Section A6.1.4		**Acute Dermal Irritation**
Annex Point IIA6.4		Section A6.1.4 : Rabbit Skin Irritation Study : Disodium Tetraborate Pentahydrate
5.1	**Materials and methods**	Skin irritation study carried out on disodium tetraborate pentahydrate to comply with US EPA-FIFRA guidelines at the time and carried out by the US Food and Drug Laboratories to GLP. Although it is not to modern protocols the data is consistent with another study on the same substance and with other borate data and further testing is not warranted in the interests of animal welfare and protecting laboratory animals
5.2	**Results and discussion**	No irritancy was observed and although not carried out to modern protocols, data from other irritation studies on boric acid confirm the results Therefore further testing is not warranted in the interest of animal welfare.
5.3	**Conclusion**	Non-irritant
5.3.1	Reliability	*2*
5.3.2	Deficiencies	

Evaluation by Competent Authorities	
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	28 February 2005
Materials and Methods	Since 2 sites per animal were treated with 0.5 g of test substance, the total amount applied was 1 g. Otherwise, the version of the applicant is acceptable.
Results and discussion	The version of the applicant is adopted.
Conclusion	The version of the applicant is adopted.
Reliability	2
Acceptability	acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.1.4	Acute Eye Irritation	
Annex Point IIA6.1.4	Disodium Tetraborate Anhydrous	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [ ]	Technically not feasible [ ] Scientifically unjustified [ ]	
Limited exposure [ ]	Other justification [x]	
Detailed justification:	Anhydrous disodium tetraborate is the anhydrous salt of disodium tetraborate decahydrate and disodium tetraborate pentahydrate. For practical purposes one part of Anhydrous Disodium tetraborate is equivalent to 1.45 parts of disodium tetraborate pentahydrate; 1.9 parts of disodium tetraborate decahydrate; 1.02 parts disodium octaborate tetrahydrate and in aqueous solution 1.23 parts of boric acid. It is hygroscopic and takes up water to form a hydrated salt and like the other borates, in solution it will exist as undissociated boric acid (see Doc IIIA A7.1.1.1 Hydrolysis and Doc IIIA Read Across Statement). Disodium tetraborate decahydrate and disodium tetraborate decahydrate are used as a buffer in eyewashes For disodium tetraborate pentahydrate a study was carried out at the request of the US EPA to confirm that the eye irritation previously seen with disodium tetraborate pentahydrate was caused by the glassy nature of the crystals of substance and not a chemical effect of irritation. To confirm this, the sample was ground to a fine powder before instillation to reduce the glassy, sharp crystals in the sample. No irritancy was observed that requires Classification under 68/548/EEC. As a result for this study the US EPA accepted that the effects were mechanical downgraded its classification according to US FIFRA to Toxicity II (40 CFR 156) by ocular administration (Corneal involvement or irritation clearing in 8-21 days). For disodium tetraborate decahydrate no new study was carried out as it was concluded that, although the substance caused slight irritation, this irritation is thought to arise from the glassy nature of the crystals of the substance and therefore classification is not relevant. This was confirmed for the closely related disodium tetraborate pentahydrate and therefore disodium tetraborate decahydrate is therefore not considered to be an eye irritant. In normal handling and use the large glassy crystals would not be able to enter the eye easily and in addition over 50 years of occupational exposure to all borate has indicated no adverse effects on the human eye While no data has been obtained for disodium tetraborate anhydrous, it the same argument can be made and therefore is assumed that disodium tetraborate anhydrous is not an eye irritant. Therefore further testing is not warranted in the interests of animal welfare and protecting laboratory animals	

Section A6.1.4	**Acute Eye Irritation**
Annex Point IIA6.1.4	**Disodium Tetraborate Anhydrous**
Undertaking of intended data submission [ ]	n.a.
	Evaluation by Competent Authorities
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	17 May 2005
Evaluation of applicant's justification	*The justification of the applicant is not acceptable.* In two eye irritation studies, both disodium tetraborate decahydrate and disodium tetraborate pentahydrate induced average scores for redness and chemosis of 2.1 and 2.8 respectively. It is suggested that this could be due to the glassy cristalline nature of these compounds. However, no convincing evidence is provided to exclude the possibility that the eye irritation is the result of a non mechanical action of these compounds. Workers exposed occupationally to borax dust (average air concentration 4.1 mg/m3) reported, among others, eye irritation (Garabrant et al., 1984, 1985). Therefore, disodium tetraborates should be classified as eye irritants. The applicant agreed with the classification as stated in the comments on the draft assessment report.
Conclusion	The justification of the applicant is not acceptable.
Remarks	
	COMMENTS FROM OTHER MEMBER STATE *(specify)*
Date	*Give date of comments submitted*
Evaluation of applicant's justification	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Decahydrate

Official use only

1 REFERENCE

1.1	**Reference**	[redacted] (1989) Primary Eye Irritation of Borax [redacted] Electronic File
1.2	**Data protection**	Yes
1.2.1	Data owner	[redacted]
1.2.2	Companies with letter of access	**Curent Access** [redacted]
1.2.3	Criteria for data protection	Data on new a.s. for first entry to Annex I/IA

2 GUIDELINES AND QUALITY ASSURANCE

2.1	**Guideline study**	Yes FIFRA (40 CFR 158, 162); TSCA (40 CFR 798). Although not carried out to an OECD protocol, the study has been carried out to an US EPA acceptable protocol and meets the requirements of OECD 405, although the report lacks detail
2.2	**GLP**	Yes
2.3	**Deviations**	Although the studies on Disodium Tetraborate Decahydrate have not been carried out to OECD protocols, two studies have been carried out to US Government Guidelines. Further testing of boric acid is therefore not justified in the interests of protecting laboratory animals.

3 MATERIALS AND METHODS

3.1	**Test material**	As given in section 2 Borax 10 Mol: Disodium Tetraborate Decahydrate
3.1.1	Lot/Batch number	8M8D
3.1.2	Specification	As given in section 2
3.1.2.1	**Description**	White powder
3.1.2.2	**Purity**	>99%
3.1.2.3	**Stability**	Stable

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Decahydrate

3.2	**Test Animals**	Non-entry field
3.2.1	Species	Rabbit
3.2.2	Strain	New Zealand White
3.2.3	Source	Approved USDA supplier
3.2.4	Sex	Male and Female
3.2.5	Age/weight at study initiation	Not reported
3.2.6	Number of animals per group	3 male; 3 female
3.2.7	Control animals	No
3.3	**Administration/ Exposure**	
3.3.1	Preparation of test substance	*Test substance was used as delivered*
3.3.2	Amount of active substance instilled	77 mg (weight of 0.1 ml volume in accordance with the guidelines)
3.3.3	Exposure period	24h followed by rinsing with physiological saline
3.3.4	Post exposure period	21 days
3.4	**Examinations**	
3.4.1	Ophthalmoscopic examination	No
3.4.1.1	**Scoring system**	Scoring in report according to Draize, but scoring reported here according to EU 67/548/EEC
3.4.1.2	**Examination time points**	60min, 24h, 48h, 72h, 4d, 7d, 21d
3.4.2	Other investigations	
3.5	**Further remarks**	
		4 RESULTS AND DISCUSSION
4.1	**Clinical signs**	Table A6_1_4E-1.
4.2	**Average score**	Non-entry field
4.2.1	Cornea	0.72
4.2.2	Iris	0.61
4.2.3	Conjunctiva	Non-entry field
4.2.3.1	**Redness**	1.50
4.2.3.2	**Chemosis**	2.11

