constant for this tpe of chemical structure. However, this compound has an amide group; C=O located at SMILES atom #: 2; hydrolysis rate extremely slow or t1/2 > 1 year.
Reliability : (2) valid with restrictions. Data were obtained by modeling.
Reference : (Klimisch et al., 1997)(Syracuse Research Corporation 1998)

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type : fugacity model level III
Media :
Air (level III) : 0.032%
Water (level III) : 45.3%
Soil (level III) : 54.5%
Sediment (level III) : 0.0757%
Method : other: Estimated by the Level III Fugacity Model (Full-Output)
Year : 2001
Remark : The fugacity calculated by an accepted method is assigned a reliability code of 2f according to the criteria established by Klimisch et al. (1997)
Reliability : (2) valid with restrictions. Data were obtained by modeling.
Reference : (Klimisch et al., 1997)(Syracuse Research Corporation 1998)

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

3.5 BIODEGRADATION

Type : Aerobic
Inoculum : Other: activated sludge bacteria from Bergen County, New Jersey
Contact time :
Degradation :

| <u>2 mg</u> | <u>5 mg</u> | <u>Duration</u> |
|-------------|-------------|-----------------|
| 7.4 % | 7.4% | After 5 day |
| 75.9% | 57 % | 15 day |
| 100% | 53.3 % | 28 day |

Result : Readily biodegradable
Kinetic of test substance : 15 day = 75.9 %
: 28 day = 100 %

Deg. Product :
Method : OECD Guide-line 301 D "Ready Biodegradability: Closed Bottle Test"
Year : 1990
GLP : No data
Test substance : Other TS: CT-444-90D: purity > 98 %
Reliability : (1) reliable without restriction
Reference : (United States Testing Company 1991)

Type : Aerobic
Inoculum : Five different soil types from Iowa
Contact time : 2-21 days
Degradation : At 3 days, degradation ranged from 11 – 71% (depending upon soil type)
: At 14-days, degradation ranged from 74-94%.
: After 21-days under waterlogged conditions, degradation ranged from 76-93%.

Result : Readily biodegradable
Kinetic of test substance : Elim
:

Deg. Product : Elim
Method : Measurement of inorganic nitrogen
Year : 1982
GLP : No data
Test substance : Analytical grade obtained from Sigma Chemical
Reliability : (1) reliable without restriction
Reference : (Abdelmagid and Tabatabai, 1982)

Type : Aerobic
Inoculum : Other: effluent from a biological sanitary waste treatment plant, adapted
Concentration : 10mg/l related to Test substance
Contact time :
Degradation : = 67 - 69 % after 5 day
Result :
Deg. Product :
Method : Other: APHA-Standardverfahren, No. 219
Year : 1971
GLP : Pre-GLP
Test substance : No data
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Bridie et al., 1979)(Winter and Wolff 1982)

Type : Aerobic

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

| | | |
|----------------------------------|---|--|
| Inoculum | : | Other: effluent from a biological sanitary waste treatment plant, non adapted |
| Concentration | : | 10mg/l related to Test substance Related to |
| Contact time | : | |
| Degradation | : | = 3 - 4 % after 5 day |
| Result | : | |
| Deg. Product | : | |
| Method | : | Other: APHA-Standardverfahren, No. 219 |
| Year | : | 1971 |
| GLP | : | Pre-GLP |
| Test substance | : | No data |
| Reliability | : | (2) reliable with restriction, information obtained from a secondary source |
| Reference | : | (Bridie et al., 1979)(Winter and Wolff 1982) |
| Type | : | Aerobic |
| Inoculum | : | Activated sludge |
| Concentration | : | 83µg/l related to Test substance Related to |
| Contact time | : | |
| Degradation | : | = 0 % after 1 hour(s) |
| Result | : | |
| Kinetic of test substance | : | 2 hour(s) = 2 % 3 hour(s) = 10 % 4 hour(s) = 25 % 24 hour(s) = 65 % |
| Deg. Product | : | |
| Method | : | Other: Biodegradation Test |
| Year | : | 1982 |
| GLP | : | Pre-GLP |
| Test substance | : | No data |
| Remark | : | The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little <i>in situ</i> degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated slude tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%) . No AMD was lost from the tapwater control. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Brown et al., 1982) |
| Type | : | Aerobic |
| Inoculum | : | Activated sludge |
| Concentration | : | 75µg/l related to Test substance related to |
| Contact time | : | |
| Degradation | : | = 2 % after 1 hour(s) |
| Result | : | |
| Kinetic of test substance | : | 2 hour(s) = 8 % 3 hour(s) = 15 % 4 hour(s) = 35 % 24 hour(s) = 100 % |
| Deg. Product | : | |
| Method | : | other: Biodegradation Test |

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Year : 1982
GLP : Pre-GLP
Test substance : no data
Remark : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little *in situ* degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated slude tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%) .No AMD was lost from the tapwater control.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1982)

Type : Aerobic
Inoculum : Activated sludge
Concentration : 50µg/l related to Test substance related to

Contact time :
Degradation : = 5 % after 1 hour(s)
Result :
Kinetic of test substance : 2 hour(s) = 25 %
3 hour(s) = 50 %
4 hour(s) = 65 %
24 hour(s) = 100 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1982
GLP : Pre-GLP
Test substance : no data
Remark : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little *in situ* degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated slude tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%) .No AMD was lost from the tapwater control.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1982)

Type : Aerobic
Inoculum : Activated sludge
Concentration : 444µg/l related to Test substance related to

Contact time :
Degradation : = 50.5 % after 48 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little *in situ* degradation of aMD in the primary or the final settlement tanks of the

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

seage works studied. The high microbial activity associated with the activated sludge tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%). .No AMD was lost from the tapwater control.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1982)

Type : Aerobic
Inoculum : Activated sludge
Concentration : 444µg/l related to Test substance related to

Contact time :
Degradation : = 55.6 % after 48 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little *in situ* degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated sludge tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%). .No AMD was lost from the tapwater control.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1982)

Type : Aerobic
Inoculum : Activated sludge
Concentration : 494µg/l related to Test substance related to

Contact time :
Degradation : = 62.3 % after 24 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little *in situ* degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated sludge tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%). .No AMD was lost from the tapwater control.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1982)

Type : Aerobic
Inoculum : Activated sludge
Concentration : 155µg/l related to Test substance

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

| | |
|-----------------------|---|
| | related to |
| Contact time | : |
| Degradation | : = 74.2 % after 24 hour(s) |
| Result | : |
| Deg. Product | : |
| Method | : other: Biodegradation Test |
| Year | : 1980 |
| GLP | : Pre-GLP |
| Test substance | : no data |
| Remark | : The biodegradation of AMD was tested in an activated sewage works, a biological filter sewage works and a river . There appeared to be little <i>in situ</i> degradation of aMD in the primary or the final settlement tanks of the seage works studied. The high microbial activity associated with the activated sludge tanks appear to result in a 50-70% reduction of dosed AMD. Both heat (70C and mercuric chloride (10mg/l) decreased AMD degradation. A rapid pass through a filter was highly efficient in reducing AMD (50%). .No AMD was lost from the tapwater control. |
| Reliability | : (1) reliable without restriction |
| Reference | : (Brown et al., 1982) |
| Type | : Aerobic |
| Inoculum | : Arthrobacter sp. (Bacteria) |
| Concentration | : 5g/l related to related to |
| Contact time | : |
| Degradation | : = 100 % after 7 day |
| Result | : |
| Deg. Product | : |
| Method | : other: Biodegradation Test |
| Year | : 1979 |
| GLP | : Pre-GLP |
| Test substance | : no data |
| Reliability | : (1) reliable without restriction |
| Reference | : (Yamada et al., 1979) |
| Type | : Aerobic and Anerobic |
| Inoculum | : other: four soil types (silt loam, clay loam, loamy fine sand and loam) Soil samples obtained in Spring and Summer |
| Concentration | : AMD (25 or 500 ppm) |
| Contact time | : Evaluated time it took to release 50% of total CO ₂ |
| Degradation | : At 22 °C (25 ppm) – 50% in 18-45 hr. Using 500 ppm – 94 hr. At 10 °C (25 ppm) – 96 hr. At 37 °C – 10 hr. |
| Result | : AMD half life was increased by decreased temperature or increasing AMD concentration. Half-life was 2.5 times greater in soil gathered in Spring compared to Summer. Anerobic metabolism was slower than aerobic metabolism. With regard to leaching, AMD is considered mobile. |
| Deg. Product | : Carbon dioxide |
| Method | : Biometer flask method |
| Year | : 1979 |
| GLP | : Pre-GLP |
| Test substance | : Synthesized by New England Nuclear |
| Reliability | : (1) reliable without restriction |
| Reference | : (Lande et al. 1979) |
| Type | : Aerobic |
| Inoculum | : other: English China Clay's Blackpool Pit effluent |

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Concentration : .5mg/l related to Test substance related to

Contact time :

Degradation : = 100 % after 6.3 day

Result :

Deg. Product :

Method : other: Biodegradation Test

Year : 1980

GLP : Pre-GLP

Test substance : no data

Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction

Reference : (Brown et al., 1980b)

Type : Aerobic

Inoculum : other: English China Clay's Blackpool Pit hosepool

Concentration : .5mg/l related to Test substance related to

Contact time :

Degradation : = 100 % after 15.6 day

Result :

Deg. Product :

Method : other: Biodegradation Test

Year : 1980

GLP : Pre-GLP

Test substance : no data

Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction

Reference : (Brown et al., 1980b)

Type : Aerobic

Inoculum : other: English China Clay's Blackpool process water

Concentration : .5mg/l related to Test substance related to

Contact time :

Degradation : = 100 % after .9 day

Result :

Deg. Product :

Method : other: Biodegradation Test

Year : 1980

GLP : Pre-GLP

Test substance : no data

Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction

Reference : (Brown et al., 1980b)

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Type : Aerobic
Inoculum : other: effluent from a waste water treatment plant
Concentration : .5mg/l related to Test substance related to
Contact time :
Degradation : = 100 % after 5.2 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: estuarine water
Concentration : .5mg/l related to Test substance related to
Contact time :
Degradation : = 100 % after 7.3 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: peat bog water
Concentration : .5mg/l related to Test substance related to
Contact time :
Degradation : = 100 % after 5.2 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: river water
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : = 100 % after 4.2 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: river water
Concentration : 5mg/l related to Test substance related to

Contact time :
Degradation : = 100 % after 75 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: sea water
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : = 100 % after 10.4 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: tap water
Concentration : .5mg/l related to related to

Contact time :
Degradation : = 0 % after 83 day
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : No data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Anaerobic
Inoculum : other: river water
Concentration : 5mg/l related to Test substance related to

Contact time :
Degradation : ca. 100 % after 100 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Anaerobic
Inoculum : other: river water
Concentration : 5mg/l related to Test substance related to

Contact time :
Degradation : ca. 100 % after 200 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

GLP : Pre-GLP
Test substance : no data
Remark : The adsorption of AMD to peat, sludges and sediments was tested. No significant adsorption was detected. No AMD was removed from sterilized river water by various clays, anionic, cationic or hydrophobic resins at various pH values and concentrations. Activated carbon was found to have a limited affinity for AMD which was not affected in the pH range tested.

Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980b)

Type : Aerobic
Inoculum : other: river water
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : = 100 % after 168 hour(s)
Result :
Kinetic of test substance : 4 hour(s) = 0 %
24 hour(s) = 0 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: estuarine water
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : = 100 % after 168 hour(s)
Result :
Kinetic of test substance : 4 hour(s) = 0 %
24 hour(s) = 0 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: estuarine water with sediment
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : 100 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: river water with sediment
Concentration : .5mg/l related to related to

Contact time :
Degradation : 100 % after 168 hour(s)
Result :
Kinetic of test substance : 24 hour(s) = 41 %
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al.,1980c)

Type : Aerobic
Inoculum : other: river water with sediment
Concentration : 10mg/l related to related to

Contact time :
Degradation : 100 % after 168 hour(s)
Result :
Kinetic of test substance : 224 hour(s) = 21 %
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sea water
Concentration : 10mg/l related to Test substance related to

Contact time :
Degradation : = 7 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sea water
Concentration : .5mg/l related to Test substance related to

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Contact time :
Degradation : = 10 % after 168 hour(s)
Result :
Kinetic of test substance : 4 hour(s) = 0 %
24 hour(s) = 0 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sea water with sediment
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : 75 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sea water with sediment
Concentration : 10mg/l related to related to

Contact time :
Degradation : 77 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sewage works effluent
Concentration : 10mg/l related to Test substance related to

Contact time :
Degradation : = 28 % after 168 hour(s)
Result :
Kinetic of test substance : 4 hour(s) = 0 %
24 hour(s) = 0 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1980

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sewage works effluent
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : = 38 % after 168 hour(s)
Result :
Kinetic of test substance : 4 hour(s) = 0 %
24 hour(s) = 0 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sewage works effluent
Concentration : 10mg/l related to Test substance related to

Contact time :
Degradation : 60 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: sewage works effluent
Concentration : .5mg/l related to Test substance related to

Contact time :
Degradation : 65 % after 168 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1980
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Brown et al., 1980c)

Type : Aerobic
Inoculum : other: Rhodococcus (10021R)
Concentration : 1300mg/l related to Test substance related to

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Contact time :
Degradation : = 100 % after 4 hour(s)
Result :
Deg. Product :
Method : other: Biodegradation Test
Year : 1981
GLP : Pre-GLP
Test substance : no data
Remark : Among the various microorganisms that biodegrade AMD, an actinomycete strains designed 10 021R was found. This organism degrades AMD to ammonia and carbon dioxide via acrylic acid.

Reliability : (1) reliable without restriction
Reference : (Arai et al., 1981)

Type : Aerobic
Inoculum : other: Rhodococcus (10021R)
Concentration : 1000mg/l related to Test substance
Related to

Contact time :
Degradation : = 85 % after 1 hour(s)
Result :
Kinetic of test substance : 2 hour(s) = 100 %
Deg. Product :
Method : other: Biodegradation Test
Year : 1981
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Arai et al., 1981)

Type : Aerobic
Inoculum : other: effluent from a biological waste water treatment plant

Contact time :
Degradation : = 34 % after 5 day
Result : readily biodegradable
Kinetic of test substance : 10 day = 57 - 84 %
20 day = 87 %

Deg. Product :
Method : other: Biodegradation Test
Year : 1975
GLP : Pre-GLP
Test substance : No data
Reliability : (2) reliable with restriction, original data source not reviewed
Reference : (DOW 1975)

Type : Aerobic
Inoculum : other: effluent from a biological waste water treatment plant

Contact time :
Degradation : = 54 % after 5 day
Result : readily biodegradable
Deg. Product :
Method : other: Biodegradation Test
Year : 1975
GLP : Pre-GLP
Test substance : No data

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Reliability : (2) reliable with restriction, original data source not reviewed
Reference : (DOW 1975)

Type : Aerobic
Inoculum : other: industrial sewage
Contact time :
Degradation : = 48 - 50 % after 5 day
Result : readily biodegradable
Kinetic of test substance : 10 day = 50 %
20 day = 50 %

Deg. Product Method :
: other: Biodegradation Test
Year : 1975
GLP : Pre-GLP
Test substance : No data
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Batchelder 1975)(DOW 1975)

Type : Aerobic
Inoculum : other: industrial sewage
Contact time :
Degradation : = 76 - 80 % after 5 day
Result : readily biodegradable
Kinetic of test substance : 10 day = 80 %
20 day >= 80 %

Deg. Product Method :
: other: Biodegradation Test
Year : 1975
GLP : Pre-GLP
Test substance : No data
Reliability : (2) reliable with restriction, information obtained from a secondary source
Reference : (Batchelder 1975)(DOW 1975)

Type : Aerobic
Inoculum : other: river water
Concentration : 8µg/l of Test substance
Contact time :
Degradation : = 90 % after 150 hour(s)
Result :
Deg. Product Method :
: Other: Biodegradation Test
Year : 1974
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Croll et al., 1974)

Type : Aerobic
Inoculum : other: river water with culture capable of degrading acrylamid
Concentration : 8µg/l of Test substance
Contact time :
Degradation : = 100 % after 38 hour(s)
Result :
Deg. Product Method :
: other: Biodegradation Test

3. Environmental Fate and Pathways

Id 79-06-1

Date 7 September 2001

Year : 1974
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Croll et al., 1974)

Type : Aerobic
Inoculum : other: effluent from a waste water treatment plant
Concentration : 170µg/l of Test Substance

Contact time :
Degradation : = 0 % after 5 day
Result :
Kinetic of test substance : 16 day = 100 %
Deg. Product :
Method : other: Open Aeration Bottle Test
Year : 1973
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Croll et al., 1974)

Type : Anaerobic
Inoculum : other: sewage effluent
Concentration : 160µg/l of Test Substance

Contact time :
Degradation : = 6 % after 5 day
Result :
Kinetic of test substance : 9 day = 100 %
Deg. Product :
Method : other: Closed Bottle Test
Year : 1973
GLP : Pre-GLP
Test substance : no data
Reliability : (1) reliable without restriction
Reference : (Croll et al., 1974)

4.1 ACUTE/PROLONGED TOXICITY TO FISH

| | |
|------------------------------|---|
| Type | : Flow through |
| Species | : Lepomis macrochirus (Bluegill Sunfish, fresh water) |
| Exposure period | : 18-96 hours |
| Unit | : mg/l |
| Analytical monitoring | : Yes |
| LC50 | : 18 hr = >350mg/l, 95%CI (---) 24 hr = 260mg/l, 95%CI (150-350) 42 hr = 190mg/l, 95%CI (150-350) 48 hr = 160mg/l, 95%CI (81-350) 66 hr = 130mg/l, 95%CI (81-150) 72 hr = 120mg/l, 95%CI (81-150) 90 hr = 100mg/l, 95%CI (81-150) 96 hr = 100mg/l, 95%CI (81-150) |
| EC50 | : 96 hr = 85 mg/l |
| NOEC | : 96 hr = 35 mg/l |
| Method | : Test performed according to APHA „Standard Method for Examination of Water and Wastewater“ and „Methods of Acute Toxicity Test with Fish, Macroinvertebrates and Amphibians“ |
| Year | : 1982 |
| GLP | : Yes |
| Test substance | : > 99 % pure |
| Test condition | : Water quality parameters of temperature, dissolved oxygen, pH and ammonia were measured throughout the test and were within acceptable limits. A flow through proportional diluter system was used to maintain constant test concentrations of acrylamide monomer. Nominal exposure concentrations were 17, 36, 69, 140, and 300 mg/l. The mean measured concentrations were 14, 35, 81, 150 and 250 mg/l. Measured concentrations were used in the LC50 determinations according to Stephan et al. 1978. |
| Result | : LC50 values presented above |
| Conclusion | : Based on the behavioral observations conducted twice daily throughout the study, it was noted that mortality was preceded by surfacing and loss of equilibrium of test fish. Taking all behavior responses into account, a 96hr EC50 was calculated to be 85 mg/l. In addition, the results indicated a 96hr NOEC at 35 mg/l, based on mortality and behavioral effects. |
| Reliability | : (1) reliable without restriction |
| Reference | : (ABC Labs 1982a) |
| Type | : flow through |
| Species | : Pimephales promelas (Fathead Minnow, fresh water) |
| Exposure period | : 18-96 hours |
| Unit | : mg/l |
| Analytical monitoring | : Yes |
| LC50 | : 18 hr = >340mg/l, 95%CI (---) 24 hr = 320mg/l, 95%CI (---) 42 hr = 230mg/l, 95%CI (160-340) 48 hr = 230mg/l, 95%CI (160-340) 66 hr = 220mg/l, 95%CI (160-340) 72 hr = 210mg/l, 95%CI (160-340) 90 hr = 130mg/l, 95%CI (77-160) 96 hr = 120mg/l, 95%CI (77-160) |
| EC50 | : 96 hr = 86 mg/l |
| NOEC | : 96 hr = 41 mg/l |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | |
|------------------------------|--|
| Method | : Test performed according to APHA „Standard Method for Examination of Water and Wastewater“ and „Methods of Acute Toxicity Test with Fish, Macroinvertebrates and Amphibians“ |
| Year | : 1982 |
| GLP | : Yes |
| Test substance | : > 99 % pure |
| Test condition | : Water quality parameters of temperature, dissolved oxygen, pH and ammonia were measured throughout the test and were within acceptable limits. A flow through proportional diluter system was used to maintain constant test concentrations of acrylamide monomer. Nominal exposure concentrations were 17, 36, 69, 140, and 300 mg/l. The mean measured concentrations were 21, 41, 77, 160, and 340 mg/l. Measured concentrations were used in the LC50 determinations according to Stephan et al. 1978. |
| Result | : LC50 values presented above |
| Conclusion | : Based on the behavioral observations conducted twice daily throughout the study, it was noted that mortality was preceded by surfacing and loss of equilibrium of test fish. Taking all behavior responses into account, a 96hr EC50 was calculated to be 86 mg/l. In addition, the results indicated a 96hr NOEC at 41 mg/l, based on mortality and behavioral effects. |
| Reliability | : (1) reliable without restriction |
| Reference | : (ABC Labs 1982b) |
| Type | : Flow through |
| Species | : Rasbora heteromorpha (Harlequin Fish, marine) |
| Exposure period | : 24-96 hours |
| Unit | : mg/l |
| Analytical monitoring | : No |
| LC50 | : 24 hr = 460mg/l 48 hr = 250mg/l 96 hr = 130mg/l |
| LC10 | : 24 hr = 390mg/l 48 hr = 220mg/l 96 hr = 103mg/l |
| Method | : UK Ministry of Agriculture, Fisheries, and Food (MAFF) procedure (Alabaster and Abram, 1965) |
| Year | : 1975 |
| GLP | : No |
| Test substance | : 100 % pure |
| Test condition | : Water hardness – 20 mg/l (calcium carbonate), pH – 8.1, test temperature - 20C. Fish were acclimatized to the dilution water for one week. LC50 was calculated using Litchfield – Wilcoxon method, which in turn was used to calculate LC10. The concentration survival time graph was extrapolated to give an estimated 3-month median lethal concentration. Water pH = 8.1, dissolved oxygen N/a, water hardness – 20 mg/l (CaCO ₂), and temp was 20C. |
| Result | : See values listed above |
| Conclusion | : The extrapolated 3 month LC50 was calculated to be 10 mg/l. |
| Reliability | : (1) reliable without restriction |
| Reference | : (Tooby and Hursey 1975) |
| Type | : Flow through |
| Species | : Salmo gairdneri (estuary, fresh water) |
| Exposure period | : 16 – 96 hours |
| Unit | : mg/l |
| Analytical monitoring | : Yes |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| LC50 | : | 16 hr = >370mg/l, 95%CI (---) 24 hr = >370mg/l, 95%CI (---) 41 hr = 240mg/l, 95%CI (150-370) 48 hr = 240mg/l, 95%CI (150-370) 66 hr = 220mg/l, 95%CI (150-370) 72 hr = 190mg/l, 95%CI (150-370) 90 hr = 120mg/l, 95%CI (74-150) 96 hr = 110mg/l, 95%CI (74-150) |
| EC50 | : | 96 hr = 88mg/l |
| NOEC | : | 96 hr = 37mg/l |
| Method | : | other: Standard Method for Acute Toxicity Test with Fish, Macroinvertebrates and Amphibians |
| Year | : | 1975 |
| GLP | : | Yes |
| Test substance | : | > 99 % pure |
| Test condition | : | Water quality parameters of temperature, dissolved oxygen, pH and ammonia were measured throughout the test and were within acceptable limits. A flow through proportional diluter system was used to maintain constant test concentrations of acrylamide monomer. Nominal exposure concentrations were 17, 36, 69, 140, and 300 mg/l. The mean measured concentrations were 17, 37, 74, 150, and 370 mg/l. Measured concentrations were used in the LC50 determinations according to Stephan et al. 1978. |
| Result | : | LC50 values presented above |
| Conclusion | : | Based on the behavioral observations conducted twice daily throughout the study, it was noted that mortality was preceded by surfacing and loss of equilibrium of test fish. Taking all behavior responses into account, a 96hr EC50 was calculated to be 88 mg/l. In addition, the results indicated a 96hr NOEC at 37 mg/l, based on mortality and behavioral effects. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (ABC Labs 1982d) |
| Type | : | Static |
| Species | : | Carassius auratus (Goldfish, fresh water) |
| Exposure period | : | 24 or 96 hours |
| Unit | : | Mg/l |
| Analytical monitoring | : | No data |
| LC50 | : | 24 hr = 460 mg/l 96 hr = 160 mg/l |
| Method | : | APHA-Guideline No. 231 – static tank acute toxicity tests |
| Year | : | 1979 |
| GLP | : | No |
| Test substance | : | No data |
| Test condition | : | 10 fish were exposed at 20C in a glass tank for the duration of the test and the solutions were aerated throughout the test period. Water pH = 7.8, dissolved oxygen 9-10 mg/l, water hardness – n/a (CaCO ₂), and temp was 20C. |
| Result | : | See values above. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Bridie et al., 1979) |
| Type | : | Static |
| Species | : | Carassius auratus (Goldfish, fresh water) |
| Exposure period | : | 72 hours |
| Unit | : | Ppm |
| Analytical monitoring | : | No |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| LC50 | : | 140 ppm |
| Method | : | Acute Toxicity Test |
| Year | : | 1974 |
| GLP | : | no data |
| Test substance | : | no data |
| Test condition | : | Water pH = n/a, dissolved oxygen n/a, water hardness – n/a (CaCO ₂), and temp was n/a. |
| Result | : | See value listed above |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, data obtained from secondary source, primary source not in English |
| Reference | : | (Paulet and Vidal 1975) |
| Type | : | Static |
| Species | : | Carassius auratus (Goldfish, fresh water) |
| Exposure period | : | 7 or 30 days |
| Unit | : | Ppm |
| Analytical monitoring | : | Yes |
| LC100 | : | 100 ppm at 7 days |
| NOEC | : | 50 ppm at 30 days |
| Method | : | According to Mattocks, 1968 |
| Year | : | 1975 |
| GLP | : | No data |
| Test substance | : | No data |
| Test condition | : | No additional information |
| Result | : | Continuous exposure of goldfish to 100 ppm killed all seven in 5-7 days. No effects were seen at 50 ppm for 30 days. |
| Conclusion | : | A variety of dosing and exposure regimes was used, but no effects could be seen with sublethal doses. |
| Reliability | : | (2) reliable with restriction, very limited information regarding testing conditions, limited information regarding exposure concentrations |
| Reference | : | (Edwards, 1975) |
| Type | : | Static |
| Species | : | Oncorhynchus mykiss (Fish, fresh water) |
| Exposure period | : | 24 – 96 hrs |
| Unit | : | mg/l |
| Analytical monitoring | : | No |
| LC50 | : | 24-hr LC ₅₀ 500 mg/l 48-hr LC ₅₀ 360 mg/l 72-hr LC ₅₀ 240 mg/l 96-hr LC ₅₀ 180 mg/l |
| LC100 | : | 1000 mg/l |
| Method | : | OECD Guide-line 203 "Fish, Acute Toxicity Test" |
| Year | : | 1990 |
| GLP | : | Yes |
| Test substance | : | > 98.5 % pure |
| Test condition | : | Information on the test substance was not available, water pH = 7.5-7.7, dissolved oxygen 8.0-8.6 mg/l, water hardness – 90 mg/l (CaCO ₂), and temp was 15C. |
| Result | : | See values above |
| Conclusion | : | |
| Reliability | : | (1) reliable without restrictions |
| Reference | : | (United States Testing Company 1990) |
| Type | : | Static |
| Species | : | Oncorhynchus mykiss (Rainbow trout, fresh water) |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | |
|------------------------------|--|
| Exposure period | : 24 – 96 hours |
| Unit | : mg/l |
| Analytical monitoring | : No data |
| LC50 | : 24-hr LC ₅₀ >300 mg/l --- 48-hr LC ₅₀ 210 mg/l Not obtainable 72-hr LC ₅₀ 170 mg/l 95%CI(137-191) 96-hr LC ₅₀ 162 mg/l 95%CI(137-191) |
| Method | : Acute Toxicity test |
| Year | : 1985 |
| GLP | : No |
| Test substance | : > 98 % pure |
| Test condition | : 10 fish/group. LC50's were calculated by Litchfield and Wilcoxon method, 1948. |
| Result | : Values listed above |
| Conclusion | : |
| Reliability | : (2) reliable with restriction, no information provided regarding test conditions for LC50 test |
| Reference | : (Petersen et al., 1985) |
| Type | : Static |
| Species | : Salmo gairdneri (Fish, fresh water) |
| Exposure period | : |
| Unit | : mg/l |
| Analytical monitoring | : |
| LC50 | : 140 mg/l |
| Method | : Acute Toxicity Test |
| Year | : 1982 |
| GLP | : No |
| Test substance | : No data |
| Test condition | : No data |
| Result | : Value is stated above. |
| Conclusion | : Authors indicate conclusions are tentative but results appear to be representative. More testing is required to develop statistical data. |
| Reliability | : (2) reliable with restriction, No details were reported as to the experimental conditions or length of exposure. |
| Reference | : (Spraggs et al., 1982) |
| Type | : Static |
| Species | : Salmo gairdneri (Fish, fresh water) |
| Exposure period | : |
| Unit | : mg/l |
| Analytical monitoring | : |
| 50% Avoidance Lvl | : 8 |
| Method | : Acute Toxicity Test |
| Year | : 1982 |
| GLP | : No |
| Test substance | : no data |
| Test condition | : No data |
| Result | : Value is stated above. |
| Conclusion | : Authors indicate conclusions are tentative but results appear to be representative. More testing is required to develop statistical data. |
| Reliability | : (2) reliable with restriction, No details were reported as to the experimental conditions or length of exposure. |
| Reference | : (Spraggs et al., 1982) |
| Type | : Static |
| Species | : Pimephales promelas (Fish, fresh water) |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

Exposure period : 24 hour(s)
Unit : mg/l
Analytical monitoring : no data
LC50 : = 411
Method : other: Acute Toxicity Test
Year : 1975
GLP : no data
Test substance : as prescribed by 1.1 - 1.4
Reliability : (2) reliable with restriction. Reported to IUCLID by Dow Deutschland Inc. Original not reviewed.
Reference : (DOW 1975)

Type : Static
Species : Pimephales promelas (Fish, fresh water)
Exposure period : 48 hour(s)
Unit : mg/l
Analytical monitoring : no data
LC50 : = 235
Method : other: Acute Toxicity Test
Year : 1975
GLP : no data
Test substance : as prescribed by 1.1 - 1.4
Reliability : (2) reliable with restriction. Reported to IUCLID by Dow Deutschland Inc. Original not reviewed.
Reference : (DOW 1975)

Type : Static
Species : Pimephales promelas (Fish, fresh water)
Exposure period : 72 hour(s)
Unit : mg/l
Analytical monitoring : no data
LC50 : = 158
Method : other: Acute Toxicity Test
Year : 1975
GLP : no data
Test substance : as prescribed by 1.1 - 1.4
Reliability : (2) reliable with restriction. Reported to IUCLID by Dow Deutschland Inc. Original not reviewed.
Reference : (DOW 1975)

Type : Static
Species : Pimephales promelas (Fish, fresh water)
Exposure period : 96 hour(s)
Unit : mg/l
Analytical monitoring : no data
NOEC : = 56
LC50 : = 124
Method : other: Acute Toxicity Test
Year : 1975
GLP : no data
Test substance : as prescribed by 1.1 - 1.4
Reliability : (2) reliable with restriction. Reported to IUCLID by Dow Deutschland Inc. Original not reviewed.
Reference : (DOW 1975)

Type : Static
Species : Salmo trutta (Brown trout, fresh water)

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| Exposure period | : | 48 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | no data |
| LC50 | : | 400 |
| Method | : | |
| Year | : | 1974 |
| GLP | : | no data |
| Test substance | : | no data |
| Test condition | : | 10 fingerling trout were maintained in an aquarium at 10C and after 48 hr emersion the dead fish were counted. Water pH = 7.6-8.0, dissolved oxygen > 50% saturation, water hardness – 210-290 mg/l (CaCO ₂), and temp was 10C. The toxicities of the effluents under test were expressed as that concentration in which 50% of the test fish were killed in 48h. This concentration was estimated by interpolation from a curve fitted by eye to the plot of the number of fish surviving in various dilutions of the effluent against the concentration. |
| Result | : | Value is provided above. |
| Conclusion | : | Results were duplicated in other laboratories. |
| Reliability | : | (1) reliable without restrictions |
| Reference | : | (Woodiwiss and Fretwell 1974) |
| Type | : | Static |
| Species | : | Heteropneustes fossilis |
| Exposure period | : | 24 – 48 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | no data |
| LC50 | : | 24 hr = 104.13 mg/l 95%CI(100.51-111.61) 48 hr = 86.81 mg/l 95%CI(80.06-94.15) |
| NOEC | : | 48 hr = 15 mg/l |
| Method | : | APHA 1975 Guideline |
| Year | : | 1986 |
| GLP | : | no data |
| Test substance | : | no data |
| Test condition | : | Mortality data were analyzed according to the method of Finney, 1952 for calculating LC50 values and confidence limits. Water pH = 7.7, dissolved oxygen 6.25 mg/l, water hardness – 129 mg/l (CaCO ₂), and temp was 20-25C. |
| Result | : | As listed above. After 10h exp to 24h LC5- the fish showed signs of excitation, irritability, gulping of air at surface and erratic swimming. Prior to death, the tails and fins appeared to be paralyzed. The dorsal fin had collapsed and there appeared some loss of sensitivity to sound and body orientation. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Shanker and Seth 1986) |

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

| | | |
|------------------------------|---|-----------------------------|
| Type | : | Flow through |
| Species | : | Daphnia magna (Water fleas) |
| Exposure period | : | 24-48 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | Yes |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|---|
| EC50 | : | 19 hr = >270mg/l, 95%CI (---) 24 hr = 230mg/l, 95%CI (---) 42 hr = 170mg/l, 95%CI (110-270) 48 hr = 160mg/l, 95%CI (110-270) |
| EC50 | : | 48 hr = 98 mg/l |
| NOEC | : | 48 hr = 60 mg/l |
| Method | : | Test performed according to APHA „Standard Method for Examination of Water and Wastewater“ and „Methods of Acute Toxicity Test with Fish, Macroinvertebrates and Amphibians“ |
| Year | : | 1982 |
| GLP | : | Yes |
| Test substance | : | > 99 % pure |
| Test condition | : | Water quality parameters of temperature, dissolved oxygen, pH and ammonia were measured throughout the test and were within acceptable limits. A flow through proportional diluter system was used to maintain constant test concentrations of acrylamide monomer. Nominal exposure concentrations were 15, 30, 60, 120, and 250 mg/l. The mean measured concentrations were 15, 25, 60, 110 and 270 mg/l. Measured concentrations were used in the LC50 determinations according to Stephan et al. 1978. |
| Result | : | LC50 values presented above |
| Conclusion | : | Based on the behavioral observations conducted twice daily throughout the study, it was noted that mortality was preceded by a migration of daphnids to the bottom of the test chambers, which remained there with little movement until death. Taking all behavior responses into account, a 48hr EC50 was calculated to be 98 mg/l. In addition, the results indicated a 48hr NOEC at 60 mg/l, based on mortality and behavioral effects. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (ABC Labs 1982e) |
| Type | : | |
| Species | : | Mysidopsis bahia (Crustacea) |
| Exposure period | : | 24 – 96 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | Yes |
| LC50 | : | 24-hr LC ₅₀ >161 48-hr LC ₅₀ 109 72-hr LC ₅₀ 94 96-hr LC ₅₀ 78 |
| NOEC | : | 96-hr NOEC 5.2 |
| Method | : | Acute Toxicity Test |
| Year | : | 1983 |
| GLP | : | |
| Test substance | : | |
| Test condition | : | Water pH = 7.7-7.8, dissolved oxygen n/a, water hardness – n/a, and temp was 23-25C. |
| Result | : | Values listed above |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, data obtained from a secondary source |
| Reference | : | (EG&G Bionomics 1983) |
| Type | : | |
| Species | : | Mysidopsis bahia (Crustacea) |
| Exposure period | : | 96 hrs - 28 days |
| Unit | : | mg/l |
| Analytical monitoring | : | Yes |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| NOEC | : | 96-hr NOEC 2.04 mg/l [Mortality in F1 generation] 28-day NOEC 2.04 mg/l [Mortality] 28-day NOEC >4.4 mg/l [Reproduction] |
| Method | : | Prolonged Toxicity Test |
| Year | : | 1985 |
| GLP | : | |
| Test substance | : | |
| Test condition | : | Water pH = 8.0-8.1, dissolved oxygen 5.3-7.2mg/l, salinity 28-31 mg/l, and temp was 23-27C. |
| Result | : | Values listed above |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, data obtained from a secondary source |
| Reference | : | (Springborn Bionomics 1985) |
| Type | : | Flow through |
| Species | : | Paratanytarsus parthenogenetica (Midge Larvae) |
| Exposure period | : | 24 – 48 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | Yes |
| LC50 | : | 17 hr = >910mg/l, 95%CI (---) 24 hr = 570mg/l, 95%CI (360-910) 42 hr = 480mg/l, 95%CI (430-530) 48 hr = 410mg/l, 95%CI (360-470) |
| EC50 | : | 48 hr = 230 mg/l |
| NOEC | : | 48 hr = 60 mg/l |
| Method | : | Test performed according to APHA „Standard Method for Examination of Water and Wastewater“, „Methods of Acute Toxicity Test with Fish, Macroinvertebrates and Amphibians“, „Methods for Conducting Acute Toxicity Tests with the Midge“, and „Chironomidae Toxicity Tests“. |
| Year | : | 1982 |
| GLP | : | Yes |
| Test substance | : | > 99 % pure |
| Test condition | : | Water quality parameters of temperature, dissolved oxygen, pH and ammonia were measured throughout the test and were within acceptable limits. A flow through proportional diluter system was used to maintain constant test concentrations of acrylamide monomer. Nominal exposure concentrations were 60, 120, 250, 500, and 1000 mg/l. The mean measured concentrations were 53, 60, 230, 360 and 910 mg/l. Measured concentrations were used in the LC50 determinations according to Stephan et al. 1978. |
| Result | : | LC50 values presented above |
| Conclusion | : | Based on the behavioral observations conducted twice daily throughout the study, it was noted that mortality was preceded by mobilization to the bottom of the test chambers, with only slight movement when prodded. Additionally, it was noted that midge larvae in exposure levels 3, 4, and 5 (230, 360, and 910 mg/l) did not produce larvae cases during the 48 hour exp. period. Taking all behavior responses into account, a 48hr EC50 was calculated to be 230 mg/l. In addition, the results indicated a 48hr NOEC at 60 mg/l, based on mortality and behavioral effects. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (ABC Labs 1982c) |

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

| | | |
|----------------|---|-----------------------------------|
| Type | : | |
| Species | : | Selenastrum capricornutum (Algae) |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | |
|------------------------------|--|
| Endpoint | : Growth Inhibition |
| Exposure period | : 72 hours |
| Unit | : mg/l |
| Analytical monitoring | : Yes |
| EC50 | : 67.7 mg/l |
| NOEC | : 32 mg/l |
| Method | : OECD Guideline 201 Algae Growth Inhibition Test |
| Year | : 1997 |
| GLP | : Yes |
| Test substance | : No data |
| Test condition | : The test has been performed to OECD 201 Guidelines and EEC Directive 92/69 Method C.3. |
| Result | : Value is stated above. |
| Conclusion | : |
| Remark | : A 72-hour EC ₅₀ of 67.7 mg/l (growth inhibition) and a NOEC of 32 mg/l (growth inhibition) are reported for the fresh water alga <i>Selenastrum capricornutum</i> with a 50% acrylamide solution. As the test was performed on a 50% acrylamide solution the EC ₅₀ and NOEC values should be divided by two to give the toxic effect due to acrylamide. This gives a 72 hour EC ₅₀ of 33.8 mg/l (growth inhibition) and a NOEC of 16 mg/l (growth inhibition). The EC ₅₀ for growth rate was found to be greater than 100 mg/l 50% acrylamide solution (50 mg/l acrylamide). |
| Reliability | : (2) reliable with restriction, data and commentary obtained from secondary source, primary source was not available |
| Reference | : (SEPC, 1997) |
| Type | : Static |
| Species | : <i>Selenastrum capricornutum</i> (Algae) |
| Endpoint | : growth inhibition |
| Exposure period | : No data |
| Unit | : mg/l |
| Analytical monitoring | : no data |
| IC50 | : 72 mg/l |
| Method | : Algae Growth Inhibition Test |
| Year | : 1982 |
| GLP | : No |
| Test substance | : No data |
| Test condition | : No data |
| Result | : Value is stated above. |
| Conclusion | : Authors indicate conclusions are tentative but results appear to be representative. More testing is required to develop statistical data. |
| Reliability | : (2) reliable with restriction, No details were reported as to the experimental conditions or length of exposure. |
| Reference | : (Spraggs et al. 1982) |

4.4 TOXICITY TO MICROORGANISMS E.G. BACTERIA

| | |
|------------------------------|--------------------------------------|
| Type | : Aquatic |
| Species | : <i>Escherichia coli</i> (Bacteria) |
| Exposure period | : 16 hours |
| Unit | : G/l |
| Analytical monitoring | : No data |
| EC100 | : 20 g/l |
| Method | : Cell Division Inhibition Test |
| Year | : 1983 |
| GLP | : no data |
| Test substance | : no data |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | |
|------------------------------|---|
| Test condition | : Cells were incubated in beef extract peptone broth and agar, containing 0.2%-5% acrylamide. The viability of the bacterial population was determined by the microculture technique (Postgate, 1969). Samples of microcultures were prepared as described (Lusta, 1978). To study the effect of osmotic fragility of bacterial cells the cells were incubated for 16h w BPB containing 1.4% acrylamide. Quantity of nucleic acids was determined according to Schmidt-Tannhauser, 1945. |
| Result | : The 16-hour EC ₁₀₀ for <i>Escherichia coli</i> was reported as 20 g/l in a cell division test. The action of acrylamide significantly decreases the viability of <i>E. coli</i> populations. Addition of acrylamide to the growth medium was found to inhibit the division of <i>E.coli</i> cells and cells of some other gram-negative bacterial species and at some concentrations to lead to their elongation. They also found that acrylamide disturbs the synthesis of DNA and to a lesser extent RNA in <i>E. coli</i> cells. The cell wall was found to be the primary target for acrylamide, which disturbs the cell envelope structure and penetrates inside the cell, thus inhibiting the synthesis of nucleic acids and disturbing the cell wall synthesis. |
| Conclusion | : The authors concluded that acrylamide is one of the major toxic factors affecting microbial cells during their immobilization in polyacrylamide. |
| Reliability | : (1) reliable without restriction |
| Reference | : (Starostina et al., 1983) |
| Type | : Static |
| Species | : Photobacterium phosphoreum (Bacteria) |
| Exposure period | : No Data |
| Unit | : Mg/l |
| Analytical monitoring | : no data |
| IC50 | : 13500 |
| Method | : |
| Year | : 1982 |
| GLP | : No |
| Test substance | : no data |
| Test condition | : No data |
| Result | : Value is stated above. |
| Conclusion | : Authors indicate conclusions are tentative but results appear to be representative. More testing is required to develop statistical data. |
| Reliability | : (2) reliable with restriction, No details were reported as to the experimental conditions or length of exposure. |
| Reference | : (Spraggs et al., 1982) |

4.5.1 CHRONIC TOXICITY TO FISH

| | |
|------------------------------|--|
| Type | : Static |
| Species | : Carassius auratus (Goldfish, fresh water) |
| Exposure period | : 7 or 30 days |
| Unit | : Ppm |
| Analytical monitoring | : Yes |
| LC100 | : 100 ppm at 7 days |
| NOEC | : 50 ppm at 30 days |
| Method | : According to Mattocks, 1968 |
| Year | : 1975 |
| GLP | : No data |
| Test substance | : No data |
| Test condition | : No additional information |
| Result | : Continuous exposure of goldfish to 100 ppm killed all seven in 5-7 days. No effects were seen at 50 ppm for 30 days. |
| Conclusion | : A variety of dosing and exposure regimes was used, but no effects could |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| Reliability | : | be seen with sublethal doses. (2) reliable with restriction, very limited information regarding testing conditions, limited information regarding exposure concentrations |
| Reference | : | (Edwards 1975) |
| Type | : | Semistatic |
| Species | : | Poecilia reticulata (Guppy, fresh water) |
| Exposure period | : | 14 day |
| Unit | : | umol/l |
| Analytical monitoring | : | No |
| LC50 | : | 2.69 umol/l (expt); 5.78 umol/l (calc) |
| Method | : | According to Konemann, 1981 |
| Year | : | 1982 |
| GLP | : | no data |
| Test substance | : | no data |
| Test condition | : | LC50's were calculated by logit transformation. All mixtures were prepared in equitoxic concentrations based on experimental LC50 values |
| Result | : | See value above |
| Conclusion | : | |
| Reliability | : | (3) invalid, because no other details on testing conditions provided. LC50 may be calculated based on a mixture of compounds and an index created by the authors, |
| Reference | : | (Hermens and Leeuwangh, 1982) |
| Type | : | Static |
| Species | : | Oncorhynchus mykiss (Rainbow Trout, fresh water) |
| Exposure period | : | 15 day |
| Unit | : | mg/l |
| Analytical monitoring | : | No data |
| EC100 | : | 50 mg/l |
| Method | : | Prolonged Toxicity Test |
| Year | : | 1987 |
| GLP | : | No data |
| Test substance | : | > 98% pure |
| Test condition | : | 10 fish/group were exposed under static conditions for 15 days to 0, 25, or 50 mg/l, followed by a 7 day depuration period. |
| Result | : | Histological lesions were observed in the gill and liver in fish exposed to 25 mg/l for 15 days. Fish exposed to 50 mg/l developed lesions in the cephalic lateral line and peripheral lateral line in addition to the gill and liver. After the depuration period additional lesions were observed in the sagittal and proximal nerve plexus (25 mg/l and 50 mg/l exposure) and in the optic nerve (50 mg/l exposure only). Swimming behavior of the fish was unaffected at exposure concentrations below 25 mg/l. At 50 mg/l fish had difficulty in orientating themselves when swimming, and based upon this effect an EC ₁₀₀ of 50 mg/l was quoted. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Petersen et al. 1987)(Petersen and Lech 1987) |

4.5.2 CHRONIC TOXICITY TO AQUATIC INVERTEBRATES

| | | |
|------------------------------|---|------------------------------|
| Type | : | |
| Species | : | Mysidopsis bahia (Crustacea) |
| Exposure period | : | 96 hrs - 28 days |
| Unit | : | mg/l |
| Analytical monitoring | : | Yes |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

NOEC : 96-hr NOEC 2.04 mg/l [Mortality in F1 generation]
28-day NOEC 2.04 mg/l [Mortality]
28-day NOEC >4.4 mg/l [Reproduction]

Method : Prolonged Toxicity Test

Year : 1985

GLP :

Test substance :

Test condition : Water pH = 8.0-8.1, dissolved oxygen 5.3-7.2mg/l, salinity 28-31 mg/l, and temp was 23-27C.

