SCCS/1512/13
Revision of 18 june 2014

Scientific Committee on Consumer Safety

SCCS

OPINION ON

Zinc pyrithione

COLIPA n° P81

- Text from previous opinion (SCCNFP/0671/03) has not been edited -

The SCCS adopted this opinion at its 2nd plenary meeting of 18 June 2013

About the Scientific Committees

Three independent non-food Scientific Committees provide the Commission with the scientific advice it needs when preparing policy and proposals relating to consumer safety, public health and the environment. The Committees also draw the Commission's attention to the new or emerging problems which may pose an actual or potential threat.
They are: the Scientific Committee on Consumer Safety (SCCS), the Scientific Committee on Health and Environmental Risks (SCHER) and the Scientific Committee on Emerging and Newly Identified Health Risks (SCENIHR) and are made up of external experts.

In addition, the Commission relies upon the work of the European Food Safety Authority (EFSA), the European Medicines Agency (EMA), the European Centre for Disease prevention and Control (ECDC) and the European Chemicals Agency (ECHA).

SCCS

The Committee shall provide opinions on questions concerning all types of health and safety risks (notably chemical, biological, mechanical and other physical risks) of non-food consumer products (for example: cosmetic products and their ingredients, toys, textiles, clothing, personal care and household products such as detergents, etc.) and services (for example: tattooing, artificial sun tanning, etc.).

Scientific Committee members

Contact
European Commission
Health & Consumers
Directorate C: Public Health
Unit C2 – Health Information/ Secretariat of the Scientific Committee
Office: HTC 03/073 L-2920 Luxembourg
SANCO-C2-SCCS@ec.europa.eu

ISSN 1831-4767 ISBN 978-92-79-30111-7
Doi 10.2772/67588 ND-AQ-13-004-EN-N

ACKNOWLEDGMENTS

SCCS members
Dr. U. Bernauer (rapporteur)
Dr. P.J. Coenraads
Prof. G. Degen
Dr. M. Dusinska
Prof. D. Gawkrodger
Dr. W. Lilienblum
Prof. A. Luch
Dr. E. Nielsen
Prof. Th. Platzek
Dr S. Ch. Rastogi (chairman)
Dr. Ch. Rousselle
Dr. J. van Benthem

External experts
Prof. V. Rogiers
Prof. T. Sanner

For the revision

SCCS members
Dr. U. Bernauer (rapporteur)
Dr. P.J. Coenraads
Prof. G. Degen
Dr. M. Dusinska
Dr. W. Lilienblum
Prof. A. Luch
Dr. E. Nielsen
Prof. Th. Platzek
Dr S. Ch. Rastogi (chairman)
Dr. Ch. Rousselle
Dr. J. van Benthem

This opinion has been subject to a commenting period of minimum four weeks after its initial publication. Comments received during this time have been considered by the SCCS and discussed in the subsequent plenary meeting. Where appropriate, the text of the relevant sections of the opinion has been modified or explanations have been added. In the cases where the SCCS after consideration and discussion of the comments, has decided to maintain its initial views, the opinion (or the section concerned) has remained unchanged. Revised opinions carry the date of revision.

Keywords: SCCS, scientific opinion, preservative, P81, Zinc pyrithione, Regulation 1223/2009.

Opinion to be cited as: SCCS (Scientific Committee on Consumer Safety), Opinion on Zinc pyrithione, 18 June 2013, SCCS/1512/13, revision of 18 June 2014.

TABLE OF CONTENTS

1. BACKGROUND

Zinc pyrithione (ZPT) (CAS 13463-41-7; EU 236-671-3) with the chemical name: Bis[(2-pyridyl-1-oxo)-thio]zinc was introduced into the Cosmetics Directive as a preservative by Directive 82/368/EEC. It was authorised as a preservative at the maximum concentration of 0.5% with the limitation "Authorized in products rinsed off after use, forbidden in products for oral hygiene".