Section 6.1.4	**Acute Eye Irritation**
Annex Point IIA6.1.4	Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Decahydrate

4.3	**Reversibility**	Yes Chemosis and Redness reversed by thirteen days
4.4	**Other**	Effects on conjunctivae redness and Chemosis were reversed by day 13. Changes included blanched, blistered and thickened appearance in conjunctiva
4.5	**Overall result**	Although the substance caused irritation, this irritation is thought to arise from the glassy nature of the crystals of the substance and therefore classification is not relevant. For the closely related disodium tetraborate pentahydrate, a study was carried out where the sample was ground to a fine powder before instillation to reduce the glassy, sharp crystals in the sample and the previously seen irritancy was not observed indicating that the irritancy is mechanical and related to the shape of the crystal. In normal handling and use the large glassy crystals would not be able to enter the eye easily and in addition over 50 years of occupational exposure to disodium tetraborate decahydrate and other borates has indicated no adverse effects on the human eye. Disodium tetraborate decahydrate is therefore not considered to be an eye irritant. Disodium tetraborate decahydrate and disodium tetraborate decahydrate are used as a buffer in eyewashes
		5 APPLICANT'S SUMMARY AND CONCLUSION
5.1	**Materials and methods**	Eye irritation study in New Zealand white rabbits to FIFRA (40 CFR 158, 162); TSCA (40 CFR 798). 77 mg substance was instilled in the eyes for 24 hours followed by rinsing with physiological saline

Section 6.1.4	**Acute Eye Irritation**
Annex Point IIA6.1.4	Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Decahydrate

5.2	**Results and discussion**	Effects on conjunctivae redness and Chemosis were reversed by day 13. Changes included blanched, blistered and thickened appearance in conjunctiva. Although the substance caused slight irritation, this irritation is thought to arise from the glassy nature of the crystals of the substance and therefore classification is not relevant. For the closely related disodium tetraborate pentahydrate, a study was carried out where the sample was ground to a fine powder before instillation to reduce the glassy, sharp crystals in the sample and the previously seen irritancy was not observed indicating that the irritancy is mechanical and related to the shape of the crystal. In normal handling and use the large glassy crystals would not be able to enter the eye easily and in addition over 50 years of occupational exposure to disodium tetraborate decahydrate and other borates has indicated no adverse effects on the human eye. Disodium tetraborate decahydrate is therefore not considered to be an eye irritant. Disodium tetraborate decahydrate and disodium tetraborate decahydrate are used as a buffer in eyewashes
5.3	**Conclusion**	Based on the small level of Irriation and the information on disodium tetraborate pentahydrate, the large glassy nature of the crystals and the fact that over 50 years of occupational exposure to disodium tetraborate decahydrate and other borates has indicated no adverse effects on the human eye, the substance is not considered to be an eye irritant.
5.3.1	Reliability	2
5.3.2	Deficiencies	

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Decahydrate

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	28 February 2005
Materials and Methods	The applicant states that the test material was used as delivered. In the study report it is reported that the material is a white powder. In the study protocol it is stated that solid or granular material will be ground to fine dust. In the study report no information is provided whether the test material was or was not ground, although in the project instructions it is stated that the study should be run strictly in accordance with the referenced protocol..
Results and discussion	The applicant considers the observed eye irritation to be caused by the glassy nature of the crystals and, since in normal handling and use the large glassy crystals would not be able to enter the eye easily, therefore discards the irritation observed in the animals as irrelevant. The RMS disagrees since, a: the test material is described as a powder (not as large, sharp glassy crystals), b: the test material may have been ground to fine dust before instillation in the eyes, according to the test protocol. In a recent study by Cerven (2000), disodium tetraborate pentahydrate appeared to be irritating to the eyes.
Conclusion	In view of the average score of 2.1 for chemosis, the test material is classified as irritant (Xi, R36).
Reliability	2
Acceptability	acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Appendix

Table A6_1_4E-1. Results of eye irritation study

Use this table, if relevant effects occur.

	Cornea	Iris	Conjunctiva	
			redness	chemosis
score (average of animals investigated)	0 to 4	0 to 2	0 to 3	0 to4
60 min	1.00	1.00	0.17	2.33
24 h	1.00	1.00	1.67	2.50
48 h	0.83	0.50	1.50	2.17
72 h	0.33	0.33	1.33	1.67
Average 24h, 48h, 72h	0.72	0.61	1.50	2.11
Area effected				
Maximum average score (including area affected, max 110)				
Reversibility*	yes	yes	yes	yes
average time for reversion	By Day 7	By Day 7	By Day 13	By Day 13

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Pentahydrate

Official use only

1 REFERENCE

1.1	**Reference**	[redacted] (2000). Acute eye irritation on rabbits: [redacted] Sodium tetraborate pentahydrate Lot #OE26. [redacted]
1.2	**Data protection**	Yes
1.2.1	Data owner	[redacted]
1.2.2	Companies with letter of access	**Curent Access** [redacted]
1.2.3	Criteria for data protection	Data on new a.s. for first entry to Annex I/IA

2 GUIDELINES AND QUALITY ASSURANCE

2.1	**Guideline study**	Yes FIFRA (40 CFR 158, 430); EPA OPPTS Series 870.2400 Final Guideline August 1998. Confirms to OECD 405
2.2	**GLP**	Yes
2.3	**Deviations**	

3 MATERIALS AND METHODS

3.1	**Test material**	As given in section 2 Borax 5 Mol: Sodium Tetraborate Pentahydrate
3.1.1	Lot/Batch number	OE26
3.1.2	Specification	As given in section 2
3.1.2.1	**Description**	White powder
3.1.2.2	**Purity**	>99%
3.1.2.3	**Stability**	Stable

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Pentahydrate

3.2	**Test Animals**	Non-entry field
3.2.1	Species	Rabbit
3.2.2	Strain	New Zealand White
3.2.3	Source	Harvey's Lake, PA
3.2.4	Sex	Male and Female
3.2.5	Age/weight at study initiation	2.4 – 2.8 kg
3.2.6	Number of animals per group	3 male; 3 female
3.2.7	Control animals	No
3.3	**Administration/ Exposure**	
3.3.1	Preparation of test substance	Test substance was ground to a fine powder
3.3.2	Amount of active substance instilled	0.08 ml equivalent.
3.3.3	Exposure period	14 days – NO rinsing
3.3.4	Post exposure period	14 days
3.4	**Examinations**	
3.4.1	Ophthalmoscopic examination	No
3.4.1.1	**Scoring system**	Scoring in report according to Draize, but scoring reported here according to EU 67/548/EEC
3.4.1.2	**Examination time points**	60min, 24h, 48h, 72h, 4d, 7d, 14 d
3.4.2	Other investigations	
3.5	**Further remarks**	
		4 RESULTS AND DISCUSSION
4.1	**Clinical signs**	Table A6_1_4E-1.
4.2	**Average score**	Non-entry field
4.2.1	Cornea	0.22
4.2.2	Iris	0.22
4.2.3	Conjunctiva	Non-entry field