Result : Values listed above

Conclusion :

Reliability : (2) reliable with restriction, data obtained from a secondary source

Reference : (Springborn Bionomics 1985)

4.6.1 TOXICITY TO SOIL DWELLING ORGANISMS

4.6.2 TOXICITY TO TERRESTRIAL PLANTS

Species : *Impatiens sultanii*

Endpoint : Emergence

Exposure period :

Unit : Ppm

NOEC : <= 2000 ppm

Method : Germination Test

Year : 1981

GLP : No

Test substance : No data

Test condition : The effect of acrylamide on pollen germination and tube growth was studied using the pollen of *Impatiens sultanii*. Pollen was transferred from the anthers of the plants to a basal medium on a depression, dispersed in the liquid and covered with a glass cover. During the experiment the slide was kept in a closed plastic box at 25C. The pollen was incubated for 15 minutes and photomicrographs taken at random periods after the incubation period. The total number of grains, the number of germinated grains and grains producing tubes longer than 40 mm were counted. To investigate the growth of pollen tubes on a solid medium 1% agar was added to the basal medium and autoclaved.

Result : When acrylamide was added to the basal medium at concentrations ranging from 10 to 2000 ppm, there was no significant effect upon germination, tube formation, or tube growth.

Conclusion :

Reliability : (1) reliable without restriction

Reference : (Bilderback 1981)

Species : *Brassica rapa* (Dicotyledon) Turnip root

Endpoint : Growth

Exposure period : 2 day

Unit : mg/l

EC50 : 220

EC39 : 100

Method : Root Elongation Test

Year : 1984

GLP : No data

Test substance : No data

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------|---|---|
| Test condition | : | The toxicity of acrylamide to higher plants was studied. About 50 seeds of turnip (<i>Brassica rapa</i> L. cv. Chuusei-kanamachi), rape (<i>Brassica rapa</i> L. cv. Tokiwa-jibai), chinese cabbage (<i>Brassica pekinensis</i>), sesame (<i>Sesamum indicum</i>) and cucumber (<i>Cucumis sativus</i>) were incubated in distilled water for 1 day at 30°C in the dark. Seeds of upland rice (<i>Oryza sativa</i>) and wheat (<i>Triticum aestivum</i>) were cultured for 2 days. 10 seedlings of similar growth rate were then transferred to a flask containing 20 ml of polymer flocculant or monomer and cultured for 2 more days. The EC ₅₀ was calculated as the concentration of flocculant or monomer where root elongation rate is equal to 50% of the control. The effect of polymer flocculants on germination was determined by dipping the seeds directly into the test solutions and shaking for 1 day. |
| Result | : | At 100 mg/l acrylamide was found to retard root elongation by 61% compared to the control and the EC ₅₀ was calculated as 220 mg/l. No significant effect on seed germination was observed. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Kuboi and Fuji 1984) |
| Species | : | Lactuca sativa (Dicotyledon) |
| Endpoint | : | Germination and growth |
| Exposure period | : | 18-23 days |
| Unit | : | |
| Method | : | |
| Year | : | 1987 |
| GLP | : | |
| Test substance | : | ¹⁴ C-Labeled acrylamide monomer |
| Test condition | : | ¹⁴ C labelled acrylamide monomer was added to 100 ml of nutrient solution and mixed with 4000 g of air dried soil to obtain a uniform concentration of 5.0 ppm. The plants were analysed for ¹⁴ C after 18 days. The roots, soil, leachate and shoots were analysed separately. |
| Result | : | They found that in those soils treated with acrylamide germination and growth were slower and the plants showed signs of necrosis. ¹⁴ C was detected in the shoots and roots of treated plants and was also present in the soil and leachate. The ¹⁴ C in the leachate and plant tissue did not appear to be acrylamide. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Hazleton Labs, 1987) |

4.6.3 TOXICITY TO OTHER NON-MAMM. TERRESTRIAL SPECIES

| | | |
|--------------------------------|---|--|
| Test substance | : | AMD, synthesized by ICN Pharmaceuticals, Plainview, NY |
| Method | : | Litchfield and Wilcoxon (1949) |
| Type | : | LD50 |
| GLP | : | Pre-GLP |
| Year | : | 1981 |
| Species and Strain | : | Japanese quail (<i>Coturnix coturnix japonica</i>) |
| Sex | : | Male |
| No. Animals/Sex/Dose | : | 8/group |
| Doses | : | 0, 150, 190, 240, 300 mg/kg |
| Vehicle | : | Distilled water |
| Route of administration | : | Injection into the crop |
| Test condition | : | Adult quail, examined pre-dosing and on day 1, 4, and 7 postdosing for lethality, body weights and neurological signs. |

4. Ecotoxicity

Id 79-06-1

Date 7 September 2001

| | | |
|------------------------------|---|--|
| Value | : | 214 mg/kg bw |
| No. Deaths/Dose Level | : | 0/0 0/150 5/190 8/240 8/300 |
| Result | : | LD50 ranged from 194-236 mg/kg. There was no affect on body weight, but right and gait were affected at all doses. |
| Conclusion | : | Neurological effects may be observable at lower doses than those employed |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Cabe and Colwell, 1981) |

4.7 BIOLOGICAL EFFECTS MONITORING

4.8 BIOTRANSFORMATION AND KINETICS

| | | |
|------------------------------|---|--|
| Type | : | Static |
| Species | : | Oncorhynchus mykiss (Rainbow trout, fresh water) |
| Exposure period | : | 24 – 96 hours |
| Unit | : | mg/l |
| Analytical monitoring | : | No data |
| LC50 | : | 24-hr LC ₅₀ >300 mg/l --- 48-hr LC ₅₀ 210 mg/l Not obtainable 72-hr LC ₅₀ 170 mg/l 95%CI(137-191) 96-hr LC ₅₀ 162 mg/l 95%CI(137-191) |
| Method | : | Uptake, deposition, elimination and Acute Toxicity test |
| Year | : | 1985 |
| GLP | : | No |
| Test substance | : | > 98 % pure |
| Test condition | : | 10 fish/group. LC50's were calculated by Litchfield and Wilcoxon method, 1948. |

| | | |
|-------------------|---|--|
| Result | : | <p>Fish were exposed to a steady state concentration of acrylamide and 2,3-¹⁴C acrylamide under static conditions for 72 hours. This was followed by a depuration period in fresh water for 96 hours. Samples of fish for analysis were taken at regular intervals. In fish exposed to 0.710 mg/l acrylamide, uptake was rapid in the carcass and viscera in the first 24 hours. Uptake was then slower until a steady concentration was reached at 72 hours.</p> <p>The BCFs calculated from these exposures were 1.44 for the carcass and 1.65 for the viscera. The elimination of acrylamide from the carcass and viscera was found to be biphasic with an initial phase being rapidly followed by a slower second phase. The t_{1/2} values for elimination of acrylamide from the viscera were 16 hours for the initial elimination and 5.7 days for the second elimination, and for the carcass, the values were 10 hours for the first elimination and 7.7 days for the second elimination. After a 96 hour depuration period acrylamide concentrations in both the carcass and viscera had declined to approximately 25% of the steady state values. Further studies indicated that acrylamide uptake was greatest in the kidneys with elimination greatest from the blood and gills and slower from the muscles and intestines. Acrylamide was found to be excreted via the gills, urine and bile, 90% of it in an unchanged form.</p> |
| Conclusion | : | |

Reliability : (1) reliable without restriction for kinetics
Reference : (Petersen et al., 1985)

4.9 ADDITIONAL REMARKS

Memo Remark Field Study
Acrylamide has been found to be effective in repelling marine bacteria and inhibiting marine organism growth. In fish exposed to acrylamide and NMA from grout application, histological changes have been observed. The *in situ* adsorption, degradation and toxicity of acrylamide in a river was studied. The effect of acrylamide in stream water on the insect fauna living on stones covered in moss was investigated as part of the study. A concentration of 50 ug/l acrylamide was maintained in a stream for 6 hours. At the end of the 6 hour period the density of insect fauna was reduced.

Reliability Reference (1) reliable without restriction
(Brown et al. 1982)(Chet and Mitchell 1976)

Memo Remark Toxicity to Amphibians
Frogs (*Rana temporaria*) were given acrylamide either by injection in saline into the dorsal sac or by exposing them to a solution containing acrylamide. A dose of 50 ug/g in 7 days killed three out of five frogs and a 2 hour exposure to a 2% (w/v) solution of acrylamide killed two out of three frogs. No ill effects were observed in the surviving frogs.

Reliability Reference (4) not assignable
(Edwards 1975)

5.1.1 ACUTE ORAL TOXICITY

Test substance : No data
Method : Weil, 1952
Type : LD50
GLP : Pre-GLP
Year : 1966
Species and Strain : Rat/ Fischer 344
Sex : Female
No. Animals/Sex/Dose : 10/group
Vehicle : Distilled water
Route of administration :
Test condition : Standard procedures, 8 week old rats, no other details provided.
Value : 203 mg/kg bw
No. Deaths/Dose Level :
Result : LD50 ranged from 166-249 mg/kg. After a single dose around the LD50, the rats developed a fine tremor lasting about 48 hours and either recovered completely or died within two to three days showing generalised weakness.
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Fullerton et al. 1966)

Test substance : No data
Method : Acute Oral Toxicity
Type : LD50
GLP : Pre-GLP
Year : 1975
Species and Strain : Rat
Sex :
No. Animals/Sex/Dose :
Vehicle :
Route of administration :
Test condition :
Value : 124 mg/kg
No. Deaths/Dose Level :
Result :
Conclusion :
Reliability : 2) reliable with restriction, conclusion obtained from secondary source, primary source not in English.
Reference : (Paulet & Vidal, 1974)

Test substance : No data
Method : Acute Oral Toxicity
Type : LD50
GLP : Pre-GLP
Year : 1979
Species and Strain : Rat/ Fischer 344
Sex : Male
No. Animals/Sex/Dose : 10/group
Vehicle : Distilled water
Route of administration :
Test condition : 10 rats/group were given 50, 100, 125,200, or 250 mg/kg dissolved in

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|-------------------------------------|---|--|
| Value | : | 175 mg/kg bw |
| No. Deaths/Dose Level Result | : | There were no mortalities at exposures up to and including 125 mg/kg. At 24 hours post-dose there were 3/10 and 7/10 mortalities at 200 and 250 mg/kg respectively. On completion of 7 days there were 8/10 mortalities and 10/10 at these 2 exposure levels. The median lethal dose (LD50) was calculated to be 175 mg/kg at 7 days. |
| Conclusion | : | Clinical signs of toxicity at 12 hours included postural and motor incoordination, hindlimb muscular dysfunction, hyperreflexia, recurrent episodes of tonic-clonic convulsions, and tremor particularly at 250 mg/kg. Details were not provided of the incidence and severity of these findings at any other exposure level. In addition, amongst high dose animals only, diarrhoea and increased urination were reported. At 12 hours post-dose the inclined screen test demonstrated increased motor dysfunction at 100 and 200 mg/kg. Decreased forelimb and hindlimb strength was noted at 200 mg/kg 12 hours after dosing, although only the hindlimb effects attained statistical significance. At 7 days, there was no significant difference in the performance of animals at 0, 50, and 100 mg/kg. The consequences of the 200 mg/kg dose could not be interpreted as there were insufficient survivors. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Tilson and Cabe, 1979b) |
| Test substance | : | 99 % pure |
| Method | : | Acute Oral Toxicity |
| Type | : | LD50 |
| GLP | : | Pre-GLP |
| Year | : | 1964 |
| Species and Strain | : | Rat |
| Sex | : | Female |
| No. Animals/Sex/Dose | : | 5/dose group |
| Vehicle | : | Water |
| Route of administration | : | |
| Test condition | : | 5 rats/group were given 126 or 252 mg/kg by gastric intubation at 2.5% acrylamide in water. |
| Value | : | 126 mg/kg/d < LD50 < 252 mg/kg/d |
| No. Deaths/Dose Level Result | : | At 126 mg/kg – 0/5 rats died and at 252 mg/kg 5/5 deaths occurred in one day in all rats. |
| Conclusion | : | The LD50 for all species appears to be in the range of 150-180 mg/kg bw. |
| Reliability | : | (2) Reliable with restriction, strain of rat not provided, LD50 not established |
| Reference | : | (McCollister et al. 1964) |
| Test substance | : | 95 % pure |
| Method | : | Weil, 1952 |
| Type | : | LD50 |
| GLP | : | Pre-GLP |
| Year | : | 1981 |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Species and Strain : Mouse/ ddY
Sex : Male
No. Animals/Sex/Dose : 4
Vehicle : Saline
Route of administration : Oral
Test condition : 4 mice/dose group, 4 unspecified dosage groups, single dose
Value : 107 mg/kg bw
No. Deaths/Dose Level :
Result :
Conclusion :
Reliability : (2) Reliable with restriction, doses not indicated, clinical signs not provided
Reference : (Hashimoto et al. 1981)

Test substance : 99% pure
Method : Acute Oral Toxicity
Type : LD50
GLP : Pre-GLP
Year : 1964
Species and Strain : Rabbit
Sex : Male/female
No. Animals/Sex/Dose : 4/dose group
Vehicle : Water
Route of administration :
Test condition : 4 rabbits/group were given 63, 126 or 252 mg/kg by gastric intubation at 2.5% acrylamide in water.
Value : 126 mg/kg/d < LD50 < 252 mg/kg/d
No. Deaths/Dose Level :
Result : At 63 mg/kg – 0/4 rabbits died, slight weight loses, at 126 mg/kg – 1/4 rabbits died, tremors and pupil dilation, and at 252 mg/kg 4/4 deaths occurred overnight
Conclusion : The LD50 for all species appears to be in the range of 150-180 mg/kg bw.
Reliability : (2) Reliable with restriction, strain of rabbit not provided, LD50 not established
Reference : (McCollister et al. 1964)

Test substance : 99% pure
Method : Acute Oral Toxicity
Type : LD50
GLP : Pre-GLP
Year : 1964
Species and Strain : Guinea pig
Sex : Male
No. Animals/Sex/Dose : 4/dose group
Vehicle : Water
Route of administration :
Test condition : 4 guinea pigs/group were given 126 or 252 mg/kg by gastric intubation at 2.5% acrylamide in water.
Value : 252 mg/kg/d > LD50 > 126 mg/kg/d
No. Deaths/Dose Level :
Result : At 126 mg/kg – 0 guinea pigs died, very slight weight loses, at 252 mg/kg – 4 guinea pigs died, death occurred overnight
Conclusion : The LD50 for all species appears to be in the range of 150-180 mg/kg bw.
Reliability : (2) Reliable with restriction, strain not provided, LD50 not established
Reference : (McCollister et al. 1964)

5.1.2 ACUTE INHALATION TOXICITY

Test substance : No data
Method : Acute Inhalation Toxicity
Type : Mortality
GLP : Unknown
Year : 2001
Species and Strain : Rat F344
 : Mouse B6C3F1
Sex : Male
No.
Animals/Sex/Dose :
Vehicle :
Route of administration : Inhalation, nose only
Test condition :
Value : 5.7 ppm for 6 hr.
No. Deaths/Dose Level :
Result :
Conclusion : No deaths following exposure to 5.7 ppm for 6 hr.
Reliability : (2) reliable with restriction, data obtained from abstract only
Reference : (Friedman, et al. 2001)

5.1.3 ACUTE DERMAL TOXICITY

Test substance : No data
Method : Acute Dermal Toxicity
Type : Acute Dermal Toxicity
GLP : Pre-GLP
Year : 1977
Species and Strain : Rabbit/ New Zealand
Sex : Male
No. Animals/Sex/Dose : 5/group
Vehicle :
Route of administration :
Test condition : 1.4, 1.7, 2.2, or 2.8 ml/kg was applied to the skin of 5 rabbits/group for an exposure period for 24 hours. The animals were observed for mortality and clinical signs for 14 days.
Value : 1.68 ml/kg bw (~1.88 mg/kg)
No. Deaths/Dose Level :
Result : Mortality was 50, 50, 100, and 100% respectively. Animals started to shake and had loss of coordination when handled. No gross findings were noted in any of the animals at necropsy. The acute dermal LD50 is 1.68 ml/kg (1.34-2.10 ml.kg, 95% CI).
Conclusion :
Reliability : (1) reliable without restriction
Reference : (Vernon et al. 1990)

Test substance : No data
Method : Acute Dermal Toxicity

5. Toxicity

Id 79-06-1

Date 7 September 2001

Type : LD50
GLP : Pre-GLP
Year : 1979
Species and Strain : Rat
Sex :
No. Animals/Sex/Dose :
Vehicle :
Route of administration :
Test condition :
Value : 400 mg/kg bw
No. Deaths/Dose Level :
Result :
Conclusion :
Reliability : (2) reliable with restriction, data obtained from secondary source, primary source was not available
Reference : (Novikova, 1979)

5.1.4 ACUTE TOXICITY, OTHER ROUTES

Test substance : No data
Method : Acute Intraperitoneal Toxicity
Type : LD50
GLP : Pre-GLP
Year : 1953
Species and Strain : Rat
Sex :
No. Animals/Sex/Dose :
Vehicle :
Route of administration : i.p.
Test condition :
Value : 120 mg/kg bw
No. Deaths/Dose Level :
Result :
Conclusion :
Reliability : (3) not reliable, value obtained from secondary source, primary source not in English. Cannot verify any information presented in this summary.
Reference : (Druckrey et al. 1953)

Test substance : No data
Method :
Type : LD50
GLP : Pre-GLP
Year : 1956
Species and Strain : guinea pig
Sex :
No. Animals/Sex/Dose : 28
Vehicle :
Route of administration : i.p. or s.c
Test condition :
Value : 170 mg/kg
No. Deaths/Dose Level :
Result :
Conclusion :

5. Toxicity

Id 79-06-1

Date 7 September 2001

Reliability : (2) reliable with restriction, data from secondary source, primary source not in English, route cannot be confirmed
Reference : (Ghiringhelli, 1956)

Test substance : No data
Method :
Type : LD50
GLP : Pre-GLP
Year : 1974
Species and Strain : Rat
Sex :
No. Animals/Sex/Dose :
Vehicle :
Route of administration : i.p.
Test condition :
Value : 90 mg/kg
No. Deaths/Dose Level :
Result :
Conclusion :
Reliability : (2) reliable with restriction, data from secondary source, primary source not in English
Reference : (Paulet & Vidal, 1974)

5.2.1 SKIN IRRITATION

Species : Rabbit
Concentration : 10%
Exposure : 10X
Exposure time : 2 weeks
Number of animals :
PDII :
Result : not irritating
EC classification : not irritating
Method : Acute Dermal Irritation
Year : 1964
GLP : Pre-GLP
Test substance : 99% pure
Test condition : 10% aqueous solution was applied to ear and abdomen of rabbit ten times over 2 weeks. An abraded area of the belly was treated for three consecutive days.

Skin absorption was determined by applying measured amounts of the acrylamide as a 12.5% solution to the skin of rabbits using the „sleeve“ technique for a 24 hour period.

Result : 10% aqueous solution applied repeatedly elicited no significant response. The abraded area showed very slight reddening and slight edema which healed satisfactorily and well upon cessation of the treatment.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (McCollister et al. 1964)

Species : Rabbit
Concentration : 12.5%
Exposure : 63, 126, 252, 500, 1000 mg/kg

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--------------------------|---|--|
| Exposure time | : | 24 hrs |
| Number of animals | : | 2-4/group |
| PDII | : | |
| Result | : | not irritating |
| EC classification | : | not irritating |
| Method | : | Acute Dermal Absorption |
| Year | : | 1964 |
| GLP | : | Pre-GLP |
| Test substance | : | 99% pure |
| Test condition | : | Skin absorption was determined by applying measured amounts of the acrylamide as a 12.5% solution to the skin of rabbits using the „sleeve“ technique for a 24 hour period. 2 rabbits rec'd 63, 126,500 and 1000 mg/kg and 4 animals rec'd 252 mg/kg |
| Result | : | One rabbit that rec'd 1000 mg/kg died within 2 days. There were no other deaths, however slight weight loss and reddening of the skin were noted in both rabbits that rec'd the application of 500 mg/kg. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (McCollister et al. 1964) |
| Species | : | Rabbit |
| Sex | : | Male |
| Strain | : | New Zealand |
| Concentration | : | 100% |
| Exposure duration | : | 4 hours |
| Number of animals | : | 3 |
| Method | : | OECD #6404 |
| Year | : | 1997 |
| GLP | : | No data |
| Test substance | : | 99% pure |
| Test condition | : | In an unpublished study conducted according to modern protocol standards 0.5g acrylamide moistened with water was applied to the shaved, intact skin of a group of 3 New Zealand White rabbits (2.47 – 2.55 kg) under a semi-occlusive dressing for 4 hours. Skin reactions were scored at 24, 48, and 72 hours post-application and graded according to the EU scheme. Examinations for moribundity/mortality were performed twice daily. |
| Result | : | There were no mortalities, and there was no indication of whether or not there were any clinical signs of toxicity. There was no erythema or edema at any of the time-points when reactions were scored. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Mercier 1997a)(Mercier 1997b) |

5.2.2 EYE IRRITATION

| | | |
|--------------------------|---|--|
| Species | : | Rabbit |
| Concentration | : | 10%, 40% |
| Dose | : | |
| Exposure Time | : | |
| Number of animals | : | 1 |
| Method | : | Eye Irritation |
| Year | : | 1964 |
| GLP | : | Pre-GLP |
| Test substance | : | 99% pure |
| Test condition | : | A small amount of the solution to be tested was dropped onto the right |

| | |
|--------------------------|--|
| | eyeball of a rabbit. Within 30 seconds, washing was performed for 2 minutes. The left eye was treated with a like amount of acrylamide solution, but left unwashed. The animal was observed for evidence of pain, and the eyes were examined within 2 or 3 minutes for conjunctival and corneal response. Similar examination was made after 1 hour and 24 hours, with and without fluorescein staining |
| Result | : The 10% aqueous solution caused a trace of pain and slight conjunctival irritation immediately following contact. There was no corneal injury at any time. The conjunctiva was completely normal within 24 hours. The 40% aqueous solution gave the following response when placed in the rabbit eye: unwashed- moderate pain, slight conjunctival irritation which was slow in healing and significant corneal injury which, however, was completely healed within 24 hours. Washed: moderate amount of pain and slight conjunctival irritation which was almost completely healed in 24 hours. There was no corneal injury. |
| Conclusion | : |
| Reliability | : (1) Reliable without restriction |
| Reference | : (McCollister et al., 1964) |
| Species | : Rabbit |
| Sex | : Male |
| Strain | : New Zealand White |
| Concentration | : 100% |
| Dose | : 82 mg |
| Exposure Time | : One application |
| Number of animals | : 3 |
| Method | : OECD #405 |
| Year | : 1997 |
| GLP | : No data |
| Test substance | : 99% pure |
| Test condition | : In an unpublished study conducted according to modern protocol standards 82mg powdered acrylamide (equivalent to a volume of about 0.1ml) was applied to one eye of each of 3 New Zealand White rabbits (2.36-2.75kg). Examination for morbidity/mortality rate were performed twice daily Reactions were scored at 24, 48, and 72 hours and at 7, 14, and 21 days post-application with grading according to the EU scheme. |
| Result | : There were no mortalities, and there was no indication of whether or not there were any clinical signs of toxicity. For iridial reactions the mean score for each animal over 24, 48 and 72 hours was 1.0; corneal opacity means ranged from 2.0-2.3; conjunctival redness 2.0 and chemosis 1.3-2.0. Iridial reactions were still apparent at day 14 post-application although there were no abnormalities noted at day 21. Overall, these results indicate that acrylamide is an eye irritant. |
| Conclusion | : |
| Reliability | : (1) Reliable without restriction |
| Reference | : (Mercier 1997a)(Mercier 1997b) |

5.3 SENSITIZATION

| | |
|--------------------------|-------------------------|
| Species | : Guinea pig |
| Sex | : Male/Female |
| Strain | : Pirbright White |
| Concentration | : 3.5% and 50% solution |
| Exposure Time | : 25 days |
| Number of animals | : 30 |
| Method | : OECD Guideline #406 |
| Year | : 1995 |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--------------------------|---|--|
| GLP | : | Yes |
| Test substance | : | No data |
| Test condition | : | Groups of 20 test and 10 control animals received dermal challenge concentrations of up to 25% aqueous acrylamide following induction of the test animals with up to 50% topically and up to 3.5% intradermally. |
| Result | : | Systemic toxic symptoms after application were not observed at any time during the study. Body weight development was positive and within normal ranges. No erythema and edema were observed at any time point after the challenge application in the control group. There were apparently no skin reactions in control animals but 85% of test animals gave a positive response. On the basis of these results, acrylamide should be considered as a skin sensitizer in animals. |
| Conclusion | : | |
| Reliability | : | (1) Reliable without restriction |
| Reference | : | (Stockhausen, 1995) |
| Species | : | Guinea pig |
| Sex | : | Female |
| Strain | : | Dunkin/Hartley - Albino |
| Concentration | : | See test conditions |
| Dose | : | See test conditions |
| Exposure Time | : | See test conditions |
| Number of animals | : | 30 |
| Method | : | OECD Guideline #406 |
| Year | : | 1995 |
| GLP | : | Yes |
| Test substance | : | CT-566-94 Acrylamide (>98% purity) |
| Test condition | : | Induction intradermal injections: 3 pairs of intradermal injections 0.5% w/v in water Topical Induction: One week following injections, a patch saturated with 25% w/v in distilled water was placed on same test area for 48 hours Challenge: Two weeks following topical induction, a patch saturated with 20% w/v in distilled water was placed on an anterior site on the flank and 10% w/v in distilled water was applied to a posterior site. Both patches were left on the test site for 24 hours. |
| Result | : | Skin reactions were observed in most control animals, but were less severe than reactions in test animals. Taking the control reactions into consideration, a positive skin response, exceeding the degree of reaction seen amongst the controls, was recorded in 40% of the test animals. A positive result was reported in the guinea pig maximization test, conducted according to modern protocol standards. On the basis of these results, acrylamide should be considered as a skin sensitizer in animals. |
| Conclusion | : | |
| Reliability | : | (1) Reliable without restriction |
| Reference | : | (Allan, 1995) |

5.4 REPEATED DOSE TOXICITY

| | | |
|-----------------------|---|------------------------|
| Test substance | : | 99 % pure |
| Method | : | Repeated Dose Toxicity |
| Type | : | |
| GLP | : | Pre-GLP |
| Year | : | 1964 |
| Species | : | Monkey |
| Strain | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Route of administration : i.p.
Duration of test : 3 days
Doses/concentration levels : 100 mg/kg
Sex : Female
Exposure period : 2 days
Frequency of treatment : Daily
Control group and treatment : No control group
Post exposure observation period : 1 day
Statistical methods :
Test condition : One monkey was given i.p. dose of acrylamide amounting to 100 mg/kg. This was repeated on the next day for a total of 200 mg/kg bw.
NOAEL (NOEL) :
LOAEL (LOEL) :
Actual dose/dose level/sex :
Toxic response/effects by dose level :
Result : On the third day, the monkey was found lying down in the cage unable to stand. The monkey had lost its sense of balance, but not its ability to use its muscles since it could still crawl around the cage. The animal died on the third day. Gross pathological examination revealed congested lungs, some congestion in the kidneys, and areas of necrosis in the liver. Microscopic examination of the liver revealed congestion of the sinusoids with fatty degeneration and necrosis. The kidneys showed degeneration of the convoluted tubular epithelium and glomerular degeneration with albuminous material in the capsular space. Considered highly toxic via i.p admin.
Conclusion :
Reliability : (2) reliable with restriction, limited reporting and the use of only one animal per dose level.
Reference : (McCollister et al., 1964)
Test substance : No data
Method :
Type : Acute Neurotoxicity
GLP : No data
Year : 1986
Species : Rat
Strain : Wistar
Route of administration : saline via i.p.
Duration of test :
Doses/concentration levels : 30 or 50 mg/kg
Sex : Female
Exposure period : 3 weeks
Frequency of treatment : 5 days/week
Control group and treatment : Control group: Yes
Post exposure observation period : :

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Statistical methods | : | |
| Test condition | : | Rats (180-220g) were given doses 5 days/week for 3 weeks. Control animals were not injected. Body weights and condition of animals were recorded daily. Animals were killed at weekly intervals. The effects on neuronal perikarya of axotomy alone, of acrylamide alone, and combined were studied by light and electron microscopy. Left and right spinal root ganglia and the lumbar spinal cord were dissected. Ganglion were examined microscopically for eccentric nucleus, crenated nuclear membrane, peripherally placed Nissl substance, mitochondria, and lysosomes. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | The responses to axotomy and to acrylamide intoxication shared certain features, namely peripheral Nissl substance and to a lesser degree nuclear eccentricity, nucleolemmal crenation and mitochondrial enlargement. Neurofilament loss was present only acrylamide. In combined axotomy and acrylamide all five features were prominent. Acrylamide appears to impede the vital neuronal responses directed towards repair of the axon. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Jones & Cavanagh 1986) |
| Test substance | : | 99 % pure |
| Method | : | Unspecified EPA Guideline |
| Type | : | |
| GLP | : | Yes |
| Year | : | 1991 |
| Species | : | Rat |
| Strain | : | Sprague-Dawley |
| Route of administration | : | Oral gavage |
| Duration of test | : | 4 weeks (3 weeks dosing + 1 week recovery + 1 week dosing) |
| Doses/concentration levels | : | 10 or 30 mg/kg/d for 3 weeks with 10 days recovery Readministration at 10 or 20 mg/kg/d for 1 week |
| Sex | : | Male/female |
| Exposure period | : | |
| Frequency of treatment | : | Daily |
| Control group and treatment | : | |
| Post exposure observation period | : | |
| Statistical methods | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Test conditions : In a study to validate a functional observational battery (FOB) conducted according to US EPA guidelines (1985) and to assess motor activity, groups of 10 male and 10 female Sprague-Dawley rats received 0, 10, or 30 mg/kg/day aqueous acrylamide (99% pure) by oral gavage 7 days/week for 3 weeks. The 3-week exposure period was followed by 10 days recovery before readministration of 0, 10, or 20 mg/kg/day acrylamide for one week (the high dose level was reduced due to 4 male and 2 female mortalities). The FOB was conducted pre-exposure, 1, 6, and 24 hours after the first administration and once per week thereafter. Parameters recorded in the FOB included physical appearance, assessment of movement, response to stimuli such as sound or tactile response, and grip strength. Other observations included measurement of food consumption and bodyweight gain, and terminal histopathology on all major organs including eyes (with optic nerve), sciatic, tibial, and sural nerves, lumbar and cervical dorsal and ventral roots, dorsal root ganglion, trigeminal ganglion, and sections from different regions of the brain and spinal cord.

NOAEL (NOEL) :

LOAEL (LOEL) :

Actual dose/dose level/sex :

Toxic response/effects by dose level :

Result :

Bodyweight gain and food consumption was statistically significantly reduced amongst animals at 30/20 mg/kg/day (22% reduction in female bodyweight and 26% in males). The onset of alterations in FOB parameters was about 2 weeks after commencement of acrylamide exposure. At 30/20 mg/kg/day the following changes were noted in the FOB: an increased incidence of rigid/difficult handling, slight ptosis, slight to moderately impaired respiration, soiled fur, increased incidence of vocalisation, increased urination, hunched posture/prostration, slight to severely impaired gait, abnormal behaviour, reduced tactile response, impaired righting reflex (also seen at 10 mg/kg/day), decreased rearing counts (also seen at 10 mg/kg/day), reduced forelimb and hind limb grip strength, reduced response to bright light, and reduced activity.

Histopathologically, examination of white matter from cervical and lumbar spinal cord sections, trigeminal and dorsal root ganglia, sciatic, tibial, and sural nerves showed altered diameter of axons (increased or decreased diameter), disruption, fragmentation and distortion of axons, and/or dilation and fragmentation of myelin sheaths, and occasionally an increased number of macrophages. The findings were more prevalent and more severe in animals at 30/20 mg/kg/day than at 10 mg/kg/day. Brain regions were not significantly affected. There was also an increased incidence of splenic pigment was observed in males and females at 30/20 mg/kg/day and also in females at 10 mg/kg/day, increased incidence of granulomatous inflammation in the lungs of animals at 30/20 mg/kg/day, and hemorrhage of the urinary bladder in several males at 30/20 mg/kg/day.

Conclusion :

Reliability : (1) reliable without restriction

Reference : (Schulze & Boysen 1991)

Test substance : 99.9 % pure

Method :

Type :

GLP : Yes

Year : 1991

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|---|
| Species | : | Rat |
| Strain | : | Fischer 344 |
| Route of administration | : | Drinking water |
| Duration of test | : | |
| Doses/concentration levels | : | males: 0, 1.4, 4.1, 12, 19, and 25 mg/kg females: 0, 1.3, 4.3, 9.0, 19, and 24 mg/kg |
| Sex | : | Male/female |
| Exposure period | : | 28 days |
| Frequency of treatment | : | Daily |
| Control group and treatment | : | Control group: Yes |
| Post exposure observation period | : | : |
| Statistical methods | : | |
| Test condition | : | 6 groups of 10 animals/sex/group (8 weeks old: males: 168.9-190.2g; females: 119.8-141.0g) were provided daily doses. Animals were checked twice daily for viability, daily clinical observations, weekly body weight msmts, weekly food and water consumption, blood was drawn for T3, T4, TSH, prolactin, and testosterone after 2 weeks of treatment and at study termination. Necropsy was performed and the following organs were preserved: adrenals, brain, mammary gland, ovaries and oviducts, sciatic nerve, testes, thyroids/parathyroids and all gross lesions. |
| | | <u>Statistical Analysis:</u> Parametric procedures were analyzed using ANOVA with Dunnett's test for significant differences. For non-parametric procedures the Kruskal Wallis test was used and significant differences were analyzed with Dunn's Summed Rank Test. |
| NOAEL (NOEL) | : | 4.1 - 4.3 mg/kg |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | Neurotoxicity (both sexes), seminal vesicle and or testes atrophy, and decreased prolactin levels observed greater or equal to 19 mg/kg/day. Little evidence of toxicity observed at 12 and 9 mg/kg/day. A nonstatistically significant decrease in male serum prolactin observed at less than or equal to 12 mg/kg/day. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Keefe, R. 1991) |
| Test substance | : | No data |
| Method | : | Repeated Dose Toxicity |
| Type | : | |
| GLP | : | No data |
| Year | : | 1985 |
| Species | : | Rabbit |
| Strain | : | Mixed |
| Route of administration | : | s.c. |
| Duration of test | : | 4 months |
| Doses/concentration levels | : | 400 mg/kg (total dose) |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Sex :
Exposure period : 4 weeks
Frequency of treatment : 2/week
Control group and treatment : Control group with concurrent vehicle
Post exposure observation period : 3 months
Statistical methods :
Test conditions : 5 rabbits weighing 2.8-4.4 kg were given acrylamide 2x/wk s.c. Total dose of 400 mg/kg was achieved over 4 weeks. 4 control animals were given saline alone. The sensitivity of the Hering Breuer reflex was compared before, during, and after the induction of acrylamide neuropathy and was measured as the tracheal pressure which produced 30 seconds of apnea.

NOAEL (NOEL) :
LOAEL (LOEL) :
Actual dose/dose level/sex :
Toxic response/effects by dose level :
Result : After 4 weeks of exposure, there was ataxia and the conduction velocity of hindlimb motor nerves was significantly reduced. There was a marked and reproducible reduction in the sensitivity of the H-B reflex. The ataxia resolved within a month of stopping acrylamide admin. Three months after the cessation of acrylamide the sensitivity of the H-B reflex had increased significantly but had not returned to normal.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Satchell, 1985)

Test substance : No data
Method : Repeated Dose Toxicity
Type :
GLP : Pre-GLP
Year : 1974
Species : Dog
Strain : Beagle
Route of administration : Oral, gelatin capsule
Duration of test :
Doses/concentration levels : 0, 5 or 15 mg/kg/day
Sex : Male/female
Exposure period : 22-60 days
Frequency of treatment : Daily
Control group and treatment :
Post exposure observation period : Up to 30 days
Statistical methods :

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|---|
| Test conditions | : | Two groups of dogs (6-12 mos old) were given acrylamide orally in gelatin capsules for 7d/wk in daily doses of 5 and 15 mg/kg/d. The high dose dogs were maintained on treatment for 22 days with recovery periods of 0, 10 or 30 days. The low dose dogs were maintained for 60 days with no recovery. Symptomology and food consumption were recorded daily and body weight 3x/wk. Blood and urine samples were taken on day 0, 18,29, and 52. A neurological exam was performed pre and post test. Motor behavior and gait were recorded on video at the end of the treatment period. Electrophysiological tests were performed at the end of the experiment. Conduction velocity and relative and absolute refractory periods were performed. |
| NOAEL (NOEL) | : | 5 mg/kg/day |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | The animals of the high dose group developed neuropathological signs consistent with peripheral neuropathy. Electrophysiological measurements on the saphenous nerve revealed abnormal nervous function at the end of the treatment period. Conduction velocity was affected to the greatest extent, followed by absolute refractory period and chronaxy. At 30 days upon cessation of treatment conduction velocity and chronaxy were restored to normal but absolute refractory period remained abnormal. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Thomann et al. 1974) |
| Test substance | : | 99 % pure |
| Method | : | |
| Type | : | |
| GLP | : | Pre-GLP |
| Year | : | 1981 |
| Species | : | Dog |
| Strain | : | Greyhound |
| Route of administration | : | Feed |
| Duration of test | : | |
| Doses/concentration levels | : | 7 mg/kg/d |
| Sex | : | |
| Exposure period | : | 8 weeks |
| Frequency of treatment | : | Daily |
| Control group and treatment | : | No control group |
| Post exposure observation period | : | |
| Statistical methods | : | |
| Test conditions | : | Fourteen dogs (1-5 yrs) received 7 mg/kg/day acrylamide by dietary admixture for about 8 weeks. There were no control animals used. Animals were examined daily and a body weight and clinical signs were recorded. The esophagus was examined radiologically. Statistical Analysis: Means were compared using Student's two tailed t test. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | Clinical signs of toxicity included severe impairment of hindlimb function: 'toe-folding' being observed from about day 30, ataxia from about day 40, clear signs of muscle weakness from around day 50, and regurgitation from around day 60. Expansion of the esophagus (megaesophagus) was noted radiologically in 3 out of 14 dogs. However, the significance of this finding is uncertain as only 3 animals were examined, there were no controls for this part of the study, and megaesophagus was reported by the authors to occur spontaneously in the dog with the etiology unknown. |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, small treatment group, no controls |
| Reference | : | (Satchell & McLeod. 1981) |
| Test substance | : | 99 % pure |
| Method | : | |
| Type | : | |
| GLP | : | No data |
| Year | : | 1989 |
| Species | : | Dog |
| Strain | : | Greyhound |
| Route of administration | : | Oral – gelatin capsules |
| Duration of test | : | |
| Doses/concentration levels | : | 5.7 mg/kg/day |
| Sex | : | |
| Exposure period | : | 6-7 weeks |
| Frequency of treatment | : | Twice daily |
| Control group and treatment | : | Control group: Yes, not specified |
| Post exposure observation period | : | up to 5 months |
| Statistical methods | : | |
| Test condition | : | In another study focusing on respiratory effects, 4 dogs (22-30 kg) received 5.7 mg/kg/day acrylamide in gelatin capsules for 6-7 weeks, with up to 8 weeks recovery. Resting respiration was measured using an intratracheal technique, and electrocardiography, electroencephalography, and heart rates were recorded. In 2 animals blood levels of CO ₂ and transcutaneous oxyhemoglobin were also recorded. The Hering-Breuer lung inflation reflex was quantified by measuring the duration of apnea produced during lung inflation and was used as an indicator of the function of the vagus nerve. Parameters for each animal were recorded pre-exposure and served as controls for this study. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | One animal was killed due to pneumonia at around week 10. Loss of use of hind limbs and 'toe-folding' were observed from about week 3 and resolved during the 5th week of recovery. Decreased respiratory frequency and slightly increased tidal volume were observed during the ACR exposure |

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| | period but were restored during the recovery period. The Hering-Breuer lung inflation reflex was impaired (as indicated by increased tidal volume and decreased respiratory frequency) Other parameters were not adversely affected. The toxicological significance of these respiratory effects is unclear, although the altered Hering-Breuer reflex could be indicative of damage to the vagus nerve. |
| Conclusion | : |
| Reliability | : (1) reliable without restriction |
| Reference | : (Hersch et al. 1989) |
| Test substance | : 99 % pure |
| Method | : Somatosensory thresholds |
| Type | : |
| GLP | : No data |
| Year | : 1983 |
| Species | : Monkey |
| Strain | : Macaca nemestrina |
| Route of administration | : Oral, drinking water |
| Duration of test | : |
| Doses/concentration levels | : 10 mg/kg/d |
| Sex | : Female |
| Exposure period | : 44-61 days |
| Frequency of treatment | : 5 days/week |
| Control group and treatment | : Control group: Yes |
| Post exposure observation period | : 146 days |
| Statistical methods | : |
| Test condition | : In an extensive study, four feral-born Macaque monkeys received 10 mg/kg/day acrylamide (>99% purity) in fruit juice for 5 days/week for 44-61 days, until the time of onset of clinical signs of toxicity. For animal welfare reasons, treatment with acrylamide was not continued beyond this point and animals were allowed to recover, with examinations still performed, for a period of up to 146 days. Two control animals received tap water only, for about 13 weeks using a similar dosing regime. Investigations included recording bodyweight, clinical signs of toxicity, a visuomotor task (time taken to pick up a food reward) performed twice per week, sensitivity to an electrical or a vibration stimulus also performed twice per week, and sural nerve histopathology performed first when vibration thresholds were elevated (about day 51-58 of acrylamide exposure) and then during the recovery phase (up to 146 days after the last acrylamide exposure). |
| NOAEL (NOEL) | : |
| LOAEL (LOEL) | : |
| Actual dose/dose level/sex | : |
| Toxic response/effects by dose level | : |
| Result | : Amongst treated animals, clinical signs of toxicity included loss of balance, decreased activity, hindlimb weakness, and forelimb tremor in the final week of acrylamide treatment for one particular animal. With the exception of forelimb tremor, which persisted for up to 4 weeks, these clinical signs of toxicity resolved within 2 weeks post-treatment. During the treatment period actual bodyweight loss (up to 30% reduction) was noted in 3/4 animals. However, one of the 2 control animals also showed bodyweight loss |

(approximately 25% reduction).