Back in 1984 (17/12/1984) the Scientific Committee on Cosmetology (SCC) concluded in its opinion XI/389/84 concerning the use of pyrithione zinc in hair-care preparations not rinsed off after use: "The Committee notes that the use of pyrithione zinc is allowed as a preservative in products rinsed off after use at a maximum concentration of 0.5% in the finished product.
The Committee finds that the substance is highly toxic, and cannot agree recommending any extension of its use unless percutaneous absorption in man can be shown not to occur in normal skin, nor under conditions of inflammation or abrasion."

Submission I for Zinc pyrithione was submitted in July 2000 by COLIPA 1.

The Scientific Committee on Cosmetic Products and Non-Food Products intended for Consumers (SCCNFP) adopted its opinion (SCCNFP/0671/03) on 17th December 2002 with the conclusion: "The SCCNFP is of the opinion that zinc pyrithione does not pose a health risk when used:

- for non-preservative purposes in cosmetic rinse-off and leave-on hair care products at a maximum concentration of 1.0 % and 0.1 %, respectively; or,
- for preservative purposes in cosmetic rinse-off hair care products at a maximum concentration of 1.0 %.

Zinc pyrithione should not be used in products for oral hygiene."

Zinc pyrithione is currently regulated as a preservative in rinse-off products (excluding oral hygiene products) in a concentration up to 0.5% in general and up to 1.0% in hair products (Annex VI/1, 8). Furthermore zinc pyrithione is also allowed in a concentration up to 0.1% in leave-on hair products (Annex III/1, 101).

In the present submission by COLIPA (submission II), which is a supplemental dossier to submission I, the applicant applies for an extension of the authorised concentration from 1.0% to 2.0% in rinse-off antidandruff hair care products.

2. TERMS OF REFERENCE

1. *Based on the scientific data provided, does the SCCS consider that zinc pyrithione, when used in a concentration up to 2.0% as an anti-dandruff agent in rinse-off hair care products, is safe for the consumer? [The request should be seen as additional to the currently authorized use].*

[1] COLIPA - European Cosmetics Toiletry and Perfumery Association

3. OPINION

3.1 Introduction

In submission II the applicant provided comprehensive toxicokinetic data, data on physiologically-based pharmacokinetic (PBPK) modelling of ZPT as well as data on a human pharmacokinetic study where disposition and excretion of ZPT after different 4-day treatment regimes with shampoo containing up to 2 % ZPT and tonic containing up to 0.25 % ZPT were investigated. Further, representative samples of HRIPT study reports were provided.

Apart from the data provided in submission II, after submission I and finalisation of opinion SCCNFP/0671/03 on 17th December 2002, further data on ZPT has become available, as the substance is also used as a biocide and as the substance has been registered under Regulation (EC) No 1907/2006 (REACH regulation) in a tonnage band of 1000 – 10000 tpa.

Information on Registered Substances is publicly available at the website of the European Chemicals Agency (ECHA) (http://echa.europa.eu/web/guest/information-on-chemicals/registered-substances) and comes from registration dossiers which have been assigned a registration number. The assignment of a registration number does not however guarantee that the information in the dossier is correct or that the dossier is compliant with Regulation (EC) No 1907/2006 (the REACH Regulation). This information has not been reviewed or verified by the Agency or any other authority.

Reproduction of information from registrations published on ECHA's website is authorised for non-commercial purposes of information provided that the ECHA is acknowledged as the source: "Source: European Chemicals Agency, http://echa.europa.eu/."

However, where copyright is vested in a third party, permission for use and reproduction must be obtained from that third party. For most toxicological information on ZPT given on ECHA's website, it is stated: "This information may not be used for any purpose other than in support of the Chemical Safety Report submitted by Arch Chemicals Inc. under Regulation EC 1907/2006."