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Pentahydrate

4.2.3.1 Redness	0.89
4.2.3.2 Chemosis	1.89
4.3 Reversibility	Yes By 14 days
4.4 Other	This study was carried out at the request of the US EPA to confirm that the eye irritation previously seen with disodium tetraborate pentahydrate was caused by the glassy nature of the crystals of substance and not a chemical effect of irritation, the sample in this study was ground to a fine powder before instillation to reduce the glassy, sharp crystals in the sample. In normal handling and use the large glassy crystals would not be able to enter the eye easily and in addition over 50 years of occupational exposure to disodium tetraborate pentahydrate as well as other borates has indicated no adverse effects on the human eye. As a result for this study the US EPA accepted that the effects were mechanical downgraded its classification according to US FIFRA to Toxicity II (40 CFR 156) by ocular administration (Corneal involvement or irritation clearing in 8-21 days). The substance is not classifiable in the EU under Directive 67/548/EEC.
4.5 Overall result	Not classifiable in the EU under Directive 67/548/EEC. Disodium tetraborate decahydrate and disodium tetraborate decahydrate are used as a buffer in eyewashes
	5 APPLICANT'S SUMMARY AND CONCLUSION
5.1 Materials and methods	Eye irritation study in New Zealand white rabbits t oFIFRA (40 CFR 158, 430); EPA OPPTS Series 870.2400 Final Guideline August 1998. Confirms to OECD 405. After grinding to a fine powder 0.08 ml substance was instilled in the eyes . No rinsing took place

Section 6.1.4	**Acute Eye Irritation**
Annex Point IIA6.1.4	Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Pentahydrate

5.2	**Results and discussion**	Not and Eye irritant This study was carried out at the request of the US EPA to confirm that the eye irritation previously seen with disodium tetraborate pentahydrate was caused by the glassy nature of the crystals of substance and not a chemical effect of irritation, the sample in this study was ground to a fine powder before instillation to reduce the glassy, sharp crystals in the sample. In normal handling and use the large glassy crystals would not be able to enter the eye easily and in addition over 50 years of occupational exposure to disodium tetraborate pentahydrate as well as other borates has indicated no adverse effects on the human eye. As a result for this study the US EPA accepted that the effects were mechanical downgraded its classification according to US FIFRA to Toxicity II (40 CFR 156) by ocular administration (Corneal involvement or irritation clearing in 8-21 days Disodium tetraborate decahydrate and disodium tetraborate decahydrate are used as a buffer in eyewashes
5.3	**Conclusion**	Non Irritant
5.3.1	Reliability	1
5.3.2	Deficiencies	

Section 6.1.4 Acute Eye Irritation

Annex Point IIA6.1.4 Section A6.1.4Rabbit Eye Irritation Study; Disodium Tetraborate Pentahydrate

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	2 March 2005
Materials and Methods	The applicant's version is incorrect on a number of points. In the study report the test substance is decribed as a white cristalline solid. Only three females were tested (no males).
Results and discussion	The scores on redness of the conjunctiva in table A6_1_4E-1 are incorrect. In the study report the average scores for redness are 2.7, 3 and 2.7 at 24, 48 and 72 h respectively. The average over these three time points is 2.8 for redness. The applicant suggests that in a previous study with disodium tetraborate dechydrate the eye irration was caused by the glassy cristalline nature of the substance. However, in the present study the test material was ground to a fine powder before instillation into the eyes.
Conclusion	In view of the average score of 2.8 for redness, the test material is classified as irritant (Xi, R36)
Reliability	2
Acceptability	acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Appendix

Table A6_1_4E-1. Results of eye irritation study

Use this table, if relevant effects occur.

	Cornea	Iris	Conjunctiva	
			redness	chemosis
score (average of animals investigated)	0 to 4	0 to 2	0 to 3	0 to4
60 min	0.00	0.00	2.33	3.00
24 h	0.00	0.67	1.00	2.00
48 h	0.00	0.00	1.00	2.00
72 h	0.67	0.00	0.67	1.67
Average 24h, 48h, 72h	0.22	0.22	0.89	1.89
Area effected				
Maximum average score (including area affected, max 110)				
Reversibility*	yes	yes	yes	yes
average time for reversion	By Day 14	By Day 14	By Day 14	By Day 14

Section A6.1.5 Annex Point IIA6.1.5	Skin sensitisation Buehler Test Disodium Tetraborate Anhydrous	
	JUSTIFICATION FOR NON-SUBMISSION OF DATA	Official use only
Other existing data [ ] Limited exposure [ ]	Technically not feasible [ ] Scientifically unjustified [ ] Other justification [x]	
Detailed justification:	Anhydrous disodium tetraborate is the anhydrous salt of disodium tetraborate decahydrate and disodium tetraborate pentahydrate. For practical purposes one part of Anhydrous Disodium tetraborate is equivalent to 1.45 parts of disodium tetraborate pentahydrate; 1.9 parts of disodium tetraborate decahydrate; 1.02 parts disodium octaborate tetrahydrate and in aqueous solution 1.23 parts of boric acid. It is hygroscopic and takes up water to form a hydrated salt and like the other borates, in solution it will exist as undissociated boric acid (see Doc IIIA A7.1.1.1 Hydrolysis and Doc IIIA Read Across Statement). Sensitisation studies on disodium tetraborate pentahydrate, disodium tetraborate decahydrate, disodium octaborate tetrahydrate and boric acid indicate that borates are not skin sensitisers. No evidence of skin sensitisation has been seen in humans exposed occupationally to sodium borates or in a human patch test with a 3% aqueous boric acid solution (Bruze et al., 1995). Bruze M, E. Hradil, I-L. Eriksohn, B. Gruvberger and L.Widstrom, Occupational allergic contact dermatitis from alkanolamineborates in metalworking fluids, Contact Dermatitis 32, 24 -27 (1995).	
Undertaking of intended data submission [ ]	n.a.	

Evaluation by Competent Authorities	
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	17May 2005
Evaluation of applicant's justification	The justification of the applicant is acceptable
Conclusion	The justification of the applicant is acceptable
Remarks	
	COMMENTS FROM OTHER MEMBER STATE *(specify)*
Date	*Give date of comments submitted*
Evaluation of applicant's justification	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

Official use only

		1 REFERENCE
1.1	**Reference**	[redacted] (1994), Dermal sensitization test-Buehler method on sodium tetraborate decahydrate. [redacted] Electronic File
1.2	**Data protection**	Yes
1.2.1	Data owner	[redacted]
1.2.2	Companies with letter of access	**Curent Access** [redacted]
1.2.3	Criteria for data protection	Data on new a.s. for first entry to Annex I/IA
		2 GUIDELINES AND QUALITY ASSURANCE
2.1	**Guideline study**	Yes OECD Guide-line 406 "Skin Sensitization"
2.2	**GLP**	Yes
2.3	**Deviations**	No
		3 MATERIALS AND METHODS
3.1	**Test material**	Sodium Tetraborate Decahydrate
3.1.1	Lot/Batch number	Lot #4J18-02271
3.1.2	Specification	As given in section 2
3.1.2.1	**Description**	White powder
3.1.2.2	**Purity**	>99%
3.1.2.3	**Stability**	Stable
3.1.2.4	**Preparation of test substance for application**	*a) <u>for induction:</u> used as delivered moistened with distilled water (95%w/v)* *b) <u>for challenge:</u> used as delivered moistened with distilled water (95%w/v)*

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

3.1.2.5	**Pre-test performed on irritant effects**	Yes
3.2	**Test Animals**	Non-entry field
3.2.1	Species	Guinea pigs
3.2.2	Strain	Hartley albino
3.2.3	Source	Davidson's Mill Farms, South Brunswick, NJ
3.2.4	Sex	
3.2.5	Age/weight at study initiation	Young adult males: 314 -411 grams; Young adult females: 256-386 grams
3.2.6	Number of animals per group	Test Group: 20 animals Naive Control: 10 animals Positive Control: 20 animals Positive Naive Control: 10 animals
3.2.7	Control animals	Yes
3.3	**Administration/ Exposure**	State study type: Non-Adjuvant
3.3.1	Induction schedule	day 0 – day –7 – day 21 *Table A6_1_5-1.*
3.3.2	Way of Induction	Topical Occlusive