Amongst treated animals, response to a 60Hz electrical stimulus was not apparently affected during or after treatment. However, there was a decreased sensitivity (as assessed by an increased time to key-pressing) towards a vibration stimulus (40Hz and 150 Hz) during the treatment phase with effects being even more pronounced in the first 10 weeks post-treatment. An increased time taken to pick up a food reward was noted in test animals towards the end of the treatment period and was more pronounced in the first 3 weeks post-treatment.

Sural nerve biopsies were prepared from 2 acrylamide-exposed animals, firstly when vibration thresholds were elevated (day 51 and 58 of treatment) and then during the recovery phase (day 146 and 136). At the first examination, in some areas there were no axons visible and myelin had formed balls or whorls although most nerve fibres appeared to be normal under light microscopy. Electron microscopy also showed that most myelinated nerve fibres were apparently normal but others showed axolemma invagination, disruption of myelin, other "severe axonal alterations" (not further described) or a loss of axons. Some Schwann cells lacked an axon and contained contorted and apparently disintegrating myelin - several Schwann cells contained lipid vacuoles. In one animal about 25% of nerve fibres were affected, but in the other, only 'occasional' fibres were affected. No abnormalities were observed in unmyelinated nerve fibres.

The second biopsy (performed during the recovery phase when no abnormalities were seen in vibration sensitivity) showed that degenerative changes were less frequent than during the treatment period and regenerative fibres were also seen. Loss of vibration sensitivity did not appear to be associated with the neuropathological findings.

| | |
|---|---|
| Conclusion | : |
| Reliability | : (1) reliable without restriction |
| Reference | : (Maurissen et al. 1983) |
| Test substance | : >99% purity |
| Method | : Other |
| Type | : |
| GLP | : Pre-GLP |
| Year | : 1980 |
| Species | : Rat |
| Strain | : Fischer 344 |
| Route of administration | : Drinking water |
| Duration of test | : |
| Doses/concentration levels | : |
| Sex | : |
| Exposure period | : 93 days |
| Frequency of treatment | : Daily |
| Control group and treatment | : 0.0, 0.05, 0.2, 1.0, 2.0, 5.0, 20.0 mg/kg |
| Post exposure observation period | : 10 males/group held for 144 day recovery period |
| Statistical methods | : |

Test conditions : Groups of 10 male and 10 female rats acrylamide (>99% purity), There were an additional 10 males per group were held for a 144 day recovery period and a further 6-9 males for interim sacrifices and electron microscopy during the 90-day exposure period. Investigations conducted weekly included: recording of bodyweight, clinical signs of toxicity, tests for peripheral neuropathy (measuring foot-splay after being dropped onto a horizontal surface from a low height), water consumption. In addition, hematology was performed on day 76, at termination, and on day 60 of the recovery period for those not sacrificed after 90 days of acrylamide exposure. Urinalysis was performed on day 76 and on completion of the exposure period. Extensive macro- and microscopic pathology examinations were performed on 59 males and 60 females after 92-93 days of acrylamide exposure and on 4 males per exposure group after 144 days of recovery. Electron microscopy was performed on males during the 90 days of acrylamide exposure (on days 7, 33, and 90) and during the 144 day recovery period (days 25, 111, and 144 of recovery).

NOAEL (NOEL) : 0.2 mg/kg

LOAEL (LOEL) :

Actual dose/dose level/sex :

Toxic response/effects by dose level :

Result : One male at 20 mg/kg/day was found dead on day 87. There were no other mortalities and the results of pathology examinations for this animal were incorporated with those sacrificed after 90 days. Reduced bodyweight gain was noted only at 20 mg/kg/day (21% reduction in males and 24% reduction in females at 90 days). Amongst those animals that were examined (males only), bodyweight was restored by day 141 of the recovery period. Significantly reduced water consumption was noted amongst females at 20 mg/kg/day (up to 40% reduction).

Statistically significant increases in landing foot-spread measurements were observed amongst males and females at 20 mg/kg/day on day 22 and effects were more pronounced at day 29 such that this test was discontinued to prevent injury. Other clinical signs of toxicity included curling of toes, splayed hindlimbs, incoordination, and hindlimb weakness. At the end of 90 days there was a loss of use of hindlimbs. The landing foot spread test was performed on males and females at 5 mg/kg/day on day 29 revealed no abnormalities.

A landing foot spread test was also conducted on day 12 of the recovery period for control males and males that had received 5 mg/kg/day. There were no abnormalities seen in this test and there were no clinical signs of toxicity at this or lower exposure levels. Males that had received 20 mg/kg/day were not subject to the landing foot spread test during the recovery period, but by day 7 of the recovery period some were able to use their hind limbs. At day 111 of the recovery period, curling of the toes was still apparent, and there were still some signs of posterior weakness. By day 144 there were no behavioural abnormalities.

Given the nature of other effects noted in this study, there were no remarkable results obtained from blood biochemistry examinations. There were no changes observed in the urinalysis parameters recorded.

Hematology examinations on day 76 and at termination showed decreased PCV, RBC, and haemoglobin (Hgb) values amongst males and females at 20 mg/kg/day. Significant decreases in these parameters were also

observed at termination amongst females at 5 mg/kg/day. Hematology performed on day 4 of the recovery period still showed a reduction in PCV, RBC, and Hgb values amongst males that had received 20 mg/kg/day. By day 60 of the recovery period a slight, but statistically significant, reduction in RBC was still observed; other values had returned to normal. There were no further hematology examinations. As with the blood biochemistry examinations, the magnitude of effects was not reported.

Increased organ weights (relative to bodyweight) were observed in males and females at 20 mg/kg/day for brain, heart, liver, and kidneys (magnitude not given). Also at this exposure level decreased relative thymus weight was observed in females, and reduced testes weights were observed in males. Increased relative liver weight was observed amongst males at 5 mg/kg/day. On day 144 of the recovery period, brain, kidney, and liver weights were still increased amongst males that had received 20 mg/kg/day.

Macroscopic examination after 90 days showed the following changes amongst males and females at 20 mg/kg/day: urogenital fur staining, decreased adipose tissue, small liver (due to reduced bodyweight), dark kidneys, foci or areas of mottled appearance in the lungs, small or flaccid testes, and small accessory genitalia in males, small uterus in females, dull appearance or loss of striated appearance of peripheral nerves, atrophy of skeletal muscle in the posterior portion of the body, distension of the bladder, and diffuse mural thickening of the stomach. There were no significant macroscopic pathology observations at lower dose levels.

Macroscopic pathology examinations performed on 4 male rats from each group after 144 days of recovery showed lesions only amongst those rats that had received 20 mg/kg/day: these were dark testes in 3/4 males and slightly distended urinary bladder in all four.

Histopathology of peripheral nerves after 90 days showed axon and myelin degeneration: both enlarged and unusually small axons were observed, others were fragmented or broken, or absent. Myelin degeneration was prominent and observed as clumping of myelin, myelin debris, vacuolization, or absence of myelin. There also appeared to be increased interstitial space between individual nerve fibres. These peripheral nerve lesions were seen to a marked extent in all animals at 20 mg/kg/day. Peripheral nerve lesions were also observed in most animals at 5 mg/kg/day but varied in severity from equivocal to very slight (focal or multifocal changes in individual nerves) in 9/10 males and 6/10 females. Spinal cord sections were taken from the cervical, thoracic, and lumbosacral regions. Equivocal to slight degenerative myelopathy (demyelination, swollen astrocytes, and swollen axons) was seen in the dorsomedial funiculi of one or all spinal cord sections in 5/10 males and 9/10 females at 20 mg/kg/day only. Transverse sections through the cerebrum, cerebellum, and midbrain did not reveal any abnormalities amongst those animals examined (control and high dose levels).

The other major pathology findings, after 90 days of treatment at 20 mg/kg/day, were atrophy of skeletal muscle in 2/10 males and 8/10 females; ulcerative gastritis or hyperkeratosis of the non-glandular stomach (4/10 males); testicular atrophy (10/10 males); mineralization of focal or multifocal seminiferous tubules of the testes (5/10 males); increased cellular debris and/or decreased spermatogenic elements in the tubular lumina of epididymides (9/10 males); vacuolization of the smooth muscle of the

bladder (1/10 males and 2/9 females); and suppurative, chronic-active or granulomatous inflammation in the lungs (3/10 males and 5/10 females).

Portions of perfused sciatic and brachial nerves from males after 25, 111, and 144 days of recovery were also examined. Nerve damage similar to that seen during the treatment phase was seen in males that had received 20 and 5 mg/kg/day only. Findings after 25 days of recovery were apparently more severe than those observed during the 90 day exposure period but subsequently gradual recovery of the nerve damage was observed such that at 144 days of recovery only very slight to slight alterations were seen in sciatic nerves of males that had received 20 mg/kg/day. However, peripheral nerve lesions (altered tinctorial properties and/or vacuolization of fibres) were still present at this dose in sciatic and brachial nerves although findings were less severe than after 90 days of treatment. There was evidence that some regeneration had occurred. There were no signs of nerve damage at this time point in other groups.

At the end of the recovery period, all four males that had received 20 mg/kg/day still had testicular lesions (slight focal or multifocal atrophy of seminiferous tubules and mineralization and cellular debris in focal or multifocal tubules). Lesions in the urinary bladder had essentially recovered by this time. Some inflammatory lesions were observed in the liver and lungs of males that had received 20 mg/kg/day although the significance of these was uncertain.

Electron microscopy of nerve tissue provided additional evidence of substantial neuropathy, with some post-exposure recovery, at 20 and 5 mg/kg/day. There were also some axolemmal invaginations at 1 mg/kg/day at 90 days. No ultrastructural changes were observed at lower doses.

In summary, this study demonstrated that oral administration of acrylamide to rats for 90 days principally resulted in severe lesions of peripheral nerves and spinal cord at 20 mg/kg/day (with associated clinical signs of toxicity); atrophy of skeletal muscle; testicular atrophy (although all of the stages of spermatogenesis were still apparent); decreased red blood cell parameters. Peripheral nerve lesions were also observed at 5 mg/kg/day, and slight changes in nerve tissue (visualised only by electron microscopy) were seen at 1 mg/kg/day. No effects were seen at 0.2 mg/kg/day or less. Where nerve damage was produced there was some, but not complete, recovery after a 144-day post-exposure recovery period.

Conclusion :
Reliability : (1) reliable without restrictions
Reference : (Burek et al., 1980)

Test substance : No data
Method :
Type :
GLP : Pre-GLP
Year : 1979
Species : Rat
Strain : Fischer 344
Route of administration : Oral gavage
Duration of test :
Doses/concentration levels : 0, 5, 10, or 20 mg/kg
Sex : Male

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Exposure period | : | 13 weeks |
| Frequency of treatment | : | 3 times/week |
| Control group and treatment | : | Control group: Yes (vehicle dosed and undosed) |
| Post exposure observation period | : | 5 weeks |
| Statistical methods | : | |
| Test condition | : | <p>5 Groups of 10 F344 male rats received 0 (dosed/undosed), 5, 10, or 20 mg/kg aqueous acrylamide by gavage 3 days/week for 13 weeks. A range of behavioral tests (hindlimb extensor response, spontaneous motor activity, forelimb grip strength) were performed predose and in weeks 1, 4, 7, 10, and 13 of acrylamide exposure. After 13 weeks, neuropathological examination (medulla oblongata, sciatic nerve at mid-thigh, branches of tibial nerve supplying calf muscles) was performed on 5 controls, all animals at 10 mg/kg and 5 out of 10 animals at 20mg/kg. The remaining animals in the control and top-dose groups were retained for further behavioral tests at weeks 1 and 5 of a recovery period followed by a neuropathological examination.</p> <p>Statistical Analysis: ANOVA was used to determine statistical significance of body weights, hindlimb and forelimb scores and motor activity. A combined control group was used to assess individual differences between treatment and controls using Dunnett -test. Pearson's product moment correlation and test for statistical association between bodyweight and hindlimb extensor response was calculated as described by Hayes.</p> |
| NOAEL (NOEL) | : | 5 mg/kg |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | <p>There was no effects at 5 mg/kg. Reduced bodyweight gain (approximately 15% reduction) was noted amongst animals receiving 10 (only up to week 7) or 20 mg/kg. Hindlimb extensor response was reduced only at 20 mg/kg in weeks 7, 10, and 13 and in week 1 of recovery. No abnormality in hindlimb response was seen after 5 weeks of recovery. Reduced spontaneous locomotor activity was noted only at 20 mg/kg in weeks 10 and 13. Recovery was complete after 5 weeks post-exposure. Forelimb grip strength was reduced at 20 mg/kg on weeks 4 and 7 and in week 1 of recovery but not at any other time point.</p> <p>After 13 weeks of exposure slight neuropathology (distal nerve fibre degeneration) was seen in 9/10 animals and moderate neuropathology (formation of Schwann cell columns) in 1/10 at 10 mg/kg. All 5 animals at 20 mg/kg that were examined showed moderate damage (fibre degeneration and Schwann cell column formation with regenerating or remyelinating fibres). After 5 weeks of recovery animals at 20 mg/kg still showed moderate neuropathology (distal degeneration of large diameter myelinated fibres with clusters of small regenerating myelinated fibres in peripheral nerves).</p> <p>In summary, peripheral neuropathy (seen histopathologically and by behavioral tests) was observed at 10 and 20 mg/kg acrylamide when administered 3 days/week for 13 weeks by the oral route. Recovery was seen by 5 weeks at 10 mg/kg but not at 20 mg/kg</p> |
| Conclusion | : | |
| Reliability | : | (1) Reliable without restriction |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Reference | : | (Tilson and Cabe 1979a) |
| Test substance | : | No data |
| Method | : | Repeated Dose Toxicity |
| Type | : | |
| GLP | : | No data |
| Year | : | 1985 |
| Species | : | Monkey |
| Strain | : | Macaca nemestrina |
| Route of administration | : | Other: oral in fruit juice |
| Duration of test | : | |
| Doses/concentration levels | : | 10 mg/kg/d |
| Sex | : | No data |
| Exposure period | : | Up to 13 weeks |
| Frequency of treatment | : | 5 days/week |
| Control group and treatment | : | Control group: Yes, not specified |
| Post exposure observation period | : | 20-30 weeks |
| Statistical methods | : | |
| Test condition | : | In a study investigating potential effects on the visual system, a group of 7 Macaque monkeys received 10 mg/kg/day acrylamide in fruit juice for 5 days/week for up to 13 weeks with approximately 20-30 weeks recovery. In addition, there were 2 control animals. Two monkeys were killed within 1 week of the intoxication period, two were killed after a comparable intoxication period, and subsequent recovery interval without receiving additional acrylamide, and three were killed after two comparable successive intoxication and recovery periods. One immediate sacrifice monkey also was subjected to intensive testing of visual capacities, before and during intoxication. Both delayed-sacrifice monkeys were tested visually before and during intoxication, as well as during the subsequent interval prior to killing. Brain, optic nerve and eyes were removed for histopathological (light and electron microscopy) examination. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | For acrylamide-treated animals sacrificed immediately after 9-13 weeks, distal axonal swelling was most prominent in distal optic tract fibres, particularly within the lateral geniculate nucleus. Myelin sheaths were disproportionately thin and degenerating myelin and occasional shrunken axons were observed. Degenerating myelin and degenerating/atrophic axons were seen in the optic nerve and the proximal optic tract. In the lateral geniculate nucleus of the brain, axonal swellings were again seen and occasional alterations in the retinal axon terminals and synapses were observed by light and electron microscopy. Dilatation of the axonal terminals, degeneration of myelin, degenerating/atrophic axons, and an increased number of astroglial processes were also seen in the lateral geniculate nucleus. No abnormalities were seen in controls. The optic nerves of acrylamide-exposed animals showed a loss of axons, and diminished numbers of fibres in the optic nerve. Electron microscopy |

5. Toxicity

Id 79-06-1

Date 7 September 2001

showed disproportionately thin myelin sheaths, densely packed astroglial processes, lipid vacuolation, and degenerating myelin fragments in the phagocytes and astrocytes.

This study shows marked effects, including loss of ganglion cells (particularly near the fovea), and axonal loss in the nerve fibres of the optic tract and in the lateral geniculate nucleus of the brain of non-human primates exposed to 10 mg/kg acrylamide, 5 days/week for up to 13 weeks.

| | | |
|---|---|--|
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Eskins et al, 1985) |
| Test substance | : | No data |
| Method | : | Neurotoxicity |
| Type | : | |
| GLP | : | No data |
| Year | : | 1983 |
| Species | : | Rat |
| Strain | : | Wistar |
| Route of administration | : | Oral, drinking water |
| Duration of test | : | |
| Doses/concentration levels | : | 0, 7.5, 12, 19, and 30 mg/kg |
| Sex | : | Male |
| Exposure period | : | 90 days |
| Frequency of treatment | : | Daily |
| Control group and treatment | : | Control group: Yes |
| Post exposure observation period | : | |
| Statistical methods | : | |
| Test condition | : | Groups of 4 male Wistar rats received 0, 52, 80, 125, or 200 mg/l acrylamide in drinking water for 90 days. Note: The published report did not state actual daily dosages but assuming a mean bodyweight of 200g and daily water consumption of 30ml; these concentrations would approximate to 0, 7.5, 12, 19, and 30 mg/kg/day. Rotarod performance according to Dunham and Miya, 1957 was recorded weekly. Light microscopy examination was performed on posterior tibial nerves and sural nerves from the lower calf muscle region. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | A slight reduction in bodyweight gain was noted amongst all treated animals (4% reduction at the lowest exposure level, 10% at the highest). Rotarod performance at day 90 showed impairment only at the two highest exposure levels (3/4 animals at approximately 19 mg/kg/day and 4/4 animals at approximately 30 mg/kg/day). No other rotarod results were available. Other clinical signs of toxicity apparently included weakness, tendency towards spreading and dragging hind limbs and occasionally, amongst more severely affected animals, urinary incontinence; however it was not clear which groups these findings were seen in. Light microscopy examination showed moderate to severe changes: shrinkage and loss of myelinated |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|---|
| Conclusion | : | fibres, myelin retraction, and corrugation of myelin sheaths at about 30 mg/kg/day. |
| Reliability | : | (2) reliable with restriction, The incidence and severity of findings at other exposure levels was not reported and hence a NOAEL was not identifiable from this study. |
| Reference | : | (Tanii and Hashimoto 1983) |
| Test substance | : | 96-99% pure |
| Method | : | Other |
| Type | : | |
| GLP | : | No data |
| Year | : | 1984 |
| Species | : | Rat |
| Strain | : | Fischer 344 |
| Route of administration | : | Drinking water |
| Duration of test | : | 2 years |
| Doses/concentration levels | : | |
| Sex | : | Male/female |
| Exposure period | : | |
| Frequency of treatment | : | Daily |
| Control group and treatment | : | 0.0, 0.01, 0.1, 0.5, 2.0 mg/kg/d |
| Post exposure observation period | : | |
| Statistical methods | : | |
| Test condition | : | 90 rats/sex/dose, approx 5 wks of age were evaluated for mortality, clinical signs, body weight, food and water consumption, clinical chemistry, hematology, urinalysis, gross pathology, organ weights, and histopathology. 10 rats/sex/dose were scheduled for interim sacrifice at 6, 12 or 18 months on study. Gross necropsy included the adrenal glands, auditory gland, aorta, bone, bone marrow, brain, cecum, cervix, coagulating glands, epididymides, esophagus, eyes, heart, liver, kidneys, large intestine, lacrimal glands, larynx, lymph node (mediastinal and mesenteric), lungs, mammary gland, ovaries, oviducts, pancreas, pituitary, prostate, salivary glands, seminal vesicles, skin, small intestine, spinal cord, spleen, stomach, testes, thymus, thyroid gland, tongue, trachea, urinary bladder, uterus, vagina, and gross lesions. |
| | | <u>Statistical Analysis:</u> Body weights by ANOVA, mortality by Wilcoxon test, and histopathological observations by Fisher's Exact Probability Test. |
| NOAEL (NOEL) | : | 0.1 mg/kg |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Results | : | A statistically significant increase in mortality was noted from 21 months onwards amongst males and females receiving 2 mg/kg/day. A slight decrease in bodyweight (up to 4%) was noted amongst males at 2 mg/kg/day and there were no significant effects on food and water consumption and no clinical signs of toxicity. There were no significant adverse effects on haematology, blood biochemistry, or urinalysis examinations, organ weights, or macroscopic pathology at 6, 12 and 18 |

months. However, at 24 months, there was an increase in the number of subcutaneous and mammary gland masses amongst females at 2 mg/kg/day. Histopathologically, there were no abnormalities at 6 months. At examinations performed from 12 months onwards there was an increase in the incidence and severity of tibial nerve degeneration amongst males at 2 mg/kg/day and from 18 months onwards in females at 2 mg/kg/day (focal swelling of nerve fibres with fragmentation of myelin and axon, and the formation of vacuoles containing small round eosinophilic globules and macrophages). There were no clear changes amongst animals at lower exposure levels or amongst other peripheral nerve samples (saphenous branch of the femoral nerve and brachial plexus).

In males, there was a statistically significantly increased incidence of benign follicular cell adenomas of the thyroid at the highest dose level (1/60, 0/58, 2/59, 1/59, 7/59). In females there was a non-significant increase in the incidence of benign follicular cell adenomas of the thyroid (0/58, 0/59, 1/59, 1/58, 3/60) and malignant adenocarcinomas (1/58, 0/59, 0/59, 0/58, 3/60). In females there was a statistically significant increase in the incidence of malignant adenocarcinomas in the uterus (1/60, 2/60, 1/60, 0/59, 5/60, or 1.7%, 3.3%, 1.7%, 0, 8.3%). The historical control range was stated to be 0-2.3%. In males there was a statistically significant increase in the incidence of malignant testicular mesothelioma at 0.5 and 2 mg/kg/day (3/60, 0/60, 7/60, 11/60, 10/60 or 5%, 0, 12%, 18%, 17%). The historical control incidence was 3.1% with a range of 2-6%.

In males there was a non-significant increase in the incidence of malignant astrocytomas in the spinal cord (1/60, 0/60, 0/60, 0/60, 3/60). There were also non-significant increases in malignant astrocytomas in the brain of females (0/60, 1/60, 0/60, 0/60, 3/60), glial proliferation in the brain suggestive of an early tumour (0/60, 0/60, 0/60, 1/60, 3/60), and malignant astrocytomas in the spinal cord (1/60, 0/59, 0/60, 0/60, 3/61). In addition, malignant astrocytomas were also observed in the brain (3/60, 0/60, 0/60, 2/60, 2/60), and glial proliferation (suggestive of an early tumour) in 0/60, 0/60, 0/60, 1/60, 1/60. The effects in astrocytomas for brain and spinal cord in males and females do not show any clear dose-response but there are some concerns as these tumours are occurring in potential target tissues and, according to the authors, the concurrent control values may have been abnormally high so trends would not have been clear. Also, the group sizes used in this study may not have been sufficiently large enough to detect clear increases. Overall, because of these limitations, the toxicological significance of the presence of these astrocytomas in this study is unclear.

For females, there was a statistically significant increase in the incidence of benign papillomas in the oral cavity at 2 mg/kg/day (0/60, 3/60, 2/60, 1/60, 7/61) and a non-significant increase in focal hyperplasia (1/60, 2/60, 1/60, 0/60, 4/61). The incidence of malignant carcinomas did not show any clear dose-response (0/60, 0/60, 0/60, 2/60, 1/61). For males, the incidence of tumour formation in the oral cavity did not show any clear exposure relationship (carcinomas 2/60, 0/60, 1/60, 0/60, 2/60, and papillomas 4/60, 7/60, 0/60, 5/60, 4/60) although there was a statistically significant increase in focal hyperplasia of the hard palate (0/60, 1/60, 1/60, 1/60, 4/60, 5/60). Again, although effects are not clear, there are some concerns as there is a possibility that hyperplasia and subsequent, but unclear, tumor formation may have arisen as a result of local effects due to the route of exposure employed.

In females there were increases in benign and malignant tumors of

5. Toxicity

Id 79-06-1

Date 7 September 2001

mammary glands (10/60, 11/60, 9/60, 19/58, 23/61 and 2/60, 1/60, 1/60, 2/58, 6/61 respectively or 17%, 18%, 15%, 33%, 38% and 3%, 2%, 2%, 3%, 10%), benign pituitary gland adenomas (25/59, 30/60, 32/60, 27/60, 32/60 or 42%, 50%, 53%, 45%, 53%), and benign tumors of the clitoral gland (0/2, 1/3, 3/4, 2/4, 5/5). In males there were increased incidences of benign tumours in the adrenal glands (pheochromocytoma) (3/60, 7/59, 7/60, 5/60, 10/60 or 5%, 12%, 12%, 8%, 17%). The increased incidences of mammary tumors, benign pituitary adenomas and adrenal pheochromocytomas are of doubtful toxicological significance due to the poor dose-response and high historical control incidence (18% for benign mammary tumours, 2% for malignant mammary tumors - NTP data only, 28-47% for pituitary adenomas, 1-14% for pheochromocytomas). For clitoral adenomas the total number of tissues examined was too small to draw any firm conclusions.

Conclusion :
Reliability : (1) Reliable without restrictions
Reference : (Johnson et al., 1984)

Test substance : 99 % pure
Method : Repeated Dose Toxicity
Type :
GLP : Pre-GLP
Year : 1964
Species : Cat
Strain : No data
Route of administration : Oral feed
Duration of test :
Doses/concentration levels : 0.0, 0.03, 0.1, 0.3, 1, 3, 10 mg/kg/d
Sex :
Exposure period : 1 year
Frequency of treatment : 5 days/week
Control group and treatment :
Post exposure observation period : :
Statistical methods :
Test conditions : Groups of 2 cats/dose of unknown origin received 0.03, 0.1, 0.3, 1, 3, or 10 mg/kg/day acrylamide by dietary administration 5 days/week for up to 1 year. Three control animals received only basic ration. The cats were weighed once weekly and were observed frequently for gross changes in appearance or behavior.

NOAEL (NOEL) : 0.3 - 1.0 mg/kg bw
LOAEL (LOEL) :
Actual dose/dose level/sex :
Toxic response/effects by dose level :
Result : Two control animals died and one was killed due to intercurrent infection after 171 days. Signs of peripheral neuropathy (loss of use of hind limbs, abnormal gait) were observed at 1 mg/kg/day and above. However, all animals at 0.03, 0.1, and 1 out of 2 cats at 0.3 mg/kg/day died apparently from intercurrent infection. It was reported that there were no pathological abnormalities attributable to acrylamide at any exposure level. However, the extent of examination was unclear and firm conclusions are hard to draw

5. Toxicity

Id 79-06-1

Date 7 September 2001

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|---|---|
| | due to the general poor condition of animals used in this study. |
| | 0.3 mg/kg/d was without evident of adverse effect in the cat. Only doubtfully significant effects at 1.0 mg/kg. |
| Conclusion | : |
| Reliability | :(2) reliable with restriction, source/condition of cats unknown |
| Reference | :(McCollister et al., 1964) |
| Test substance | : 99 % pure |
| Method | : Repeated Dose Toxicity |
| Type | : |
| GLP | : Pre-GLP |
| Year | : 1964 |
| Species | : Monkey |
| Strain | : |
| Route of administration | : Oral feed |
| Duration of test | : |
| Doses/concentration levels | : 0.0, 0.03, 0.1, 0.3, 1, 3, 10 mg/kg/d |
| Sex | : |
| Exposure period | : 363 days |
| Frequency of treatment | : 5 days/week |
| Control group and treatment | : Control group: Yes |
| Post exposure observation period | : |
| Statistical methods | : |
| Test conditions | : One female monkey (unspecified species) per dose level received either 0, 0.03, 0.1, 0.3 (2 animals at this exposure level), 1, 3, or 10 mg/kg/day aqueous acrylamide by oral gavage or dietary administration 5 days/week for up to 1 year. After acrylamide exposure hematology, blood cholinesterase measurements, and macro- and microscopic pathology were conducted. There were no details available regarding these examinations. |
| NOAEL (NOEL) | : 1-3 mg/kg bw |
| LOAEL (LOEL) | : |
| Actual dose/dose level/sex | : |
| Toxic response/effects by dose level | : |
| Result | : Clear and severe clinical signs of neuropathy were apparent at 10 mg/kg/day. At 3 mg/kg/day occasional abnormalities were observed; reduced knee jerk reaction, reduced pupillary reflexes (response to bright light), and lethargic behavior. At 0.1, 0.3 and 1 mg/kg/day acrylamide exposure for 1 year there were no apparent effects on bodyweight, no clinical signs of toxicity, no changes in hematology (although it is unclear which parameters were recorded), liver and kidney weights, and no macroscopic or microscopic pathology abnormalities (extent of examination unclear, but probably included at least the brain and spinal cord). |
| Conclusion | : It is difficult to draw firm conclusions from this study due to limited reporting, and the use of only one animal per dose level. |
| Reliability | :(2) reliable with restriction, limited reporting and the use of only one animal per dose level |
| Reference | :(McCollister et al., 1964) |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Test substance | : | No data |
| Method | : | |
| Type | : | |
| GLP | : | No data |
| Year | : | |
| Species | : | Rodent |
| Strain | : | |
| Route of administration | : | Oral unspecified |
| Duration of test | : | |
| Doses/concentration levels | : | ≥ 12.5 mg/kg |
| Sex | : | No data |
| Exposure period | : | |
| Frequency of treatment | : | |
| Control group and treatment | : | |
| Post exposure observation period | : | : |
| Statistical methods | : | |
| Test conditions | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | Histopathologically, no effects ere seen in the nerves of animals exposed to 12.5 mg/kg |
| Source | : | Acrylamide Monomer Producers Association, Classification and labelling of acrylamide position paper on repeated dose toxicity (oral) |
| Reliability | : | (2) reliable with restrictions, data obtained from secondary source, primary source was not available |
| Reference | : | (Newton et al., 1993) |
| Test substance | : | 99 % pure |
| Method | : | |
| Type | : | |
| GLP | : | Pre-GLP |
| Year | : | 1982 |
| Species | : | Mouse |
| Strain | : | BALB/c |
| Route of administration | : | Oral, drinking water |
| Duration of test | : | Treatment 12 days/+44 days free/+19 days treatment/+31 days recovery |
| Doses/concentration levels | : | 26 mg/kg/d, after return to baseline 20 mg/kg/d |
| Sex | : | Female |
| Exposure period | : | 12 days/+19 days |
| Frequency of treatment | : | daily |
| Control group and treatment | : | Control group: Yes |
| Post exposure observation period | : | 44 days/+ 31 days |

| | | |
|---|---|---|
| Statistical methods | : | |
| Test condition | : | Groups of 5 female BALB/c mice received 0 or 26 mg/kg/day acrylamide (99% pure) in drinking water for 12 days. Following a recovery period of 44 days treated animals then received 20 mg/kg/day acrylamide for 19 days. An additional control group received 4-6% saccharin in order to mimic the reduction in water consumption in acrylamide-exposed animals, which may have affected performance in some of the tests conducted. Another 'control' group was given a restricted amount of food each day for similar reasons. Rotarod tests were conducted twice per week and were repeated 3 times on each of those occasions and a landing foot-spread test was conducted once per week and was repeated 5 times. |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Actual dose/dose level/sex | : | |
| Toxic response/effects by dose level | : | |
| Result | : | Hind limb foot-splay was increased from 6 days, and rotarod retention time decreased from 8 days after initial acrylamide exposure. Water consumption was reduced from day 1, although this was probably related to unpalatability. Actual bodyweight loss was noted from day 2 giving an overall loss by day 12 of 19% compared to pre-exposure weight, which could be partially attributed to reduced water consumption. After the 44th day recovery period, bodyweight values, water consumption, hind limb foot-splay, and rotarod retention were all apparently restored to control values. A similar pattern of effects and time taken to the onset of effects was noted for this second exposure period. Bodyweight, water consumption, and rotarod retention values were restored by day 31 of the recovery period following the second acrylamide exposure. The hind limb foot-splay effects were still unresolved after the 31st day recovery period. |
| | | Animals receiving distilled water, saccharin, or restricted food intake group showed no obvious changes in rotarod performance or hind limb foot-splay demonstrating that the impairment in performance in test animals was due to acrylamide. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Gilbert and Maurissen, 1982) |

5.5 GENETIC TOXICITY 'IN VITRO'

| | | |
|--------------------------------|---|---|
| Test substance | : | > 99% pure |
| Method | : | Ames et al. 1975, Maron and Ames, 1983 |
| Type | : | Ames test |
| System of testing | : | Bacterial |
| GLP | : | No data |
| Year | : | 1987 |
| Species and strain | : | Salmonella typhimurium TA 97, TA 98, TA 100, TA 1535, TA 1537 |
| Metabolic activation | : | with and without |
| Concentrations tested | : | ≤ 10000 ug/plate |
| Statistical methods | : | |
| Test conditions | : | According to standard procedures |
| Cytotoxic concentration | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--------------------------------|---|---|
| Genotoxic effects | : | |
| Statistical results | : | |
| Result | : | Lab1 (CWR): ambiguous Lab2 (SRI): negative |
| | | Lab1: Results were weakly mutagenic using <i>Salmonella typhimurium</i> tester strains TA98, TA 100, and TA 1535, and TA1537 in the presence and absence of metabolic activation. |
| | | Lab2: Results were negative using <i>Salmonella typhimurium</i> tester strains TA 97, TA98, TA 100, and TA 1535, in the presence and absence of metabolic activation. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Zeiger et al. 1987) |
| Test substance | : | 99% pure |
| Method | : | |
| Type | : | Ames test |
| System of testing | : | Bacterial |
| GLP | : | No data |
| Year | : | 1984 |
| Species and strain | : | Salmonella typhimurium TA 98, TA 100, TA 1535, TA 1537 |
| Metabolic activation | : | with and without |
| Concentrations tested | : | 3-30 mg/plate |
| Statistical methods | : | |
| Test condition | : | Assays were conducted with half log dose levels ranging from 0.001 to 3 mg in an initial assay and from 3 to 30 mg in a repeat assay. Duplicate platings were made at each test dose. S-9 fraction from Arochlor induced male SD rats was used for activation. Negative and positive controls were included with each assay. Sodium azide for strains TA1535 and TA100, 2-nitrofluorene for TA98 and 9 aminoacridine for TA1537 with activation, 2 aminoanthracene for all strains. |
| Cytotoxic concentration | : | |
| Genotoxic effects | : | |
| Statistical results | : | |
| Result | : | Negative Acrylamide failed to give a response in any strain with or without the addition of S-9, and with up to 30 mg added per plate. Negative mutagenicity results are apparently not attributable to cytotoxic interferences. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Bull et al., 1984) |
| Test substance | : | ≥ 98 % pure |
| Method | : | Ames et al. 1975, Maron and Ames, 1983 |
| Type | : | Ames test |
| System of testing | : | Bacterial |
| GLP | : | No data |
| Year | : | 1988 |
| Species and strain | : | Salmonella typhimurium TA 98, TA 100, TA 102, TA 1535, TA 1537 |
| Metabolic activation | : | with and without |
| Concentrations tested | : | ≥ 100000 ug/plate |
| Statistical methods | : | |
| Test conditions | : | According to standard procedures |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Negative

Results were consistently negative using *Salmonella typhimurium* tester strains TA 98, TA 100, TA 102, TA 1535, TA 1537, and TA 1538, in the presence and absence of metabolic activation.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Knaap et al. 1988)

Test substance : No data
Method : Ames et al. 1975
Type : Ames test
System of testing : Bacterial
GLP : No data
Year : 1985
Species and strain : *Salmonella typhimurium* TA 98, TA 100, TA 1535, TA 1537, TA1538
Metabolic activation : with and without
Concentrations tested : ≤ 5000 ug/plate
Statistical methods :
Test conditions : According to standard procedures.
Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Negative

Results were consistently negative using *Salmonella typhimurium* tester strains TA 98, TA 100, TA 1535, TA 1537, and TA 1538, in the presence and absence of metabolic activation.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Hashimoto & Tanii 1985)

Test substance : 99.9% pure
Method : Ames et al. 1975
Type : Ames test
System of testing : Bacterial
GLP : No data
Year : 1993
Species and strain : *Salmonella typhimurium* TA 98, TA 100, TA 1535, TA 1537
Metabolic activation : with and without
Concentrations tested : ≤ 50000 ug/plate
Statistical methods :
Test conditions : According to standard procedures
Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Negative

Results were consistently negative using *Salmonella typhimurium* tester strains TA 98, TA 100, TA 1535, and TA 1537, in the presence and absence of metabolic activation.

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--------------------------------|---|---|
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Tsuda et al 1993) |
| Test substance | : | ≥ 99 % pure |
| Method | : | Chromosome Aberration Test and SCE's |
| Type | : | Cytogenetic assay |
| System of testing | : | |
| GLP | : | No data |
| Year | : | 1993 |
| Species and strain | : | V79 Chinese Hamster Cells |
| Metabolic activation | : | without |
| Concentrations tested | : | ≤ 355 ug/ml |
| Statistical methods | : | |
| Test conditions | : | Standard procedures according to Dean and Danforde, 1984. V79H3 Chinese hamster cells were exposed to 0-5 mM (0-355 µg/ml) acrylamide without metabolic activation. The exposure period was 24 hours and fixation followed a further 20 or 40 hours later. |
| | | SCE: V79H3 Chinese hamster cells were exposed to 0-3 mM (0-213 µg/ml) acrylamide without metabolic activation for 24 hours followed by an additional 28 hours for bromodeoxyuridine (BrdU) incorporation; 50 cells per exposure level were counted. |
| Cytotoxic concentration | : | |
| Genotoxic effects | : | |
| Statistical results | : | |
| Result | : | Positive |
| | | There was a dose-related, statistically significant increase in the number of metaphases with chromosome aberrations. There was also a dose-related, statistically significant increase in polyploid cells at 20 and 40 hours (29% and 24% respectively at 4mM, 284 µg/ml). The incidence of chromosome aberrations and polyploid cells was less than 2% in negative controls, and the frequencies amongst acrylamide-exposed cells exceeded those of the positive control, N-methyl-N'-nitro-N-nitrosoguanidine (MNNG). There was no clear information available regarding cytotoxicity. |
| | | SCE: A slight, but dose-related and statistically significant increase in SCE was seen (50% increase compared to an 800% increase by the positive control, mitomycin C). |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Tsuda et al. 1993) |
| Test substance | : | ≥ 98 % pure |
| Method | : | |
| Type | : | Bacterial gene mutation assay |
| System of testing | : | Bacterial |
| GLP | : | No data |
| Year | : | 1988 |
| Species and strain | : | Klebsiella pneumoniae |
| Metabolic activation | : | Without |
| Concentrations tested | : | 2000, 5000, or 10000 ug/ml |
| Statistical methods | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Test conditions : Mutations to streptomycin resistance was assessed using *Klebsiella pneumoniae*. Tubes containing test compound and bacteria in nutrient broth were incubated at 37C. After 20h the bacteria were plated on streptomycin-supplemented agar and incubated for three days at 37C. Mutation frequencies were calculated based on the fraction of plates without mutants.

Cytotoxic

concentration :

Genotoxic effects :

Statistical results :

Result: Negative

In a fluctuation test (to determine mutations in genes conferring resistance to streptomycin) in *Klebsiella pneumoniae* the mutation frequency was not significantly altered by 2000-10000 µg/ml acrylamide

Conclusion :

Reliability : (1) reliable without restriction

Reference : (Knaap et al. 1988)

Test substance : ≥ 98 % pure

Method : Chromosome Aberration Test and SCE's

Type : Cytogenetic assay

System of testing :

GLP : No data

Year : 1988

Species and strain : V79 Chinese Hamster Cells

Metabolic activation : with and without

Concentrations tested : ≤ 3000 µg/ml

Statistical methods :

Test conditions : Standard procedures according to Dean and Danforde, 1984. V79 Chinese hamster cells exposed to 0-3000 µg/ml acrylamide with and without metabolic activation. The exposure period was for 3 hours with fixation following a further 15 hours.