SCCS contacted ECHA in order to be able to cite third party data on ZPT published on ECHA's website. ECHA advised the SCCS to directly contact the owner of the data. Subsequently, the SCCS requested Cosmetic Europe to get additional Zn-pyrithione data, which was submitted to ECHA but not to the SCCS. However, these data were not provided to the SCCS before publication of the opinion in summer 2013.

In part, studies published on ECHA's website had been described and taken up in two publicly available scientific opinions on ZPT:

1) HSE (The Health and Safety Executive) (2003): Advisory committee on pesticides No 208. Evaluation on: Zinc pyrithione: use as a booster biocide in antifouling products (available at: http://www.pesticides.gov.uk) (reference D8)

2) MAK (2012): Zinkpyrithion. The MAK Collection for Occupational Health and Safety (available at http://onlinelibrary.wiley.com/doi/10.1002/3527600418.mb1346341d0052/pdf) (reference D19).

Therefore, SCCS utilised information given in these two documents to supplement the toxicological information on ZPT.

After commenting period, further full study reports have been made available to the SCCS. Furthermore, a dermal PBPK model has been submitted.

The present evaluation considers the data submitted during/after the commenting period.

3.2. Chemical and Physical Specifications

3.2.1. Chemical identity

3.2.1.1. Primary name and/or INCI name

Zinc pyrithione (INCI name)

3.2.1.2. Chemical names

Bis [1-hydroxy-2(1 H)-pyridinethionato-O,S] (T-4) zinc (IUPAC)
Pyrithione zinc
Zinc bis(2-pyridylthio)-N-oxide
Zinc pyridinethione
Zinc 2-pyridinethione-l-oxide
Bis (N-oxopyridine-2-thionato) zinc (II)
ZP, ZnPT, ZnPTO, BOTZ

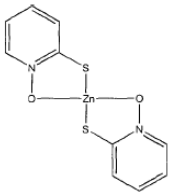
3.2.1.3. Trade names and abbreviations

Zinc Omadine
Vancide ZP

3.2.1.4. CAS / EINECS number

CAS: 13463-41-7
EINECS: 236-671-3

3.2.1.5. Structural formula



3.2.1.6. Empirical formula

Formula: $C_{10}H_8N_2O_2S_2Zn$

3.2.2. Physical form

White to slightly yellow crystals

SCCS comment
Additional information is available on ECHA's website (http://echa.europa.eu/)

3.2.3. Molecular weight

Molecular weight: 317.7 g/mol

3.2.4. Purity, composition and substance codes

Zinc pyrithione is commercially supplied as a 24-26 % aqueous solution

3.2.5. Impurities / accompanying contaminants

No data submitted

3.2.6. Solubility

Very low solubility in most solvents

Water:	0.0015 w/w %
Ethanol:	0.031 w/w %
Acetone:	0.07 w/w %
Chloroform:	0.34 w/w %
Mineral oil, light:	0.0001 w/w %

Water solubility at 20°C, reported at ECHA Website: 4.93 mg/L (EU Method A.6),
6.3 ppm (OECD 105)

SCCS comment
Concerning solubility in ethanol and acetone, further information is available on ECHA's website (http://echa.europa.eu/). HSE gives a water solubility of 8 g/l.

3.2.7. Partition coefficient (Log Pow)

Log P_{ow}: 0.9 (HSE, 2003)
0.97 (MAK, 2012)
0.883 (EU Method A.8, ECHA Website) 0.9 (OECD 107, ECHA Website)

3.2.8. Additional physical and chemical specifications

Melting point:	240 °C (Decomposition at 240°C)
Boiling point:	/

Flash point:	/
Vapour pressure:	< 0.000001 Pa at 25°C (OECD 104)
Density:	1.782 at 25 °C
Viscosity:	1.76 g/cm^3 (OECD 109)
pKa:	/
Refractive index:	/
UV_Vis spectrum (200-800 nm):	/

SCCS comment
Concerning melting point, ECHA's website (http://echa.europa.eu/) gives a slightly different value, whereas in HSE 2003 it is stated that the substance decomposes before melting (200°C).
HSE (2003) gives a vapour pressure of < 0.532 Pa at 21°C, MAK (2012) gives a vapour pressure of 2.49 x 10^{-9} hPa.