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

3.3.3	Concentrations used for induction	0.4 g 95% w/w substance moistened with distilled water to enhance skin contact
3.3.4		
3.3.5	Challenge schedule	Day 28; Table A6_1_5-1.
3.3.6	Concentrations used for challenge	95% w/w substance moistened with distilled water to enhance skin contact
3.3.7	Rechallenge	No
3.3.8	Scoring schedule	24h, 48h after challenge
3.3.9	Removal of the test substance	After 6 hours test substance wiped off with water
3.3.10	Positive control substance	Dinitrochlorobenzene
3.4	**Examinations**	Non-entry field
3.4.1	Pilot study	No
3.5	**Further remarks**	
		4 RESULTS AND DISCUSSION
4.1	**Results of pilot studies**	No pilot study
4.2	**Results of test**	See Table A6_1_5-2
4.2.1	24h after challenge	0/20
4.2.2	48h after challenge	0/20
4.2.3	Other findings	
4.3	**Overall result**	Non -sensitiser
		5 APPLICANT'S SUMMARY AND CONCLUSION
5.1	**Materials and methods**	OECD Guide-line 406 "Skin Sensitisation" method (Buehler test) using 95% w/w disodium tetraborate decahydrate moistened with distilled water to enhance skin contact
5.2	**Results and discussion**	No irritation was observed
5.3	**Conclusion**	Non-sensitiser
5.3.1	Reliability	1
5.3.2	Deficiencies	No

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5

Buehler Test

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	4 March 2005
Materials and Methods	Induction dose was administered at days 0, 7 and 14. Otherwise the version of the applicant is acceptable.
Results and discussion	The version of the applicant is adopted.
Conclusion	The version of the applicant is adopted.
Reliability	1
Acceptability	Acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

Table A6_1_5-1. Detailed information including induction/challenge/scoring schedule for skin sensitisation test

Treatments	Buehler test	Observations/Remarks *give information on irritation effects*
	day of treatment	
Induction 1	day 0	No irritation observed
Induction 2	7	No irritation observed
Induction 3	14	No irritation observed
challenge	28	No irritation observed
(rechallenge)		
scoring 1	29	No irritation observed
scoring 2	30	No irritation observed

Table A6_1_5-2. Result of skin sensitisation test

	Number of animals with signs of allergic reactions / number of animals in group		
	Negative control	**Test group**	**Positive control**
scored after 24h	***0 / 10***	***0 / 20***	***10/20***
scored after 48h	***0 / 10***	***0 / 20***	***7/20***

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

Official use only

1 REFERENCE

1.1	**Reference**	[redacted] (1994), Dermal sensitization test-Buehler method on sodium tetraborate pentahydrate. [redacted] Electronic File
1.2	**Data protection**	Yes
1.2.1	Data owner	[redacted]
1.2.2	Companies with letter of access	**Current Access** [redacted]
1.2.3	Criteria for data protection	Data on new a.s. for first entry to Annex I/IA

2 GUIDELINES AND QUALITY ASSURANCE

2.1	**Guideline study**	Yes OECD Guide-line 406 "Skin Sensitization"
2.2	**GLP**	Yes
2.3	**Deviations**	No

3 MATERIALS AND METHODS

3.1	**Test material**	Sodium Tetraborate Pentahydrate
3.1.1	Lot/Batch number	Lot #4ho2-2471
3.1.2	Specification	As given in section 2
3.1.2.1	**Description**	White powder
3.1.2.2	**Purity**	>99%
3.1.2.3	**Stability**	Stable
3.1.2.4	**Preparation of test substance for application**	*c) <u>for induction:</u> used as delivered moistened with distilled water (95%w/v)* *d) <u>for challenge:</u> used as delivered moistened with distilled water (95%w/v)*

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

3.1.2.5	**Pre-test performed on irritant effects**	Yes
3.2	**Test Animals**	Non-entry field
3.2.1	Species	Guinea pigs
3.2.2	Strain	Hartley albino
3.2.3	Source	Davidson's Mill Farms, South Brunswick, NJ
3.2.4	Sex	
3.2.5	Age/weight at study initiation	Young adult males: 314 -411 grams; Young adult females: 256-441 grams
3.2.6	Number of animals per group	Test Group: 20 animals Naive Control: 10 animals Positive Control: 20 animals Positive Naive Control: 10 animals
3.2.7	Control animals	Yes
3.3	**Administration/ Exposure**	State study type: Non-Adjuvant
3.3.1	Induction schedule	day 0 – day –7 – day 21 *Table A6_1_5-1.*
3.3.2	Way of Induction	Topical Occlusive

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

3.3.3	Concentrations used for induction	0.4 g 95% w/w substance moistened with distilled water to enhance skin contact
3.3.4		
3.3.5	Challenge schedule	Day 28; Table A6_1_5-1.
3.3.6	Concentrations used for challenge	95% w/w substance moistened with distilled water to enhance skin contact
3.3.7	Rechallenge	No
3.3.8	Scoring schedule	24h, 48h after challenge
3.3.9	Removal of the test substance	After 6 hours test substance wiped off with water
3.3.10	Positive control substance	Dinitrochlorobenzene
3.4	**Examinations**	Non-entry field
3.4.1	Pilot study	No
3.5	**Further remarks**	
		4 RESULTS AND DISCUSSION
4.1	**Results of pilot studies**	No pilot study
4.2	**Results of test**	See Table A6_1_5-2
4.2.1	24h after challenge	0/20
4.2.2	48h after challenge	0/20
4.2.3	Other findings	
4.3	**Overall result**	Non -sensitiser
		5 APPLICANT'S SUMMARY AND CONCLUSION
5.1	**Materials and methods**	OECD Guide-line 406 "Skin Sensitisation" method (Buehler test) using 95% w/w disodium tetraborate pentahydrate moistened with distilled water to enhance skin contact
5.2	**Results and discussion**	No irritation was observed
5.3	**Conclusion**	Non-sensitiser
5.3.1	Reliability	1
5.3.2	Deficiencies	No

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5

Buehler Test

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	4 March 2005
Materials and Methods	Induction dose was administered at days 0, 7 and 14. Otherwise the version of the applicant is acceptable.
Results and discussion	The version of the applicant is adopted.
Conclusion	The version of the applicant is adopted.
Reliability	1
Acceptability	Acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.1.5 Skin sensitisation

Annex Point IIA6.1.5 Buehler Test

Table A6_1_5-1. Detailed information including induction/challenge/scoring schedule for skin sensitisation test

Treatments	Buehler test	Observations/Remarks *give information on irritation effects*
	day of treatment	
Induction 1	day 0	Very faint erythema (0.5) observed at three sites at 24 hours after first induction dose. No other irritation observed
Induction 2	7	No irritation observed
Induction 3	14	No irritation observed
challenge	28	No irritation observed
(rechallenge)		
scoring 1	29	No irritation observed
scoring 2	30	No irritation observed

Table A6_1_5-2. Result of skin sensitisation test

	Number of animals with signs of allergic reactions / number of animals in group		
	Negative control	**Test group**	**Positive control**
scored after 24h	***0 / 10***	***0 / 20***	***10/20***
scored after 48h	***0 / 10***	***0 / 20***	***7/20***

Section A6.2 Percutaneous absorption (in vivo test)

Annex Point IIA6.2

Section A6.2 Human In vivo

Official use only

1 REFERENCE

1.1 Reference

[redacted] (1996). In Vivo Percutaneous Absorption of Boric Acid, Borax and Octaborate Tetrahydrate (DOT) in Man. [redacted]