Cytotoxic

concentration :

Genotoxic effects :

Statistical results :

Result : Positive

A dose-related significant increase in the number of metaphases with chromosome aberrations was observed with and without metabolic activation. There was no clear information available regarding cytotoxicity.

There was a slightly significant increase in SCE's at concs tested.

Conclusion :

Reliability : (1) reliable without restriction

Reference : (Knaap et al. 1988)

Test substance : No data

Method : Escherichia coli Reverse Mutation Assay

Type : Escherichia coli reverse mutation assay

System of testing :

GLP : No data

Year : 1981

Species and strain : Escherichia coli CR 63

Metabolic activation : Without

Concentrations tested : 100 µg/ml

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--------------------------------|---|--|
| Statistical methods | : | |
| Test conditions | : | Performed according to Mandel & Higa, 1970. Liver microsomes fractions were prepared according to Rajamanickam et al. 1975. |
| Cytotoxic concentration | : | |
| Genotoxic effects | : | |
| Statistical results | : | |
| Result | : | Positive |
| Conclusion | : | In a bacterial transfection assay using <i>E.coli</i> CR63 cells, a linear increase in percentage inhibition of transfection (apparently indicative of mutagenic potential in this assay system) was noted using up to 10 µg acrylamide. The significance of this finding is uncertain, particularly in view of the negative results in standard bacterial assay systems. |
| Reliability | : | (2) reliable with restriction, see conclusion |
| Reference | : | (Vasavada & Padayatty 1981) |
| Test substance | : | ≥ 99 % pure |
| Method | : | HPRT-Test |
| Type | : | HPRT assay |
| System of testing | : | |
| GLP | : | No data |
| Year | : | 1988 |
| Species and strain | : | L5178Y Mouse Lymphoma Cells |
| Metabolic activation | : | with and without |
| Concentrations tested | : | ≤ 10000 µg/ml |
| Statistical methods | : | |
| Test conditions | : | In a mouse lymphoma L5178Y TK +/- assay, cells were exposed to 0-10000 µg/ml acrylamide with and without metabolic activation for 2, 4, or 20 hours. Colonies were counted and the mutant frequencies were calculated. |
| Cytotoxic concentration | : | |
| Genotoxic effects | : | |
| Statistical results | : | |
| Result | : | Positive |
| Conclusion | : | At each of the time-points, with and without metabolic activation (+ or - S9), there was a slight dose-related increase in mutant frequency: 19-36 mutants/10 ⁶ survivors in controls; with 2 hours exposure 39/10 ⁶ at 2500 µg/ml -S9 and 119/10 ⁶ at 7500 µg/ml +S9, with 4 hours exposure 160/10 ⁶ at 2000 µg/ml -S9 and 189/10 ⁶ at 2500 µg/ml +S9; with 20 hours exposure 89/10 ⁶ at 300 µg/ml -S9 and 57-95/10 ⁶ at 300 µg/ml in the presence of metabolic activation (from primary rat liver or Syrian hamster ovary cells). These increases were associated with low survival (usually less than 30% survival). A positive result was obtained in this assay. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Knaap et al. 1988) |
| Test substance | : | ≥ 99 % pure |
| Method | : | HPRT-Test |
| Type | : | HPRT assay |
| System of testing | : | |
| GLP | : | No data |
| Year | : | 1993 |
| Species and strain | : | V79H3 Chinese Hamster Cells |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Metabolic activation : Without
Concentrations tested : ≤ 500 ug/ml
Statistical methods :
Test conditions : In an assay using Chinese hamster V79H3 cells (HPRT locus), cells were exposed without metabolic activation to 0-7 mM (0-500 µg/ml) acrylamide for 24 hours. Colonies were counted and the mutant frequencies were calculated.

Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Negative

The number of mutants per 106 survivors was 9, 8, 6, 5, 1, 2 at 0, 71, 213, 355, 416, 500 µg/ml respectively. At 213 µg/ml survival was 59%, at 355 µg/ml or more, survival was 21% or less. Acrylamide was not mutagenic in this assay.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Tsuda et al. 1993)

Test substance : > 99 % pure
Method : Chromosome Aberration Test
Type : Mouse lymphoma assay
System of testing :
GLP : No data
Year : 1987
Species and strain : L5178Y Mouse Lymphocyte
Metabolic activation : Without
Concentrations tested : 500, 600, 750, 850 ug/ml
Statistical methods :
Test conditions : Cells were treated using 0-850 µg/ml acrylamide (>99% pure) for 4 hours without metabolic activation (Clive et al., 1979) studying large and small colonies. After 9-11 days incubation at 37C, colonies were counted and the colony size distribution was determined. BrdUrd was added for cytogenetic analysis. 100 metaphase spreads were analysed for aberrations.

Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Positive

Increases associated with low survival were noted. An increased mutant frequency (70-400 mutants/10⁶ survivors, approximately 20/10⁶ in controls) was observed at 500 µg/ml acrylamide and above, and percentage survival was 40% at 500 µg/ml, 10% at 850 µg/ml. There was a dose-related increase in the frequency of small colonies and at 750 µg/ml and above mutants were mainly small colonies. The increase in small colony formation is suggestive of clastogenicity, and a separate assay for chromosome aberrations confirmed the increase (27% cells had chromosome and chromatid breaks or rearrangements).

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Moore et al. 1987)

Test substance : > 99 %

5. Toxicity

Id 79-06-1

Date 7 September 2001

Method : other: DNA Damage and Repair/Unscheduled DNA-Synthesis
Type : Unscheduled DNA synthesis
System of testing :
GLP : No data
Year : 1986
Species and strain : Primaere Rattenhepatozyten
Metabolic activation : Without
Concentrations tested : ≤ 711 ug/ml
Statistical methods :
Test conditions : The effect of acrylamide on unscheduled DNA synthesis using hepatocyte primary culture (HPC)/DNA repair test was examined. An *in vitro* UDS assay was available in which 0-50 mM (0-3.55 mg/ml) acrylamide was incubated with rat hepatocytes for 18 hours. Incorporation of 3HTdR into DNA was determined by autoradiography.

Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Negative

Cytotoxicity (cell death) was observed at the highest concentration. A negative result was obtained in this and in a repeat experiment in that there was not an increase of 5 net nuclear grain counts over negative control values and there was no clear dose-response in the counts observed. The positive control 2-aminofluorene gave a clear increase in UDS.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Miller & McQueen 1986)

Test substance : No data
Method : DNA Damage and Repair/Unscheduled DNA-Synthesis
Type : Unscheduled DNA synthesis
System of testing :
GLP : No data
Year : 1983
Species and strain : SV 40DNA Amplification Chinese Hamster CO60
Metabolic activation : Without
Concentrations tested : 50 -150 ug/ml
Statistical methods :
Test conditions : In an SV40-DNA amplification study CO60 Chinese hamster cells were exposed to 0-150 μ g/ml acrylamide. Cells were incubated with acrylamide for 24 hours before hybridization with 32 P-labelled SV40-DNA.

Cytotoxic concentration :
Genotoxic effects :
Statistical results :
Result : Positive

Increased synthesis of SV40 DNA (DNA amplification) was seen at acrylamide concentrations of >50 μ g/ml, concentrations that were associated with cell survival of 57% or less. The weak SV40 DNA amplification observed might result from a weak DNA damaging activity as biologically illustrated by the capacity of high concentrations of acrylamide to irreversibly inhibit the DNA synthesis rate of CO60 cells. The authors considered this result to indicate that acrylamide had little or no ability to induce SV40-DNA amplification (i.e. little or no potential to directly damage DNA in this system). The true significance of the findings in this invalidated

Conclusion : system is unclear.
Reliability : (2) reliable with restriction, invalidated system
Reference : (Van Horick & Moens 1983)

5.6 GENETIC TOXICITY 'IN VIVO'

Test substance : No data
Method : Chromosome Aberration Test
Type : Cytogenetic assay
GLP : No data
Year : 1989
Species : Mouse
Strain : C57NL/6J
Sex : Male
Route of administration : i.p.
Doses/concentration levels : 50, 100, 125 mg/kg
Exposure period : See test condition
Statistical methods :
Test condition : A) Groups of 4 male mice (8-10wk old) received single ip. injections of 0, 50, 100, or 125 mg/kg acrylamide. Positive controls received cyclophosphamide. Mitoses were analysed from 100 spleen lymphocytes at each exposure level 24 hours post-administration only.
 B) Groups of 4 male mice (8-10 wk old) received single ip. injections of 0, 50, 100, or 125 mg/kg acrylamide. Chromosome and chromatid aberrations were scored in spermatogonia and spermatocytes 24 hours post-administration only.
 Statistical Analysis: A modified Jonckherre-Terpstra analysis was used to determine at each endpoint whether or not acrylamide induced an effect.

Effect on mitotic index or PCE/NCE ration by dose level :
Genotoxic effect :
NOAEL (NOEL) :
LOAEL (LOEL) :
Statistical results :
Result : A) There was no clear increase in the frequency of metaphases with chromosome aberrations but there was a non-statistically significant increase in chromatid aberrations: 5% at 125 mg/kg compared to 2% in negative controls at 125 mg/kg. There was also a significant dose-related increase in the frequency of SCE/cell.
 B) There were no significant changes in the number of chromosome/chromatid aberrations or hyperploidy compared to negative controls.

Conclusion :
Reliability : (2) reliable with restriction, results from study A were not validated and cho ab study is limited by the use of only one sampling time.
Reference : (Backer et al. 1989)
Test substance : No data

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|--|
| Method | : | Chromosome Aberration Test |
| Type | : | Cytogenetic assays |
| GLP | : | Pre-GLP |
| Year | : | 1978 |
| Species | : | Mouse |
| Strain | : | DDY |
| Sex | : | Male |
| Route of administration | : | Oral feed |
| Doses/concentration levels | : | See test condition |
| Exposure period | : | 1 - 3 weeks |
| Statistical methods | : | |
| Test condition | : | <p>A) Groups of 5 male mice (4 weeks old, 25g) received 500 ppm acrylamide (approximately 60 mg/kg/day assuming 25g bodyweight and food consumption of 3g/day) by dietary administration for 1, 2 or 3 weeks, or a single ip. injection of 0 or 100 mg/kg. At least 100 bone marrow cells were scored for chromosome breaks per animal at 1/2, 1, 11 and 12 days post-administration in the case of ip. treated mice, and immediately after sacrifice at weeks 1, 2, and 3 for animals receiving dietary acrylamide (animals were pre-treated with colchicine). There was no mention of the use of positive controls in this study.</p> <p>B) Groups of 5 male mice (4 weeks old, 25g) received 500 ppm acrylamide (approximately 60 mg/kg/day assuming 25g body weight and food consumption of 3g/day) by dietary administration for 1, 2 or 3 weeks or a single ip. injection of 0 or 100 mg/kg. At least 100 metaphases from spermatogonia were scored for chromosome aberrations per animal at 12 and 24 hours, and 11 and 12 days post-administration in the case of ip. Treated mice and immediately after sacrifice for animals receiving dietary acrylamide. In addition, 50-100 spermatocytes in the diakinesis-metaphase / stage were examined from each animal.</p> |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | <p>A) Following single-exposure there was an increase in metaphases with chromosome breaks (2.7% in negative controls, 3.5-7% in treated animals). There was an increase in the frequency of cells with aneuploidy or polyploidy (3.7% in negative controls, 5-10.5% in treated animals). For animals treated by the dietary route there were similar slight increases. In addition, there was also a slight, but not statistically significant increase in SCE/cell (2.9 in controls and 3.5-3.7 in treated animals).</p> <p>B) An increased incidence of spermatogonia with aneuploidy, chromosome breaks, and sister chromatid exchanges was seen using both exposure regimes. Similarly, amongst primary spermatocytes there was a marked increase in sex-chromosome and autosomal univalents, fragments and rearrangements observed in both exposure regimes.</p> |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction; It was not clear from the results that were presented whether or not results at each time point were compared with control groups or if any statistical comparisons had been performed. The author concluded these changes to be negative responses. Based on a |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|--|
| Reference | : | secondary source, there are some doubts about the validity of this assertion due to the limitations in the presentation of information. The values presented for chromosome breaks would, by current standards, indicate a positive result. (Shiraishi, 1978) |
| Test substance | : | 99 % pure |
| Method | : | Chromosome Aberration Test |
| Type | : | Cytogenetic assay/Micronucleus assay |
| GLP | : | No data |
| Year | : | 1988 |
| Species | : | Mouse |
| Strain | : | ICR |
| Sex | : | Male |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 100 mg/kg bw |
| Exposure period | : | One time |
| Statistical methods | : | |
| Test condition | : | A single i.p injection of 100 mg/kg was given to adult ICR mice. Groups of 5 males were used in each treatment/time group. Negative control was used. At 6, 18, 24, and 48 h after injection metaphase analysis was prepared. Fifty mitoses per mouse were analyzed for the incidence of structural chromosome aberrations. Breaks and gaps were distinguished using criteria published by Savage. Statistical analysis of the results was performed by the Fisher exact test. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | The single ip injection elevated the frequency of chromosome aberrations and micronucleated polychromatic erythrocytes in the bone marrow of male mice. A positive relationship between frequency of micronuclei and dose of acrylamide was obtained in the dose range of 2 x 25, 2 x 50, and 2 x 100 mg/kg. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Cihak & Vontorkova, 1988) |
| Test substance | : | No data |
| Method | : | Other: Chromosome Aberration Test/Micronucleus Assay |
| Type | : | Cytogenetic assay |
| GLP | : | No data |
| Year | : | 1988 |
| Species | : | Mouse |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|--|
| Strain | : | C101/E1xC3H/E1)F1 |
| Sex | : | male/female |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0 - 150 mg/kg bw |
| Exposure period | : | |
| Statistical methods | : | |
| Test condition | : | <p>Assay 1: Groups of 5 male and 5 female mice (10-14 weeks old, 25-29g) received single ip. injections of 0 or 100 mg/kg acrylamide in saline. Samples of bone marrow were taken at 12, 18, 24, 30, and 36 hours for analysis of chromosome aberrations.</p> <p>Assay 2: An additional dose-response assay was performed with mice receiving 0, 50, 100, and 150 mg/kg acrylamide with a sampling time of 18 hours. A positive control group received cisplatin.</p> <p>In the micronucleus assay bone marrow samples were taken 18, 24, and 30 h after a dose of 100 mg/kg and the dose response study was performed at the 24 h interval with 4 doses (50-150 mg/kg). Additionally differentiating spermatogonia were scored for chromosomal aberrations in a subset of 5 treated males per dose at the 24 h interval only.</p> <p>Statistically significant differences calculated by the Mann-Whitney-Wilcoxon test. The Cochran Armitage test for linear trend was used on the dose response data in the bone marrow chromosomal assay. The dose-response relationship for the micronucleus test data was estimated by least squares regression.</p> |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | <p>Assay 1: Statistically significant increases in the number of metaphases with chromosome and chromatid breaks were observed (2.6-4.4% excluding gaps compared to 0.7% in controls). The maximal response was at 18 hours in this assay.</p> <p>Assay 2: Statistically significant increases in the frequency of aberrant cells were seen at all exposure levels (2.1%-4.1% excluding gaps compared to 0.3% in the negative control and 3.6% for the positive controls). Mitotic index was reduced by up to 27% compared with negative controls.</p> <p>In the chromosomal bone marrow and the micronucleus assay positive results were obtained with acrylamide, and in the latter test the effect increased linearly with dose. Chromosomal aberrations were not induced in differentiating spermatogonia by the acute acrylamide treatment.</p> |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Adler et al. 1988) |
| Test substance | : | >= 98 % pure |
| Method | : | Sex-Linked Recessive Lethal Test in Drosophila melanogaster |
| Type | : | Drosophila SLRL test |
| GLP | : | No data |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Year : 1988
Species : *Drosophila melanogaster*
Strain : Berlin K
Sex : Male
Route of administration : abdominal injection
Doses/concentration levels : 0, 40 or 50 mM
Exposure period : one time
Statistical methods :
Test condition : 1-2 day old male flies were injected .After treatment were individually mated to virgin females of the Basc for 3 days and subsequently remated twice with fresh virgins for 3 and 4 days duration, to obtain three broods. Following procedures by Wurgler et al, 1977, the F2 generation was scored for the occurrence of sex-linked recessive lethals. For confirmation of putative lethals, F2 cultures containing less than three round eyed males were re-tested.

Effect on mitotic index or PCE/NCE ration by dose level :
Genotoxic effect :
NOAEL (NOEL) :
LOAEL (LOEL) :
Statistical results :
Result : Both test groups showed a small enhancement of the mutation rate in the first broods which did not reach significance given the sample size.

Conclusion :
Reliability : (2) reliable with restriction, small sample size
Reference : (Knaap et al. 1988)

Test substance : > 99 % pure
Method : Mouse Heritable Translocation Assay
Type : Mammalian germ cell cytogenetic assay
GLP : No data
Year : 1987
Species : mouse
Strain : (C3H x 101)F1
Sex : male
Route of administration : i.p.
Doses/concentration levels : 0, 40, or 50 mg/kg bw
Exposure period : 5 days
Statistical methods :
Test condition : Either dose was injected to 120 mice/dose on 5 consecutive days. The lower dose was used in the second experiment to increase the number of progeny by reducing the dominant lethal effect.

Effect on mitotic index or PCE/NCE ration by dose level :
Genotoxic effect :
NOAEL (NOEL) :
LOAEL (LOEL) :
Statistical results :
Result : Mating on days 7-10 following the last injection yielded a high frequency of translocation carriers in the F1 male population, which demonstrated that acrylamide is an effective inducer of tranlocations in postmeiotic germ cells.

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| Conclusion | : | As an inducer of both dominant lethals and heritable translocations in late spermatids and early spermatozoa, acrylamide is similar to alkylating agents such as ethyl-methanesulfonate and thylene oxide. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Shelby et al. 1987) |
| Test substance | : | > 99 % pure |
| Method | : | Rodent Dominant Lethal Test |
| Type | : | Mammalian germ cell cytogenetic assay |
| GLP | : | No data |
| Year | : | 1986 |
| Species | : | Mouse |
| Strain | : | other: (C3H x 101)F1 |
| Sex | : | Male |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0 or 125 mg/kg bw |
| Exposure period | : | one time |
| Statistical methods | : | |
| Test condition | : | Male mice, 12 weeks old, received a single ip. injection of 125 mg/kg (60 animals) or 5 daily injections of 50 mg/kg acrylamide (1) prior to (20 male mice) mating with females. In addition there was an assay performed with mating over a limited period (days 6-10 after acrylamide-treatment) such that the available sperm would derive from cells exposed as late spermatids or epididymal sperm. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | Dominant lethality, observed as an increased frequency of dead implants particularly between days 4-12 post-administration, was noted following single and repeated exposure. The early increase was suggestive of an effect on late spermatids and early spermatozoa. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Shelby et al, 1986) |
| Test substance | : | > 99.9 % pure |
| Method | : | Dominant Lethal Test |
| Type | : | Mammalian germ cell cytogenetic assay |
| GLP | : | No data |
| Year | : | 2000 |
| Species | : | Rat |
| Strain | : | Fischer 344 |
| Sex | : | Male |
| Route of administration | : | Drinking water. |
| Doses/concentration levels | : | 0.5, 2.0 or 5.0 mg/kg bw |
| Exposure period | : | 64 days |
| Statistical methods | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Test condition : Fo males were held for a maximum of one week and exposed to the concentrations of acrylamide in the water for a total of 64 days. The Fo males were then removed from acrylamide exposure for 2 days and cohabited with naive females with normal estrous cyclicity dedicated to the dominant lethal portion of this study. On gestational day 14, each female was euthanized and examined for gross lesions, ovarian corpora lutea, number and distribution of total uterine implantation sites, resorption sites, and live and dead implants. Frequency of dominant lethal factors were calculated.

Effect on mitotic index or PCE/NCE ration by dose level :

Genotoxic effect :

NOAEL (NOEL) :

LOAEL (LOEL) :

Statistical results :

Result : In this assay, total and live implants were reduced, pre and post implantation loss was increased, and the frequency of dominant lethal factors was increased at 5.0 mg/kg/d.

Conclusion :

Reliability : (1) reliable without restriction

Reference : (Tyl et al, 2000a)

Test substance : > 99 % pure

Method : Rodent Dominant Lethal Test

Type : Mammalian germ cell cytogenetic assay

GLP : No data

Year : 1989

Species : Rat

Strain : Long-Evans

Sex : Male

Route of administration : oral gavage

Doses/concentration levels : 30, 45, 60 mg/kg

Exposure period : 5 days

Statistical methods :

Test condition : As part of a dominant lethal assay in which groups of 15 male Long-Evans rats received 0, 5, 15, 30, 45, or 60 mg/kg/day acrylamide by oral gavage for 5 days.

Following this dominant lethal assay, groups of 10 males received up to 45 mg/kg/day by oral gavage for 5 days. Mating behavior and evaluation of sperm were assessed in weeks 1, 2, 3, and 4 using ovariectomised, hormonally primed females which were sacrificed 15 minutes after ejaculation. In addition, further groups of 15 males were mated with proestrus females at 4 one-weekly intervals after acrylamide exposure for examination of ovaries, oviducts, and uteri.

Effect on mitotic index or PCE/NCE ration by dose level :

Genotoxic effect :

NOAEL (NOEL) :

LOAEL (LOEL) :

Statistical results :

Result : There was a marked, statistically significant, reduction in male fertility at 15 mg/kg/day or more. At 15 and 30 mg/kg/day the fertility index (number of

pregnant/number of sperm positive females) was reduced, only in week 1 post-administration, to 46% and 17% respectively. At 45 mg/kg/day, reductions were only significant in weeks 1 and 3 (15% and 67 % respectively), and at 60 mg/kg/day reductions were observed in the first 4 weeks (7% in weeks 1 and 3, 53-60% in weeks 2 and 4).

Copulatory behavior of males (mount and ejaculation latency, number of mounts and intromissions) was unaffected by acrylamide exposure. In week 1 after exposure of males to 15 and 45 mg/kg/day an increased number of females did not have sperm in the uterus (percentages with sperm were 100%, 60%, and 20 % at 0, 15, and 45 mg/kg/day respectively). In subsequent weeks, there were no clear effects on uterine sperm.

Sperm samples from males receiving 45 mg/kg/day were not located for analysis in week 1 - the reason for this was not clear. Examinations were further limited by the use of only two exposure levels, 0 and 45 mg/kg/day. The only statistically significant effects on sperm were observed in week 3 post-administration. There were slight reductions in sperm count (61×10^6 at 45 mg/kg/day compared with 82×10^6 in controls), percentage motility (58% at 45 mg/kg/day, 75% in controls), curvilinear velocity (122 $\mu\text{m/s}$ versus 132 $\mu\text{m/s}$), linearity and straight line velocity. These changes, in themselves, are of limited significance, but support the observation of reduced male fertility in the dominant lethal assay. In addition, there was a statistically significant reduction in the percentage of ova fertilised by males exposed to 15 and 45 mg/kg/day in week 1 (29% and 41% compared to 84% in controls), and at 45 mg/kg/day in week 3 (12% compared to 65% in control). Overall, this study demonstrated reduced fertility in male rats in an exposure regime, which did not apparently affect mating performance through neurotoxicity; male fertility was impaired at exposure levels of 15 mg/kg/day or more for 5 days.

| | |
|--|---|
| Conclusion | : |
| Reliability | : |
| Reference | : |
| Test substance | : |
| Method | : |
| Type | : |
| GLP | : |
| Year | : |
| Species | : |
| Strain | : |
| Sex | : |
| Route of administration | : |
| Doses/concentration levels | : |
| Exposure period | : |
| Statistical methods | : |
| Test condition | : |
| Effect on mitotic index or PCE/NCE ration by dose level | : |
| Genotoxic effect | : |
| NOAEL (NOEL) | : |

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | No increase in structural aberrations was observed on completion of 80 days although a slight increase in reciprocal translocations was noted amongst treated animals (0, 1, 1, and 2 in each of the groups respectively). The significant increase in pre-implantation loss would suggest that an adequate exposure level was used. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Smith et al., 1986) |
| Test substance | : | 99.9 % pure |
| Method | : | Unscheduled DNA-Synthesis |
| Type | : | Mammalian germ cell cytogenetic assay |
| GLP | : | No data |
| Year | : | 1990 |
| Species | : | Mouse |
| Strain | : | (C3H x BL10)F1 |
| Sex | : | Male |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 8, 16, 31, 63, or 125 mg/kg bw |
| Exposure period | : | one time |
| Statistical methods | : | |
| Test condition | : | Groups of 4-6 male mice (12-18 wks old) received single ip. injections of 0, 8, 16, 31, 63, or 125 mg/kg acrylamide. Tritiated thymidine was injected into the testes 0-48 hours after acrylamide administration and sperm from the caudal epididymides were recovered 16 days post-administration for UDS analysis. In addition, groups of 4-6 male mice received a single ip. injection of 0 or 125 mg/kg acrylamide with tritiated thymidine injected into the testes 6 hours later and sperm from caudal epididymides (spermatozoal to early spermatocytes at the time of treatment) was recovered at 2-3 day intervals for up to 30 days post-administration for UDS analysis. Also, groups of 4 male mice received ip. injections of 46 mg/kg [¹⁴ C]-acrylamide. DNA was extracted from liver and testes samples 1-24 hours post-administration and analysed for radioactivity. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | In the first experiment, there was a clear increase in UDS, the maximum response of one order of magnitude greater than that of controls occurring 6 hours after tritiated thymidine injection. This peak response related to sperm, which would have been in the early spermatid stage at the time of acrylamide exposure. For the second experiment, no significant amounts of tritiated thymidine were incorporated during the first 10 days following exposure to 125 mg/kg acrylamide but from days 12-27 a positive UDS response was seen. |

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| | In the third experiment DNA alkylation was observed, which reached maximum levels 4-6 hours post-administration in the testes and 1-2 hours post-administration in the liver, with levels being substantially (10-fold) lower in testes than liver. |
| Conclusion | : |
| Reliability | : (1) reliable without restriction |
| Reference | : (Sega et al. 1990) |
| Test substance | : \geq 98 % pure |
| Method | : Micronucleus Test |
| Type | : Micronucleus assay |
| GLP | : No data |
| Year | : 1988 |
| Species | : Mouse |
| Strain | : Swiss-NIH |
| Sex | : Male/female |
| Route of administration | : i.p. |
| Doses/concentration levels | : 136 mg/kg bw with positive and negative controls |
| Exposure period | : one time |
| Statistical methods | : |
| Test condition | : Groups of 5 males/5females (7 week old mice (25g)) were injected with 136 mg/kg. DMBA was used as a positive control at 30 mg/kg. Mice were killed at 24, 48 and 72 hr intervals and 2000 PCE's were scored for micronuclei. The criterion used for the minimum size of a micronucleus was 1/10 of the cell diameter. The ratio of polychromatic versus normochromatic was used to assess the cytotoxicity of the acrylamide. A chi square test was used to determine the stat. sig. between groups. |
| Effect on mitotic index or PCE/NCE ration by dose level | : |
| Genotoxic effect | : |
| NOAEL (NOEL) | : |
| LOAEL (LOEL) | : |
| Statistical results | : |
| Result | : There was 2-2.5 fold increase in micronuclei. In this micronucleus assay in male and female mice stat. sig. positive results were seen using cells taken from bone marrow at 136 mg/kg aqueous acrylamide administered by the intraperitoneal route singly. |
| Conclusion | : |
| Reliability | : (1) reliable without restriction |
| Reference | : (Knaap et al. 1988) |
| Test substance | : No data |
| Method | : Mouse Spot Test/Teratogenicity |
| Type | : Somatic mutation assay |
| GLP | : No data |
| Year | : 1989 |
| Species | : Mouse |
| Strain | : T and HT stock |
| Sex | : Female |
| Route of administration | : i.p. |
| Doses/concentration levels | : 0, 50, 75 mg/kg |
| Exposure period | : Single or 3x |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|--|
| Statistical methods | : | |
| Test condition | : | As part of a study investigating potential mutagenic and developmental effects, groups of 31-93 pregnant female mice (10-14 wks old) received single or 3 daily ip. injections of 0, 50 or 75 mg/kg aqueous acrylamide on day 12 or days 10, 11, and 12. Approximately 220-300 offspring per dose level were available for examination. Spot test according to Neuhauser-Klaus and Chauhan, 1987. Compared by Fischers exact test or Pearson's chi square test. Teratogenicity: Fetuses were examined degenerative changes. Number of implantation sites, resorptions, and living fetuses were counted in each litter from each pregnant female. The frequency of fetuses w. histological abnormalities was analysed using the chi square test. The significance of the difference between mean fetal weights of each group was determined using the t test. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | A positive result (doubling in the number of spots of genetic relevance compared to negative controls) was reported following single exposure to either 50 or 75 mg/kg and to repeated exposure to 50 mg/kg/day. Repeated exposure to 75 mg/kg resulted in increased embryotoxicity and cytotoxicity. |
| Conclusion | : | |
| Reliability | : | (1) Reliable without restriction |
| Reference | : | (Neuhauser-Klaus & Schmahl, 1989) |
| Test substance | : | No data |
| Method | : | Positive Selective Assay |
| Type | : | other: Transgenic Mouse Assay |
| GLP | : | No |
| Year | : | 1997 |
| Species | : | Mouse |
| Strain | : | other: Muta™ Mouse |
| Sex | : | no data |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0, 50 or 100 mg/kg bw |
| Exposure period | : | Single dose |
| Statistical methods | : | |
| Test condition | : | Groups (number varies) of mice were given a single injection of 50 or 100 kg/bw and tissues were prepared 3, 10, or 100 days post treatment. <u>Statistical Analysis</u> of mutation frequency was performed with the Fisher's Exact Test. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | Negative |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | |
|--|---|
| | <p>There was a slight increase due to treatment of the observed mutation frequencies in the acrylamide liver group for all three assay times, slightly more pronounced in 50 mg//kg group. From the day 3 group to the day 100 group a time dependent decrease in all the absolute mutant frequencies was detectable but not significantly different from control. No meaningful results from germ assay.</p> |
| Conclusion | : Test performed to determine activity of acrylamide in liver and germ cells. |
| Reliability | : (2) reliable with restriction, TMA is inappropriate to assess mutagenic activity of germ cells. |
| Reference | : (Krebs and Favor, 1997) |
| Test substance | : No data |
| Method | : other: Detection of germ-line gene conversion events |
| Type | : other: Transgenic Mouse Assay |
| GLP | : No data |
| Year | : 1994 |
| Species | : mouse |
| Strain | : other: COR3 and OPP2 |
| Sex | : male |
| Route of administration | : i.p. |
| Doses/concentration levels | : 0, 50 mg/kg |
| Exposure period | : 5 days |
| Statistical methods | : |
| Test condition | : 60-90 day old mice were treated with 5 daily injections and with assays performed on testicular cell preparations examining the <i>LacZ</i> mutation system 21-23 days after last administration. Spermatogenic cells were prepared according to Romrell et al 1976. PCR of sperm DNA was performed according to Cui et al 1989. Conversion events were visualized by histochemical staining or flow cytometric analysis of transgenic spermatids. |
| Effect on mitotic index or PCE/NCE ration by dose level | : |
| Genotoxic effect | : |
| NOAEL (NOEL) | : |
| LOAEL (LOEL) | : |
| Statistical results | : |
| Result | : Negative |
| | <p>Produced no marked increase in converted spermatids. Microscopic examination showed an increase in the number of "unusually large cells" which were speculated, but not proven by the authors, to be due to interkinetic delay during meiosis caused by acrylamide. May be due to complicating factor of possible meiotic arrest being induced by the high levels of test substance administered. Or the system is not sufficiently sensitive to detect marginal genotoxins.</p> |
| Conclusion | : Utilize a transgenic mouse mutation system for efficient detection of germ line gene conversion events as a mutagenic screening tool. Test is a viable option for inexpensive and rapid whole animal mutagen testing. Overall, no firm conclusions can be drawn on the nature and significance of effects seen. |
| Reliability | : (1) reliable without restriction |
| Reference | : (Murti et al., 1994) |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|---|
| Test substance | : | No data |
| Method | : | |
| Type | : | other: Heritable translocation test |
| GLP | : | No data |
| Year | : | 1994 |
| Species | : | mouse |
| Strain | : | C3H/EI |
| Sex | : | male/female |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0, 50 and 100 mg/kg (males only) |
| Exposure period | : | Single/repeated dose |
| Statistical methods | : | |
| Test condition | : | 10-12 wk old male mice (25-28g) were treated with single dose (50 animals/dose) or with 5 daily injections (50 mg/kg) and mated 7-16 days after treatment to untreated female 102/EI mice. Translocations carriers among the F1 progeny were selected by a sequential procedure of fertility testing and cytogenetic analysis including G-band karyotyping to determine the chromosomes involved in the respective translocations. Statistical Analysis: Student's t test was used to compare litter sizes. Fishers Exact Test was used to detect differences in translocation frequencies. Dose response tested with Cochran-Armitage test for trend in binomial proportions. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | Dominant lethal effects were observed at the highest exposure level and there was an exposure-related increase in heritable translocations; 3/8700 (0.04%) in controls, 2/362 (0.6%) at 50 mg/kg, 10/367 (2.7%) at 100 mg/kg, 23/105 (22%) at 5x50 mg/kg. The results of these two studies demonstrate that acrylamide caused heritable translocations in mice. |
| Conclusion | : | If late spermatids of mouse and man are equally sensitive to translocation induction by Acrylamide, and assuming a spontaneous translocation frequency of 1/1000 newborns for humans, the doubling dose for translocation induction by Acrylamide in human spermatids would be about 25 mg/kg of acrylamide. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Adler et al., 1994) |
| Test substance | : | No data |
| Method | : | Chromosomal aberration |
| Type | : | Cytogenetic assay |
| GLP | : | No data |
| Year | : | 1998 |
| Species | : | Mouse |
| Strain | : | 101/E1 – C3H/E1 |
| Sex | : | Male/female |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0, 100 mg/kg |
| Exposure period | : | Single dose |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|--|---|---|
| Statistical methods | : | |
| Test condition | : | Study A: Groups of 5 male and 5 female mice received single ip. injections of 0 or 100 mg/kg acrylamide in saline. Samples of bone marrow were taken at 12, 18, 24, 30, and 36 hours for analysis of chromosome aberrations. Study B: An additional dose-response assay was performed with mice receiving 0, 50, 100, and 150 mg/kg acrylamide with a sampling time of 18 hours. A positive control group received cisplatin. Statistical Analysis: differences between individual treatment and control groups was calculated by Mann-Whitney-Wilcoxon test. The Cochran-Armitage test for linear trend was used on the dose response data in the bone marrow chromosomal assay. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | Study A: Statistically significant increases in the number of metaphases with chromosome and chromatid breaks were observed (2.6-4.4% excluding gaps compared to 0.7% in controls). The maximal response was at 18 hours in this assay. Study B: Statistically significant increases in the frequency of aberrant cells were seen at all exposure levels (2.1%-4.1% excluding gaps compared to 0.3% in the negative control and 3.6% for the positive controls). Mitotic index was reduced by up to 27% compared with negative controls. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restrictions |
| Referenc | : | (Adler et al., 1998) |
| Test substance | : | Analytical grade |
| Method | : | |
| Type | : | other: Transgenic Mouse Assay |
| GLP | : | No data |
| Year | : | 1993 |
| Species | : | Mouse |
| Strain | : | other: Muta™ Mouse |
| Sex | : | Male |
| Route of administration | : | i.p. |
| Doses/concentration levels | : | 0, 50 mg/kg/day |
| Exposure period | : | 5 days |
| Statistical methods | : | |
| Test condition | : | 8-10 wk old mice were given 5 daily injections. Animals were killed at 3, 7, and 10 days after the last dose and the LacZ mutation system was used to determine mutation frequency. Bone marrow tissue was examined for mutation. |
| Effect on mitotic index or PCE/NCE ration by dose level | : | |
| Genotoxic effect | : | |
| NOAEL (NOEL) | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|----------------------------|---|---|
| LOAEL (LOEL) | : | |
| Statistical results | : | |
| Result | : | Small increases in mutant frequency with no dependence on harvesting times showed mutation in bone marrow cells (An increase in mutation frequency was noted ($62-89 \times 10^6$ compared to $15-26 \times 10^6$ in controls), although with no clear pattern with respect to sampling time). |
| Conclusion | : | A transgenic mouse strain was evaluated for its effectiveness in detecting <i>lacZ</i> - mutations in selected tissues in this new test method. The results of this demonstrate the applicability of this transgenic mouse as an effective model to detect and analyze gene mutation in selected organs including germinal tissues. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Hoorn et al., 1993) |

5.7 CARCINOGENICITY

| | | |
|-------------------------------|---|---|
| Species | : | mouse |
| Sex | : | female |
| Strain | : | Sencar |
| Route of admin. | : | Gastric intubation, i.p. injection, dermal |
| Exposure period | : | 2 weeks |
| Frequency of treatment | : | 6 applications |
| Post. obs. period | : | |
| Doses | : | 12.5, 25.0, 50.0 mg/kg |
| Control group | : | yes, concurrent vehicle |
| Method | : | Skin Initiation-Promotion Assay |
| Year | : | 1984 |
| GLP | : | No data |
| Test substance | : | 99% pure |
| Test condition | : | 40 mice/dose (6-8 weeks old) were dosed by 3 separate routes. Two weeks following these tumor initiating doses, a tumor promotion regimen was begin where 1.0 ug TPA in 0.2 ml acetone was applied to the shaved back of each animal 3x/week for 20 weeks. Control groups (20.group) for promotion rec'd 0.2 ml acetone at the same frequency and duration of treatment. Tumor incidences were charted from weekly observation. All animals were sacrificed at 52 weeks in the study and histopathological evaluations were done on all gross lesions. Dose response was statistically analyzed by Cox regression analysis. |
| Result | : | An acrylamide dose-related increase in tumor formation was noted for all routes of acrylamide administration when TPA was administered subsequently but there was no increase in tumor incidence in mice treated with acrylamide but not subsequently with TPA. These results suggest that acrylamide was "initiating" tumor formation. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Bull et al., 1984) |
| Species | : | Rat |
| Sex | : | Male/female |
| Strain | : | Fischer 344 |
| Route of admin. | : | Drinking water |
| Exposure period | : | 2 years |
| Frequency of treatment | : | Daily |
| Post. obs. period | : | No |
| Doses | : | 0.01, 0.1, 0.5, 2.0 mg/kg/day |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Control group : Yes, concurrent vehicle
Method : Other: Carcinogenicity Study
Year : 1986
GLP : No data
Test substance : 96-99% pure
Test condition : 90 rats/sex/dose, approx 5 wks of age were evaluated for mortality, clinical signs, body weight, food and water consumption, clinical chemistry, hematology, urinalysis, gross pathology, organ weights, and histopathology. 10 rats/sex/dose were scheduled for interim sacrifice at 6, 12 or 18 months on study. Gross necropsy included the adrenal glands, auditory gland, aorta, bone, bone marrow, brain, cecum, cervix, coagulating glands, epididymides, esophagus, eyes, heart, liver, kidneys, large intestine, lacrimal glands, larynx, lymph node (mediastinal and mesenteric), lungs, mammary gland, ovaries, oviducts, pancreas, pituitary, prostate, salivary glands, seminal vesicles, skin, small intestine, spinal cord, spleen, stomach, testes, thymus, thyroid gland, tongue, trachea, urinary bladder, uterus, vagina, and gross lesions.

Results

Statistical Analysis: Body weights by ANOVA, mortality by Wilcoxon test, and histopathological observations by Fisher's Exact Probability Test.

A statistically significant increase in mortality was noted from 21 months onwards amongst males and females receiving 2 mg/kg/day. A slight decrease in bodyweight (up to 4%) was noted amongst males at 2 mg/kg/day and there were no significant effects on food and water consumption and no clinical signs of toxicity. There were no significant adverse effects on haematology, blood biochemistry, or urinalysis examinations, organ weights, or macroscopic pathology at 6, 12 and 18 months. However, at 24 months, there was an increase in the number of subcutaneous and mammary gland masses amongst females at 2 mg/kg/day. Histopathologically, there were no abnormalities at 6 months. At examinations performed from 12 months onwards there was an increase in the incidence and severity of tibial nerve degeneration amongst males at 2 mg/kg/day and from 18 months onwards in females at 2 mg/kg/day (focal swelling of nerve fibres with fragmentation of myelin and axon, and the formation of vacuoles containing small round eosinophilic globules and macrophages). There were no clear changes amongst animals at lower exposure levels or amongst other peripheral nerve samples (saphenous branch of the femoral nerve and brachial plexus).

In males, there was a statistically significantly increased incidence of benign follicular cell adenomas of the thyroid at the highest dose level (1/60, 0/58, 2/59, 1/59, 7/59). In females there was a non-significant increase in the incidence of benign follicular cell adenomas of the thyroid (0/58, 0/59, 1/59, 1/58, 3/60) and malignant adenocarcinomas (1/58, 0/59, 0/59, 0/58, 3/60). In females there was a statistically significant increase in the incidence of malignant adenocarcinomas in the uterus (1/60, 2/60, 1/60, 0/59, 5/60, or 1.7%, 3.3%, 1.7%, 0, 8.3%). The historical control range was stated to be 0-2.3%. In males there was a statistically significant increase in the incidence of malignant testicular mesothelioma at 0.5 and 2 mg/kg/day (3/60, 0/60, 7/60, 11/60, 10/60 or 5%, 0, 12%, 18%, 17%). The historical control incidence was 3.1% with a range of 2-6%.

In males there was a non-significant increase in the incidence of malignant astrocytomas in the spinal cord (1/60, 0/60, 0/60, 0/60, 3/60). There were also non-significant increases in malignant astrocytomas in the brain of females (0/60, 1/60, 0/60, 0/60, 3/60), glial proliferation in the brain suggestive of an early tumour (0/60, 0/60, 0/60, 1/60, 3/60), and malignant

astrocytomas in the spinal cord (1/60, 0/59, 0/60, 0/60, 3/61). In addition, malignant astrocytomas were also observed in the brain (3/60, 0/60, 0/60, 2/60, 2/60), and glial proliferation (suggestive of an early tumour) in 0/60, 0/60, 0/60, 1/60, 1/60. The effects in astrocytomas for brain and spinal cord in males and females do not show any clear dose-response but there are some concerns as these tumours are occurring in potential target tissues and, according to the authors, the concurrent control values may have been abnormally high so trends would not have been clear. Also, the group sizes used in this study may not have been sufficiently large enough to detect clear increases. Overall, because of these limitations, the toxicological significance of the presence of these astrocytomas in this study is unclear.