3.2.9. Homogeneity and Stability

Homogeneity and stability of Zn pyrithione in test solutions has been reported for some of the studies performed. In the toxicokinetic investigations described in section 3.3.9.3., stability of zinc pyrithione in frozen rat plasma was evaluated by analyzing stability samples stored under the same conditions as study samples. Results indicate a frozen-state stability of approximately 377 days at -70 °C.

SCCS general comments to physico-chemical characterisation

Concerning some physico-chemical properties, diverging values are available from different sources of information.

3.3. Function and uses

Zinc pyrithione (ZPT) is currently regulated as a preservative in rinse-off products (excluding oral hygiene products) in a concentration up to 0.5% in general and up to 1.0% in hair products (Annex VI/1, 8). Furthermore zinc pyrithione is also allowed in a concentration up to 0.1% in leave-on hair products (Annex III/1, 101).

In the present submission, the applicant applies for an extension of the authorised concentration from 1.0% to 2.0% in rinse-off antidandruff hair care products.

According to the EC Commission Regulation (No. 1451/2007), Zink pyrithione is also used as a biocide in biocidal product categories 2, 6, 7, 9, 10, 11, 12 and 22 of Annex V of the EU Biocide Directive (Directive 98/8/EC).

3.4. Toxicological Evaluation

3.4.1. Acute toxicity

3.4.1.1. Acute oral toxicity

Ingredient based data taken from SCCNFP/0671/03
LD_{50} values for zinc pyrithione have been determined in various species after oral administration. The values in the rat ranged from 92 to 266 mg/kg and in the mouse from 160 to 1000 mg/kg. Six hundred mg/kg was found to be the LD_{50} when administered orally to dogs.

Ref.: B6, B10, B33, B57, B71, B73

New ingredient based data
Information on further acute oral studies performed with ZPT is available on ECHA's website (http://echa.europa.eu/).

Product derived data taken from SCCNFP/0671/03
The oral LD_{50} values for shampoo formulations containing zinc pyrithione have been established in rats as 2.5 g/kg for a cream shampoo and 3.0 ml/kg for a lotion shampoo.
In addition, Snyder et al (1965) studied the acute oral toxicity of the cream shampoo product with higher levels of ZPT and estimated the LD_{50}. The results showed that increasing the level of ZPT increases acute oral toxicity.

Ref.: B61

Emetic studies in dogs and pigeons showed that zinc pyrithione in a cream shampoo product is a potent emetic (ED_{50} app. 0.05 g/kg). In the emetic studies with dogs, the emesis typically occurred within 60 minutes of dosing, the average being 30 minutes, and involved two to four episodes. Occasional bloody vomitus was seen, indicating gastric irritation. The ratios of ED_{50} to LD_{50} for both forms of the product are 1:125 for the cream shampoo and 1:42 for the lotion shampoo; therefore it was concluded in SCCNFP/0671/03 that it is unlikely that a human accidentally ingesting shampoo could retain a hazardous amount.

Ref.: B14, B15, B38.

SCCS comment
In addition to acute oral toxicity studies evaluated in SCCNFP/0671/03 further studies have been performed. The data is not available for evaluation. SCCS notes, however, that classification as Acute Tox 3; H301 (toxic if swallowed) according to CLP (Regulation (EC) No 1272/2008 of the European Parliament and of the Council of 16 December 2008 on classification, labelling and packaging of substances and mixtures is suggested by the registrant(s) under REACH (ECHA website (http://echa.europa.eu/).

3.4.1.2. Acute dermal toxicity

Ingredient-based data taken from SCCNFP/0671/03
Dermal LD_{50} values for albino rabbits range from < 2,000 mg/kg to 10,000 mg/kg.