Also published as Wester RC, Hui X, Hartway T, Maibach HI, Bell K, Schell MJ, Northington DJ, Strong P and Culver, BD. In vivo percutaneous absorption of boric acid, Borax and disodium octaborate tetrahydrate in humans compared to in vitro absorption in human skin from infinite to finite doses. Toxicol Sciences 45 42-51 (1998)

1.2 Data protection Yes

1.2.1 Data owner [redacted]

1.2.2 Companies with letter of access **Curent Access** [redacted]

1.2.3 Criteria for data protection *Data on new a.s. for first entry to Annex I/IA*

2 GUIDELINES AND QUALITY ASSURANCE

2.1 Guideline study No

Human Study specifically designed and therefore no specific guidelines available, but designed to comply with US 40 CFR, 160

2.2 GLP Yes

2.3 Deviations Not relevant

(If yes, describe deviations from test guidelines or refer to respective field numbers where these are described, e.g. "see 3.x.y")

3 MATERIALS AND METHODS

Section A6.2 Percutaneous absorption (in vivo test)

Annex Point IIA6.2 Section A6.2 Human In vivo

3.1	**Test material**	As given in section 2
3.1.1	Lot/Batch number	
3.1.2	Specification	As given in section 2
3.1.2.1	**Description**	White powder
3.1.2.2	**Purity**	>99%
3.1.2.3	**Stability**	Stable
3.1.2.4	**Radiolabelling**	^{10}B
3.2	**Test Animals**	Non-entry field
3.2.1	Species	Humans
3.2.2	Strain	
3.2.3	Source	
3.2.4	Sex	Male & female
3.2.5	Age/weight at study initiation	Age 22 -50
3.2.6	Number of animals per group	8/groups
3.2.7	Control animals	Internal controls (i.e. baseline boron measured)
3.3	**Administration/ Exposure**	Dermal both intact and abraded skin
3.3.1	Preparation of test site	Skin was washed and a 30 cm x 30 cm area marked on back
3.3.2	Concentration of test substance	5% Boric acid ; 5% Borax or 10% DOT in distilled water
3.3.3	Specific activity of test substance	
3.3.4	Volume applied	3 ml/900 cm²
3.3.5	Size of test site	900 cm²

Section A6.2 **Percutaneous absorption (in vivo test)**

Annex Point IIA6.2 Section A6.2 Human In vivo

3.3.6	Exposure period	After 5 days during which urine samples were collected the test substance was applied topically; air-dried and a commercial white T-shirt worn for 24 hours during which time urine was collected. At 24 hours the T-shirt was removed and analysed. The exposed areas were analysed for transepidermal water loss (TEWL) and then washed carefully with soap and distilled deionised water and all washing analysed. On day 11 the TEWL was measured and the treatment site dosed with 1.8 ml of 2% SDS (sodium lauryl sulphate) to cause irritation. On day 12 the TEWL was measured and the test substance was applied again topically; air-dried and a commercial white T-shirt worn for 24 hours during which time urine was collected. At 24 hours the T-shirt was removed and analysed. The exposed areas were analysed for transepidermal water loss (TEWL) and then washed carefully with soap and distilled deionised water and all washing analysed.
3.3.7	Sampling time	See above – Sample time 24 hours
3.3.8	Samples	Urine sampled as well as T-shirts worn and skin washings samples – see above

4 RESULTS AND DISCUSSION

4.1	**Toxic effects, clinical signs**	No adverse effects
4.2	**Dermal irritation**	No skin Irriation observed
4.3	**Recovery of labelled compound**	BA -76.5%; Borax 72%; DOT 78.5% Since the skin was washed 10 times and less 1 % was found I the last wash, it is assumed that most of the substance unaccounted for was in lost to outside clothing (over the T-shirt) an bedding during the 24 hour dosing period

4.4 Percutaneous absorption

Substance	% Dose Absorbed (95% CI)	Flux $\mu g/cm^2/hr$	Permeability Kp cm/hr.
5 % Boric Acid	0.226 ± 0.125	0.009	1.8×10^{-7}
5 % Borax[1]	0.210 ± 0.194	0.009	1.8×10^{-7}
10% DOT[2]	0.122 ± 0.10	0.010	1.0×10^{-7}

[1] Disodium tetraborate decahydrate

[2] Disodium octaborate tetrahydrate

5 APPLICANT'S SUMMARY AND CONCLUSION

Section A6.2 Percutaneous absorption (in vivo test)

Annex Point IIA6.2

Section A6.2 Human In vivo

5.1 Materials and methods

This study was designed to address absorption of typical solutions used in wood preservation and other biocidal uses.

Human Volunteers (8 per group) Group I, group II, and group III received two separate topical application of B^{10}-enriched 5% Boric Acid, 5% Borax, and 10% DOT solutions on their back skin, respectively and the in vivo percutaneous absorption was determined for a 24-hour dosing period. One dose was applied on day 5 under normal skin conditions and the other on day 12 under irritated skin conditions created by applying 2% SLS solution. Twenty- four hours after each topical dose, residual chemical on the dosed skin site was removed by skin wash. Urine samples were collected every 24 hours for 17 days. Urine samples from day 1 to day 4 were used to establish base boron levels and isotope ratios in the urine. The samples from day 5 to day 11 and day 12 to the end were used to compare absorbed level under normal skin and irritated skin conditions. To evaluate the dosing site skin condition, TEWL measurement and skin visual scoring were taken each time before dosing (including SLS treatment) and washing. To control any boron intake some food/beverage restrictions were instituted and daily detailed records were required. Boron analysis was done using inductively coupled mass spectrometry

5.2 Results and discussion

Approximately one-half of the administered topical dose was recovered after 24 hours in the T-shirt covering the dosed skin area and the skin washes. Pre-treatment with the potential skin irritant 2~ sodium lauryl sulphate had no effect on boron skin absorption for all three different dosage forms. No skin irritation was noted for any of the dosage forms.

Substance	% Dose Absorbed (95% CI)	Flux $\mu g/cm^2/hr$	Permeability Kp cm/hr.
5 % Boric Acid	0.226 ± 0.125	0.009	1.8×10^{-7}
5 % Borax[1]	0.210 ± 0.194	0.009	1.8×10^{-7}
10% DOT[2]	0.122 ± 0.10	0.010	1.0×10^{-7}

[1] Disodium tetraborate decahydrate

[2] Disodium octaborate tetrahydrate

5.3 Conclusion

Low skin absorption. For risk assessment where an absorbed dose is used the mean plus the standard deviation is used as a conservative absorption figure Boric acid = 0.351% absorption; Borax = 0.404; DOT = 0.132

The percutaneous absorption of disodium tetraborate decahydrate can be read across to disodium tetraborate pentahydrate and disodium tetraborate anhydrous

Disodium tetraborate pentahydrate only slightly less hydrated than the decahydrate. Anhydrous disodium tetraborate is the anhydrous salt of disodium tetraborate decahydrate and disodium tetraborate