For females, there was a statistically significant increase in the incidence of benign papillomas in the oral cavity at 2 mg/kg/day (0/60, 3/60, 2/60, 1/60, 7/61) and a non-significant increase in focal hyperplasia (1/60, 2/60, 1/60, 0/60, 4/61). The incidence of malignant carcinomas did not show any clear dose-response (0/60, 0/60, 0/60, 2/60, 1/61). For males, the incidence of tumor formation in the oral cavity did not show any clear exposure relationship (carcinomas 2/60, 0/60, 1/60, 0/60, 2/60, and papillomas 4/60, 7/60, 0/60, 5/60, 4/60) although there was a statistically significant increase in focal hyperplasia of the hard palate (0/60, 1/60, 1/60, 1/60, 4/60, 5/60). Again, although effects are not clear, there are some concerns as there is a possibility that hyperplasia and subsequent, but unclear, tumor formation may have arisen as a result of local effects due to the route of exposure employed.

In females there were increases in benign and malignant tumors of mammary glands (10/60, 11/60, 9/60, 19/58, 23/61 and 2/60, 1/60, 1/60, 2/58, 6/61 respectively or 17%, 18%, 15%, 33%, 38% and 3%, 2%, 2%, 3%, 10%), benign pituitary gland adenomas (25/59, 30/60, 32/60, 27/60, 32/60 or 42%, 50%, 53%, 45%, 53%), and benign tumors of the clitoral gland (0/2, 1/3, 3/4, 2/4, 5/5). In males there were increased incidences of benign tumors in the adrenal glands (pheochromocytoma) (3/60, 7/59, 7/60, 5/60, 10/60 or 5%, 12%, 12%, 8%, 17%). The increased incidences of mammary tumors, benign pituitary adenomas and adrenal pheochromocytomas are of doubtful toxicological significance due to the poor dose-response and high historical control incidence (18% for benign mammary tumors, 2% for malignant mammary tumors - NTP data only, 28-47% for pituitary adenomas, 1-14% for pheochromocytomas). For clitoral adenomas the total number of tissues examined was too small to draw any firm conclusions.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Johnson et al., 1986)

Species : Mouse
Sex : Male/female
Strain : A/J
Route of admin. : p.o./i.p
Exposure period : 8 weeks
Frequency of treatment : 3 times/week
Post. obs. period :
Doses : Study performed at two locations (EPA and Medical College of Ohio):
 EPA: 6.25, 12.5, 25.0 mg/kg/day
 Med Coll Ohio: 1, 3, 10, 30, 60 mg/kg/day

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|-----------------------|---|--|
| Control group | : | yes, concurrent vehicle and positive control |
| Method | : | Lung Adenoma Bioassay |
| Year | : | 1984 |
| GLP | : | No data |
| Test substance | : | 99% pure |
| Test condition | : | EPA Lab: 40 male/40female mice (8 weeks old) received p.o. doses 3x/wk for 8 weeks. Control group received distilled water at the same frequency/duration. MCO lab: 16 male/16 female mice (8 weeks old) received i.p. doses 3x/wk for 8 weeks. For comparison with the EPA study a 30 mg/kg p.o. group was included. Two baseline control groups were included: an untreated group and a vehicle control group that rec'd i.p. injections of distilled water. Positive control groups rec'd a single i.p. injection of ethyl carbamate at dose levels of 500 and 1000 mg/kg. These animals were sacrificed after 8-9 months. Tumor incidence was statistically analysed using a logit regression model analysis. |
| Result | : | In the adenoma bioassay, there was an stat. significant exposure-related increase in the formation of lung tumors in both sexes via both routes of administration. Acrylamide was 1/7 th as potent as ethyl carbamate in the induction of lung adenomas. These data confirm the hypothesis that acrylamide possesses carcinogenic properties similar to EC. |
| Conclusion | : | The enhancement of benign lung tumor incidence by acrylamide in a mouse strain showing a high background incidence of such tumors is of doubtful significance in relation to human health. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Bull et al., 1984) |

5.8 REPRODUCTION TOXICITY

| | | |
|---|---|---|
| Test substance | : | No data |
| Method | : | Reproductive Toxicity |
| Type | : | Fertility |
| GLP | : | No data |
| Year | : | 1986 |
| Species | : | Rat |
| Strain | : | Long-Evans |
| Route of administration | : | drinking water |
| Doses | : | Male: 4, 8, 10 mg/kg/day Female: 5, 10, 15 mg/kg/d |
| Sex | : | Male/female |
| Control group | : | yes, concurrent vehicle |
| Frequency of treatment | : | Daily |
| Duration of test | : | 11 weeks |
| Exposure period | : | 10 weeks |
| Premating exposure period – Male | : | 10 weeks |
| Premating exposure period – Female | : | 2 weeks |
| Statistical methods | : | |
| Test condition | : | In a one-generation reproduction study, groups of 15 male Long-Evans rats received 0, 50, 100, or 200 ppm acrylamide in drinking water for up to 10 weeks, and females received 0, 25, 50, and 100 ppm for 2 weeks prior to mating, during gestation, and also during lactation. <For males and |

females, the mean exposure levels, based on bodyweight and water consumption values presented were approximately 0, 4, 8 and 10 mg/kg/day and 0, 5, 10, and 15 mg/kg/day respectively, although for pregnant and lactating females values would have been somewhat higher.>

During the 10-week exposure period copulatory activity and sperm parameters were assessed in males. In week 10 males from the control and high exposure groups were mated with untreated females. Males were subsequently sacrificed and their organs were examined macroscopically, with histopathological examination of testes and counts made of spermatids and epididymal sperm. The untreated females were sacrificed on day 17 of pregnancy and the number of foetuses and implantation sites were recorded.

To assess female reproductive performance, in the third week of treatment acrylamide-exposed females were paired with untreated males for up to 7 days. Females were allowed to deliver and pups were eventually sacrificed on day 42.

Statistical Analysis: ANOVA was used to analyze body weight, fluid consumption, copulatory behaviors, dam/litter body weight and ejaculated semen parameters. Square root transformations were applied to the frequency data for the semen parameters, while temporal indices were subjected to logarithmic transformation. Ratios for fertility and postimplantation loss were contrasted using chi-square analysis. Linear regression techniques were applied to delineate the contribution of body weight, cumulative acrylamide uptake, and fluid consumption and the respective interactions on litter birth and weaning weights. ANOVA was applied to variables of litter size, survival rate, and day of vaginal patency.

| | | |
|------------------------------|---|--|
| NOAEL Parental | : | 5 mg/kg bw |
| Actual dose/level/sex | : | |
| Parental and F1 data | : | |
| Offspring toxicity | : | |
| Statistical results | : | |
| Result | : | Signs of toxicity amongst males receiving 200 ppm (approximately 10 mg/kg/day) were so severe (including one mortality, loss of use of hind limbs, marked bodyweight loss, and reduced water consumption) that these were sacrificed for humane reasons after about 5-6 weeks of exposure. Mortality was not observed amongst other groups although loss of use of hind limbs was observed from week 8 amongst males at 100 ppm (approximately 8 mg/kg/day). |

In the copulatory activity assessment, a marked decrease in the number of intromissions was seen during week 6 for males at 200 ppm, which was presumably related to their poor clinical condition. The mount latency was apparently not affected by acrylamide, although in their final week only 4/12 males at 200 ppm and 11/15 at 100 ppm managed to ejaculate within a 30 minute allotted time span.

Evaluation of ejaculated semen was conducted in week 9 amongst males receiving 0, 50, or 100 ppm. There was a statistically significant reduction in sperm count at 100 ppm (67% reduction). Sperm motility and morphology could not be adequately assessed at 100 ppm because semen was recovered from the uterus of only one female despite the fact that 11/15 males at 100 ppm ejaculated. For the other females, semen was found only in the vagina. Sperm count, motility and morphology, and seminal plug weight were not affected for males in other groups. Only 33% of females that were paired with males receiving 100 ppm were pregnant compared to

79% impregnation of females mated with control males. There was also a statistically significant increase in the incidence of post-implantation loss amongst females impregnated by males receiving 100 ppm (8% in controls, 32% at 100 ppm). This parameter was not assessed for males that received 50 or 200 ppm.

For males at 200 ppm that were sacrificed after 6 weeks there were apparently no significant effects on organ weight, or sperm parameters. At the terminal kill for the other groups (after 11 weeks) there were no significant changes in organ weights (liver, brain, kidney, adrenals, spleen, heart, testes, prostate, vas deferens, epididymides) or sperm counts. Overall, copulatory ability (reduced intromission and impaired ejaculation) was altered at 100 and 200 ppm although it is likely to have been influenced by the impaired hind limb function.

There were no mortalities amongst females, although there was a loss of use of hind limbs during weeks 1 and 2 of the gestation period for females receiving 100 ppm. There was also a significant reduction in bodyweight (approximately a 10% reduction compared to control) at this exposure level at the end of the second week prior to gestation, which remained throughout the rest of the study. At 50 ppm bodyweight gain was reduced (approximately a 10% reduction) throughout lactation.

Reduced birth weight and subsequently reduced bodyweight gain was noted amongst male and female pups born to females that received 50 or 100 ppm acrylamide. The reduced birth weight of pups was indicative of a slight retardation in pup development which was also suggested by a delay in vaginal opening (about 36 days at 100 ppm, about 33 days in all other groups). As there was no cross-fostering in this study it was difficult to determine whether or not the subsequent effects on pup weight were as a result of direct exposure to acrylamide *in utero*, from maternal milk or indirectly related to poor milk production and impaired nursing ability of acrylamide-affected mothers.

| | |
|-----------------------|---|
| Conclusion | : Overall, this study indicates that male copulatory activity was impaired at 100 ppm (approximately 8 mg/kg/day) or more. However, in this study, it is likely that impaired mating ability was been secondary to neurotoxic effects (such as hind limb splaying). There was a marked reduction in sperm count at 100 ppm although sperm motility and morphology could not be adequately assessed. This effect does not provide conclusive evidence but suggests that male fertility might be impaired. In addition, reduced birth weights and delayed vaginal opening were suggestive of retarded development of offspring. The increased incidence of post-implantation loss may have been connected with dominant-lethal mutations, which is in agreement with studies reported in section 4.1.2.7 and with other developmental studies reported later in this section. There was evidence of generalised toxicity at 50 and 100 ppm, seen as impaired bodyweight gain during pregnancy and lactation. The retardation in pup development may be a secondary effect related to maternal toxicity. |
| Reliability | : (1) Reliable without restriction |
| Reference | : (Zenick et al., 1986) |
| Test substance | : > 95% pure |
| Method | : Fertility Test |
| Type | : Fertility |
| GLP | : Pre-GLP |
| Year | : 1981 |
| Species | : Mouse |

5. Toxicity

Id 79-06-1

Date 7 September 2001

Strain : Ddy
Route of administration : gavage
Doses : 35.5 mg/kg/day
Sex : Male
Control group : yes
Frequency of treatment : 2/week
Duration of test : 8 weeks
Premating exposure period :
Test condition : Groups of 5-7 male mice (5-6 wks of age, avg 29g) received 0 or approximately 36 mg/kg acrylamide (>95% purity) in saline by oral gavage twice weekly for 8 weeks. Testicular weight was taken at necropsy. Student's t test used for comparisons.
NOAEL Parental :
Actual dose/level/sex :
Parental and F1 data :
Offspring toxicity :
Statistical results :
Result : Relative testicular weight was reduced (83% of control value).

Light microscopy of the testes showed "degeneration of epithelia in spermatids and spermatocytes" (presumably meaning that a reduced number of spermatids and spermatocytes were observed in the epithelium when compared with controls), reduction in spermatozoa, the presence of multinucleate giant cells. Sertoli cells and interstitial cells were apparently unaffected. In addition, the epididymides were apparently unaffected. No further histopathological investigations were performed.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Hashimoto et al 1981)

Test substance : > 95% pure
Method : Reproductive toxicity
Type : Fertility
GLP : No data
Year : 1986
Species : Mouse
Strain : other: ddY
Route of administration : drinking water
Doses : 21, 43, 64, or 85 µg/ml
Sex : male/female
Control group : Yes
Frequency of treatment : Daily
Duration of test : 4 weeks
Premating exposure period - Male :
Premating exposure period - Female :
Statistical methods :
Test condition : Groups of 9-24 male and female ddY mice received 0, 21, 43, 64, or 85 µg/ml acrylamide in drinking water for 4 weeks. Assuming a bodyweight of 35g and 5 ml/day water consumption this would result in dose levels of about 0, 3, 6, 9, and 12 mg/kg/day. Water and food consumption were

5. Toxicity

Id 79-06-1

Date 7 September 2001

measured weekly. On completion of the dosing period, half of the treated males and all of the females were mated with untreated controls of the opposite sex. Uterine contents were examined on day 13 of gestation for implants and resorptions except for half of the females at the highest exposure level, which were allowed to complete their gestation period and deliver pups which were examined for a further 4 weeks for any abnormalities. Immediately after the 4-week exposure period, the males not being used for mating were killed and organ weights were determined (liver, testes and seminal vesicles) with further examinations for sperm count and sperm cell morphology.

NOAEL Parental :
Actual dose/level/sex :
Parental and F1 data :
Offspring toxicity :
Statistical results :
Result : The highest exposure level (about 12 mg/kg/day) was associated with loss of use of hind limbs amongst treated males and females on completion of 4 weeks exposure. Bodyweight, food and water consumption was unaffected.

The fertility rate, assessed on day 13 and also on the day of delivery was clearly affected only at the highest exposure level when treated males were mated with control females (2/9 pregnant compared with 8/9 in controls on day 13, and 3/15 versus 12/15 on the day of delivery). In addition, there were statistically significant reductions in the number of foetuses per dam at the highest exposure level and also at the next highest treatment level (~9 mg/kg/day).

At week 4, there was a slight, but not statistically significant, reduction in testes weight of males receiving approximately 12 mg/kg/day (about a 10% reduction). Also, epididymal sperm count was significantly reduced from males receiving the highest exposure of acrylamide (23×10^5 /mg epididymis compared to 36×10^5 /mg in controls) and there was an increase in head and tail abnormalities in sperm (8% versus 4%). Overall, this study showed clear effects on epididymal sperm count following exposure of males to approximately 12 mg/kg/day for 4 weeks and reduced fertility after 2 or 4 weeks. However, the effects on fertility were observed at a level that was associated with impaired hind limb function. It is unclear from this study whether or not the impaired fertility was secondary to neurotoxicity.

Conclusion :
Reliability : (1) Reliable without restriction
Reference : (Sakamoto and Hashimoto, 1986)

Test substance : > 99% pure
Method :
Type : Fertility
GLP : No data
Year : 1989
Species : Rat
Strain : Long Evans hooded
Route of administration : Gavage
Doses : 5, 15, 30, 45, or 60 mg/kg/day
Sex : Male/female
Control group : Yes
Frequency of treatment : Daily
Duration of test : 3 weeks

5. Toxicity

Id 79-06-1

Date 7 September 2001

Premating exposure

period – Male

: 5 days

Premating exposure

period – Female

: 5 days

Test condition

: Groups of 15 male Long-Evans rats received 0, 5, 15, 30, 45, or 60 mg/kg/day acrylamide by oral gavage for 5 days.

Groups of 10 males received up to 45 mg/kg/day acrylamide by oral gavage for 5 days. Mating behavior and evaluation of sperm were assessed in weeks 1, 2, 3, and 4 using ovariectomised, hormonally primed females which were sacrificed 15 minutes after ejaculation. In addition, further groups of 15 males were mated with proestrus females at 4 one-weekly intervals after acrylamide exposure for examination of ovaries, oviducts, and uteri.

Statistical Analysis: Chi square statistics were used to analyze fertility rates. Pre- and post implantation loss and fertilization rates were analyzed by ANOVA. Sperm measures were evaluated by the Mann-Whitney U-Test.

NOAEL Parental

:

Actual dose/level/sex

:

Parental and F1 data

:

Offspring toxicity

:

Statistical results

:

Result

: There was a marked, statistically significant, reduction in male fertility at 15 mg/kg/day or more. At 15 and 30 mg/kg/day the fertility index (number of pregnant/number of sperm positive females) was reduced, only in week 1 post-administration, to 46% and 17% respectively. At 45 mg/kg/day, reductions were only significant in weeks 1 and 3 (15% and 67 % respectively), and at 60 mg/kg/day reductions were observed in the first 4 weeks (7% in weeks 1 and 3, 53-60% in weeks 2 and 4).

Copulatory behavior of males (mount and ejaculation latency, number of mounts and intromissions) was unaffected by acrylamide exposure. In week 1 after exposure of males to 15 and 45 mg/kg/day an increased number of females did not have sperm in the uterus (percentages with sperm were 100%, 60%, and 20 % at 0, 15, and 45 mg/kg/day respectively). In subsequent weeks, there were no clear effects on uterine sperm.

Sperm samples from males receiving 45 mg/kg/day were not located for analysis in week 1 - the reason for this was not clear. Examinations were further limited by the use of only two exposure levels, 0 and 45 mg/kg/day. The only statistically significant effects on sperm were observed in week 3 post-administration. There were slight reductions in sperm count (61×10^6 at 45 mg/kg/day compared with 82×10^6 in controls), percentage motility (58% at 45 mg/kg/day, 75% in controls), curvilinear velocity (122 $\mu\text{m/s}$ versus 132 $\mu\text{m/s}$), linearity and straight line velocity. These changes, in themselves, are of limited significance, but support the observation of reduced male fertility in the dominant lethal assay. In addition, there was a statistically significant reduction in the percentage of ova fertilised by males exposed to 15 and 45 mg/kg/day in week 1 (29% and 41% compared to 84% in controls), and at 45 mg/kg/day in week 3 (12% compared to 65% in control).

Conclusion

: Overall, this study demonstrated reduced fertility in male rats and an increase in pre and post implantation losses in an exposure regime, which did not apparently affect mating performance through neurotoxicity; male fertility was impaired at exposure levels of 15 mg/kg/day or more for 5

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Reliability | : | days. |
| Reference | : | (1) reliable without restriction (Sublet et al., 1989) |
| Test substance | : | > 99.9 % pure |
| Method | : | |
| Type | : | Two generation study |
| GLP | : | Yes |
| Year | : | 1998 |
| Species | : | Rat |
| Strain | : | Fischer 344 |
| Route of administration | : | Drinking water |
| Doses | : | 0.5, 2.0, or 5.0 mg/kg |
| Sex | : | male/female |
| Control group | : | Yes, concurrent no treatment |
| Frequency of treatment | : | Daily |
| Duration of test | : | Premating (F0) through weaning (F2) |
| Premating exposure period - Male | : | 10 weeks, F0 |
| Premating exposure period - Female | : | 10 weeks, F0 |
| Test condition | : | 30 rats/sex/group were dosed for 10 weeks and then mated. Exposure of the Fo females continued through gestation, parturition and lactation of the F1 litters. The Fo males, after completion of the mating were removed from acrylamide exp. And mated to detect possible dominant lethal effects. 30 F1 weanlings/sex/group randomly selected to produce the f2 generation were exposed for 11 weeks to the same drinking water dose levels of acrylamide as their parents and then paired. Gestation, lactation, necropsy of F1 parents and selected F2 weanlings were performed. |
| NOAEL Parental | : | 0.5 mg/kg bw |
| Actual dose/level/sex | : | |
| Parental and F1 data | : | |
| Offspring toxicity | : | |
| NOAEL F1 Offspr. | : | 2 mg/kg bw |
| NOAEL F2 Offspr. | : | 2 mg/kg bw |
| Statistical results | : | |
| Result | : | Pre-breeding exposure of the Fo animals resulted in evidence of toxicity to both sexes: reduced body weights for Fo rats of either sex given 2.0 or 5.0 mg/kg/d and males at 0.5 mg/kg/d. At 5.0 mg/kg/d, head tilt and leg splay were observed in Fo males and leg splay was observed in Fo females. Reproductive indices and gestational length from the Fo mating (to produce F1 offspring) were equivalent for all groups. The number of implantations per dam and thenumber of live pups per litter at birth were significantly reduced at 5.0 mg/kg/d. Fo females exhibited reduced weights and weight gain during gestation at 5.0 mg/kg/d. During lactation, maternal body weights remained lower at 5.0 mg/kg/d than in controls. However, lactational weight gain was increased at 5.0 mg/kg/d. In the dominant lethal evaluation, at .5.0 mg/kg/d, the number of total and live implants were reduced, non-live implants and pre- and post implantations loss were increased and calculated frequency of dominant lethal factors (F1%) was 20%. Lower dose groups were unaffected. Survival of F1 pups was unaffected by treatment. During the latter half of the lactation period during the time when pups begin to self feed and drink, male pup weight per litter was significantly reduced at 5.0 mg/kg/d. No treatment-related histopathological lesions were seen in the Fo parents of |

5. Toxicity

Id 79-06-1

Date 7 September 2001

the F1 weanlings.

F1 animals, selected as parents for F2 generation, had reduced body weights at 2.0 and 5.0 mg/kg/d for both sexes during the 11 week pre breed exposure. Head tilt (but not leg splay) was observed in F1 males only at 5.0 mg/kg/d. F1 breeding (to produce F2 offspring) produced results essentially identical to those observed in the Fo breeding.

Reproductive indices and gestational length were unaffected. The number of implantations and live pups per litter were decreased with the percent post-implantation loss therefore increased at the 5.0 mg/kg/d dose level. Maternal gestational weights were depressed at 5.0 mg/kg/d and gestational weight gain was depressed at 2.0 and 5.0 mg/kg/d. Maternal lactational body weight gain was increased at 5.0 mg/kg/d. F2 pup survival indices were unaffected by treatment. F2 litter sizes were reduced at 5.0 mg/kg/d due entirely to the initial reduction observed at birth. Gross necropsy of selected F2 weanlings and all F1 adults indicated no treatment related effects. Peripheral nerve sections (sciatic and tibial) from randomly selected F1 males from the high dose group revealed minimal to mild axonal fragmentation and/or swelling. Spinal cord sections from three high dose females did not reveal any abnormal findings. Therefore, the „NOEL“ for prenatal toxicity was 2.0 mg/kg/d and the NOEL for adult toxicity was 0.5 mg/kg/d.

| | | |
|----------------------------------|---|--|
| Conclusion | : | The parental NOEL was based on depression in body weight gain and neurotoxicity observed at higher doses. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Tyl et al., 2000a) |
| Test substance | : | > 99.7% pure |
| Method | : | |
| Type | : | Reproductive/Neurotoxicity |
| GLP | : | No data |
| Year | : | 2000 |
| Species | : | Rats |
| Strain | : | Long Evans |
| Route of administration | : | Oral gavage |
| Doses | : | 5, 15, 30, 45, or 60 mg/kg/d |
| Sex | : | Male |
| Control group | : | Yes |
| Frequency of treatment | : | Daily |
| Duration of test | : | 9 days |
| Premating exposure period | : | 5 days |
| Test condition | : | To determine whether there is a relationship between reproductive and neurotoxic effects of acrylamide, Long Evans male rats were gavaged (25/group), at 0, 5, 15, 30, 45, and 60 mg/kg/d for 5 days (similar to Sublet et al, 1989). On Day 8, males were paired overnight with untreated virgin females (1:1) in proestrus/estrus. On Day 9, males were evaluated for forelimb and hindlimb grip strength. Five males/group were perfusion fixed, 20/group were used for andrologic assessment and all were necropsied. Sciatic nerves were examined histologically. Sperm positive females were examined for preimplantation and postimplantation loss at midpregnancy. |
| NOAEL Parental | : | |
| Actual dose/level/sex | : | |
| Parental and F1 data | : | |
| Offspring toxicity | : | |
| Statistical results | : | |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
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| Result | : | At 15 - 60 mg/kg/d, males exhibited significantly reduced weight gain, reduced mating, fertility, and pregnancy indices by trend analysis (significant at the high dose) and increased postimplantation loss and dominant lethal factor at 45 and 60 mg/kg/d. At 60 mg/kg/d, the sperm beat cross frequency was increased with no significant effects on epididymal sperm motility or concentration, and hindlimb grip strength was decreased, with no pathologic lesions in sciatic nerves. Therefore, epididymal sperm, mating, and neurotoxic effects were observed at doses that also resulted in increased postimplantation loss. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Tyl et al., 2000b) |
| Test substance | : | No data |
| Method | : | |
| Type | : | |
| GLP | : | No data |
| Year | : | 1984 |
| Species | : | Rat |
| Strain | : | Fischer 344 |
| Route of administration | : | drinking water |
| Doses | : | 0.5, 2, or 5 mg/kg |
| Sex | : | male/female |
| Control group | : | Yes |
| Frequency of treatment | : | Daily |
| Duration of test | : | exposure period: females: 10 weeks throughout gestation and lactation; males: 10 weeks |
| Premating exposure period – Male | : | |
| Premating exposure period – Female | : | |
| Statistical methods | : | |
| Test condition | : | |
| NOAEL | : | Male/Female: 0.5 mg/kg |
| LOAEL | : | Male/Female: 2 mg/kg |
| Actual dose/level/sex | : | |
| Parental and F1 data | : | |
| Offspring toxicity | : | |
| Statistical results | : | |
| Result | : | Male rats experienced lower body weight gain and ataxia; decreases in body weight and body weight gain and an increase in preimplantation loss occurred in the highest dose level; decreased fecundity was observed in the 2 mg/kg/day dose, but no adverse effects were observed in the lower dose groups |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, data came from secondary source, primary source was not available |
| Reference | : | (Johnson et al., 1984) |
| Test substance | : | 99% pure |
| Method | : | |
| Type | : | |
| GLP | : | No data |
| Year | : | 1986 |
| Species | : | Rat |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---|---|--|
| Strain | : | Long Evans |
| Route of administration | : | Oral, drinking water |
| Doses | : | 1.5, 2.8, or 5.8 mg/kg |
| Sex | : | Male/Female |
| Control group | : | Yes |
| Frequency of treatment | : | Daily |
| Duration of test | : | 80 days |
| Premating exposure period – Male | : | |
| Premating exposure period – Female | : | |
| Statistical methods | : | |
| Test condition | : | Groups of 10-11 male rats were given 0, 1.5, 2.8, or 6 mg/kg/day acrylamide in drinking water for a total of 80 days. After 72 days males were mated with untreated females. Number of corpora lutea and living and dead fetal implants were examined for. Fertility rates and % of pre and post implantation losses were calculated. Fertility rates were analysed via chi square statistics. ANOVA was used for pre and post implantation comparisons with a Mann whitney U test for post-hoc comparisons. |
| NOAEL | : | 1.5 mg/kg |
| LOAEL | : | 2.8 mg/kg |
| Actual dose/level/sex | : | |
| Parental and F1 data | : | |
| Offspring toxicity | : | |
| Statistical results | : | |
| Result | : | Significantly increased pre-implantation loss was noted amongst females mated with high dose males. Post-implantation loss was increased at 2.8 and 6 mg/kg/day. There were no clinical or histopathological signs of neurotoxicity which may have affected male fertility. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Smith et al., 1986) |

5.9 DEVELOPMENTAL TOXICITY/TERATOGENICITY

| | | |
|--------------------------------|---|--|
| Test substance | : | No data |
| Method | : | |
| GLP | : | Pre-GLP |
| Year | : | 1979 |
| Species | : | Developing chick embryos |
| Strain | : | White Leghorn SK 12 |
| Route of administration | : | Injection |
| Doses | : | 0.007, 0.07 or 0.7 mg (0.1, 1.0, 10 µmol) |
| Sex | : | No data |
| Exposure period | : | |
| Frequency of treatment | : | Single |
| Control group | : | Yes |
| Duration of test | : | 14 days |
| Statistical methods | : | |
| Test condition | : | Eggs were injected on day 3 of incubation and examined on day 14 of incubation. Injection site #1 into airspace of egg: dose groups 0.1, 1.0, and 10 µmol; number of eggs/dose group 30, 30, and 25, respectively. Injection |

5. Toxicity

Id 79-06-1

Date 7 September 2001

site #2 into yoke sac of the egg: dose groups 0.1, 1.0, and 10 μmol ; number of eggs in all dose groups was 10

Maternal toxicity :
Developmental toxicity :
Actual dose/level/sex :
Maternal data :
Fetal data :
Statistical results :
Result : Mortality was elevated (40% mortality in highest dose) but no malformations occurred in eggs at concentrations up to 10 μmol in the living embryos. LD50 in the range of 1-10 (approximately 5 μmol /egg).

Conclusion : The authors state that the embryos were examined for macroscopic malformations but no evidence on induced teratogenicity was shown.

Reliability : (1) reliable without restriction

Reference : (Kankaanpaa et al., 1979)

Test substance : No data

Method :
GLP : Pre-GLP
Year : 1976
Species : Rat
Strain : Porton

Route of administration : Oral feed

Doses : 200 or 400 ppm in feed or 100 mg/kg in water given intravenously

Sex : Female

Exposure period : Gestational days 1-20

Frequency of treatment : Daily

Control group : Yes

Duration of test :
Statistical methods :
Test condition : Group 1: 8 pregnant Porton rats received 200 ppm acrylamide by diet from day of mating until parturition. Group 2: 6 pregnant Porton rats received 400 ppm acrylamide by dietary admixture on days 1-20 of gestation. [Assuming a bodyweight of 300 g and food consumption of 20 g/day, these dietary concentrations would result in dose levels of about 15, and 30 mg/kg/day]. Group 3: 4 pregnant Porton rats were given one dose of 100 mg/kg acrylamide in water intravenously on day 9 of gestation. For all groups: The endometria were examined for evidence of resorption, and fetuses weighed and examined for visceral and skeletal abnormalities.

Maternal toxicity :
Developmental toxicity :
Actual dose/level/sex :
Maternal data :
Fetal data :
Statistical results :
Result : At 200 and 400 ppm, there were signs of loss of use of hind limbs amongst the pregnant females and reduced food consumption at 400 ppm (approximately 48% reduction). Amongst progeny, bodyweight was reduced (by about 25%) in Group 2 (400 ppm) only, although this could be attributed to reduced maternal food consumption and was therefore of minimal significance with respect to developmental effects. There were no other effects on development. No effects on development in Group 3 rats or progeny.

Overall, in this study continuous exposure of pregnant female rats to dietary

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| | concentrations of 400 ppm acrylamide (approximately 30 mg/kg/day) was associated with minor effects on development seen only at maternally toxic concentrations. |
| Conclusion | : |
| Reliability | : (1) reliable without restriction |
| Reference | : (Edwards, 1976) |
| Test substance | : 95% pure |
| Method | : |
| GLP | : No data |
| Year | : 1990 |
| Species | : Rats/mice |
| Strain | : Rat: Sprague-Dawley Mice: Swiss |
| Route of administration | : Gavage |
| Doses | : Rat: 2.5, 7.5, or 15 mg/kg mice: 3, 15, or 45 mg/kg |
| Sex | : Male/female |
| Exposure period | : Rat: gestational days 6 to 20 mice: gestational days 6 to 17 |
| Frequency of treatment | : Once/day |
| Control group | : Yes |
| Duration of test | : |
| Statistical methods | : |
| Test condition | : Rat: Groups of 29-30 mated Sprague-Dawley rats (201-263 g) received 0, 2.5, 7.5, or 15 mg/kg/day aqueous acrylamide (about 98% pure) by oral gavage on gestation days 6-20. Animals were observed daily for clinical signs. On day 20, pregnant females were sacrificed, uteri examined (number of implantation sites, resorptions, live and dead fetuses). Fetuses were thoroughly examined macroscopically for visceral and skeletal abnormalities (including the head). Mice: Groups of 30 pregnant Swiss mice (24.7-34.1 g) received 0, 3, 15, or 45 mg/kg/day aqueous acrylamide (~98% pure) by oral gavage on gestation days 6-17. Animals were observed daily for clinical signs. On day 17, pregnant females were sacrificed, uteri examined (number of implantation sites, resorptions, live and dead fetuses). Fetuses were thoroughly examined macroscopically for visceral and skeletal abnormalities (including the head). Statistical Analysis: GLM procedures were applied for the ANOVA of maternal and fetal parameters. GLM analysis determined the significance of dose-response relationships and the significance of dose effects, replicate effects and dose x replicate interactions. If the ANOVA was significant, William's or Dunnett's Multiple Comparison Tests compared ACRL exposed to control groups. One tailed tests were used for all pairwise comparisons except maternal body and organ weights and fetal body weight. Nominal scale measures were analysed by chi square test for independence and by a test for linear trend on proportions. When a chi square showed significant group differences, a one-tailed Fisher's exact probability test was used for pairwise comparisons of ACRL and control groups. |
| Maternal Toxicity (NOAEL) | : Rat: 2.5 mg/kg Mice: 15 mg/kg |

5. Toxicity

Id 79-06-1

Date 7 September 2001

| | | |
|---------------------------------------|---|--|
| Developmental Toxicity (NOAEL) | : | Rat: 15 mg/kg Mice: 15 mg/kg |
| Actual dose/level/sex | : | |
| Maternal data | : | |
| Fetal data | : | |
| Statistical results | : | |
| Result | : | <p>Rat:</p> <p>There were no maternal mortalities and no clear clinical signs of toxicity. When corrected for gravid uterine weight, maternal bodyweight gain was decreased amongst animals receiving 7.5 and 15 mg/kg/day (12% and 18% reductions respectively). There were no apparent effects on embryo/fetal viability, growth or malformations. There was a slight, but not statistically significant, increase in the incidence of skeletal variations (percentage of litters with variations - 61% in controls, 92% at 15 mg/kg/day, and percentage of fetuses with variations per litter - 14% in controls, 24% at 15 mg/kg/day). The most frequently observed variation was the presence of a rudimentary extra lumbar rib. This finding is considered likely to be an indirect consequence of maternal toxicity or stress and is of limited toxicological importance.</p> <p>2.5 mg/kg/d was considered to be the maternal NOAEL and 15 mg/kg/d was considered to be the NOAEL for developmental toxicity.</p> <p>Mice:</p> <p>There were no exposure-related maternal mortalities, although clinical signs of toxicity (loss of use of hind limbs) were observed amongst females at 45 mg/kg/day. Maternal bodyweight was statistically significantly reduced at 15 and 45 mg/kg/day (9% and 16% reduction respectively), which was directly related to the statistically significant reduction in gravid uterine weight.</p> <p>Mean fetal bodyweight was reduced amongst male and female offspring from mice that had received 45 mg/kg/day (15% reduction for both sexes). As with the accompanying rat developmental study, there was also a slight, but not statistically significant, increase in the incidence of skeletal variations amongst all acrylamide-exposed mice (percentage of litters with variations - 44% in controls, 64% at 15 mg/kg/day, and percentage of fetuses with variations per litter - 7% in controls, 15% at 15 mg/kg/day). The most frequently observed variations were the presence of an extra lumbar rib or rudimentary extra rib; findings considered being of limited toxicological importance. No other evidence of developmental toxicity was apparent.</p> <p>Overall, the results of this study indicate that, in mice, there was reduced fetal bodyweight only at a dose level (45 mg/kg/day) resulting in significant maternal toxicity. There was no evidence of significant developmental toxicity at 15 mg/kg/day and below. 15 mg/kg/d was considered to be the maternal and developmental NOAEL..</p> |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Field et al., 1990) |
| Test substance | : | No data |
| Method | : | |
| GLP | : | Pre-GLP |
| Year | : | 1981 |
| Species | : | Rat |
| Strain | : | Fischer 344 |

5. Toxicity

Id 79-06-1

Date 7 September 2001

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| Route of administration | : | Oral gavage |
| Doses | : | 20 mg/kg/d |
| Sex | : | Female |
| Exposure period | : | gestational days 6 to 17 |
| Frequency of treatment | : | Daily |
| Control group | : | Yes, vehicle only |
| Duration of test | : | 60 days post exposure |
| Statistical methods | : | |
| Test condition | : | To measure prenatal and lactational exp on the development of intestinal enzymes pregnant rats (8 weeks old) were exposed to acrylamide over 10 days. On Day 1 of age, the pups were pooled into two groups (treated and control) and then cross-fostered into four experimental groups: treated dams with treated pups (t/t), treated dams with control pups (t/c), control dams with treated pups (c/t), and control damss with control pups (c/c). 4 randomly selected pups from each group were sacrificed at 14, 21, and 60 days after birth to examine the development of five marker enzymes in the small intestine: acid phosphatase(ACP), alkaline phosphatase (ALP), B-gluconidase(B-GLU), citrate synthase (CS), and lactate dehydrogenase (LDH). Difference in enzyme activities were indicative of a prenatal effect (C/t values compared to c/c values), a lactational effect (t/c to c/c), and an enhancement of prenatal effects (t/t to c/t). These differences were either increases or decreases. No information on statistical analysis used. |
| Maternal Toxicity | : | |
| Developmental Toxicity | : | |
| Actual dose/level/sex | : | |
| Maternal data | : | |
| Fetal data | : | |
| Statistical results | : | |
| Result | : | ALP activities showed a prenatal effect at 14, 21, and 60 days, a lactational effect at 20 and 60 days, and an enhancement of prenatal effects at 14, 21, and 60 days. CS activities were not affected in any of the treatment groups. The only change in LDH was a lactational effect at 21 days. ACP activities exhibited a prenatal effects at 14 and 60 days, a lactatational effect at 14 days and an enhancement effect of prenatal effects at 21 and 60 days. B-GLU activities showed a prenatal effect at 21 days, a lactaional effect at 21 days, and an enhancement of prenatal effects at 21 and 60 days. Measurements of maternal enzyme levels in each of the groups showed no significant change. These measurements were made after weaning pups. No overt signs of maternal toxicity. Litter sizes, number and sex ratio of pups were not affected by the dose administered. |
| Conclusion | : | Prenatal and lactational exposure to acrylamide does significantly change intestinal enzyme levels during the early stages of development. |
| Reliability | : | (2) reliable with restriction, no info on statistical analysis |
| Reference | : | (Walden et al., 1981) |

5.10 OTHER RELEVANT INFORMATION

| | | |
|----------------------|---|--|
| Type | : | Formation of Hemoglobin Adducts |
| Methodology | : | 3 Sprague Dawley rats (130-260g)/group were injected ip with acrylamide or glycidamide in doses ranging from 0 0.5, 1.0, 5.0, 10.0, 50.0, and 100 mg/kg body wt. and the hemoglobin adduct levels were determined. |
| Result/Remark | : | The hemoglobin binding index of acrylamide to cysteine was found to be 6400 pmol (g Hb)-1/umol (kg body wt)-1. In rats injected with acrylamide, formation of adducts of the parent compound was approximately linear with |

| | | |
|----------------------|---|--|
| Conclusion | : | dose (0-100 mg/kg), whereas adducts of the epoxide metabolite glycidamide generated a concave curve. The first order rates of elimination of acrylamide and glycidamide from the blood compartment of rats were estimated to be 0.37 and 0.48/hr, respectively. Subchronic treatment of rats with 10 mg/kg/d for 10 days or 3.3. mg.kg.d for 30 days confirmed that the conversion rate of acrylamide to glycidamide as determined from hemoglobin adduct formation, is higher at lower administered doses. Findings suggest that dose-rate effects may significantly affect risk estimates of this compound and that different low dose extrapolation procedures should be employed for effects induced by the parent compound acrylamide and those induced by the metabolite glycidamide. |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Bergmark et al., 1991) |
| Type | : | other: Neuro- and Reproductive Toxicity |
| Methodology | : | <p>In initial neurotoxicological experiments (rotarod performance, hindlimb splay), the effects of the parent compound (8-14 days, 25 and 50 mg/kg/day) and the metabolite (8-14 days, 50 and 100 mg/kg/day) were compared. 5 groups of Sprague Dawley rats (270-310g) were i.p. injected once a day for eight days with acrylamide or its metabolite. Control rats were injected with vehicle.</p> <p>For the repro tox experiment, SD rats (350g) were administered acrylamide (50 mg/kg/d for seven days) or glycidamide (50 mg/kg/d for 14 days). 24 hrs after last injection, testis, epididymis and vas deferens were dissected and weighed. Protein content in testis was determined according to Bradford, 1976. The head, body and tail of epididymis were minced and homogenized and the number of spermatozoa was determined. Sperm viability was measured as described by Bishop and Smiles, 1957.</p> |
| Result/Remark | : | <p>Statistical Analysis: Results were analyzed by ANOVA followed by Fisher exact test for comparison between groups.</p> <p>While at the higher dose both compounds affected the rats' performance on the rotarod, only acrylamide had a significant effect in the hindlimb splay test, which is considered a more sensitive indicator of peripheral neuropathy.</p> <p>On the other hand, a stronger effect was seen for glycidamide than for acrylamide on the male reproductive system, especially on sperm cell viability.</p> <p>Preliminary results suggest that while the parent compound appears to be primarily responsible for the induction of peripheral neuropathy, other toxic effects associated with acrylamide exposure, such as reproductive toxicity, may be attributed to glycidamide.</p> |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| Reference | : | (Costa et al., 1992) |
| Type | : | Metabolism |
| Methodology | : | Contradictory results as to the role of cytochrome P-450 mediated metabolism of acrylamide in the induction of neurotoxic effects prompted the investigation of the possible formation of glycidamide, a reactive epoxide metabolite. |
| Remark | : | The formation of this epoxide was strongly indicated by the identification by means of gas chromatography – mass spectrometry of derivatized S-(2-carboxy-2-hydroxyethyl)cysteine in hydrolyzed hemoglobin samples from |

5. Toxicity

Id 79-06-1

Date 7 September 2001

- rats treated with acrylamide in vivo and in microsomal suspensions of acrylamide with cysteine in vitro. This amino acid was found to be present in uninduced and phenobarbital-induced SD rats and absent in controls, but occurred in lower amounts than the adduct derived from the parent compound, S-(2-carboxy-2-hydroxyethyl)cysteine.
- Reliability Reference** : (1) reliable without restriction
: (Calleman et al., 1990)
- Type Remark** : other: Formation of Hemoglobin Adducts
: Hemoglobin (Hb) adducts, formed by the neurotoxic agent acrylamide (AA) and its genotoxic metabolite glycidamide(GA), were measured in the rat by means of a method for simultaneous determination of the adducts formed to cysteine. A novel, nonlinear dosimetric model was developed to describe Hb adduct formation. This model incorporates the saturable kinetics of the metabolic conversion in vivo of AA to GA. The pharmacokinetic parameters Vmax and Km and the first order rates of elimination, k, and k2, for AA and GA from all processes except conversion of AA to GA, were estimated directly from Hb adduct data to 19 M hr⁻¹, 66 uM, 0.21 hr⁻¹, and 0.48 hr⁻¹, respectively. At low concentrations, approximately 60% of AA was metabolized to GA.
- Reliability Reference** : (1) reliable without restriction
: (Calleman et al., 1993)
- Type Methodology** : other: Kinensin based - Microtubule Motility Assay
: Kinensin and microtubules (MT) were evaluated as molecular sites of action using an in vitro MT motility assay. MT motility assays were performed 3 to 6 times for each concentration (0.1, 0.5 or 1.0 mM) preincubation with kinensin or tubulin as well as for the different temperatures and the non-neurotoxic analogue. ANOVA was used to determine the rat eof microtubule movement. Stat. Sig differences in microtubule detachments.were determined using ANOVA also.Tukey's post-hoc test was used to determine differences from control.
- Result Remark** : The number of locomoting MT which lifted from a bed of kinensin, increase from 7% in controls to 80, 89 and 100) following preincubation of kinensin with 0.1, 0.5 or 1.0 mM acrylamide, respectively; rates were variably reduced by as much as 20%. It is concluded that kinensin and MT are covalently modifiedby ACR resulting in reduced affinity for each other. The greater sensitivity of kinensin indicates that a primary cause of transient, ACR induced reductions in fast anterograde axonal transport may be sufficient to produce axonal degeneration.
- Conclusion Reliability Reference** :
: (1) reliable without restriction
: (Sickles et al., 1996)
- Type Methodology** : other: Effect of Acrylamide Mitotic Chromosomal Migration
: Cultured HT 1080 fibrosarcoma cells were exposed to acrylamide at concentrations of 1.0, 2.5, 5.0 and 10 mM, to determine if mitosis was adversely affected. Nonneurotoxic analogue, methylene bisacrylamide, was dosed at same concentrations. Two control dishes without ACR and two with 30 uM colchicine (positive control) were processed similarly. Four hours after introduction into culture, cells were processed.
Statistical Analysis: The number of mitotic cells for each concentration of each toxicant was compared to control with ANOVA and Bartlett's variance check followed by Tukey's HSD post hoc test.
- Result/Remark** : The number of cells arrested in mitosis increased in a concentration dependent manner, from 1 to 10 mM. A 4 hr exp. to 10 mM acrylamide increased the mitotic index by 4.5 fold over control comparable to the arrest

5. Toxicity

Id 79-06-1

Date 7 September 2001

caused by colchicine. In mitotic acrylamide exposed cells, the chromosomes remained at the metaphase plate; no changes in spindle microtubules, as seen with tubulin immunofluorescence, were observed. The distance between spindle poles was the same in control and experiment cells. The nonneurotoxic analogue methylene bisacrylamide had no effect in the same concentration range. The data suggest potential molecular mechanisms of action for general toxicity and neurotoxicity to be disruption in MT disassembly of MT kinetochore interactions and/or cellular homeostatis.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Sickles et al., 1994)

Type : other: Fast axonal transport deficiencies
Methodology : To determine the effects of acrylamide (at concs. of 0.25, 0.5, and 1 mM) and analogues on the number of vesicles moving within the neurite processes of cultured rat (Sprague-Dawley) embryonic neurons. Stat. sig. differences in transport were determined using ANCOVA. The ANCOVA was necessary to control for baseline transport quantity variations, since larger neurites were observed to transport greater numbers of vesicles per unit time. Two group effects, toxicant and dosage, and a trail effect, time were evaluated.