Ref.: B66*

New ingredient based data:
Information on further acute dermal studies performed with ZPT is available on ECHA's website (http://echa.europa.eu/). HSE (2003) describes an acute dermal toxicity study

(limit study) performed in 5 male and 5 female New Zealand White rabbits. The animals received a single dermal application of 2000 mg/kg ZPT moistened with water which was held in place for 24 hrs under semi-occlusive patch. The LD_{50} was > 2000 mg/kg. A single death was noted but it was unclear whether this was treatment-related.

Product derived data taken from SCCNFP/0671/03
A shampoo containing 2% ZPT at levels of 2.5, 5.0, 10.0, and 20.0 g/kg was tested on rabbits. The shampoo was occluded with a rubber sleeve and left in place for 24 hours. There were no observable systemic effects in animals treated with 2.5, 5.0, or 10.0 g/kg. Two of the four animals dosed with 20 g/kg showed a slight temporary depression. There were no deaths at any level. These data are in line with a study on ZPT alone indicating that its incorporation into a shampoo formulation does not significantly enhance penetration.

Ref.: B61, B67

3.4.1.3. Acute inhalation toxicity

Guideline:	OPPTS. 870.1300
Species/strain/sex:	Rat, Sprague-Dawley, both sexes
Group size:	5 animals/sex/dose
Test substance:	Zinc Omadine® 48% Dispersion (FPS) Master Log: 74-01B10CuZPT Sample 0104241211
Dose levels:	0.68, 1.19 and 2.25 mg/l
Exposure:	4 hrs, nose only
GLP statement:	Yes
Date:	2003- 2004

Five male and 5 female animals were nose-only exposed for 4 hours to an aerosol generated from the undiluted liquid test substance (dose levels: 0.68, 1.19 and 2.25 mg/l, respective MMADs: 3.2, 2.5 and 2.5 µm). Animals were observed until day 14 after exposure. Clinical signs included activity decrease, crusted eyes, piloerection, ptosis, respiratory gurgle and sensitivity to touch/sound which were no longer evident in surviving animals by day 10. Emaciation was found in one animal that died on test. Body weights of several surviving animals were affected by exposure. Abnormal necropsy findings occurred in animals that died on test and consisted of red crust around mouth and dark red and swollen lungs. Deaths occurred as follows: 1 of 5 males and 0 of 5 females at 0.68 mg/l; 2 of 5 males and 0 of 5 females at 1.19 mg/l; 2 of 5 males and 1 of 5 females at 2.25 mg/kg. From these data the acute inhalation LC_{50} was calculated to be 5.08 mg/L.

Ref.: E3

Guideline:	OECD TG 403 (1981), EC B.2
Species/strain/sex:	Rat, Sprague Dawley, both sexes
Group size:	5 / sex / dose
Test substance:	Zinc Pyrithion Label: Zinc-Pyrion, Batch TOX 1000
Purity:	not given
Dose levels:	0.53, 0.95, 1.82 mg/l (aerosol dust)
Exposure:	4 hrs, nose only
GLP statement:	Yes
Date:	1996

Male and female Sprague-Dawley rats were nose-only exposed to a dust atmosphere of concentrations of 0.53, 0.95 and 1.82 mg/l (respective MMADs were 3.3 µm at 0.53 mg/l,

3.5 µm at 0.95 mg/l and 3.8 µm at 1.82 mg/l). Animals were observed until day 14 after exposure. All deaths occurred on day 1 after exposure as follows: 1 of 5 males and 0 of 5 females at 0.53 mg/l; 3 of 5 males and 2 of 5 females at 0.95 mg/l; 5 of 5 males and 3 of 5 females at 1.82 mg/kg. All surviving animals showed signs of toxicity (one of the two surviving females exposed to 1.82 mg/l showed stiffness in the hind legs) which improved until the end of the observation period. Except one female exposed to 0.53 mg/l, normal bodyweight development was observed during the second week. Lung abnormalities (swelling, pallor, abnormally dark appearance, dark patches and foci) and liver changes (dark appearance) were observed in animals that died. Except one female exposed to 0.53 mg/l, no abnormalities were observed in animals that were killed at the end of the observation period. The following LC_{50} values were derived: males: 0.84 mg/l, females: 1.34 mg/l, males and females combined: 1.03 mg/l.
The results of this study lead to classification as Acute Tox 3; H331 (toxic if inhaled) according to CLP (Regulation (EC) No 1272/2008) by the registrant.