Section A6.2 **Percutaneous absorption (in vivo test)**

Annex Point IIA6.2 Section A6.2 Human In vivo

		pentahydrate. For practical purposes one part of anhydrous disodium tetraborate is equivalent to 1.45 parts of disodium tetraborate pentahydrate; 1.9 parts of disodium tetraborate decahydrate; 1.02 parts disodium octaborate tetrahydrate and in aqueous solution 1.23 parts of boric acid. Anhydrous disodium tetraborate is hygroscopic and takes up water to form a hydrated salt and like the other borates, in solution it will exist as undissociated boric Acute dermal limit studies carried out on both hydrated forms and disodium octaborate tetrahydrate indicated the LD_{50} to be > 2000 mg/kg bw. In these studies, limited symptoms were seen with the tetraborates and no symptoms with disodium octaborate tetrahydrate suggesting minimal dermal absorption. In an acute dermal limit study on boric acid, the rabbit skin was abraded to increase the absorption. Even in this study there was limited symptoms observed and the acute dermal LD_{50} was > 2000 mg/kg bw. This data supports minimal absorption, which is supported by the results of the human percutaneous absorption study Since anhydrous disodium tetraborate and disodium tetraborate pentahydrate will form the various similar borates in the moistened form that it is applied to the skin, they are unlikely to be absorbed at any greater rate than the other borates tested.
5.3.1	Reliability	1
5.3.2	Deficiencies	No

Section A6.2 Percutaneous absorption (in vivo test)

Annex Point IIA6.2

Section A6.2 Human In vivo

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	7 March 2005
Materials and Methods	The version of the applicant is accepted.
Results and discussion	In the studies total recovery of the applied dose ranged from 48.8-63.6%. Accordingly 36.4-51.2% of the applied dose is not accounted for. This may be due to loss to outside clothing and bedding, as suggested by the study authors. However, part of the lost dose may be located in the body or in the skin at the application site, which in that case should be considered as being absorbed. As such, the absorption estimates from this study are unreliable. On the other hand, toxicokinetic studies also indicate that borates have a low dermal absorption and low potential for accumulation in the body. In this respect the present data are in line with dermal absorption data from other studies. Therefore, based on this study and other data a dermal absorption borates of 0.5% can be assumed as a reasonable worst case estimate.
Conclusion	Reasonable worst case estimate for dermal absorption of borates is 0.5%.
Reliability	3
Acceptability	acceptable
Remarks	
	COMMENTS FROM ...
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.2-A10 **Doc III A Read Across to Boric Acid**

Annex Point Section A6.2-A10

6 APPLICANT'S SUMMARY AND CONCLUSION

Section A6.2-A10 Doc III A Read Across to Boric Acid

Annex Point Section A6.2-A10

Conversion factors to Boron Equivalents

		Conversion Factor for Equivalent dose of B
Boric acid	H_3BO_3	0.175
Boric oxide	B_2O_3	0.311
Disodium tetraborate decahydrate (Borax)	$Na_2B_4O_7 \bullet 10H_2O$	0.113
Disodium tetraborate pentahydrate	$Na_2B_4O_7 \bullet 5H_2O$	0.148
Disodium tetraborate anhydrous	$Na_2B_4O_7$	0.215
Disodium octaborate tetrahydrate	$Na_2B_8O_{13} \bullet 4H_2O$	0.210

Therefore, regardless of whether the boron source is boric acid or one of the other borates (such as boric oxide or a sodium borate), monomeric species are predominant in most biological fluids as well as under environmental conditions (Maeda, 1979). This was verified in an independent study aimed at identifying the species present in systems under typical biologically active conditions, i.e. pH 6.5-pH7.5 and <0.02M. Raman spectroscopy confirmed the presence of undissociated boric $B(OH)_3$ as the species present (Doc IIIA 7.1.1.1 Hydrolysis Boric Acid.doc) (de Vette et al., 2001)

B2O3 + 3H2O = 2(H3BO3)

Na2B8O13.4H2O + H2O = Na2O + 4(B2O3).4H2O = 2NaOH + 8(H3BO3)

Na2B4O7 xH2O (x = 0, 5 or 10) = 2Na2O + B2O3 x H2O = 4NaOH + 2 (H3BO3)

At the levels of substance introduced in toxicological studies the rates of dissolution may vary with tetraborate anhydrous and disodium tetraborate pentahydrate being slower. The main concerns could be local irritation at the site of contact. However, local irritation is not observed acute studies.

In long-term oral studies of boric acid and disodium tetraborate decahydrate, the test substance was administered in the diet (over a period of roughly 8 hours when rodents eat) and therefore the test material was given over an extended period of time, not as a bolus. Any differences in the dissolution rates of borates should make no difference in their systemic toxicity, which is the primary concern. No evidence of local GI tract irritation has been seen in any study carried out.

For dermal studies, absorption is very low (>0.4%) in human studies and there is no evidence to indicate that any borate is absorbed significantly more than another borate.

Absorption by inhalation is expected to be 100%. Since respirable particles are very small, they are likely to be dissolved very quickly on

the lung surfaces. The data from acute inhalation limit studies indicates some minor effects of dust exposure, but there does not appear to be a significant difference in effects between the borates tested.

In genotoxicity *in vitro* studies the borates will not be bioavailable unless dissolved in the test media. Once absorbed in the cells they will exist as undissociated boric acid. In *in vivo* studies, bioavailability will also be dependent on dissolution and the same arguments apply as for long-term toxicity.

The equivalence between the borates is indicated by studies in dogs and rats where both boric acid and disodium tetraborate decahydrate were tested. The results indicate that the effects seen are equivalent on boron basis indicating that the bioavailability of the two borates is similar.

Comparison of Toxicology of Boric Acid and Borax (Disodium Tetraborate Decahydrate)

Dose in diet	Dose	Symptoms
*****2 Year Dog Studies** Doc IIIA A_6_4.1 Subchronic 2 year Dogs Boric acid.doc & Doc IIIA A_6_4.1 Subchronic 2 year Dogs Tetraborates BA Entry.doc		
0.2 % BA	***10.9 mg B/kg (8.8)	No symptoms
0.309 % Borax	***9.6 mg B/kg (8.8)	No symptoms
0.67 % BA	***40.8 mg B/kg (39)	3/8 diarrhoea on 2-4 occasions. Soft stools in all dogs on 2 –5 occasions. 1/8 exema after 19 weeks ↓ Testicular weight; 2 dogs azospermic; 2
1.03 % Borax	***38.1 mg B/kg (39)	6/8 diarrhoea 1-6 occasions. Instances of soft stools in all dogs up to 26 weeks. ↓ Testicular weight; generalised spermatic arrest; atrophy of the seminiferous
2 Year Rat Studies Doc IIIA A_6_5 Chronic rat Boric acid.doc & Doc IIIA A_6_5 Chronic Rat Tetraborates BA Entry.doc		

Section A6.2-A10 Doc III A Read Across to Boric Acid

Annex Point Section A6.2-A10

2 Year Rat Studies Doc III A A_6_5 Chronic rat Boric acid.doc & Doc III A A_6_5 Chronic Rat Tetraborates BA Entry.doc		
0.2 % BA	17.5 mg B/kg	No effects
0.308% Borax	17.5 mg B/kg	No effects
0.67 % BA	58.5 mg B/kg	Clinical signs of Coarse hair coats, hunched position, swollen pads and inflamed bleeding eyes ↓ Body weight gain ↓ Red cell volume and haemoglobin Testicular degeneration Testicular atrophy No viable sperm in atrophied testis Complete sterility ↓ Body weight gain in females ↓ Reduced food intake
1.03 % Borax	58.5 mg B/kg	Clinical signs of Coarse hair coats, hunched position, swollen pads and inflamed bleeding eyes ↓ Body weight gain ↓ Red cell volume and haemoglobin Testicular degeneration Testicular atrophy No viable sperm in atrophied testis Complete sterility ↓ Reduced food intake

* There are a number of flaws in these studies that make them unsuitable for evaluation for risk assessment see Doc III Dog 90 day studies