Result/Remark Acrylamide produced severe, concentration dependent and time dependent (0-60min) reduction in the quantity of vesicles translocated in both the anterograde and retrograde directions. Glycidamide, a potential neurotoxic metabolite of acrylamide, produced a time dependent but not a concentration dependent reduction bidirectional transport. Based on inhibition at 60 min, glycidamide was estimated to be 4 times more potent than acrylamide in altering transport.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Harris et al., 1994)

Type : other: Neurotoxicity
Methodology : Male, Sprague-Dawley rats (225-250 g) were anesthetized and underwent a laminectomy to expose the L5 dorsal root ganglion. To differentiate a toxicant-induced compromise in the capacity of the fast anterograde axonal transport system from a neuron cell body processing effect, selective exposure of either the L5 dorsal root ganglion or sciatic nerve to 0.7 mM acrylamide or 4 mM 2,5-hexanedione was performed during in vitro transport. Experiment was solely performed to determine site of effect. Statistical Analysis: Differences between control and experimental groups were determined with a two tailed ANOVA and post hoc Tukey's test; homogeneity of variances were verified using Bartlett's test.

Result/Remark : Nerve exposure to acrylamide decreased the quantity of transport by 32%, 2,5-hexanedione reduced the quantity by 44%. Ganglion exposure produced no significant changes. It was concluded that both toxicants penetrate the nerve barriers and act directly and/or indirectly on the axonal transport mechanisms to cause the reductions in transport.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Sickles, 1992)

Type : other: Neurotoxicity
Methodology : The effects of a single exposure to acrylamide on the rate and quantity of protein transported in the rat sciatic nerve was measured to determine whether fast axonal transport is compromised by the toxicant. A single i.p.

injection of 50, 75, or 100 mg/kg was given 20 minutes after the 2H-leucine injection into the dorsal root ganglion (4 rats/group).

Statistical Analysis: The rate and capacity of protein transport in the sciatic nerves of treated rats were compared to control with a two-tailed ANOVA and Dunnett's T-test. The quantity of protein synthesis in controls was compared with those expected to acrylamide and to cycloheximide with a two tailed ANOVA and Tukey's test. A Bartlett's test verified that the group variances were homogenous for all cases.

Result/Remark : Using the segmental analysis of radioactive label of proteins (Ochs and Ranish, 1970) following 3H-leucine injections into the dorsal root ganglion, acrylamide (50-100 mg/kg) significantly reduced the rate of fast anterograde transport by 9.3 to 20.8% but also reduced the quantity of transported protein by 42.4 to 51.3%. Fast anterograde transport was significantly affected by a single exposure to acrylamide in the same magnitude as retrograde transport. Discovery of these dramatic changes was due to differences from previous studies in the time frame of study of transport in relation to toxicant injection and to measurements of the quantity of protein transported rather only the rate. These changes may be significant in terms of the pathogenesis of distal nerve degeneration.

Conclusion :
Reliability : (1) reliable without restriction
Reference : (Sickles, 1989)

Type : Comparative Distribution in Foetuses
Methodology : Rats: Osborne-Mendel
 Rabbits: New Zealand white
 Dogs: beagle
 Minature Pig: Hormel
 Single iv dose 1-2 days [C^{14} AMD) before parturition
 Dose: 10 mg/kg (rats), 5 mg/kg other species.
 Sacrifice: 1 hr (rats) and 2 hr. (other species) following injection

Result/Remark : Neither sex nor position of fetus affected uptake
Conclusion : In all tissues, AMD present in higher levels in pig compared to dog.
Reliability : (1) reliable without restriction
Reference : (Ikeda et al. 1983)

Type : Comparative Distribution in Foetuses
Methodology : Dogs: beagle
 Minature Pig: Hormel
 Single iv dose, 5 mg/kg, on gestation day 60 (dogs) or 109 (pigs).
 Dose: 5 mg/kg
 Sacrifice: 2 hr. following injection

Result/Remark : In the dog, distribution in the maternal tissue was 1.1-1.2 times higher than in the fetus. Kidney (both maternal and foetal) contained 1.3-1.6 times that found in the blood. Liver contained the highest levels and was 2.5 x (maternal) or 1.7 x (fetus) blood. Placental barrier in the dog was 17.7%. In the pig, maternal blood was 1.45 x than in the fetus. Kidney and liver had the highest concentration. The placental barrier was 31%. There was negligible blood-brain barrier affects in either species, adult or fetus.

Conclusion : Tissue distribution is the same in dam and fetus, although lower in the latter. Blood-brain barrier affects were insignificant.
Reliability : (1) reliable without restriction
Reference : (Ikeda et al. 1985)
 :

5. Toxicity

Id 79-06-1

Date 7 September 2001

Type : Comparative Distribution and Excretion
Methodology : Dogs: beagle
Minature Pig: Hormel
Dose: subjects received dietary AMD for 3-4 wk. (1 mg/kg/day) followed by a single oral dose of radiolabelled AMD (1 mg/kg). Sacrifice: animals were sacrificed at 6 hr., 1, 2, 4, or 14-days thereafter. day 60 (dogs) or 109 (pigs).
Result/Remark : Muscle was the major site of deposition in both species (31-35% at 6 hr. and 5-7% at 14-days). Less than 1% was found in the brain of either species. Approx. 60% of the dose was excreted in the urine (both species), smaller amounts in feces (approx. 25% in pigs and 7% in dogs)
Conclusion : Absorption of AMD administered in the diet is more rapid and more extensive in the dog than in the pig. Distribution is the same.
Reliability : (1) reliable without restriction
Reference : (Ikeda et al. 1987)

Type : Comparative Metabolism and Distribution
Methodology : Rats: male F344
Mice: male B6C3F1
Route: inhalation, nose only
Dose: 5.7 and 2.9 ppm
Duration: 6 hr.
Samples: urine, feces, expired air and tissues (obtained from the 2.9 ppm exposure)
Route: dermal application
Dose: 162 mg/kg
Duration: 24 hr.
Samples: same as above
Result/Remark : Inhalation: urine, feces, and washes accounted for most of the recovered dose (42% and 51%, rats and mice, respectively) with tissues accounted for 56% and 46% (rats and mice, resp.)
Dermal application: 22% of the dose was absorbed; 44% of the dose recovered in urine, feces and washes, 53% recovered in the tissues.
Conclusion : Route differences in the metabolism and distribution of AMD are important considerations in the development of models of assessing the potential risk from human exposure.
Reliability : (2) reliable with restrictions, information obtained from a published abstract
Reference : (Friedman et al. 2001)

5.11 EXPERIENCE WITH HUMAN EXPOSURE

Memo : Worker Exposure
Test condition : A cohort of 8854 men, 2293 of whom were exposed to acrylamide, was examined from 1925 to 1983 for mortality. This cohort consisted of four plant populations in two countries: the United States and The Netherlands. Exposure estimates for all jobs at each location were developed using monitoring data and by working with plant persons who had knowledge of past jobs and processes. Exposure to acrylamide was defined as 0.3 mg/m³/d. Standard mortality ratios and relative risk were employed to estimate risk. For the SMR, 95% confidence intervals were calculated using the method of Bailar and Ederer and a chi-square test was used to evaluate statistical significance of trends. For RR, 95% confidence intervals were based on the procedure of Miettinen and the test for significance of trend is based on a method developed by Mantel.

5. Toxicity

Id 79-06-1

Date 7 September 2001

- Result/Remark** : Analysis of trends by cumulative exposure and duration of exposure indicated no increased risk of mortality with increased level of exposure (accounting for smoking). No statistically significant excess of all-cause or cause-specific mortality was found among acrylamide workers. Analysis by acrylamide exposure levels showed no trend (adjusted for confounding factors) of increased risk mortality from 26 cancer sites. These results do not support the hypothesis that acrylamide is a human carcinogen.
- Conclusion** :
Reliability : (1) reliable without restriction
Reference : (Collins et al., 1989)
- Memo** : Worker Exposure: 1994 Follow-up to Collins et al. 1989
Test condition : Analyses of SMR using national and local rates and relative risk regression modeling were performed according to site specific cancer risks by demographic and work history factors, and exposure indicators for AMD and muriatic acid.
- Result/Remark** : For the 1925-94 study period, excess and deficit overall mortality risks were observed for cancer sites of a priori interest: brain and other CNS (SMR=0.65, 95% CI=0.36-1.09), thyroid gland (SMR=2.11, 95% CI=0.44-6.17), testis and other male genital organs (SMR=0.28, 95% CI=0.01-1.59), and respiratory system cancer (SMR=1.10, 95% CI=0.99-1.22); however, none were statistically significant or associated with acrylamide exposure. A previously reported excess mortality risk of RSC cases at one plant remained elevated among workers with potential exposure to muriatic acid (RR=1.50, 95% CI=0.86-2.59), but was only slightly elevated among workers exposed or unexposed to acrylamide. In an exploratory exposure response analysis of rectal, esophageal, pancreatic, and kidney cancer, an elevated SMR for some exposure categories but little evidence of an exposure response relationship. A stat. Sig. 2.26 fold risk (95% CI=1.03-4.29) was observed for pancreatic cancer among workers with cumulative exposure to acrylamide greater than 0.30 mg/m³-years; however, we detected no consistent exposure response relationships with the exposure measures considered when relative risk regression models were adjusted for time since first AMD exposure. The contribution of 1115 additional deaths and nearly 60,000 person years over the 11 yr update period corroborate the original cohort study findings of little evidence for a causal relationship between AMD exposure and mortality from any cancer sites, including those of a priori interest.
- Conclusion** :
Reliability : (1) reliable without restriction
Reference : (Marsh et al., 1998, unpublished)
- Memo** : Determination of Hemoglobin Adducts in Humans Occupationally Exposed to Acrylamide
Test condition : Blood samples were obtained from a group of 41 workers occupationally exposed to acrylamide at a factory in China and hemoglobin was extracted for analysis of acrylamide (AA) and glycidamide (GA) adducts. Workers were potentially exposed to acrylamide by inhalation and dermal routes - air concentrations ranged from 0.11-8.8 mg/m³ (8-hour TWA) with an occasional peak value of up to 153 mg/m³; skin peeling was observed on the hands indicative of significant dermal exposure. 10 nonexposed workers used for control group.
- Result** : The following valine adducts were released by acid hydrolysis of hemoglobin: N-(2-carboxyethyl)valine and N-(2-carboxy-2-hydroxyethyl)valine. The latter being indicative of epoxide formation and is consistent with earlier work in rats which also indicated the formation of glycidamide following acrylamide exposure. In the group of control workers;

a very low level of N-(2-carboxyethyl)valine was found in the blood sample of one of these individuals, who was a smoker (0.01 nmol/g hemoglobin, compared to 0.3-34 nmol/g in acrylamide-exposed workers).

There was a linear relationship between the AA and GA adduct levels ($r=0.96$) and the ratio of the in vivo doses of GA and AA was 3:10. These results suggest that AA is metabolized to GA in humans, as had previously been shown in the rat. The average daily in vivo doses of AA and GA in the highest exposed workers were comparable to the in vivo doses in rats injected with 3 mg/kg AA. Since a regimen of 2 mg/kg/day is known to cause a significant increase of tumors in rats, preventive measures may be necessary for humans exposed to high levels of AA in industrial settings.

Conclusion
Reliability
Reference

- :
: (1) reliable without restriction.
: (Bergmark et al., 1993)

Memo

- : Relationship b/w Biomarkers of Exposure and Neurological Effects in Humans Occupationally Exposed to Acrylamide

Test condition

- : 41 workers were exposed to a mixture of acrylamide (0.58-5.95 mg/m³) in a plant in China. The workers underwent a complete medical and neurological examination (vibration threshold, electroneuromyography) and provided blood and urine for the determination of several biomarkers of exposure (mercapturic acid, hemoglobin adducts).

Statistical Analysis: The chi square test was used to analyze the symptoms and signs, and Student's t test was used in the analysis of the ENMG parameters. Variance analysis and the Q test were used in the comparison of vibration thresholds between the reference group and the exposed group. Univariate and multivariate linear regression analysis was used to estimate correlation coefficients and levels of stat.sig. for the biomarkers of exposure.

Result

- : Among the exposed workers, signs and symptoms indicating peripheral neuropathy were found with stat. sig. increased frequencies compared to a group of controls from the same city. Based on neuropathic signs and symptoms and quantifiable indicators of peripheral nerve dysfunction, such as vibration thresholds and electroneuromyography measurements, a neurotoxicity index (Nin) specific for acrylamide-induced peripheral neuropathy was designed. The Nin was significantly correlated with the levels of mercapturic acids in 24hr urine, hemoglobin adducts of acrylamide, accumulated in vivo doses of acrylamide, employment time, and vibration sensitivity. The results seem to indicate that hemoglobin adducts are useful as predictors of acrylamide induced peripheral neuropathy and the measurements of vibration thresholds are useful for identifying early neurotoxic effects in workplaces with hazardous exposures to acrylamide.

Conclusion
Reliability
Reference

- :
: (1) reliable without restriction.
: (Calleman et al 1994)

6. References

Id 79-06-1

Date 7 September 2001

- ABC Labs (1982a): Dynamic 96-Hour Acute Toxicity of Acrylamide Monomer to Bluegill Sunfish (*Lepomis macrochirus*) Final Report December 28, 1982.
- ABC Labs (1982b): Dynamic 96-Hour Acute Toxicity of Acrylamide Monomer to Fathead Minnow (*Pimephales promelas*) Final Report December 28, 1982.
- ABC Labs (1982c): Dynamic 48-Hour Acute Toxicity of Acrylamide Monomer to Midge Larvae (*Paratanytarsus parthenogenetica*) Final Report December 28, 1982.
- ABC Labs (1982d): Dynamic 96-Hour Acute Toxicity of Acrylamide Monomer to Rainbow Trout (*Salmo gairdneri*) Final Report December 28, 1982.
- ABC Labs (1982e): Dynamic 48-Hour Acute Toxicity of Acrylamide Monomer to Water Fleas (*Daphnia magna*) Final Report December 28, 1982.
- Abdelmagid, H.M. and Tabatabai, M.A. (1982) Decomposition of acrylamide in soils. J. Environ. Qual. 11(4), 701-704
- Adler, I.-D., Ingwersen, I., Kliesch, U. and El Tarras, A. (1988) Clastogenic effects of acrylamide in mouse bone marrow cells. Mutat. Res. 206, 379 – 385
- Adler, I.D., Reitmeir, P, Scholler, R., Schriever-Schwemmer, G. (1994) Dose response for heritable translocations induced by acrylamide in spermatids of mice. Mutat. Res. 309, 285-291.
- Allan S. (1995) CT-566-94 Acrylamide skin sensitisation in the guinea pig. Huntingdon Research Centre Ltd., Huntingdon, Cambs. England. Report no. CTI 2/940899/55.
- Anbar, M. and Neta, P. (1967) A compilation of specific bimolecular rate constants for the reactions of hydrated electrons, hydrogen atoms and hydroxyl radicals with inorganic and organic compounds in aqueous solution. Int. J. Appl. Radiation Isotopes 18, 493 - 523
- Arai, T., Kuroda, S. and Watanabe, I. (1981) Biodegradation of acrylamide monomer by a *Rhodococcus* strain. In: Actinomycetes, Schaal, K.P. und Pulverer, G. (Eds.), Gustav Fischer Verlag, Stuttgart, New York, 297 – 307
- Backer, L.C., Dearfield, K.L., Erexson, G.L., Campbell, J.A., Westbrook-Collins, B. and Allen, J.W. (1989) The effects of acrylamide on mouse germ-line and somatic cell chromosomes. Environ. Mol. Mutagen. 13, 218 – 226
- Batchelder, T.L. (1975): NTIS/OTS 0206135 Doc. # 878210963 US Department of Commerce, Springfield, VA
- Bergmark, E., Calleman, C.J. and Costa, L.G. (1991) Formation of hemoglobin adducts of acrylamide and its epoxide metabolite glycidamide in the rat. Toxicol. Appl. Pharmacol. 111, 352 – 363
- Bergmark, E., Calleman, C.J., He, F. and Costa, L.G. (1993) Determination of hemoglobin adducts in humans occupationally exposed to acrylamide. Toxicol. Appl. Pharmacol. 120, 45-54
- Bilderback, D.E. (1981) Impatiens pollen germination and tube growth as a bioassay for toxic substances. Environ. Health Perspect. 37, 95 – 103

Bridie, A.L., Wolff, C.J.M. and Winter, M. (1979) The acute toxicity of some petrochemicals to goldfish. *Water Res.* 13, 623 – 626

Brown, L., Rhead, M.M. and Bancroft, K.C.C. (1980) Case studies of acrylamide pollution resulting from the industrial use of polyacrylamides. *Water Pollut. Control* 79, 507 – 510

Brown, L., Rhead, M.M., Bancroft, K.C.C. and Allen, N. (1980b) Model studies of the degradation of acrylamide monomer. *Water Res.* 14, 775 – 778

Brown, L., Bancroft, K.C.C. and Rhead, M.M. (1980c) Laboratory studies on the adsorption of acrylamide monomer by sludge sediments, clays, peat and synthetic resins. *Water Res.* 14, 779 – 781

Brown, L., Rhead, M.M., Hill, D. and Bancroft, K.C.C. (1982) Qualitative and quantitative studies on the *in situ* adsorption, degradation and toxicity of acrylamide by the spiking of the waters of two sewage works and a river. *Water Res.* 16, 579 – 591

Budavari, S. (ed.) *The Merck Index – Encyclopedia of Chemicals, Drugs and Biologicals*. Rahway, NJ: Merck and Co., Inc., 1989. 23

Bull, R.J., Robinson, M., Laurie, R.D., Stoner, G.D., Greisiger, E., Meier, J.R. and Stober, J. (1984) Carcinogenic effects of acrylamide in Sencar and A/J mice. *Cancer Res.* 44, 107 – 111

Burek, J.D., Albee, R.R., Beyer, J.E., Bell, T.J., Carreon, R.M., Morden, D.C., Wade, C.E., Hermann, E.A., Gorzinski, S.J. (1980) Subchronic toxicity of acrylamide administered to rats in the drinking water followed by up to 144 days of recovery. *J. Environm. Path. Toxicol.* 4, 157 – 182

Cabe, P.A. and Colwell, P.B. (1981) Toxic effects of acrylamide in Japanese quail (*Coturnix coturnix Japonica*). *J. Toxicol. Environ. Health* 7, 935-940

Calleman, C.J., Bergmark, E. and Costa, L.G. (1990) Acrylamide is metabolized to glycidamide in the rat: Evidence from hemoglobin adduct formulation. *Chem. Res. Toxicol.* 3(5), 406-412

Calleman, C.J., Bergmark, E., Stern, L.G. and Costa, L.G. (1993) A nonlinear dosimetric model for hemoglobin adduct formation by the neurotoxic agent acrylamide and its genotoxic metabolite glycidamide. *Environ. Health Perspectives* 99, 221-223

Calleman, C.J., Wu, Y. He, F., Tian, G., Bergmark, E., Zhang, S., Deng, H., Wang, Y., Crofton, K.M., Fennell, T. and Costa, L. (1994) Relationships between biomarkers of exposure and neurological effects in a group of workers exposed to acrylamide. *Toxicol. Appl. Pharmacol.* 126, 361-371

Carpenter, E.L. and Davis, H.S. (1957) Acrylamide. Its preparation and properties. *J. Appl. Chem.* 7, 671 – 676

Chet, I. and Mitchell, R. (1976) Control of marine fouling by chemical repellants. *Proc. Int. Biodegradation Symp.* 3rd ed., 515 – 521

Cihak, R. and Vontorkova, M. (1988) Cytogenetic effects of acrylamide in the bone marrow of mice. *Mutat. Res.* 209, 91 – 94

Collins, J.J., Swaen, G.M.H., Marsh, G.M., Utidjian, H.M.D., Caporossi, J.C. and Lucas, L.J. (1989) Mortality patterns among workers exposed to acrylamide. *J. Occup. Med.* 31, 614 – 617

Costa, L.G., Deng, H., Gregotti, C., Manzo, L., Faustman, E.M., Bergmark, E. and Calleman, C.J. (1992) Comparative studies on the neuro-and reproductive toxicity of acrylamide and its epoxide metabolite glycidamide in the rat. *NeuroToxicity* 13, 219-224

Croll, B.T., Arkell, G.M. and Hodge, R.P.J. (1974) Residues of acrylamide in water. *Water Res.* 8, 989 – 993

DOW (1975): DOW Chemical co., Midland, Michigan, NTIS/OTS 0206715 Doc. # 878214928. US Department of Commerce, Springfield, VA

DOW (1989): DOW Deutschland Inc., Werk Stade, Unveroeffentlichter Bericht vom 08.05.1989

Druckrey, H., Consbruch, U. and Schmaehl, D. (1953) Wirkungen von monomerem Acrylamid auf Proteine. *Z. Naturforsch.* 8b, 145 – 150

Edwards, P.M. (1975) Neurotoxicity of acrylamide and its analogues and effects of these analogues and other agents on acrylamide neuropathy. *Br. J. Ind. Med.* 32, 31 – 38

Edwards, P.M. (1976) The insensitivity of the developing rat fetus to the toxic effects of acrylamide. *Chem.-Biol. Inter-act.* 12, 13-18

EG & G Bionomics (1983): NTIS/OTS 0510507 Doc. # 40-8631566, US Department of Commerce, Springfield, VA

Eskin, T.A., Lapham, L.W., Maurissen, J.P.J. and Merigan, W.H. (1985) Acrylamide effects on the Macaque visual system. II. Retinogeniculate morphology. *Invest. Ophthalmol. Vis. Sci.* 26, 317 – 329

Field, E., Price, C.J., Sleet, R.B., Marr, M.C., Schwetz, B.A. and Morrissey, R.E. (1990) Developmental toxicity evaluation of acrylamide in rats and mice. *Fundamental and Applied Toxicology* 14, 502-512

Friedman, M., Fennell, T.R., Asgharian, B., Williams, C. and Sumner, S.J. (2001) Metabolism and distribution of acrylamide in rats and mice following inhalation exposure or dermal application. Society of Toxicology Annual Meeting 2001, Abstract No. 444, pg 93

Fujisawa, S. and Masuhara, E. (1980) Binding of methyl methacrylate to bovine serum albumin. *J. Dent. Res.* 59, 2056 – 2061

Fujisawa, S. and Masuhara, E. (1981) Determination of partition coefficients of acrylates, methacrylates, and vinyl monomers using high performance liquid chromatography (HPLC). *J. Biomed. Mater. Res.* 15, 787 – 793

Fullerton, P.M. and Barnes, J.M. (1966) Peripheral neuropathy in rats produced by acrylamide. *Brit. J. Industr. Med.* 23, 210 – 221

GEMS; Graphical Exposure Modeling System. FAP, Fate of Atmos. Pollut (1986).

Ghiringhelli, L. (1956) Studio comparativo sulla tossicita di alcuni nitrili e di alcune amidi. *Med. Lav.* 47, 192 – 199

Gilbert, S.G. and Maurissen, J.P.J. (1982) Assessment of the effects of acrylamide, methylmercury, and 2,5-hexanedione on motor functions in mice. *J. Toxicol. Environ. Health* 10, 31-41

Hansch, C. and Leo, A. (1979): Cited in: US EPA (1980): U.S. Environ. Prot. Agency, EPA-560/11-80-016, Washington, D.C., Order No. PB 80-220312, 1 – 33

Harris, C.H., Gulati, A.K., Friedman, M.A. and Sickles, D.W. (1994) Toxic neurofilamentous axonopathies and fast axonal transport. V. Reduced bidirectional vesicle transport in cultured neurons by acrylamide and glycidamide. *J. Toxicol. Environ. Health* 42, 343-356

Hashimoto, K. and Tanii, H. (1985) Mutagenicity of acrylamide and its analogues in *Salmonella typhimurium*. *Mut. Res.* 158, 129-133

Hashimoto, K., Sakamoto, J. and Tanii, H. (1981) Neurotoxicity of acrylamide and related compounds and their effect on male gonads in mice. *Arch. Toxicol.* 47, 179 – 189

Hazleton Laboratories America (1987) Plant growth study to estimate the potential for the uptake and accumulation of residual acrylamide monomer in plant tissue. HLA Study No. 6015-310.

Hermens, J. and Leeuwangh, P. (1982) Joint toxicity of 8 and 24 chemicals to the guppy (*Poecilia reticulata*). *Ecotoxicol. Environ. Safety* 6, 302 – 310

Hersch, M.I., McLeod, J.G., Satchell, P.M., Early, R.G. and Sullivan, C.E. (1989) Breathing pattern, lung inflation reflex and airway tone in acrylamide neuropathy. *Respir. Physiol.* 76, 257 – 276

Hoorn, A.J.W., Custer, L.L., Myhr, B.C., Brusick, D., Gossen, J. and Vijg, J. (1993) *Mutagenesis* 8(1), 7-10

Ikeda, G.J., Miller, E., Sapienza, P.P., Michel, T.C., King, M.T., Turner, V.A., Blumenthal, H., Jackson III, W.E. and Levin, S. (1983) Distribution of ¹⁴C-labelled acrylamide and betaine in foetuses of rats, rabbits, beagle dogs and miniature pigs. *Fd. Chem. Tox.* 21(1), 49-58

Ikeda, G.J., Miller, E., Sapienza, P.P., Michel, T.C., King, M.T. and Sager, A.O. (1985) Maternal-foetal distribution studies in late pregnancy. II. Distribution of [1-¹⁴C]Acrylamide in tissues of beagle dogs and miniature pigs. *Fd. Chem. Tox.* 23(8), 757-761

Ikeda, G.J., Miller, E., Sapienza, P.P., Michel, T.C. and Inskeep, P.B. (1987) Comparative tissue distribution and excretion of [1-¹⁴C]Acrylamide in tissues of beagle dogs and miniature pigs. *Fd. Chem. Tox.* 25(11), 871-875

Johnson, K.A., Gorzinski, S.J., Bodner, K.M. and Campbell, P.A. (1984) Acrylamide: A two-year drinking water chronic toxicity-oncogenicity study in Fischer 344 rats. Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, USA, Dow Chemical USA, Final Report, September 21, 1984.

Johnson, K.A., Gorzinski, S.J., Bodner, K.M., Campbell, R.A., Wolf, C.H., Friedman, M.A. and Mast, R.W. (1986) Chronic toxicity and oncogenicity study on acrylamide incorporated in the drinking water of Fischer 344 rats. *Toxicol. Appl. Pharmacol.* 85, 154 – 168

Jones, H.B. and Cavanagh, J.B. (1984) The axon reaction in spinal ganglion neurons of acrylamide-treated rats. *Neuropathol. Appl. Neurobiol.* 10, 101 – 121

Jung, D. et al. (1980): In: Ullmanns Enzyklopaedie der technischen Chemie, 4. Aufl., Bd. 19, 1 – 30

Kankaanpaa, J., Elovaara, E., Hemminki, K. and Vainio, H. (1979) Embryotoxicity of acrolein, acrylonitrile and acrylamide in developing chick embryos. *Toxicol. Lett.*, 4, 93-96.

Keefe, R.T. (1991) 28-day subchronic toxicity study in rats. EXXON Biomedical Sciences, Inc. No. 234870, Final Report April 2, 1991

Kirk-Othmer (1991): *Encyclopedia of Chemical Technology*, 4th ed. John Wiley & Sons, Vol. 1, 252-253

Klimisch, H.J., Andreae, M and Tillman, U. (1997) A systemic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology*. 25:1-5

Knaap, A.G.A.C., Kramers, P.G.N., Voogd, C.E., Bergkamp, W.G.M., Groot, M.G., Langebroek, P.G., Mout, H.C.A., van der Stel, J.J. and Verharen, H.W. (1988) Mutagenic activity of acrylamide in eukaryotic systems but not in bacteria. *Mutagenesis* 3, 263 – 268

Krebs, O and Favor, J. (1997) Somatic and germ cell mutagenesis in lambda *lacZ* transgenic mice treated with acrylamide or ethylnitrosourea. *Mutat. Res.* 388, 239-248

Kuboi, T. and Fujii, K. (1984) Toxicity of cationic polymer flocculants to higher plants. I. Seedling assay. *Soil Sci. Plant Nutr.* 30, 311 – 320

Lande, S.S., Bosch, S.J. and Howard, P.H. (1979) Degradation and leaching of acrylamide in soil. *J. Environ. Quality* 8(1) 133-137

Lewis, R.J. (2000): *Sax's Dangerous Properties of Industrial Materials*, 10th Edition. John Wiley & Sons, Inc., New York, Chichester, Weinheim, Brisbane, Singapore, Toronto.

Lipnick, R.L. , Watson, K.R., and Strausz, A.K. (1987) A QSAR study of the acute toxicity of some industrial organic chemicals to goldfish. Narcosis, electrophile and proelectrophile mechanisms. *Xenobiotica* 17, 1011 – 1025

MacWilliams, D.C. (1978): In: Kirk-Othmer: *Encyclopedia of Chemical Technology*, 3rd ed., Vol. 1, 298 – 311

Marsh, G.M., Lucas, L.J., Youk, A.O. and Schall, L.C. (1998) Mortality patterns among workers exposed to acrylamide: 1994 Follow-up. Unpublished

Matthews, R.W. and Sangster, D.F. (1965) Measurement by benzoate radiolytic decarboxylation of relative rate constants for hydroxyl radical reactions. *J. Phys. Chem.* 69, 1938 – 1946

Maurissen, J.P.J., Weiss, B. and Davis, H.T. (1983) Somatosensory thresholds in monkeys exposed to acrylamide. *Toxicol. Appl. Pharmacol.* 71, 266-279

McCollister, D.D., Oven, F. and Rowe, V.K. (1964) Toxicology of acrylamide. *Toxicol. Appl. Pharmacol.* 6, 172 – 181

Mercier, O. (1997a) Acrylamide – Primary cutaneous irritation and corrosivity test in the rabbit (P.C.I.C.) – 3 rabbits. *Pharmakon Europe Report No.* 59996

Mercier, O. (1997b) Acrylamide – Ocular irritation and reversibility test in the rabbit (O.I.R.) – 3 rabbits. Pharmakon Europe Report No. 60096, February 26, 1997

Miller, M.J. and McQueen, C.A. (1986) The effect of acrylamide on hepatocellular DNA repair. *Environ. Mutagen.* 8, 99 – 108

Moens, J. and Smets, G. (1957) Alkaline and acid hydrolysis of polyvinylamides. *J. Polymer Sci.* 23, 931 – 948

Moore, M.M., Amtower, A., Doerr, C., Brock, K.H. and Dearfield, K.L. (1987) Mutagenicity and clastogenicity of acrylamide in L5178Y mouse lymphoma cells. *Environ. Mutagen.* 9, 261 – 267

Murti, J. R., Schimenti, K. J. and Schimenti, J.C. (1994) A recombination –based transgenic mouse system for genotoxicity testing. *Mutat. Res.* 307, 583-595

Neuhaeuser-Klaus, A. and Schmahl, W. (1989) Mutagenic and teratogenic effects of acrylamide in the mamalian spot test. *Mutat. Res.* 226, 157 – 162

Newton, P.E., Schroeder, R.E., Sullivan, J.B., Busey, W.M. and Banas, D.A. (1993) Inhalation toxicity of phosphine in the rat: acute, subchronic, and developmental. *Inhalation Toxicology* 5(2), 223-239

Novikova, E.E. (1979) Toxic effect of acrylamide entering through skin surfaces (Russ.) *Gig. Sanit.* 10, 73 - 74. Cited in: IARC (1986): IARC Monogr. Eval. Carcinog. Risk Chem. Hum. 39, 41 - 66.

Paulet, G. and Vidal M. (1975) De la toxicite de quelques esters acryliques et methacryliques de l'acrylamide et des polyacrylamides. *Arch. Mal. Prof. Med. Trav.* 36, 58 – 60

Petersen, D.W. and Lech, J.J. (1987) Hepatic effects of acrylamide in rainbow trout. *Toxicol. Appl. Pharmacol.* 89, 249 – 255

Petersen, D.W., Cooper, K.R., Friedman, M.A. and Lech, J.J. (1987) Behavioral and histological effects of acrylamide in rainbow trout. *Toxicol. Appl. Pharmacol.* 87, 177 - 184

Petersen, D.W., Kleinow, K.M., Kraska, R.C. and Lech, J.J. (1985) Uptake, disposition, and elimination of acrylamide in rainbow trout. *Toxicol. Appl. Pharmacol.* 80, 58 – 65

Sakamoto, J. and Hashimoto, K. (1986) Reproductive toxicity of acrylamide and related compounds in mice – effects on fertility and sperm morphology. *Arch. Toxicol.* 59, 201-205

Satchell, P. (1985) Reversible abnormalities of the Hering Breuer reflex in acrylamide neuropathy. *J. Neurol. Neurosurg. Psychiat.* 48, 670 – 675

Satchell, P.M. and McLeod, J.G. (1981) Megaesophagus due to acrylamide neuropathy. *J. Neurol. Neurosurg. Psychiat.* 44, 906-913

Schulze, G.E. and Boysen, B.G. (1991) A neurotoxicity screening battery for use in safety evaluation: effects of acrylamide and 3',3'-Iminodipropionitrile. *Fundamental and Applied Toxicology* 16, 602-615

Sega, G.A., Generoso, E.E., and Brimer, P.A. (1990) Acrylamide exposure induces a delayed unscheduled DNA synthesis in germ cells of male mice that is correlated with the temporal pattern of adduct formation in testis DNA. *Environ. Mol. Mutagen.* 16, 137 – 142

- SEPC (1997) Inhibition test (72 hours) in freshwater unicellular algae *Selenastrum capricornutum*. Company report G104
- Shanker, R. and Seth, P.K. (1986) Toxic effects of acrylamide in a freshwater fish, *Heteropneustes fossilis*. Bull. Environ. Contam. Toxicol. 37, 274 – 280
- Shelby, M.D., Cain, K.T., Cornett, C.V. and Generoso, W.M. (1987) Acrylamide: Induction of heritable translocations in male mice. Environ. Mutagen. 9, 363 – 368
- Shelby, M.D., Cain, K.T., Hughes, L.A., Braden, P.W. and Generoso, W.M. (1986) Dominant lethal effects of acrylamide in male mice. Mutat. Res. 173, 35 – 40
- Shiraishi, Y. (1978) Chromosome aberrations induced by monomeric acrylamide in bone marrow and germ cells of mice. Mutat. Res. 57, 313 – 324
- Sickles, D.W. (1989) Toxic neurofilamentous axonopathies and fast anterograde axonal transport. I. The effects of single doses of acrylamide on the rate and capacity of transport. NeuroToxicology 10, 91 – 101
- Sickles, D.W. (1992) Toxic neurofilamentous axonopathies and fast anterograde axonal transport. IV. In vitro analysis of transport following acrylamide and 2,5-hexanedione. Toxicology Letters 61, 199-204
- Sickles, D.W., Brady, S.T., Testino, A., Friedman, M.A. and Wrenn, R.W. (1996) Direct effect of the neurotoxicant acrylamide on kinesin-based microtubule motility. J of Neurosci Res 46, 7-17
- Sickles, D.W., Welter, D.A. and Friedman, M.A. (1994) Acrylamide arrests mitosis and prevents chromosome migration in the absence of changes in spindle microtubules. Accepted 17 June 1994, 73-86
- Smith, M.K., Zenick, H., Preston, R.J., George, E.L. and Long, R.E. (1986) Dominant lethal effects of subchronic acrylamide administration in the male Long-Evans rat. Mutat. Res. 173, 273 – 277
- Spraggs, L.D., Gehr, R. and Hadjinicolaou, J. (1982) Polyelectrolyte toxicity tests by fish avoidance studies. Water Sci. Tech. 14, 1564 – 1567
- Springborn Bionomics (1985): NTIS/OTS 0510508 Doc. # 40-8631565, US Department of Commerce, Springfield, VA
- Starostina, N.G., Lusta, K.A. and Fikhte, B.A. (1983) Morphological and physiological changes in bacterial cells treated with acrylamide. Eur. J. Appl. Microbiol. Biotechnol. 18, 264 – 270
- Stockhausen (1995) Skin sensitisation of acrylamide (50%) on guinea pigs (Pirbright White). Final Report No. 12006, June 30, 1995.
- Sublet, V.H., Zenick, H. and Smith, M.K. (1989) Factors associated with reduced fertility and implantation rates in females mated to acrylamide treated rats. Toxicology 55, 53 – 67
- Syracuse Research Corporation (1998) Syracuse, NY, Pollution Prevention (P2) Assessment Framework, U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics (Draft) – EPIWIN Generated Printout

Tanii, H. and Hashimoto, K. (1983) Neurotoxicity of acrylamide and related compounds in rats: Effects on rotarod performance, morphology of nerves and neurotubulin. Arch. Toxicol. 54, 203-213

The Merck Index (1996) An Encyclopedia of Chemicals, Drugs, and Biologicals, 12th ed. Merck & Co., Inc. Whitehouse Station, NJ, 23-25

Thomann, P., Koella, W.P., Krinke, G., Petermann, H., Zak, F. and Hess, R. (1974) The assessment of peripheral neurotoxicity in dogs: Comparative studies with acrylamide and clioquinol. Agents Actions 4, 47 – 53

Thomas, W.M. (1964): In: Encyclopedia of Polymer Science and Technology, John Wiley & Sons, Vol. 1, 177 - 181, 195 – 197

Tilson, H.A. and Cabe, P.A. (1979a) The effects of acrylamide given acutely or in repeated doses on fore- and hindlimb function of rats. Toxicol. Appl. Pharm. 47, 253 – 260

Tilson, H.A. and Cabe, P.A. (1979b) Acrylamide neurotoxicity in rats: A correlated neurobehavioral and pathological study. Neurotoxicology 1, 89 – 104

Tooby, T.E. and Hursey, P.A. (1975) The acute toxicity of 102 pesticides and miscellaneous substances to fish. Chem. Ind., Heft 12, 523 – 526

Tsuda, H., Shimizu, C.S., Taketomi, M.K., Hasegawa, M.M., Hamada, A., Kawata, K.M. and Inui, N. (1993) Acrylamide induction of DNA damage, chromosomal aberrations and cell transformation without gene mutations. Mutagenesis 8(1), 23-29

Tyl, R.W., Friedman, M.A., Losco, P.E., Fisher, L.C., Johnson, K.A., Strother, D.E. and Wolf, C.H. (2000a) Rat two-generation reproduction and dominant lethal study of acrylamide in drinking water. Reproductive Toxicol. 14, 000-000

Tyl, R.W., Marr, M.C., Myers, C.B., Ross, W.P. and Friedman, M.A. (2000b) Relationship between acrylamide reproductive and neurotoxicity in male rats. Reproductive Toxicol. 14, 147-157

United States Testing Company, Inc. (1990) Aquatic toxicity tests verses *Onchorhynchus mykiss*. Report of Test No. 063102-4

United States Testing Company, Inc. (1991) Modified OECD test for ready biodegradability. Test Report No. 063102-4

US EPA (1980) TSCA Chemical Assessment Series: Assessment of Testing Needs: Acrylamide. EPA-560/11-80-016, U.S. Environ. Prot. Agency, Washington, D.C., Order No. PB 80-220312, 1 – 33

Van der Burg, J.H.N. (1922) Sur la preparation de l'acide acrylique et de quelques-uns de ses derives. Rec. Trav. Chim. Pays-Bas 41, 21 – 23

Vanhorick, M. and Moens, W. (1983) Carcinogen-mediated induction of SV40 DNA amplification is enhanced by acrylamide in Chinese hamster CO60 cells. Carcinogenesis 4, 1459 – 1463

Vasavada, H.A. and Padayatty, J.D. (1981) Rapid transfection assay for screening mutagens and carcinogens. Mutat. Res. 91, 9 – 14

6. References

Id 79-06-1

Date 7 September 2001

Vernon, P., Dulak, L, and Deskin, R. (1990) Acute toxicologic evaluation of acrylamide 50% solution. J. Am. Coll. Toxicol. 1(2) Part B, 115-116

Walden, R., Squibb, R.E., and Schiller, C.M. (1981) Effects of prenatal and lactational exposure to acrylamide on the development of intestinal enzymes in the rat. Toxiol. Appl. Pharmacol. 58, 363-369

Winter, M. and Wolff, C.J.M. (1982): NTIS/OTS 0206200 Doc. # 878210096, US Department of Commerce, Springfield, VA.

Woodiwiss, F.S. and Fretwell, G. (1974) The toxicities of sewage effluents, industrial discharges and some chemical substances to brown trout (*Salmo trutta*) in the Trent River Authority Area. Water Pollut. Control 73, 396 – 405

Yamada, H., Asano, Y., Hino, T. and Tani, Y. (1979) Microbial utilization of acrylonitrile. J. Ferment. Technol. 57, 8 – 14

Zeiger, E., Anderson, B., Haworth, S., Lawlor, T., mortelmans, K. and Speck, W. (1987): *Salmonella* mutagenicity tests: III. Results from the testing of 255 chemicals. Environ. Mutagen. 9, Supplement 9, 1 - 3, 11 - 12, 19, 29 - 30

Zenick, H., Hope, E. and Smith, M.K. (1986) Reproductive toxicity associated with acrylamide treatment in male and female rats. J. Toxicol. Environ. Health 17, 457 – 472

7.1 END POINT SUMMARY

7.2 HAZARD SUMMARY

7.3 RISK ASSESSMENT

7 SEPTEMBER 2001

Acrylamide, N-(hydroxymethyl)-

NMA

Data Set

New Chemical : Acrylamide N-(hydroxymethyl)-
CAS No. : 924-42-5
Molecular Formula : C₄H₇NO₂
Molecular Weight : 101.10

Printing Date : 7 September 2001
Originally prepared in IUCLID 3.1

1.2 SYNONYMS

Acrylamide, N-(Hydroxymethyl)-

N-(Hydroxymethyl)-2-Propenamide

N-Methanolacrylamide

N-Methylolacrylamide

Monomethylolacrylamide

NCI-C60333

2-Propenamide, N-(Hydroxymethyl)-

Uramine T 80

NMA

2.1 MELTING POINT

Value : = 69.5 ° C
Sublimation :
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : The melting point is estimated by the EPIWIN/MPBPWIN model, using Joback, Gold and Ogle methods
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997)(Syracuse Research Corporation, 1998)

Value : = 74 - 75 ° C
Reliability : (1) valid without restrictions
(Feurr and Lynch 1953)

2.2 BOILING POINT

Value : = 276.50° C
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(hydroxymethyl)-
Remark : The boiling point is estimated by the EPIWIN/Stein and Brown Method.
Reliability : (2) valid with restrictions. Data were obtained by modeling
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

Value : = 100 ° C
Test substance : Other TS: 40-44% NMA Monomer, 48% Aqueous, Inhibited
Reliability : (2) valid with restrictions. Details on experimental conditions are not present.
(Cyttec MSDS No. 05741)

2.4 VAPOUR PRESSURE

Value : 0.00023 mg Hg at 25 ° C
Decomposition :
Method : other (calculated): Estimated by the MPBPWIN Program (v.1.40), using various methods (Antoine, Modified Grain, and Mackay Methods)
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : Other VP = 0.000207 mm Hg (Antoine Method) and 0.00448 mm Hg (Mackay Method) The vapor pressure calculated by an accepted methods assigned a reliability code of 2f according to the criteria established by Klimisch et al. (1997). These values were determined using an estimated boiling point of 276.50 ° C and an estimated melting point of 69.46 ° C.
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997)(Syracuse Research Corporation, 1998)

2. Physico-Chemical Data

Id 924-42-5

Date

Value : 23.76 mm Hg @ 25 ° C
Test substance : Other TS: 40-44% NMA Monomer, 48% Aqueous, Inhibited
Reliability : (2) valid with restrictions. Details on experimental conditions are not present.

(Cytec MSDS No. 05741)

2.5 PARTITION COEFFICIENT

Log pow : = -1.81
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : The log Kow was estimated using EPIWIN/KOWWIN based on molecular structure and functionality.
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997)(Syracuse Research Corporation, 1998)

2.6.1 WATER SOLUBILITY

Value : = 122 g/100 ml at 10 ° C – Water
: = 188 g/100 ml at 20 ° C – Water
: = 354 g/100 ml at 40 ° C – Water
: = 755 g/100 ml at 60 ° C – Water
: = 149 g/100 ml at 30 ° C – Methanol
: = 77 g/100 ml at 30 ° C – Ethanol, Absolute
: = 116 g/100 ml at 30 ° C – Ethanol – 90%
: = 53 g/100 ml at 30 ° C – Isopropanol
: = 42 g/100 ml at 30 ° C – n-Butanol
: = 12 g/100 ml at 30 ° C – Acrylonitrile
: = 2 g/100 ml at 30 ° C – Vinyl Acetate
: = 3 g/100 ml at 30 ° C – Methyl Methacrylate
: = 3 g/100 ml at 30 ° C – Ethyl Acrylate
Year : 1995
GLP : No data
Test substance : N-methylolacrylamide
Reliability : (1) valid without restrictions

(Cytec Industries, Inc., 1995)

Value : = 188 g/100 ml at 20 ° C – Water
: = 149 g/100 ml at 30 ° C – Methanol
: = 116 g/100 ml at 30 ° C – 9% Ethanol
: = 53 g/100 ml at 30 ° C – Isopropanol
: = 42 g/100 ml at 30 ° C – n-Butanol
Year : 1990
GLP : No data
Test substance : Acrylamide N-(hydroxymethyl)- 48% aqueous solution
Reliability : (1) valid without restrictions

(American Cyanamid Company, 1990)

3.1.1 PHOTODEGRADATION

Deg. Product : Not specified
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : The photodegradation rate was estimated by EPIWIN/AOP program which estimates rate constants and half-lives of atmospheric reactions of organic compounds with hydroxyl radicals and ozone in the atmosphere.
Result : Overall OH Rate Constant = 28.2479 E-12 cm³/molecule-sec;
Half-Life = 0.379 days (12-hr day- 1.5E6 OH/cm³); Half Life = 4.544 Hours
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997)(Syracuse Research Corporation, 1998)

3.1.2 STABILITY IN WATER

Type : Abiotic
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : Estimated by the EPIWIN/HYDROWIN program.
This program was not able to estimate a hydrolysis rate constant for this type of chemical structure. However, this compound has an amide group: C=O located at SMILES atom #: 2; Hydrolysis rate extremely slow or t_{1/2} > 1 year.
Result : No estimate available.
Reliability : (2) valid with restrictions. Data were obtained by modeling
(Syracuse Research Corporation, 1998)

Type : Abiotic
Year : 1953
GLP : Pre-GLP
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : Aqueous solutions are highly reactive upon heating in the presence of acids, they are rapidly polymerized to infusible resins.
Result :
Reliability : (1) valid without restrictions
(Feurr and Lynch, 1953)

Type : Abiotic
Year : 1990
GLP : No data
Test substance : Acrylamide N-(hydroxymethyl)- 48% Aqueous solution
Remark : The stability of solutions is dependent mainly on oxygen level, contaminants, storage temperature, and pH.
Result :
Reliability : (1) valid without restrictions
(American Cyanamid Company, 1990)

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

| | |
|-----------------------------|--|
| Type | : Fugacity model level III |
| Media | : |
| Air (level III) | : 0.000307 |
| Water (level III) | : 45.3 |
| Soil (level III) | : 54.6 |
| Sediment (level III) | : 0.0755 |
| Method | : Other: Estimated by the Level III Fugacity Model (Full-Output) |
| Year | : 2001 |
| GLP | : No |
| Remark | : Level III fugacity was estimated using the EPIWIN/Fugacity Model (Full-Output) |
| Reliability | : (2) valid with restrictions. Data were obtained by modeling (Klimisch et al., 1997)(Syracuse Research Corporation, 1998) |

3.5 BIODEGRADATION

| | |
|----------------------------------|--|
| Type | : Aerobic |
| Inoculum | : activated sludge |
| Contact time | : 28 day |
| Degradation | : = 51.9 % after 28 day |
| Result | : not readily biodegradable |
| Kinetic of test substance | : 5 day 0 % 15 day 16.7 % 28 day 51.9 % |
| Control substance | : Aniline |
| Kinetic | : 5 day 18.5 % 15 day 66.7 % 28 day 98.1 % |
| Method | : OECD Guide-line 301 D "Ready Biodegradability: Closed Bottle Test" |
| Year | : 1991 |
| GLP | : Yes |
| Test substance | : other TS |
| Result | : The sample stock solution fell within the organic content range stated in the formula. The sample containing 1 mg/l degraded by 0%, 0% and 40.7% over 5, 15 and 28 days, respectively. The sample containing 2 mg/l degraded by 0%, 16.7% and 51.9% over 5, 15 and 28 days, respectively. The sample containing 5 mg/l degraded by 0%, 14.8% and 40.7% over 5, 15 and 28 days, respectively. The average degradation for the three concentrations was 0%, 10.5% and 44.43% over 5, 15 and 28 days, respectively. Aniline degraded by 18.5, 66.7 and 98.1% over the three time periods. Because a level of 70% was not reached, the test substance is not "Readily Biodegradable" by this test procedure. |
| Test condition | : Stock solution was prepared by adding 2 g of sample to 1 liter of distilled water. The stock solution was screened to determine if it had a similar percent carbon content as stated in the formula provided by the supplier. Stock solution was diluted to 100 ppm as carbon after analysis. The diluted stock was then added to BOD bottles at 3.33 ml, 6.67 ml and 16.65 ml to yield test concentrations of 1 mg, 2 mg and 5 mg as carbon, respectively. Test solutions were inoculated with a low concentration of microorganisms from a mixed population and kept in closed bottles in the dark at a constant temperature of 20 ± 1° C. The activated sludge bacteria was from Bergen Co., New Jersey. The degradation was followed by oxygen analyses with |

the YSI Dissolved Oxygen Analyzer 54A over a 28-day period. Degradability was based on a comparison of readings of actual oxygen demand to the theoretically expected oxygen demand. A parallel control with inoculum, but without test material, was run as a blank correction factor. The procedure was validated by means of a reference substance (aniline, 2 mg/l) of known biodegradability.

Test Substance : H₂O (~50-54%), C₄H₇O₂N (~40-44%), C₃H₅NO (<5%), CH₂O (<3%), C₇H₈O₂ (~0.003%), Cu.H₂O₄S (~0.0002%). Carbon content was 23.7%.

Reliability : (1) valid without restriction (Wang 1991)

Deg. Product :
Method : Other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide N-(hydroxymethyl)-
Remark : The biodegradation rate was estimated using EPIWIN/STP Fugacity Model: Predicted Fate in a Wastewater Treatment Facility.
Result : Total biodegradation is predicted to be 0.09%. The material is not considered readily biodegradable.
Reliability : (2) valid with restrictions. Data were obtained by modeling (Klimisch et al., 1997)(Syracuse Research Corporation, 1998)

4.1 ACUTE TOXICITY TO FISH

| | |
|------------------------------|--|
| Type | : Static |
| Species | : Rainbow trout |
| Exposure period | : 96 hours |
| Concentrations | : 0, 625, 1,250, 2,500, 5,000, 10,000 ppm |
| Analytical monitoring | : |
| LC50 | : 890 ppm |
| Method | : OECD method 203 |
| Statistical method | : ASTM STP634, Thompson 1947, Litchfield and Wilcoxin, 1949 |
| Year | : 1990 |
| GLP | : Yes |
| Test substance | : 40-44% pure |
| Test condition | : This test was conducted in EPA moderately hard reconstituted water. Ten fish (5/replicate) were exposed to the following nominal test concentrations: 0, 625, 1,250, 2,500, 5,000, or 10,000 ppm. Water temperature, dissolved oxygen and pH were measured throughout the test and were within acceptable limits. Temperature was maintained at 15°C |
| Result | : At 96 hours, there was no mortality in the control and 635 ppm group. All fish died in the other dose groups. Using the Acute-Toxicity Rating Scale published by the US Fish and Wildlife Service, this is practically non-toxic. The NOEL is 625 ppm. The 24-hour LC50=2,900 ppm, 48-hour LC50=1,350 ppm, the 72-hour and 96-hour LC50 = 890 ppm |
| Reliability | : (1) reliable without restriction (Cooke 1990) |

| | |
|------------------------------|--|
| Type | : Static |
| Species | : Rainbow trout |
| Exposure period | : 96 hours |
| Concentrations | : 0, 12.5, 25, 50, 100 |
| Analytical monitoring | : |
| LC50 | : >100 ppm |
| Method | : |
| Statistical method | : Harris, 1959, Litchfield and Wilcoxin, 1948, Hamilton et al, 1977 |
| Year | : 1990 |
| GLP | : Yes |
| Test substance | : 100% pure |
| Test condition | : This test was conducted in chemically dechlorinated Milwaukee tap water. The mean weight of the fish was 1.64 g (sd=0.0209 g). EPA moderately hard reconstituted water. Ten fish (5/repliacte) were exposed to the following nominal test concentrations: 0, 12.5, 25, 50, or 100 ppm. Water temperature, dissolved oxygen and pH were measured throughout the test and were within acceptable limits. |
| Result | : At 96 hours, there was no mortality in any of the groups. The LC50 is estimated to be higher than the highest does (100 ppm). The NOEC = 100 ppm. |
| Reliability | : (1) reliable without restriction (Medical College of Wisconsin Great Lakes Research Facility, 1990) |

4.2 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

No data

4.3 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

No data

5.1.1 ACUTE ORAL TOXICITY

Type : LD50
Species : Mouse
Strain : DdY
Sex : Male
Number of animals : 4
Vehicle : Saline
Route of admin. : Oral
Exposure time : Single dose
Value : 677 mg/kg bw
Method : Weil, 1952
Year : 1981
GLP : No data
Test substance : 95 % pure
Test condition : 4 mice/dose group, 4 unspecified dosage groups
Result : LD50 is 677 mg/kg
Conclusion :
Reliability : (2) Reliable with restriction, doses not indicated, clinical signs not provided (Hashimoto et al. 1981)

Type : LD50
Species : Rat
Strain : Fischer 344
Sex : Male/female
Number of animals : 50 (5 sex/group)
Vehicle : Water
Doses : 50, 100, 200, 400, 800 mg/kg
Method : Acute Oral Toxicity
Route of Administration : Gavage
Year : March 16, 1981
GLP : Yes
Test substance : >98% pure
Test condition : Groups of 5 rats per sex were dosed with 50, 100, 200, 400 or 800 mg/kg. Male rats ranged from 101-128 g starting body weight, females weighed from 84-102 g at the study initiation. Animals received a single dose in a concentration 5.0 ml/kg. Animals were observed twice a day for clinical signs. After 14 days observation, surviving animals were terminated without necropsy or histopathology. No clinical pathology was performed.
Result : All rats in the 800 mg./kg group died on or before Day 2. In the 400 mg/kg group, four males and three females died during the first week of observation. No abnormal clinical signs were noted. LD50 = 400 mg/kg
Conclusion :
Reliability : (1) reliable without restriction (Batelle 1981a)

Type : LD50
Species : Rat
Strain : No data
Sex : No data
Number of animals : No data
Vehicle : No data
Method : Acute Oral Toxicity
Year : 1979
GLP : No data

5. Toxicity

Id 924-42-5

Test substance : No data
Test condition : No data
Result : LD50 = 474 mg/kg
Conclusion :
Reliability : (2) reliable with restriction, primary source not in English, details of study were not available for review
02.05.01 (Japanese Journal of Hygiene 1979)

Type : LD50
Species : Mouse
Strain : B6C3F₁
Sex : Male/Female
Number of animals : 50 (5/sex/group)
Doses : 50, 100, 200, 400, 800 mg/kg
Vehicle : Water
Route of Administration : Gavage
Value : LD50 = 400 mg/kg bw
Method : Acute Oral Toxicity
Year : 1981
GLP : Yes
Test substance : >98 % pure
Test conditions : Groups of 5 mice per sex were dosed with 50, 100, 200, 400 or 800 mg/kg. Male mice ranged from 20 to 24 g starting body weight, females weighed from 18-19 g at the study initiation. Animals received a single dose in a concentration 5.0 mL/kg. Animals were observed twice a day for clinical signs. After 14 days observation, surviving animals were terminated without necropsy or histopathology. No clinical pathology was performed.
Result : All male mice in the 400 and 800 mg/kg dose group died by or on Day 2 of the study. The remaining males survived to Day 14. All females in the 800 mg/kg group died on or before Day 2. One female died on Day 2 in the 400 mg/kg dose group. All other females survived to Day 14. LD50 = 400 mg/kg.
Conclusion :
Reliability : (1) reliable without restriction (Batelle 1981b)

Type : LD50
Species : Mouse
Strain : Albino
Sex : Male
Number of animals : No data
Doses : No data
Vehicle : No data
Route of Administration : Gavage
Value : LD 50 = 420 mg/kg
Method : Acute Oral Toxicity
Year : 1953
GLP : Pre-GLP
Test substance : 97.5% pure
Test conditions : Test article was administered in a single dose as a 1% aqueous solution to male albino mice

Result : Most deaths occurred within 48 hours of administration of the dose. Following administration symptoms in the range of the LD50 included ataxia, loss of pain and righting reflexes and mild tonic and clonic convulsions prior to death. Examination of animals that died revealed hemorrhagic or hyperemic lungs and irritated intestines. All survivors had normal behavior after 48 hours of administration. After a one-week observation period, no significant gross pathology was noted.

Conclusion Reliability : (2) reliable with restriction. Based on summary report. No data on doses, strain, or number of animals was available
(Cyanamid Report 53-82, 1953)

5.1.2 ACUTE INHALATION TOXICITY

Type : Acute
Species : Mice
Strain : No data
Sex : No data
Number of animals : 7
Vehicle :
Exposure time : 6 hour(s)
Concentration : 9.5 ppm (39 mg/m³)
Value :
Method : Acute inhalation toxicity
Year : 1953
GLP : Pre-GLP
Test substance : 97.5% pure
Test condition : Mice were exposed for 6 hours to near saturated vapor of the compound produce by passing air at a rate of 12 L/min. through a column packed with the compound. This air-stream was led into an exposure chamber with a 160 L capacity.

Result : No clinical signs of toxicity were noted. At necropsy, no significant gross or histopathological findings were noted.

Conclusion Reliability : (2) Reliable with restriction, information on strain not provided, LC50 not established.
(Vernon et al 1990)

Type : Acute
Species : Rat
Strain : No data
Sex : No data
Number of animals : 7
Vehicle : No data
Exposure time : 6 hour(s)
Concentration : 9.5 ppm (39 mg/m³)
Value : No data
Method : No data
Year : 1953
GLP : Pre-GLP
Test substance : 97.5% pure
Test condition : Rats were exposed for 6 hours to near saturated vapor of the compound produce by passing air at a rate of 12 L/min. through a column packed with the compound. This air-stream was led into an exposure chamber with a 160 L capacity.

Result : No clinical signs of toxicity were noted. At necropsy, no significant gross or histopathological findings were noted.

Conclusion :
Reliability : (2) Reliable with restriction, information on strain not provided, LC50 not established

(Vernon et al 1990)

Type : Acute
Species : Guinea pig
Strain : No data
Sex : No data
Number of animals : 7
Vehicle : No data
Exposure time : 6 hour(s)
Value : No data
Concentration : 9.5 ppm (39 mg/m³)
Method : No data
Year : 1953
GLP : Pre-GLP
Test substance : 97.5% pure
Test condition : Guinea pigs were exposed for 6 hours to near saturated vapor of the compound produce by passing air at a rate of 12 L/min. through a column packed with the compound. This air-stream was led into an exposure chamber with a 160 L capacity.

Result : No clinical signs of toxicity were noted. At necropsy, no significant gross or histopathological findings were noted.

Conclusion :
Reliability : (2) Reliable with restriction, information on strain not provided, LC50 not established

(Vernon et al 1990)

5.1.3 ACUTE DERMAL TOXICITY

Type : LD50
Species : Rabbit
Strain : Albino
Sex : No data
Number of animals : 16 (4 groups of 4)
Vehicle : Water
Value : Moderate dermal irritant
Method
Year : 1953
GLP : Pre GLP
Test substance : 97.5% pure
Test condition : 2, 4, 8, or 16 g/kg of test material was moistened with water to form a paste. The material was occlusively patched to intact skin for 24 hours. Following 24-hour administration, the patch was removed and any remaining test article was removed. The animals were observed for mortality and clinical signs for 5-7 days following dose administration. One animal in the high-dose group died shortly after exposure period. Tremors and hind limb impairment was observed prior to death. No other mortality, clinical signs or gross necropsy findings were noted. LD50 = >16 g/kg

Result : One animal in the high-dose group died shortly after exposure period. Tremors and hind limb impairment was observed prior to death. No other mortality, clinical signs or gross necropsy findings were noted. LD50 = >16 g/kg

Conclusion
Reliability : (1) reliable without restriction

(Vernon et al 1990)

5.1.4 ACUTE TOXICITY, OTHER ROUTES

5.2.1 SKIN IRRITATION

| | | |
|--------------------------|---|--|
| Type | : | Dermal irritation |
| Species | : | Rabbit |
| Strain | : | Albino |
| Sex | : | No data |
| Number of animals | : | 16 (4 groups of 4) |
| Vehicle | : | Water |
| Value | : | Moderate dermal irritant |
| Method | : | Occlusive patch |
| Year | : | 1953 |
| GLP | : | Pre GLP |
| Test substance | : | 97.5% pure |
| Test condition | : | 2, 4, 8, or 16 g/kg of test material was moistened with water to form a paste. The material was occlusively patched to intact skin for 24 hours. Following 24-hour administration, the patch was removed and any remaining test article was removed. The animals were observed for mortality and clinical signs for 5-7 days following dose administration. |
| Result | : | One animal in the high-dose group died shortly after exposure period. Tremors and hind limb impairment was observed prior to death. No other mortality, clinical signs or gross necropsy findings were noted. There was some incidence of skin irritation at all dose levels (erythema, edema, and desquamation). The highest dose produced caustic burns. The material is moderately irritating |
| Conclusion | : | |
| Reliability | : | (2) reliable with restriction, individual irritation scores not available (Vernon et al., 1990) |

5.2.2 EYE IRRITATION

| | | |
|--------------------------|---|--|
| Type | : | Primary Eye Irritation |
| Species | : | Rabbit |
| Strain | : | Albino |
| Sex | : | No data |
| Number of animals | : | Three |
| Concentration | : | No data |
| Dose | : | 3 mg |
| Exposure Time | : | 5 days |
| Number of animals | : | 3 |
| Method | : | No data |
| Value | : | Minimal eye irritant |
| Year | : | 1953 |
| GLP | : | Pre-GLP |
| Test substance | : | 97.5% pure |
| Test condition | : | 3 mg of test material (a dry powder) was placed into the left eye of 3 rabbits. The eyes remained unwashed. The right eye was not treated. The eyes were examined over a 5-day observation period. |
| Result | : | Mild irritation was noted immediately following application. Within 1 hour of application all eyes were free of irritation. |
| Conclusion | : | |
| Reliability | : | (1) reliable with restriction (Vernon et al., 1990) |

5.3 SENSITIZATION

No data found

5.4 REPEATED DOSE TOXICITY

Species : Rat
Sex : Male/Female
Strain : F344/N
Route of admin. : Gavage
Exposure period : 16 days
Frequency of treatment : 12 doses over 16 days
Post obs. period :
Doses : 0, 25, 50, 100, 200, 400 mg/kg/day
Control group : Yes, concurrent control
NOEL : 50 mg/kg/day
Method :
Year : 1981
GLP : Yes
Test substance : 98% pure
Test Condition : Animals received 12 doses over 16 days with 0, 25, 50, 100, 200, 400 mg/kg/day (5/sex/group). All animals were observed twice daily. At the end of treatment all surviving animals were subject to complete necropsy.

Result : NOEL = 50 mg/kg. In the top dose group (400 mg/kg) all male and female animals died within the first 4 days of dosing. Before death all animals showed increased motor activity and startle reactivity. In the 200 mg/kg dose group, 3 males died during the study. Both sexes exhibited ataxia, muscle tremors and hyperirritability to handling and noise beginning on Day 3. The animals that died also exhibited abnormal motor reflexes. In the 100 mg/kg group, beginning on Day 7 the animals became ataxic after gavage. Final mean body weight of males receiving 100 or 200 mg/kg was 10% or 27% lower than controls. Females in the 200 mg/kg group had a final mean body weight that was 20% lower than controls. No abnormal clinical observations were seen in the 25 or 50 mg/kg groups.

Conclusion :
Reliability : (1) reliable without restriction
(NTP TR-352, 1989)

Species : Mouse
Sex : Male/Female
Strain : B6C3F₁
Route of admin. : Gavage
Exposure period : 16 days
Frequency of treatment : 12 doses over 16 days
Post obs. period :
Doses : 0, 25, 50, 100, 200, 400 mg/kg/day
Control group : Yes, concurrent control
NOEL : 100 mg/kg/day
Method :
Year : 1981
GLP : Yes
Test substance : 98% pure
Test Condition : Animals were dosed daily with 0, 25, 50, 100, 200, 400 mg/kg/day

Result : (5/sex/group). All animals were observed twice daily. At the end of treatment all surviving animals were subject to complete necropsy. NOEL = 100 mg/kg. In the top dose group (400 mg/kg) all male and 4 female animals died within the 24 hours of the first dose. The single female survivor and the males and females in the 200 mg/kg/ group were ataxic after dosing beginning on Day 2. Final mean body weight of control males was lower than their initial weight. The initial mean body weight of the control females was approximately 3 g lower than those of the dosed groups. Therefore, weight changes could not be interpreted. Males (4/5) and females (2/5) in the 400 mg/kg group exhibited mild bronchial epithelial hyperplasia as did females (2/5) in the 200 mg/kg group and control males (1/5) and control females (1/5). All males and 3/5 females in the top dose group exhibited sinusoidal congestion of the liver. Vacuolar degeneration of the myocardial fibers was seen in 1/5 males and 2/5 females in top dose group. There were no abnormal clinical observations for 100, 50 or 25 mg/kg dose groups

Conclusion :
Reliability : (1) reliable without restriction
27.04.01 (NTP TR-352 1989)

Species : Rat
Sex : Male
Strain : Albino
Route of admin. : Diet
Exposure period : 28 days
Frequency of treatment : Daily
Post obs. period : 4 weeks
Doses : 0, 4, 46, 247 mg/kg/day
Control group : Yes, concurrent control
NOEL : 4 mg/kg
Method :
Year : 1953
GLP : Pre-GLP
Test substance : 97.5% pure
Test condition : Animals were dosed daily with 0, 4, 46 or 247 mg/kg/day (10/group).

Result : Rats receiving 4 mg/kg showed no toxic effects and food consumption and weight gain were comparable to that of the controls. No significant gross pathology was found in the 5 animals sacrificed at the end of 4 weeks. The remaining 5 animals were maintained for an additional 4 weeks on the control diet. Rats receiving 46 mg/kg showed signs of growth retardation, decreased food consumption. Three animals in this group were sacrificed at the end of 4 weeks. Four of the remaining 7 animals continued on the test diet, while 3 of the remaining 7 were shifted to the control diet. Those rats receiving 46 mg/kg for 8 weeks showed more severe signs during the period from 4 to 8 week including weakness of the hind limbs. Two of the animals appeared slightly ataxic. Those rats that discontinued treatment after 4 weeks showed improvement in their condition while on the control diet during the recovery period. The 247 mg/kg group started out at a higher dosage, but after one week it was reduced to this level due to marked weight loss and signs of toxicity which included tremors, slow righting reflexes, and hyperexcitability. Animals in this group showed signs of growth retardation, decreased food consumption, weakness of the hind limbs, and swaying movements, customarily described as "waltzing" during locomotion. Five animals were sacrificed at the end of the 4 week period, while the remaining 5 were transferred to the control diet for an additional 4 weeks. Following removal to the control diet, the animals increased their food consumption, gained weight, and the signs of systemic toxicity showed definite abatement. No characteristic gross pathological findings were found at autopsy in any group.

Conclusion :
Reliability : (2) reliable with restriction. Based on summary report.
(Cyanamid Report 53-82, 1953)

Species : Rat
Sex : Male/Female
Strain : F344/N
Route of admin. : Gavage
Exposure period : 90 days
Frequency of treatment : 5 days/week for 13 weeks
Post obs. period :
Doses : 0, 12.5, 25, 50, 100, 200 mg/kg/day
Control group : Yes, concurrent control
NOEL : 12.5 mg/kg/day
Method :
Year : 1981
GLP : Yes
Test substance : 98% pure
Test Condition : Animals were dosed 5 days/week for 13 weeks with 0, 12.5, 25, 50, 100 or 200 mg/kg/day (10/sex/group). Neurobehavioral tests were performed on all animals during week 6 and 13. Tests performed included motor activity, fore limb/hind limb grip strength, acoustic startle reflex measurement, and landing foot spread. Animals were observed twice daily, individual animal weights were recorded weekly. Special perfusion techniques were used to examine the plantar and tibial nerves to allow fixation of the pelvic limbs without compromising organ weights. Light microscopy was employed.

Result : Complete mortality occurred in the groups administered 100 and 200 mg/kg/day. Animals in groups receiving 50 mg/kg/day or higher developed hind limb ataxia which progressed to hind limb paralysis. Decreased forelimb and/or hind limb grip strength were seen at 100 mg/kg/day at 6 weeks and at doses down to 25 mg/kg/day for female rats and 12.5 mg/kg/day for male rats after 13 weeks of treatment. Landing foot spread was increased at 6 weeks only for rats in the 50 mg/kg/day group. This group was not tested at 13 weeks due to hind limb paralysis. No other group showed effects on landing foot spread. There were no consistent effects on motor activity, although it appeared reduced at 6 weeks for female rats given 100 mg/kg. There was a dose-related decrease in final body weight in surviving animals. The final mean body weight of male rats in 25 mg/kg or 50 mg/kg dose group was 8% or 16% lower than controls and 6% or 10% lower for females. There no changes in organ weights (liver, thymus, kidney, heart, brain, lungs or testes) except for an increase in testes weight in the males in 50 mg/kg/day group and the relative kidney weight in females given 50 mg/kg. Inflammation and/hemorrhage and edema of the mucosal cells lining the urinary bladder were seen in males given 25 mg/kg group (1/10), females given 50 mg/kg (3/10), males (5/10) and females (1/10) given 100 mg/kg and females (1/10) given 200 mg/kg. Degeneration of peripheral nerves was observed at doses of 50 mg/kg/day or higher using light microscopy and at doses of 25 mg/kg/day and higher using electron microscopy. NOEL = 12.5 mg/kg

**Conclusion
Reliability**

:
: (1) reliable without restriction

(NTP TR 352, 1989)

Species : Mouse
Sex : Male/Female
Strain : B6C3F₁
Route of admin. : Gavage
Exposure period : 90 days
Frequency of treatment : 5 days/week for 13 weeks
Post obs. period :
Doses : 0, 12.5, 25, 50, 100, 200 mg/kg/day
Control group : Yes, concurrent control
NOEL : 12.5 mg/kg
Method :
Year : 1981
GLP : Yes
Test substance : >98% pure
Test condition : Mice received 0, 12.5, 25, 50, 100 or 200 mg/kg/day (10/sex/group). Neurobehavioral tests were preformed on all animals during week 6 and 13. Tests performed included motor activity, fore limb/hind limb grip strength, acoustic startle reflex measurement, and landing foot spread. Animals were observed twice daily, individual animal weights were recorded weekly. Light microscopy was employed.

- Result** : There was complete mortality in the group receiving 200 mg/kg/day. These animals developed hind limb paralysis prior to their death. Behavioral tests performed after 6 or 13 weeks of treatment showed dose-related decreases in forelimb grip strength on both sexes at doses as low as 25 mg/kg/day. Motor activity was not significantly different at any dose group. Other tests (rotarod performance and startle response) did not show consistent dose-related changes. There were no significant effects on body weight. Final group mean body weights were comparable. A decreased relative testicular weight was seen in mice receiving 12.5 mg/kg/day or more. Kidney weights were increased at doses of 50 mg/kg/day or higher. Hepatocellular necrosis was observed in mice in the 200 mg/kg/day group, but not at lower doses. Adrenal cortex cytoplasmic vacuolization was observed in all female mice receiving 100 mg/kg/day. There was no nervous system lesions observed using light microscopy.
- Conclusion** :
Reliability : (1) reliable without restriction
(NTP TR 352, 1989)
- Species** : Rat
Sex : Male
Strain : Wistar
Route of admin. : Oral
Exposure period : 90 days
Frequency of treatment : Daily
Post obs. period :
Doses : 0, 3.36, 5.41, 8.65, 13.8 mM
Control group : Yes, concurrent control
Method : Neurotoxicity
Year : 1983
GLP : No data
Test substance : No data
Test condition : Groups of 4 male Wistar rats received 0, 3.36, 5.41, 8.65, 13.8 mM in drinking water for 90 days. This is equivalent to approximately 0, 33.9, 54.6, 87.4, and 139.4 mg/mL, respectively. Rotarod performance according to Dunham and Miya, 1957 was recorded weekly. Light microscopy examination was performed on posterior tibial nerves and sural nerves from the lower calf muscle region.
- Result** : Significant reductions in body weight gain was noted amongst all treated animals (6% reduction at the lowest exposure level, 43% at the highest). Rotarod performance at day 90 showed impairment only at the two highest exposure levels (3/4 animals at 8.65 mM and 4/4 animals at 13.8 mM). No other rotarod results were available. Other clinical signs of toxicity apparently included weakness, tendency towards spreading and dragging hind limbs and occasionally, amongst more severely affected animals, urinary incontinence; however it was not clear which groups these findings were seen in. Light microscopy examination showed moderate to severe changes: shrinkage and loss of myelinated fibers, myelin retraction, and corrugation of myelin sheaths. A significant reduction of the colchicine-binding was also detected in the spinal cord of both cervical and lumbar regions. However this reduction in colchicine binding was not seen in brain or the cerebellum.
- Conclusion** :
Reliability : (2) reliable with restriction, The incidence and severity of findings at other exposure levels was not reported and hence a NOAEL was not identifiable from this study.
(Tanii and Hashimoto 1983)

5. Toxicity

Id 924-42-5

Species : Rat
Sex : Male
Strain : Porton
Route of admin. : Gavage
Exposure period : 24 days
Frequency of treatment :
Post obs. period : 13 days
Doses : 7 doses of 100 mg/kg over 12 days; followed by 200 mg/kg/day for 2 days
Control group : No
Method :
Year : 1963
GLP : Pre-GLP
Test substance : No data
Test condition : Six male rats received 7 doses of 100 mg/kg over 12 days. On Days 23 and 24 an additional 200 mg/kg was given. Animals were killed on Day 37. Animals were weighed weekly and their gait was observed when walking on a non-slippery surface including an ascent up a slopping wooden board.

Result : The rats developed fine tremors. The authors state they were generally affected but did not develop the gross picture of clinical weakness. None showed typical bladder changes associated with urinary retention.

Conclusion :
Reliability : (2) reliable with restriction, detailed information on effects was not available. No data on compound purity. The authors state that neurotoxic effects may have been due to the presence of acrylamide in the test article.
(Barnes 1970)

Species : Rat
Sex : Male
Strain : Porton
Route of admin. : Oral (diet)
Exposure period : 27 weeks
Frequency of treatment :
Post obs. period : 3 months
Doses : Seven
Control group : No
Method :
Year : 1963
GLP : Pre-GLP
Test substance : No data
Test condition : Six male rats were fed a diet containing 400 ppm for 14 weeks followed by 7 weeks at 800 ppm and the six weeks at 1600 ppm. Animals were weighed weekly and their gait was observed when walking on a non-slippery surface including an ascent up a slopping wooden board.

Result : At the end of test article administration (27 weeks) all rats exhibited definite weakness which recovered somewhat during the 3 months on a normal diet.

Conclusion :
Reliability : (2) reliable with restriction, detailed information on effects not available and no data on compound purity. The authors state that neurotoxic effects may have been due to the presence of acrylamide in the test article.
(Barnes 1970)

Species : Rat
Sex : Male

Strain : Porton
Route of admin. : Oral dietary
Exposure period : 5 weeks
Frequency of treatment : Daily
Post obs. period :
Doses : 1800 ppm (daily intake 27 mg/rat) for one week and thereafter 900 ppm (daily intake 18.6 mg/rat) for four weeks, in addition to dosing via the diet, for 2 weeks, 4 i.p. doses (50 mg/kg) were given.
Control group :
Method :
Year : 1975
GLP : No data
Test substance : Less than 0.05% acrylamide was present
Remark : Slight ataxia in male Porton rats after 5 weeks; moderate disability at 7 weeks
Reliability : (1) reliable without restriction

(Edwards 1975a)

5.5 GENETIC TOXICITY 'IN VITRO'

Type : Ames test
System of testing : Salmonella typhimurium strains TA97, TA98, TA100, and TA1535
Concentration : 0, 100, 333, 1,000, 3,333, 10,000 µg/plate
Cycotoxic conc. :
Metabolic activation : With and without
Method :
Year : 1987
GLP : Yes
Test substance : >98% pure
Test condition : Cells were incubated with either the test article or solvent (dimethyl sulfoxide) in the absence of metabolic activation or with Aroclor 1254-induced S9 from male Syrian hamster liver or male Sprague Dawley rat liver
Result : Results were consistently negative using *Salmonella typhimurium* tester strains TA97, TA98, TA 100 and TA 1535 in the presence and absence of metabolic activation.
Conclusion :
Reliability : (1) without restriction

(Zeiger 1988)

Type : Ames test
System of testing : Salmonella typhimurium TA 98, TA 100, TA 1535, TA 1537, TA1538
Concentration : <= 5000 ug/plate
Cycotoxic conc. :
Metabolic activation : With and without
Result : Negative
Method : Ames et al. 1975
Year : 1985
GLP :
Test substance :
Test condition : According to standard procedures.

| | | |
|-----------------------------|---|---|
| Result | : | Results were consistently negative using <i>Salmonella typhimurium</i> tester strains TA 98, TA 100, TA 1535, TA 1537, and TA 1538, in the presence and absence of metabolic activation. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction (Hashimoto and Tanii 1985) |
| Type | : | Cytogenic Assay |
| System of testing | : | Tested in the BALB/3T3 cell transformation assay in the presence of an Aroclor-induced rat liver S-9 reaction mixture. |
| Concentration | : | 125, 250, 500, 1000 µg/mL. |
| Cycotoxic conc. | : | |
| Metabolic activation | : | S-9 |
| Result | : | Negative |
| Method | : | OCED |
| Year | : | 1986 |
| GLP | : | Yes |
| Test substance | : | No data |
| Test Condition | : | The test article dosing solutions were prepared in distilled and diluted to approximately twice of the concentration in phosphate buffered saline (PBS). BALB/3T3 cells were exposed to solvent alone at the final concentration of 2 µg/mL and to seven concentrations of test article ranging from 1 – 1000 µg/mL. All concentrations were soluble. Based upon these findings final dose levels were selected. Both solvent and positive (bezno(a)pyrene) controls were employed in the transformation test. In the first test, cells were treated for 2 hours in suspension at a density of 2×10^6 cells/ 4 mL PBS and the appropriate concentration of the test article. The treatment medium was removed and the cells were cultured at a density of 250 cells/60 mm dish. After 7-10 days incubation, the cells were fixed, stained and scored for colony formation. In the transformation test, the cells were prepared in the same manner, except the cell were cultures at a density of 10^4 cells/60 mm dish. These cells were incubated for 4-6 weeks and then fixed, stained and scored for morphological transformation. |
| Result | : | No significant transforming activity was observed in any of the doses tested. As compared to solvent control, in a suspension culture in the presence of exogenous metabolic activation, the cell survival was approximately 84%, 100%, 97%, and 105% at 1000, 500, 250 and 125 µg/mL, respectively. Three spontaneous Type II and 1 Type III foci were observed on the solvent control for a background frequency of 0.19×10^{-4} . Two type II and 2 Type III foci were observed at 1000 µg/mL, 3 Type II foci were scored at 500 µg/mL, 3 Type III foci were observed at 125 µg/mL and 2 Type III foci were seen at 125 µg/ml. None of the transformation frequencies in the treated groups were statistically increased ($p > 0.05$, Modified Poisson Distribution) relative to solvent control. The positive control induced 9 Type II and 7 Type III foci for a transformation frequency of 1.63×10^{-4} , which was statically increased. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction (Microbiological Associates 1986) |
| Type | : | Cytogenic Assay |
| System of testing | : | Chinese hamster ovary cells |
| Concentration | : | 16.7, 50, 125, 166.7, 250, 500, 1,700 µg/mL |
| Cycotoxic conc. | : | |
| Metabolic activation | : | |
| Result | : | Positive |

| | |
|------------------------|--|
| Method | : Chromosome Aberration Test and SCE's as described in Galloway 1985, 1987 |
| Year | : No data |
| GLP | : Yes |
| Test substance | : > 98% pure |
| Test Conditions | : Chinese hamster cells were incubated with the study compound at doses of 16.7, 50, 125, 166.7, 250, 500, 1,700 µg/mL or solvent (dimethyl sulfoxide) and cultured for enough time to reach second metaphase division. Cells were collected by mitotic shake-off, fixed, air-dried and stained. |
| Results | : A dose-related increase in chromosomal aberrations and sister chromatid exchanges were seen both with and without activation using rat liver S9. |
| Reliability | : (1) reliable without restriction |

(NTP TR-352, 1989)

5.6 GENETIC TOXICITY 'IN VIVO'

| | |
|-----------------------------|---|
| Type | : Micronucleus test |
| System of testing | : |
| Concentration | : 37.5, 75, 150 mg/kg 5 animals per group |
| Cycotoxic conc. | : |
| Metabolic activation | : |
| Result | : Negative |
| Method | : |
| Year | : No data |
| GLP | : Yes |
| Test substance | : > 98% pure |
| Test Condition | : Male mice were given two ip injections, at 24 hour intervals of the test article dissolved in corn oil. Bone marrow smears were prepared 24 hours after the second injection. 2,000 PCEs were scored for the incidence of micronuclei in each of the five animals per dose group. Both solvent control and positive control (dimethylbenzanthracene) were employed. |
| Result | : No increase in micronucleated polychromatic erythrocytes (PCEs) was observed in the bone marrow of B6C3F1 mice after intraperitoneal injection of the test material. |
| Reliability | : (1) reliable without restriction |

(NTP 1989)

5.7 CARCINOGENITY

| | |
|-------------------------------|--------------------|
| Species | : Rat |
| Sex | : male/female |
| Strain | : Fischer 344 |
| Route of admin. | : Gavage |
| Exposure period | : 103 weeks |
| Frequency of treatment | : 5 days/week |
| Post. obs. period | : |
| Doses | : 0, 6 or 12 mg/kg |
| Control group | : Yes |
| Method | : |
| Year | : 1984 |
| GLP | : Yes |
| Test substance | : ~ 98 % |