** Dose estimated form actual dietary intake. Dose in Brackets based on standard assumptions regarding body weight and food consumption and as reported in the study reports

*** There are a number of flaws in these studies that make them unsuitable for evaluation for risk assessment see Doc IIIA Dog 2 year studies

References

1. Farmer, 1982 Structural Chemistry in the Borate Industry., Chem and Ind.,
2. Ingri, N Sven. Kem. Tidskr. 75(4), 199 (1963)
3. Kirk – Othmer Encyclopedia of Chemical Technology, V4, 1992, pp 378-380
4. Holleman, 1995. Lehrbuch der anorganischen Chemie. 101st ed de Gruyter, Berlin, copyright

Section A6.2-A10 Doc III A Read Across to Boric Acid

Annex Point Section A6.2-A10

5. De Vette, [redacted] 2001 Hydrolysis as a function of pH and identification of breakdown products. A study on the identification and comparison of the dissociation products of [redacted] Borax [redacted] and Boric Acid [redacted] Aqueous Solution using Raman spectrometry. [redacted]
6. Maeda M, Raman Spectra of polyborate ions in aqueous solution. J Inorg. Nucl. Chem., Vol 41, pp 1217-1220 (1979)
7. Rickards, [redacted] 2004 [redacted]

Section A6.2-A10 Doc III A Read Across to Boric Acid

Annex Point Section A6.2-A10

	Evaluation by Competent Authorities
	Use separate "evaluation boxes" to provide transparency as to the comments and views submitted
	EVALUATION BY RAPPORTEUR MEMBER STATE
Date	9 March 2005
Materials and Methods	Not applicable.
Results and discussion	In aqueous solutions at physiological and acidic pH, simple borates such as disodium tetraborate decahydrate ($Na_2B_4O_7 \bullet 10H_2O$; borax), disodium tetraborate pentahydrate ($Na_2B_4O_7 \bullet 5H_2O$; borax pentahydrate), boric oxide (B_2O_3) and disodium octaborate tetrahydrate ($Na_2B_8O_{13} \bullet 4H_2O$) will exist as undissociated boric acid. Therefore, the toxicokinetics and toxicological effects of boric acid, disodium tetraborate decahydrate, boric oxide (B2O3) and disodium octaborate tetrahydrate are likely to be similar on a boron equivalents basis. Therefore, it is justified to draw conclusions on disodium tetraborates on the basis of data on studies on toxicokinetics and toxicity of other simple borates such as boric acid.
Conclusion	It is justified to draw conclusions on disodium tetraborates on the basis of data on studies on toxicokinetics and toxicity of other simple borates such as boric acid. The effects assessment of borates, as described above by the applicant, is not adopted. For a detailed evaluation of the toxicokinetics and toxicology of borates see DOC II A of the disodium tetraborates evaluation.
Reliability	Not applicable.
Acceptability	
Remarks	
	COMMENTS FROM ... *(specify)*
Date	*Give date of comments submitted*
Materials and Methods	*Discuss additional relevant discrepancies referring to the (sub)heading numbers and to applicant's summary and conclusion.* *Discuss if deviating from view of rapporteur member state*
Results and discussion	*Discuss if deviating from view of rapporteur member state*
Conclusion	*Discuss if deviating from view of rapporteur member state*
Reliability	*Discuss if deviating from view of rapporteur member state*
Acceptability	*Discuss if deviating from view of rapporteur member state*
Remarks	

Section A6.4.1

Annex Point IIA6.4

Subchronic toxicity

Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier

Official use only

7 REFERENCE

7.1 Reference

[redacted] (1966) Two-Year Dietary Feeding -- Dogs. Borax (Sodium Tetraborate Decahydrate). Final Report. [redacted]

This study was published in summary form in Weir RJ and Fisher RS. (1972) Toxicological studies on borax and boric acid. Toxicol Appl Pharmacol. 23(3):351-64. Unfortunately, the published version does not always accurately reflect the original study reports. Thus, it is necessary to evaluate the original study reports to appreciate the limitations of these studies.

7.2 Data protection

Yes

7.2.1 Data owner

[redacted]

7.2.2

7.2.3 Criteria for data protection

Data on new a.s. for first entry to Annex I

8 GUIDELINES AND QUALITY ASSURANCE

Section A6.4.1	**Subchronic toxicity**
Annex Point IIA6.4	*Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier*

8.1	**Guideline study**	No. No guidelines available at the time the study was conducted.
8.2	**GLP**	No. GLP was not compulsory at the time the study was performed.
8.3	**Deviations**	No guidelines available at the time the study was conducted.
		9 MATERIALS AND METHODS
9.1	**Test material**	Borax was provided [redacted]
9.1.1	Lot/Batch number	Not stated.
9.1.2	Specification	Not stated.
9.1.2.1	**Description**	"Fine, white powder without noticeable odor"
9.1.2.2	**Purity**	Boron content of the test substance varied between 103.2 and 105.4% of the theoretical value. Analysis performed monthly by [redacted]
9.1.2.3	**Stability**	Test material expected to be stable.
9.2	**Test Animals**	
9.2.1	Species	Dog
9.2.2	Strain	Purebred Beagle
9.2.3	Source	Unknown
9.2.4	Sex	Male and female
9.2.5	Age/weight at study initiation	Body weights of the dogs ranged from 4.2-11.5 kg during the first week of the study. The dogs were described as "young," but no specific age was provided.
9.2.6	Number of animals per group	4 males and 4 females per group at the start of the study. However, these were sacrificed at different time intervals. Consequently, the number of dogs per sex per group varied from 1-2 at any given time of sacrifice.
9.2.7	Control animals	Yes, the control group was employed as a common control with studies of borax. Separate control groups were used for the 2-year and 38-week studies, since they were started at different times.
9.3	**Administration/ Exposure**	Oral (diet)

Section A6.4.1

Annex Point IIA6.4

Subchronic toxicity

Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier

9.3.1	Duration of treatment	2 years, except the highest dose (38 weeks). Initially, groups of 4 male and 4 female dogs were fed diets containing 0, 0.051, 0.103, or 0.309% borax for up to 2 years. Because no effects were observed in this initial portion of the study, additional groups of 4 male and 4 female dogs were fed diets containing 0 or 1.03% borax for up to 38 weeks. Dogs were sacrificed at various time intervals, so the duration of exposure varied depending on the time of sacrifice of each dog. In the initial portion of the study, one male and one female from each group were sacrificed at one year. The remaining dogs were sacrificed at 2 years, except for one male and one female in the control and 0.309% groups, which were sacrificed 3 months post-exposure (2 years). In the second portion of the study, two males and two females were sacrificed at 26 weeks in the high dose (1.03%) and its concurrent control group. At 38 weeks, the one male and one female were sacrificed in the high dose and two male and two female concurrent control group dogs were sacrificed. One male and one female dog at the high dose were sacrificed 25 days post-exposure (38 weeks) to study recovery.
9.3.2	Frequency of exposure	Seven days per week
9.3.3	Postexposure period	One male and one female from the control and 0.309% groups were placed on the control diet for 3 months postexposure to evaluate recovery and disappearance of stored boron.
9.3.4	**Oral**	
9.3.4.1	**Type**	In food
9.3.4.2	**Concentration**	0, 0.051, 0.103, 0.309, 1.03% borax in the diet These concentrations provided doses of 0, 1.9, 3.6, 9.6, and 38 mg B/kg/day, based on the actual body weight and food consumption data in the study. Others in the scientific literature have described these doses as 0, 1.5, 2.9, 8.8, and 29 mg B/kg/day, but these calculations are based on standard assumptions regarding body weight and food consumption, not on the actual data from this study. Food consumption was *ad libitum*.
9.3.4.3	**Vehicle**	
9.3.4.4	**Concentration in vehicle**	
9.3.4.5	**Total volume applied**	