- Test condition** : Groups of 50 rats/sex were administered 0, 6, or 12 mg/kg in water via gavage, 5 days per week for 103 weeks. All animals were observed two times per day. Body weights were recorded once per week for the first 13 weeks of the study and once a month thereafter. Necropsy was performed on all animals. All organs and tissues were examined for grossly visible lesions. In accordance with the "inverse pyramid" design (McConnell 1983), complete histopathological exams were performed on all high dose and control animals and on low dose animals dying through month 21 of the study. And on all grossly visible lesions in all dose groups. If mortality in the highest dose group was 15% greater than in controls, complete histopathological exam was performed on all animals in the second highest dose group as well. Probability of survival was estimated according to the method of Kaplan and Meier (1958). Dose related survival statistical analysis was evaluated according to the method of Cox (1972) and life table test for a dose-related trend according to the method of Tarone (1975). Tumor incidence was analyzed using life table tests, logistic regression and Fisher Exact/Cochran-Armitage trend analyses.
- Result** : Mean body weight of dosed animals was within 6% of controls. Survival of female rats in the low dose group was significantly lower than controls after day 550. However survival in females in the 12 mg/kg/day group did not differ from controls (control 35/50; low dose 22/50; high dose 33/50). There were no other differences in survival in any other group (males 28/50; 22/50; 27/50). No biological important non-neoplastic or neoplastic were attributed to the test compound.
- Conclusion** : No evidence of carcinogenic activity for male or female rats receiving 6 or 12 mg/kg/day
- Reliability** : (2) reliable with restriction, the authors concluded that higher doses might have increased the sensitivity of the studies to determine the presence or absence of a carcinogenic response
(NTP TR-352, 1989)
- Species** : Mouse
Sex : Male/Female
Strain : B6C3F1
Route of admin. : Gavage
Exposure period : 103 weeks
Frequency of treatment : 5 days/week
Post. obs. period :
Doses : 0, 25 or 50 mg/kg
Control group : Yes, concurrent control
Method :
Year : 1984
GLP : Yes
Test substance : ~ 98 %

- Test condition** : Groups of 50 mice/sex were administered 0, 25, or 50 mg/kg in water via gavage, 5 days per week for 103 weeks. All animals were observed two times per day. Body weights were recorded once per week for the first 13 weeks of the study and once a month thereafter. Necropsy was performed on all animals. All organs and tissues were examined for grossly visible lesions. In accordance with the "inverse pyramid" design (McConnell 1983), complete histopathological exams were performed on all high dose and control animals and on low dose animals dying through month 21 of the study. And on all grossly visible lesions in all dose groups. If mortality in the highest dose group was 15% greater than in controls, complete histopathological exam was performed on all animals in the second highest dose group as well. Probability of survival was estimated according to the method of Kaplan and Meier (1958). Dose related survival statistical analysis were evaluated according to the method of Cox (1972) and life table test for a dose-related trend according to the method of Tarone (1975). Tumor incidence was analyzed using life table tests, logistic regression and Fisher Exact/Cochran-Armitage trend analyses.
- Result** : Mean body weight of females was as much as 25% greater than controls. Males mean body weight was up to 13% greater than controls. There were no other differences in survival in any other group (male mice: 30/50; 20/50; 21/50; female mice: 41/50; 35/50; 33/50). Incidence of adenomas of the Harderian gland was increased in treated males and in females in the top dose group (male: control 1/48; low dose 14/49; high dose 29/50; female: 5/47; 8/45; 20/48). Incidences of carcinoma of the Harderian gland were not significantly increased (male 1/48; 0/49; 2/50; female: 0/47; 3/45; 2/48). Incidences of hepatocellular adenomas were increased in males and females in the high dose group (male: 8/50; 4/50; 19/50; female: 0/47; 3/45; 2/49). Hepatocellular adenomas and carcinomas (combined) occurred with positive treatment. Incidences in males and females in the top dose group were increased compared to controls (male: 12/50; 17/50; 26/50; female: 6/50; 7/50; 17/49). Females exhibited increased incidence of ovarian atrophy (3/50; 39/45; 38/47) and benign granulose cell tumors (0/50; 5/45; 5/47). High dose females had a lower incidence of adenomas of the pars distalis than controls (13/49; 5/14; 4/43). Incidence of chronic inflammation and alveolar epithelial hyperplasia of the lung were observed. Sentinel mice were seropositive for Sendai virus at 18 months. Males in the top dose group had increased incidence of alveolar/bronchiolar adenomas (3/49; 6/50; 11/50) and carcinomas (2/49; 4/50; 10/50). Females in the top dose group exhibited increases in the incidence of alveolar/bronchiolar adenomas or carcinomas (combined) (6/50; 8/50; 13/49).
- Conclusion** : Clear evidence of carcinogenic activity for male B6C3F₁ mice based on increased incidences of neoplasms of the Harderian gland, liver and lung. Clear evidence of carcinogenic activity for female B6C3F₁ mice based on increased incidences of neoplasms of the Harderian gland, liver, lung and ovary.
- Reliability** : (1) reliable without restriction

(NTP TR-352, 1989)

5.8 REPRODUCTION TOXICITY

- Type** : Fertility
Species : Mouse
Sex : Male
Strain : Ddy
Route of admin. : Gavage

| | | |
|----------------------------------|---|---|
| Exposure period | : | 8 weeks |
| Frequency of treatment | : | 2/week |
| Duration of test | : | 8 weeks |
| Doses | : | 0, 2.9 mmol/kg (292 mg/kg) |
| Control group | : | Concurrent control |
| Method | : | Fertility Test |
| Year | : | 1981 |
| GLP | : | |
| Test substance | : | > 95% pure |
| Test condition | : | Groups of 5-7 male mice (5-6 wks of age, avg 29g) received 0 or approximately 2.9 mmol/kg (>95% purity) in saline by oral gavage twice weekly for 8 weeks. Testicular weight was taken at necropsy. Student's t test used for comparisons. Light microscopy was employed. |
| Result | : | Relative testicular weight was reduced (55% of control value). Light microscopy of the testes showed "degeneration of epithelia in spermatids and spermatocytes" (presumably meaning that a reduced number of spermatids and spermatocytes were observed in the epithelium when compared with controls), reduction in spermatozoa, the presence of multinucleate giant cells. Sertoli cells and interstitial cells were apparently unaffected. In addition, the epididymides were apparently unaffected. No further histopathological investigations were performed. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| | | (Hashimoto et al 1981) |
| Type | : | Fertility |
| Species | : | Mouse |
| Sex | : | Male/female |
| Strain | : | DdY |
| Route of admin. | : | Drinking water |
| Exposure period | : | 6 weeks |
| Frequency of treatment | : | Daily |
| Premating exposure period | : | 6 weeks |
| Male | : | |
| Female | : | |
| Duration of test | : | |
| Doses | : | 0, 4.3 mM |
| Control group | : | |
| Method | : | Reproductive toxicity |
| Year | : | 1986 |
| GLP | : | No data |
| Test substance | : | > 95% pure |
| Test condition | : | Groups of 14 male and 24 female ddY mice received 0, or 4.3 mM N-hydroxymethylacrylamide in drinking water for 6 weeks. Water and food consumption were measured weekly. On completion of the dosing period, half of the treated males and all of the females were mated with untreated controls of the opposite sex. Uterine contents were examined on day 13 of gestation for implants and resorptions except for half of the females at the highest exposure level, which were allowed to complete their gestation period and deliver pups which were examined for a further 4 weeks for any abnormalities. Immediately after the 6-week exposure period, the males not being used for mating were killed and organ weights were determined (liver, testes and seminal vesicles) with further examinations for sperm |

| | |
|----------------------------------|---|
| Result | <p>count and sperm cell morphology.</p> <p>: The exposure level was associated with slight signs of hind limbs weakness amongst treated females on completion of 6 weeks exposure. Bodyweight, food and water consumption was unaffected.</p> <p>The fertility rate, assessed on day 13 and also on the day of delivery was slightly affected only when treated males were mated with control females (11/15 pregnant compared with 12/15 in controls on the day of delivery). There was no affect on fertility when treated males were mated with control females on Day 13. The bodyweight of the offspring was within normal limits. No abnormalities in body weight gain and general behavior were observed up to 4 weeks after birth.</p> <p>At week 6, there were significant decreases in relative weights of the testis and a reduction, but not statistically significant, in seminal vesicles. Also, epididymal sperm count was significantly reduced from males and there was an increase in head and tail abnormalities in sperm. Overall, this study showed clear effects on epididymal sperm count following exposure of males to 4.3 mM for 6 weeks. It is unclear from this study whether or not the impaired fertility was secondary to neurotoxicity.</p> |
| Conclusion | : |
| Reliability | <p>: (1) Reliable without restriction</p> <p style="text-align: right;">(Sakamoto and Hashimoto 1986)</p> |
| Type | : One generation study |
| Species | : Mouse |
| Sex | : Male/female |
| Strain | : CD-1 (Swiss) mice |
| Route of admin. | : Drinking water |
| Exposure period | : 27 weeks during continuous breeding phase |
| Frequency of treatment | : Daily |
| Premating exposure period | : |
| Male | : |
| Female | : |
| Duration of test | : |
| Doses | : 0, 60 ppm (~11 mg/kg/day), 180 ppm (~ 30-40 mg/kg/day), 360 ppm (~ 115 mg/kg/day) |
| Control group | : |
| Method | : Modified Reproductive Assessment by Continuous Breeding (RACB) |
| Year | : 1990-1991 |
| GLP | : Yes |
| Test substance | : $\geq 97\%$ |
| Test Condition | <p>: In the continuous breeding phase there was a control group (40 breeding pairs) and 3 dose groups (20 pairs/group). Top dose level was expected to cause neurotoxicity half-way through administration with greater than 90% survival; the middle dose was expected to produce little or no systemic toxicity, the low dose was expected to be a no effect level. The animals were housed as breeding pairs for 98 days, following 7 days of pre-mating exposure to the test article while house singly. Endpoints for this phase included clinical signs, parental body weight, fertility, (number producing a litter/number of breeding pairs), litters per pair, live pups per litter, proportion of pups born alive, sex of live pups, pup body weight within 24 hours of birth, feed and water consumption, forelimb and hindlimb grip strength assessments. At the end of 98 days the pairs were separated and housed individually with continued dosing. Any litters born (F1) after</p> |

continuous breeding phase were reared by dam until weaning. Selected weanlings were housed in same sex groups until 74 ± 10 days of age. Lactating females continued to receive the same doses. Their F1 offspring were used for assessment of 2nd generation fertility. A one-week crossover mating trial was performed on the parental animals from the control and high dose groups to determine the affected sex. Low and mid-dose males and females continued on treatment after separation of the breeding pairs and necropsied with the animals from the cross-over mating trial. During the crossover breeding trial administration of the test article was suspended. Following crossover breeding, vaginal cytology was evaluated for control and high dose females 12 days prior to necropsy. At necropsy, all females from all dose groups were sacrificed. Body and organ weights were recorded. In selected males, an endocrine challenge test (Noden et al., 1984; Grizzle et al., 1989; Fail et al., 1992) was administered to assess the response of the pituitary and testis to a tropic stimulus of control and high dose males. Males from all dose groups were sacrificed on the same morning. Body and organ weights were recorded and an epididymal sperm evaluation was conducted. One testis was frozen for quantification of homogenization-resistant spermatid heads and intratesticular testosterone. Selected organs were examined microscopically.

Assessment of the F1 generation was conducted using offspring from all four dose groups. Animals were maintain on the same dose as their parents until sexual maturity (74 ± 10 days). Twenty control animals of each sex and 20 treated animals of each sex in each treatment group were selected. Animals not selected were euthanized. Males and females within treatment groups were randomly assigned to breeding pairs and housed, one breeding pair per cage. Breeding pairs were cohabited for 7 days or until vaginal copulatory plug was found, whichever was less, then separated and housed singly. After delivery of all the litters and collection of vaginal smears from the females, all animals were euthanized and necropsied. Grip strength evaluations and an endocrine challenge test were also preformed. Body weight and selected organ weights were recorded. Data collected included epididymal and testicular spermatozoa evaluations, concentrations of peripheral testosterone, luteinizing hormone (LH), follicle-stimulating hormone (FSH) and intratesticular testosterone. Selected organs were examined microscopically.

Result

: Exposure to the test article resulted in moderate reproductive toxicity. Decreased pups per litter were seen at 360 ppm in F0 mice and at 60, 180, and 360 ppm in F1 mice. F1 mice had lowered body weights and increased liver and kidney/adrenal weight at all doses. Testicular toxicity was present in F0 animals at 360 ppm as indicated by decreased weight. Histopathology was not evident. F1 animals were more susceptible than F0 animals to the neuromuscular effects of the test material at all doses. The compound consistently decreased grip strength in both limbs of both sexes in F1 animals during post-weaning development, as opposed to sporadic effects in the hind limbs and forelimbs of F0 animals: more often in females.

The F0 females given at 60, 180, and 360 ppm had a longer litter interval. Increased postimplantation loss (dominant lethal effect) was seen at the at 180 and 360 ppm dose levels for untreated females mated to F0 males, in the absence of any clear-cut systemic toxicity in F0 mice.

The test material had dominant lethal effects, which reduced live litter size by 26% in the F0 mice and by as much as 55% for the F1 mice at the high dose. The reduced number of pups per litter was equivalent in pairs fed

the test material to those after exposed males were mated to non-treated females in the dominant lethal test. Thus, the dominant lethal effects may account for all the F0 reproductive toxicity.

Reliability : (1) Reliable without restriction (NTP 1993)

5.9 DEVELOPMENTAL TOXICITY/TERATOGENICITY

5.10 OTHER RELEVANT INFORMATION

Type Remark : Other: absorption, distribution and excretion
 : N-hydroxymethylacrylamide iv injected into rats (140 mg/kg) was distributed throughout the total body water within a few minutes. Breakdown of N-hydroxymethylacrylamide to acrylamide was negligible in vitro and in vivo. It caused a rapid decrease in liver glutathione in vivo and glutathione conjugates were excreted in the bile. Neurotoxicity of acrylamide was five times greater than that observed with N-hydroxymethylacrylamide. (Edwards, 1975b)

Type Remark : Metabolism,
 : All analogs studied, including N-hydroxymethylacrylamide, were metabolized by hepatic glutathione S-transferase. (Tanii and Hashimoto, 1981)

Type Remark : Metabolism and reactions with normal carbonyl compounds
 : N-Methylolacrylamide is an unsaturated carbonyl compound that reacts with nucleophilic atoms in Michael-type additions. It modified glycolytic enzymes in brain *in vitro*. (Sakamoto and Hashimoto, 1985)

Type Remark : Biochemical studies
 : Hashimoto and Aldridge (1970) found similar rates for the reaction of N-Methylolacrylamide and acrylamide with glutathione *in vitro*; they also found that both compounds react with protein sulfhydryls and haemoglobin in rats *in vivo*. The patterns of distribution of the two compounds between different tissues and subcellular organelles were also similar following oral administration to rats of equal doses of substances labelled in the carbonyl carbon. (Hashimoto and Aldridge, 1970)

Type Remark : Distribution and metabolism
 : Male Porton Rats were given test substance 27 mg/kg (1st week) and 18.6 mg/kg (2nd week) dietary and 4 injections of 50mg/kg during weeks 3-4 i.p. Total time: 7 weeks. N-Hydroxymethylacrylamide, N-methylacrylamide, and N,N-diethylacrylamide produce peripheral neuropathy in rats. Seven other compounds related to acrylamide do not produce neuropathy. Rats given one of the three neurotoxic compounds are more susceptible to acrylamide. A regime for testing acrylamide analogues for neurotoxicity is suggested. DDT, phenobarbitone, or high dietary concentrations of vitamin A or E have no effect on the development of acrylamide neuropathy in rats. Acrylamide produces neuropathy in hens but not in frogs or goldfish. (Edwards 1975a)

5.11 EXPERIENCE WITH HUMAN EXPOSURE

6.1 REFERENCES

- American Cyanamid Company. 1990. *Product Bulletin: CYLINK® NMA Monomer N-Methylol Acrylamide: Applications- Processes- Products- References* (PRT-708-A), Wayne, NJ [now Cytec Industries, Linden, NJ].
- Barnes, J.M. 1970. Observations on the effects on rats of compounds related to acrylamide. *Brit. J. Industr. Med.* 27, 147-149.
- Batelle's Columbus Laboratories. Acute toxicity study: N-methylolacrylamide (C60333), Fischer 344 rats. March 16, 1981a.
- Batelle's Columbus Laboratories. Acute toxicity study: N-methylolacrylamide (C60333), B6C3F1 mice. March 16, 1981b.
- Cooke, D. Aquatic toxicity tests versus *Onchorhynchus mykiss*. United States Testing Company, Inc., January 21, 1990.
- Cyanamid Report 53-82: Methylolacrylamide, summarization of toxicity data from Hazleton reports, May 7 to Sept. 25, 1953. Inter-office correspondence from H. H. Golz to N.B. Somer, April 26, 1954.
- Cytec Industries, Inc. 1995. *Product Bulletin: CYLINK® NMA Monomer N-Methylol Acrylamide* (PRT-707-B), West Paterson, NJ.
- Cytec MSDS No. 05741, CYLINK NMA Monomer, 48% Aqueous, Inhibited, Date: December 20, 1999, Cytec Industries, Inc. (40-44 % NMA (CAS 924-42-5), <6 % Acrylamide (CAS 79-06-1), <2 % Formaldehyde (CAS 50-00-0)).
- Edwards, P.M. 1975a. Neurotoxicity of acrylamide and its analogues and effects of these analogues and other agents on acrylamide neuropathy. *British Journal of Industrial Medicine* 32, 31-38.
- Edwards, P.M. 1975b Distribution and metabolism of acrylamide and its neurotoxic analogs in rats. *Biochem Pharmacol* 24 (13-14) 1277.
- Feurr, H. and Lynch, U. E. 1953. The synthesis and reactions of unsaturated N-Methylolamides. *J. Amer. Chem. Soc.* 75, 5027-5029.
- Hashimoto, K. and Aldridge, W. N. 1970. Biochemical studies on acrylamide, a neurotoxic agent. *Biochem. Pharmacol.* 19, 2591-2604.
- Hashimoto, K. and Tanii, H. 1985. Mutagenicity of acrylamide and its analogues in *Salmonella typhimurium*. *Mutation Research* 158, 129-133.
- Hashimoto, K., Sakamoto, J., and Tanii, H. 1981. Neurotoxicity of acrylamide and related compounds and their effects on male gonads in mice. *Arch. Toxicol.* 47(3) 179.
- Japanese Journal of Hygiene* 34(1):183, 1979.
- Klimisch, H.J., Andreae, M and Tillman, U. 1997. A systemic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology* 25:1-5.
- Medical College of Wisconsin Great Lakes Research Facility, LC50 – Cyanamid n-methylolacrylamide, Paper Product 2851, CT-270-86 in Rainbow trout (*Salmo gairdineri*), 1990.

NTP 1993. National Toxicology Program, Final report on the reproductive toxicity of N-(hydroxymethyl)acrylamide (HACR) (CAS No. 924-42-5) in CD-1 (Trade Name) Swiss mice. NIH Publication No. RACB90017 U.S. Department of Health and Human Services, National Toxicology Program, National Institute of Environmental Health Sciences, Research Triangle Park, NC 27709. January 1993.

NTP TR-352, 1989. National Toxicology Program, Toxicology & Carcinogenesis Studies of N-Methyloacrylamide in F344/N Rats and B6C3F1 Mice. Technical Report Series No. 352 (1989) NIH Publication No. 89-2807 U.S. Department of Health and Human Services, National Toxicology Program, National Institute of Environmental Health Sciences, Research Triangle Park, NC 27709.

Putman, D. Morphological transformation of BALB/3T3 mouse embryo cells in the presence of exogenous metabolic activation. Microbiological Associates, March 31, 1986.

Sakamoto, J. and Hashimoto, K. 1985. Effect of acrylamide and related compounds on glycolytic enzymes in mouse brain *in vitro*. *Arch. Toxicol.* 57, 276-281.

Sakamoto, J. and Hashimoto, K. 1986. Reproductive toxicity of acrylamide and related compounds in mice – effects on fertility and sperm morphology. *Arch. Toxicol.* 59, 201-205.

Syracuse Research Corporation, 1998. Syracuse, NY pollution prevention (P2) assessment framework, U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics (Draft) – EPIWIN Generated Printout.

Tanii H and Hashimoto K. 1981. Studies on *in vitro* metabolism of acrylamide and related compounds. *Arch. Toxicol.* 48(2-3) 157.

Tanii, H. and Hashimoto, K. 1983. Neurotoxicity of acrylamide and related compounds in rats: effects on rotarod performance, morphology of nerves and neurotubulin. *Arch. Toxicol.* 54:203-213.

Vernon, P. A., Dulak, L. H., and Deskin R. 1990. Acute toxicologic evaluation of N-methylolacrylamide. *J. Am. Coll. Toxicol.* 1(2) Part B, 111-112.

Wang, X.M., Modified OECD Test for ready biodegradability of CT-444-90E. United States Testing Company, Inc., February 20, 1991.

Zeiger, E. Anderson, B., Haworth, S., Lawlor, T., and Mortelmans, K. 1988. Salmonella mutagenicity tests: IV. results from the testing of 300 chemicals. *Environmental and Molecular Mutagenesis* 11(Suppl. 12) 1-158.

7.1 END POINT SUMMARY

7.2 HAZARD SUMMARY

7.3 RISK ASSESSMENT

7 SEPTEMBER 2001

Acrylamide, N-(butoxymethyl)-

NBMA

Data Set

New Chemical : Acrylamide, N-(butoxymethyl)-
CAS No. : 1852-16-0
Molecular Formula :
Molecular Weight :

Printing Date : 7 September 2001
Originally prepared in IUCLID 3.1

1.2 SYNONYMS

Acrylamide, N-Butoxymethyl-

N-Butoxymethylakrylamid (Czech)

2-Propenamide, N-(Butoxymethyl)-

N(Isobutoxymethyl) Acrylamide

N(Methoxymethyl) Acrylamide

NBMA

IBMA

2. Physico-Chemical Data

Id 1852-16-0

Date 10/05/01

2.1 MELTING POINT

Value : = 79.9 ° C
Sublimation :
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : The melting point is estimated by the EPIWIN/MPBPWIN model using the average of values obtained from the Gold and Ogle method and the Joback, Gold, and Ogle method.
Reliability : (2) valid with restrictions. Data were obtained by modeling (Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

2.2 BOILING POINT

Value : = 296.53° C
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : The boiling point is estimated by the EPIWIN/Stein and Brown Method.
Reliability : (2) valid with restrictions. Data were obtained by modeling (Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

Value : = 118 - 143 ° C
Test substance : Other TS: N-Butoxymethyl Acrylamide Solution (>51 % N-Butoxymethyl acrylamide)
Reliability : (2) valid with restrictions. Details on experimental conditions are not present.
(Cytec MSDS No. 4500)

2.3 DENSITY

Value : = .889 - .923
Test substance : Other TS: N-Butoxymethyl Acrylamide Solution (>51 % N-Butoxymethyl acrylamide)
Reliability : (2) valid with restrictions. Details on experimental conditions are not present.
(Cytec MSDS No. 4500)

2.4 VAPOUR PRESSURE

Value : 0.000704 mm Hg at 25 ° C
Decomposition Method : other (calculated): Evaluated by the MPBPWIN Program (v.1.40), using various methods (Antoine, Modified Grain, and Mackay Methods).
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-

2. Physico-Chemical Data

Id 1852-16-0

Date 10/05/01

Result : All values were determined using an estimated boiling point of 296.53 deg C and an estimated melting point of 79.65 deg C.
Selected VP (25 deg C): 0.000704 mm Hg (Modified Grain Method)

Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

2.5 PARTITION COEFFICIENT

Log pow : = .92
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : The log Kow was estimated using EPIWIN/KOWWIN based on molecular structure and functionality.

Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

2.6.1 WATER SOLUBILITY

Value : = .00012 mg/l at 25 ° C
PH :
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : The water solubility is estimated by the EPIWIN/WSKOW model based on log Kow.

Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

Value :
Qualitative : not soluble
PH :
Test substance : Other TS: N-Butoxymethyl Acrylamide Solution (>51 % N-Butoxymethyl acrylamide)

Reliability : (2) valid with restrictions. Details on experimental conditions are not present.
(Cytec MSDS No. 4500)

3. Environmental Fate and Pathways

Id 1852-16-0

Date 10/05/01

3.1.1 PHOTODEGRADATION

Deg. Product :
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : The photodegradation rate was estimated by the EPIWIN/AOP program which estimates rate constants and half-lives of atmospheric reactions of organic compounds with hydroxyl radicals and ozone in the atmosphere.
Result : Overall OH Rate Constant = 50.4091 E-12 cm³/molecule-sec;
Half-life = 2.546 Hours
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

3.1.2 STABILITY IN WATER

Type : Abiotic
Method : other: calculated
Year : 2001
GLP : No
Test substance : Acrylamide, N-(butoxymethyl)-
Remark : Estimated by the EPIWIN/HYDROWIN program.
This program was not able to estimate a hydrolysis rate constant for this type of chemical structure. However, this compound has an amide group: C=O located at SMILES atom #: 2;
Hydrolysis rate extremely slow or t_{1/2} > 1 year.
Result : No estimate available.
Reliability : (2) valid with restrictions. Data were obtained by modeling.
(Syracuse Research Corporation, 1998)

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type : fugacity model level III
Media :
Air (level III) : 0.177
Water (level III) : 44.5
Soil (level III) : 55.3
Sediment : 0.0794
Method : other: calculated
Year : 2001
Remark : Level III fugacity was estimated using the EPIWIN/Fugacity Model (Full-Output)
Reliability : (2) valid with restrictions. Data were obtained by modeling
(Klimisch et al., 1997) (Syracuse Research Corporation, 1998)

3.5 BIODEGRADATION

Type : Aerobic
Inoculum : activated sludge
Contact time : 28 day
Degradation : = 79.6% after 28 day
Result : not readily biodegradable
Kinetic of test : 5 day 14.8 %

3. Environmental Fate and Pathways

Id 1852-16-0

Date 10/05/01

| | |
|--------------------------|--|
| substance | 15 day 5.6 % 28 day 79.6 % |
| Control substance | : Aniline |
| Kinetic | : 5 day 18.5 % 15 day 66.7 % 28 day 98.1 % |
| Method | : OECD Guide-line 301 D "Ready Biodegradability: Closed Bottle Test" |
| Year | : 1991 |
| GLP | : Yes |
| Test substance | : other TS |
| Result | : The sample stock solution fell within the organic content range stated in the formula. The sample containing 1 mg/l degraded by 0%, 3.7% and 77.8% over 5, 15 and 28 days, respectively. The sample containing 2 mg/l degraded by 14.8%, 5.6% and 79.6% over 5, 15 and 28 days, respectively. The sample containing 5 mg/l degraded by 39.3%, 50.4% and 45.9% over 5, 15 and 28 days, respectively. The average degradation for the three concentrations was 18.0%, 19.9% and 67.8% over 5, 15 and 28 days, respectively. Aniline degraded by 18.5, 66.7 and 98.1% over the three time periods. Because a level of 70% was reached, the test substance is "Readily Biodegradable" by this test procedure. |
| Test condition | : Stock solution was prepared by adding 2 g of sample to 1 liter of distilled water. The stock solution was screened to determine if it had a similar percent carbon content as stated in the formula provided by the supplier. Stock solution was diluted to 100 ppm as carbon after analysis. The diluted stock was then added to BOD bottles at 3.33 ml, 6.67 ml and 16.65 ml to yield test concentrations of 1 mg, 2 mg and 5 mg as carbon, respectively. Test solutions were inoculated with a low concentration of microorganisms from a mixed population and kept in closed bottles in the dark at a constant temperature of $20 \pm 1^\circ \text{C}$. The activated sludge bacteria was from Bergen Co., New Jersey. The degradation was followed by oxygen analyses with the YSI Dissolved Oxygen Analyzer 54A over a 28-day period. Degradability was based on a comparison of readings of actual oxygen demand to the theoretically expected oxygen demand. A parallel control with inoculum, but without test material, was run as a blank correction factor. The procedure was validated by means of a reference substance (aniline, 2 mg/l) of known biodegradability. |
| Test Substance | : $\text{C}_8\text{H}_{15}\text{NO}_2$ (80-84%), $\text{C}_3\text{H}_5\text{NO}$ (<6%), $\text{C}_4\text{H}_7\text{NO}_2$ (<6%), $\text{C}_4\text{H}_{10}\text{O}$ (<5%), $\text{C}_{11}\text{H}_{21}\text{NO}_3$ (1-4%), CH_2O (<0.5%), $\text{C}_7\text{H}_{10}\text{N}_2\text{O}_2$ ($\leq 0.2\%$), $\text{C}_7\text{H}_8\text{O}_2$ (0.02%). Carbon content was 61.1%. |
| Reliability | : (1) valid without restriction (Wang, 1991) |
| Deg. Product | : |
| Method | : other: calculated |
| Year | : 2001 |
| GLP | : No |
| Test substance | : Acrylamide, N-(butoxymethyl)- |
| Remark | : The biodegradation rate was estimated using EPIWIN/STP Fugacity Model: Predicted Fate in a Wastewater Treatment Facility. |
| Result | : Total biodegradation is predicted to be 0.09%. |
| Reliability | : (2) valid with restrictions. Data were obtained by modeling (Klimisch et al., 1997) (Syracuse Research Corporation, 1998) |

4.1 ACUTE/PROLONGED TOXICITY TO FISH

| | | |
|------------------------------|---|---|
| Type | : | Static |
| Species | : | Rainbow trout |
| Exposure period | : | 96 hours |
| Concentrations | : | 0, 62.5, 125, 250, 500, 1000 ppm |
| Analytical monitoring | : | |
| LC50 | : | 75 ppm |
| Method | : | OECD method 203 |
| Statistical method | : | ASTM STP634, Thompson 1947, Litchfield and Wilcoxin, 1949 |
| Year | : | 1990 |
| GLP | : | Yes |
| Test substance | : | 80-84% pure |
| Test condition | : | This test was conducted in EPA moderately hard reconstituted water. Ten fish (5/replicate) were exposed to the following nominal test concentrations: 0, 62.5, 125, 250, 500, or 1000 ppm. Water temperature, dissolved oxygen and pH were measured throughout the test and were within acceptable limits. Temperature was maintained at 15°C |
| Result | : | At 96 hours, there was complete mortality in all groups except for the low dose and control groups. There was 10% mortality in the 62.5 ppm group. Using the Acute-Toxicity Rating Scale published by the US Fish and Wildlife Service, this is slightly toxic to Rainbow Trout. The NOEC is <62.5 ppm. The LC50 is calculated to be 75 ppm. The 24-hour LC50=440 ppm, 48-and 72-hour LC50=175 ppm, the 96-hour LC50 = 75 ppm |
| Reliability | : | (1) reliable without restriction |

(Cooke, 1990)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

No data

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

No data

5. Toxicity

Id 1852-16-0

Date 10/05/01

5.1.1 ACUTE ORAL TOXICITY

Type : LD50
Species : Rat
Strain : Harlan-Wistar
Sex : Male
Number of animals : 17 (3 groups of 5, 1 group of 2)
Vehicle :
Route of admin. : Gavage
Doses : 0.625, 1.25, 2.5, 10 mL/kg
Exposure time :
Value : LD50=1.02 mL/kg
Method : Acute Oral Toxicity
Year : 1971
GLP : Pre-GLP
Test substance : 80% pure
Test condition : Three groups of 5 young adult, male rats were given 0.625, 1.25 or 2.5 mL/kg in a single gavage dose. One group of 2 rats were given 10 mL/kg.
Result : Mortalities were 0, 80% and 100% for the 0.625, 1.25, 2.5 mL/kg groups respectively. In the 10mL/kg group, both rats died within 6 hours of test article administration. All deaths occurred by Day 2. The majority of animals exhibited labored breathing and sluggishness. Necropsy of the animals that died revealed hemorrhaging of the lungs, stomach and intestines. Survivors had slight congestion of the adrenals and kidneys. Based on these data the LD50 is calculated to be 1.02 mL/kg
Conclusion :
Reliability : (1) reliable without restriction
Reference (Carpenter 1971)

Type : LD50
Species : Rat
Strain :
Sex :
Number of animals :
Vehicle :
Value : = 630 mg/kg bw
Method :
Year :
GLP : No data
Test substance : No data
Reliability : (2) reliable with restriction, no data on compound purity, strain, doses tested, or individual results. Original data source not reviewed, information obtained from Material Safety Data Sheet MSDS (Cytec MSDS No. 4500)

Species : Rat
Strain :
Sex :
Number of animals :
Vehicle :
Value : LD50 = 1030 mg/kg
Remark : (2) reliable with restriction, primary source is not in English. Data obtained from RTECS (Marhold 1986) (RTECS 2001)

5. Toxicity

Id 1852-16-0

Date 10/05/01

5.1.2 ACUTE INHALATION TOXICITY

No data

5.1.3 ACUTE DERMAL TOXICITY

| | | |
|--------------------------|---|--|
| Value | : | LD50 = 0.884 mL/kg |
| Type | : | Acute Dermal Toxicity |
| Species | : | Rabbit |
| Strain | : | Albino |
| Sex | : | Male |
| Number of animals | : | 12 (2 groups of 4, 2 groups of 2) |
| Vehicle | : | |
| Route of admin. | : | Dermal |
| Doses | : | 0.625, 1.25, 2.5 10.0 mL/kg |
| Exposure time | : | |
| Value | : | |
| Method | : | Acute Dermal Toxicity |
| Year | : | 1971 |
| GLP | : | Pre-GLP |
| Test substance | : | 80% pure |
| Test condition | : | Two groups of 4 male rabbits were given doses of 0.625 or 1.25 mL/kg. Two groups of 2 received |
| Result | : | The mortality was in rabbits given 0.625 or 1.25 mL/kg was .25 and 75% respectively. All animals that received either 2.5 or 10 mL/kg died. In the 1.25 mL/kg group I was observed, while in the 2.5 ml/kg group twitching of the ears and tremors was observed. Necropsy on the dead rabbits revealed congested lungs and pale kidneys. Based on these data the LD50 is calculated to be 0.884 mL/kg. |
| Conclusion | : | |
| Reliability | : | (1) reliable without restriction |
| | : | (Carpenter, 1971) |

5.1.4 ACUTE TOXICITY, OTHER ROUTES

5.2.1 SKIN IRRITATION

| | | |
|--------------------------|---|---|
| Type | : | Dermal irritation |
| Species | : | Rabbit |
| Strain | : | |
| Sex | : | No data |
| Number of animals | : | 6 |
| Vehicle | : | |
| Dose | : | 0.1 mL |
| Value | : | Primary Irritation Index = 1.8, Mild skin irritant |
| Method | : | FHSA (covered patch) |
| Year | : | 1971 |
| GLP | : | Pre-GLP |
| Test substance | : | 80% pure |
| Test condition | : | 0.1 ml of the test article was administered to one intact and one abraded site on each of 6 rabbits. Erythema and edema were scored at 24 and 72 hours using the Draize method. |

5. Toxicity

Id 1852-16-0

Date 10/05/01

Result Erythema, which was very slight to well defined to well-defined at 24 hours in the intact skin, cleared up by 72 hours. Edema which was non-existent in the intact skin, ranged from very slight to slight in the abraded skin of the rabbit. The Primary Irritation Index was calculated to be 1.8. based on these data, the test article was a mild skin irritant.

Conclusion :
Reliability : (1) reliable without restriction (Carpenter, 1971)

5.2.2 EYE IRRITATION

Type : Primary Eye Irritation
Species : Rabbit
Strain :
Sex : No data
Number of animals : Three
Concentration : No data
Dose : 0.5 mL
Exposure Time :
Number of animals : 12 (2 groups of 6)
Method : FHSA
Value : Mild eye irritant
Year : 1971
GLP : Pre-GLP
Test substance : 80% pure
Test condition : The test article was administered into the eyes of 2 groups of 6 rabbits. Corneal opacity iritis and conjunctival irritation was scored at 24, 48 and 72 hours
Result : Three of the 6 rabbits in each group had corneal damage. Based on these data the test article is a mild eye irritant
Conclusion :
Reliability : (1) reliable without restriction (Carpenter, 1971)

5.4 REPEATED DOSE TOXICITY

Species : Rats
Sex : Male/female
Strain : Charles River Albino
Route of admin. : Oral (diet)
Exposure period : 42 Days
Frequency of treatment : Daily
Post obs. period :
Doses : 0, 0.062, 0.125, 0.25% of diet (approximately 0, 0.072, 0.140, 0.306 gm/kg [males], 0, 0.076, 0.157, 0.294 [females])
Control group : Yes, concurrent control
NOAEL :
Method :
Year : 1976
GLP :

5. Toxicity

Id 1852-16-0

Date 10/05/01

| | | |
|-------------------------------|---|---|
| Test substance | : | The commercial product contains approximately 5% acrylamide and 5% N-methylolacrylamide. The commercial material was washed repeatedly with water to remove all water-soluble components. Examination by HPLC showed no other acrylamide homologs. |
| Test Condition | : | Forty rats, weighing not more than 100g were observed for 1 week. Test animals were randomized by weight stratification, 5/sex/group. All animals were observed daily for signs of toxicity, as well as possible changes in appearance, behavior, gait and excretory function. Food intake was measured weekly. Body weight was measured initially and then weekly for the duration of the study. All animals were subject to complete necropsy. Two-way analysis of variance (random block design) was employed to access the possible statistical differences of means. If any difference was noted, Dunett's t-test was used. The G.E. DATANET Time Sharing Computer System was employed using Fortran IV; for significance the value of $p < 0.05$ was employed. |
| Result | : | No deaths occurred during the study. Ataxia occurred in the 0.25% group, beginning at week 4 and persisting to Week 5. Improvement was noted on 3 of 5 males at termination. Those improving showed only slight signs of ataxia. All females at 0.25% improved during week 6, when only slight lack of coordination was apparent at termination. There was no significant change in body weight, although there was a trend toward lower body weight as the study progressed. Only the animals in the high dose group were contributing to this trend, which was apparent beginning at Week 1. Food consumption was significantly decreased. All dose groups consumed less the food the first week. By the end of the second week the 0.062% group was eating approximately the same amount as the control animals. |
| Conclusion Reliability | : | (1) reliable without restriction (Huntingdon Research Center, 1976) |

5.5 GENETIC TOXICITY 'IN VITRO'

| | | |
|-------------------------------|---|---|
| Type | : | Ames test |
| System of testing | : | <i>Salmonella typhurium</i> strains TA98, TA100, TA1535 TA1537 and TA1538 |
| Concentration | : | 0, 333, 667, 1,000, 3,330, 6,670, 10,000 µg/plate |
| Cytotoxic conc. | : | |
| Metabolic activation | : | With and without |
| Method | : | Ames, 1975 |
| Year | : | 1990 |
| GLP | : | Yes |
| Test substance | : | CT452-90 |
| Test condition | : | Cells were incubated with either the test article or vehicle (dimethyl sulfoxide) in the absence of metabolic activation or with Aroclor 1254-induced S9 from male Sprague Dawley rat liver |
| Result | : | Results were consistently negative using <i>Salmonella typhimurium</i> tester strains TA97, TA98, TA 100 and TA 1535 in the presence and absence of metabolic activation. |
| Conclusion Reliability | : | (1) without restriction (Hazleton Laboratories America, Inc., 1990a) |
| Type | : | Cytogenic Assay |
| System of testing | : | Chinese hamster ovary cells |
| Concentration | : | 98.5, 197, 296, 394, 493 µg/mL without activation; 246, 369, 493, 788, 1180, 1580, and 1970 µg/mL with activation (20 hour harvest); 502, 804, 1210, 1610, and 2010 µg/mL with activation (30 hour harvest) |

5. Toxicity

Id 1852-16-0

Date 10/05/01

Cycotoxic conc. :
Metabolic activation : With and without S9
Result : Positive
Method : Chromosome Aberration Test
Year : 1990
GLP : Yes
Test substance : CT452-90
Test Conditions : Chinese hamster cells were incubated with the study compound at selected doses or solvent (dimethyl sulfoxide) and cultured for enough time to reach second metaphase division. Cells were collected by mitotic shake-off, fixed, air-dried and stained.
Results : A dose-related increase in chromosomal aberrations were seen both with and without activation using rat liver S9.
Reliability : (1) reliable without restriction
(Hazleton Laboratories America, Inc.,1990b)

5.6 GENETIC TOXICITY 'IN VIVO'

No data available

5.7 CARCINOGENITY

No data available

5.8 REPRODUCTION TOXICITY

No data available

5.9 DEVELOPMENTAL TOXICITY/TERATOGENICITY

No data available

5.10 OTHER RELEVANT INFORMATION

Not required

5.11 EXPERIENCE WITH HUMAN EXPOSURE

Not required

6. References

Id 1852-16-0

Date 10/05/01

Carpenter, C. Range-Finding Toxicity Tests: Single Oral, Single Dermal, Skin Irritation (FHSA), and Eye Irritation (FHSA). Carnegie-Mellon University, November 24, 1971.

Cooke, D. Aquatic Toxicity Tests versus *Onchorhyncus mykiss*, United States Testing Company, Inc. January 21, 1990.

Cytec MSDS No. 4500: N-Butoxymethyl Acrylamide Solution, Date: July 01, 1997, Cytec Industries, Inc. (>51 % N-Butoxymethyl acrylamide (CAS 1852-16-0), <2.3 % N-Methylolacrylamide (CAS 924-42-5), <3.3 % Acrylamide (CAS 79-06-1), <0.6 % N,N'-Methylene-bisacrylamide (CAS 110-26-9), <30.0 % Butanol (CAS 71-36-3), and <10 % Xylene (CAS 1330-20-7))

Huntingdon Research Center, 6-Week Feeding Study with Isobutoxymethylacrylamide. Report No.: R-8236-14 (1-352), December 1, 1976.

Hazleton Laboratories America, Inc., Mutagenicity Test on CT # 452-90 In the Salmonella/Mammalian – Microsome Reverse Mutation Assay (Ames Test) with a Confirmatory Assay. Report No.: 12442-0-401R, December 11, 1990a.

Hazleton Laboratories America, Inc., Mutagenicity Test on CT # 452-90 In an in vitro Cytogenetic Assay Measuring Chromosomal Aberration Frequencies in Chinese Hamster Ovary (CHO) Cells with Multiple Harvests under Conditions of Metabolic Activation. Report No. 12442-0-437C, November 1, 1990b.

Klimisch, H.J., Andreae, M and Tillman, U. A systematic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology*. 25:1-5, 1997.

Marhold, J. (1986) *Prehled Prumyslove Toxikologie; Organické Latky. Avicenum* (Prague, Czechoslovakia) 706.

RTECS (2001) U.S. Department of Health and Human Services, Public Health Service, Center for Disease Control, National Institute for Occupational Safety Health. Registry of Toxic Effects of Chemical Substances (RTECS). CODEN: 85JCAE, August 14, 2001.

Syracuse Research Corporation, Syracuse, NY, Pollution Prevention (P2) Assessment Framework, U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics (Draft), 1998 – EPIWIN Generated Printout.

Wang, X.M. Modified OECD Test for Ready Biodegradability of CT-444-90F, United States Testing Company, Inc. February 20, 1991.

7.1 END POINT SUMMARY

7.2 HAZARD SUMMARY

7.3 RISK ASSESSMENT

Klimisch, H.J., Andreae, M and Tillman, U. A systematic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology*. 25:1-5, 1997.

RTECS (2001) U.S. Department of Health and Human Services, Public Health Service, Center for Disease Control, National Institute for Occupational Safety Health. Registry of Toxic Effects of Chemical Substances (RTECS). National Library of Medicine's current MEDLARS file., p. 82/8010

Syracuse Research Corporation, Syracuse, NY, Pollution Prevention (P2) Assessment Framework, U.S. Environmental Protection Agency, Office of Pollution Prevention and Toxics (Draft), 1998.

Wang, X.M. Modified OECD Test for Ready Biodegradability of CT-444-90F, United States Testing Company, Inc. February 20, 1991.