Section A6.4.1

Annex Point IIA6.4

Subchronic toxicity

Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier

9.3.4.6 Controls	Plain diet (Wayne Dog Meal). The diet was not analyzed for the background level of boron. Boron is an essential plant element, and plant-derived foods (such as fruits and vegetables) are significant dietary sources of boron. The background level of boron in the control diet was not considered in the calculations of the dose levels.
9.4 Examinations	
9.4.1 Observations	
9.4.1.1 Clinical signs	Yes. The dogs were observed daily for appearance, behaviour, and gross signs of systemic toxicity or pharmacological effects.
9.4.1.2 Mortality	Yes. Daily throughout the study.
9.4.2 Body weight	Yes. Weekly.
9.4.3 Food consumption	Yes. Weekly.
9.4.4 Water consumption	No.
9.4.5 Ophthalmoscopic examination	No.
9.4.6 Haematology	Yes. Except for the high dose and its concurrent control group, all surviving dogs initially and at 1, 3, 6, 12, 18, 24, and in some cases, 27 months. At the high dose (1.03%) and its concurrent control group, all dogs initially and at 4, 12, 26, and 38 weeks. Parameters: Haematocrit, haemoglobin concentration, erythrocyte count, total and differential leukocyte count, sedimentation rate.
9.4.7 Clinical Chemisty	Yes. Except for the high dose and its concurrent control group, all surviving dogs initially and at 1, 3, 6, 12, 18, 24, and in some cases, 27 months. At the high dose (1.03%) and its concurrent control group, all dogs initially and at 4, 12, 26, and 38 weeks. Parameters: glucose, blood urea nitrogen. In addition, serum glutamic-pyruvic transaminase and serum glutamic oxaloacetic transaminase were done on all dogs sacrificed at the one year interval and on the control and 0.309% groups sacrificed after two years of exposure.
9.4.8 Urinalysis	Yes. Except for the high dose and its concurrent control group, all surviving dogs initially and at 1, 3, 6, 12, 18, 24, and in some cases, 27 months. At the high dose (1.03%) and its concurrent control group, all dogs initially and at 4, 12, 26, and 38 weeks. Parameters: appearance, specific gravity, pH, protein, glucose, acetone, blood, and microscopic findings.
9.5 Sacrifice and pathology	Except for the high dose and its concurrent control group, one male and one female from each group were sacrificed at one year. The remaining dogs were sacrificed at 2 years, except for one male and one female in the control and 0.309% groups, which were sacrificed 3 months post-exposure (2 years). Two males and two females were sacrificed at 26 weeks in the high dose (1.03%) and its concurrent control group. At 38 weeks, the one

male and one female were sacrificed in the high dose and two male and two female concurrent control group dogs were sacrificed. One male and one female dog at the high dose were sacrificed 25 days post-exposure (38 weeks) to study recovery.

Section A6.4.1 **Subchronic toxicity**

Annex Point IIA6.4 *Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier*

9.5.1	Organ Weights	Yes. All dose groups. Organs: liver, kidneys, adrenals, testes, thymus, spleen, brain, heart, thyroid
9.5.2	Gross and histopathology	Yes. All dose groups. Organs: brain, thyroid, stomach, small and large intestines, liver, pancreas, kidneys, adrenals, spleen, heart, lungs, testes
9.5.3	Other examinations	Ejaculate specimens were collected from two male dogs each from the control group and the 0.309% group prior to the 2-year sacrifice for determination of sperm counts and motility. Blood and urine samples were taken from each dog initially and at 1, 3, 6, 12, 18, 24, and 27 months for subsequent analysis for boron content. Samples of various organs were frozen for subsequent analysis for boron content. One male and one female per group were selected for this determination at 12, 24, and 27 months. These organs included: brain, liver, kidney, body fat, and muscle. Pharmacokinetic Study. A single male dog from the control and 0.309% groups were placed in a metabolism cage for one month at the beginning of the study, two weeks at the one-year interval, and three months at the two-year interval to study the pharmacokinetics of boron. One female dog from both of these groups was added for the three-month terminal pharmacokinetic study. With the exception of the terminal balance study, urine and faeces were collected at 24-hour intervals; blood was collected weekly. In the terminal study, urine and faeces were collected 24-hours prior to compound removal and daily thereafter for a period of five weeks; blood was also collected 24 hours prior to compound removal, three times per week for the next 5 weeks, and weekly thereafter until termination (13 weeks). During the 28th week, one male from the control and the test groups (except the high dose) was selected for a 6-day study to determine the pH and various electrolytes in the blood and urine for the purpose of evaluating the acid-base balance.
9.5.4	Statistics	No.
9.6	**Further remarks**	This study was conducted in a parallel with a 2-year and 38-week study of boric acid in dogs. The studies were identical in design, and they employed a common control group.

10 RESULTS AND DISCUSSION

(Describe findings. If appropriate, include table. Sample tables are given below.)

10.1 Observations

10.1.1	Clinical signs	No effects attributable to the test material except for diarrhoea and soft stool at the high dose (6/8 diarrhoea 1-6 occasions; Instances of soft stools in all dogs up to 26 week).
10.1.2	Mortality	No mortalities at any dose.

Section A6.4.1

Annex Point IIA6.4

Subchronic toxicity

Section A6.4.1 Subchronic (2 year) – Oral Dogs Borax (Sodium Tetraborate Decahydrate. Also submitted in Boric Acid Dossier

10.2	**Body weight gain**	No effects attributable to the test material.
10.3	**Food consumption and compound intake**	No effects.
10.4	**Ophtalmoscopic examination**	
10.5	**Blood analysis**	
10.5.1	Haematology	No effects.
10.5.2	Clinical chemistry	No effects.
10.5.3	Urinalysis	No effects.
10.6	**Sacrifice and pathology**	
10.6.1	Organ weights	Testis weight and testis/body weight ratios were lower than controls at the high dose (1.03%) at both the 26- and 38-week sacrifices. The testicular weight and testis/body weight ratio of the one male on the 3-week recovery phase was less than the controls, which were sacrificed at the earlier time intervals. (There was no concurrent control male for the one male on the 3-week recovery phase.) No other treatment-related effects.
10.6.2	Gross and histopathology	At 1.03%, generalized spermatogenic arrest, progressing to complete atrophy of the seminiferous epithelium in various number of tubules was observed in the two male dogs sacrificed at 26 weeks. One of the two control males sacrificed at 26 weeks exhibited atrophy of the seminiferous epithelium in a few scattered tubules. In contrast, at 38 weeks, degenerative changes in the single male given 1.03% in the diet was not as severe as those found in the two control dogs sacrificed at 38 weeks. The authors concluded: "Despite the testicle changes in the control animals sacrificed at 38 weeks being somewhat greater than in the test animal and the small number of dogs sacrificed at each interval, the ingestion of the test material and testicular atrophy seems to be related since the two males sacrificed at the 26-week interval were severely affected." No histological effects at concentrations below 1.03% were considered compound-related.
10.7	**Other**	The authors reported that it was not possible to obtain sperm samples from any of the four test dogs at 1.03%. Two of four concurrent control dogs had normal sperm counts, one had a low sperm count, and the investigators were unable to obtain a sample from one control dog. Boron levels in blood, urine and feces were within background levels within four days after the dogs were removed from the test die (1.03%)t; this time may have been shorter but smaller intervals were not explored.