SCCS comment
This study shows that systemic effects on hind legs might occur after inhalation exposure.

Ref.: E2

Guideline:	US EPA 81-3 (complies with OECD 403)
Species/strain/sex:	Rat, Sprague-Dawley, both sexes
Group size:	5 / sex / dose
Test substance:	Zinc Omadine® Powder; Id.-No. 10215B
Batch:	ORC-152-011 DP
Purity:	95%
Dose levels:	0.24 and 0.61 mg/l (aerosol dust)
Exposure:	4 hrs, nose-only
GLP statement:	Yes
Date:	1991

Male and female Sprague-Dawley rats were nose-only exposed to a dust atmosphere of concentrations of 0.24 and 0.61 mg/l (respective MMADs were 1.9 µm at 0.24 mg/l and 2.3 µm at 0.61 mg/l). Animals were observed until day 14 after exposure. All deaths occurred on day 1 after exposure as follows: 1 of 5 males and 0 of 5 females at 0.24 mg/l; 1 of 5 males and 2 of 5 females at 0.61 mg/l. Surviving animals showed signs of toxicity post exposure (decreased activity, salivation, laboured breathing, tremours, staining around the mouth). No visible abnormalities were observed after post-exposure day 5. Necropsy of animals which died on study revealed congestion or discoloured lungs: animals necropsied at study termination showed no abnormalities.
An LC50 value could not be derived from the study but it is assumed to be < 0.6 mg/l.

Ref.: E25

SCCS comment
In HSE (2003), a further whole body inhalation study performed in male and female Sprague-Dawley rats is mentioned, in which a LC_{50} value of 0.14 mg/l was derived.

SCCS notes that classification as Acute Tox 3; H331 (toxic if inhaled) according to CLP (Regulation (EC) No 1272/2008 of the European Parliament and of the Council of 16 December 2008 on classification, labelling and packaging of substances and mixtures) is suggested by the registrant(s) under REACH (ECHA website (http://echa.europa.eu/).

3.4.1.4. Acute toxicity – other routes

Ingredient-based data taken from SCCNFP/0671/03

Intraperitoneal
Intraperitoneal injection of ZPT resulted in LD_{50} values of 36 mg/kg for rats and 500 mg/kg for mice.

Ref.: B71, B73

Intravenous
Generally, 25 mg/kg of ZPT was fatal to both dogs and monkeys within 24 hours and produced cholinergic-like effects prior to death. Doses of 15 and 20 mg/kg produced slight cholinergic stimulation in dogs but death did not result. One of two Yorkshire pigs died when injected intravenously with 20 mg/kg and 10 mg/kg was a lethal dose for rabbits. Intravenous doses of 5 mg/kg or less produced only transient effects.

Ref.: B2, B7, B72

3.4.2 Irritation and corrosivity

3.4.2.1. Skin irritation

Product derived data taken from SCCNFP/0671/03

A study evaluated the effect of ZPT in a marketed shampoo base on human skin pigmentation at sub-irritating levels. Product was applied daily at 0.2, 0.4, and 2.0% under non-occluded dressings to each of eight Caucasians and eight black males for 64 consecutive days. Under the experimental conditions used, the cream and lotion shampoos did not produce any skin irritation, nor did they change the skin pigmentation level in Caucasian or black skin.

Ref.: B37

A case report described a reaction by a patient to a shampoo containing 2% ZPT. The patient had had a similar reaction after using a hair cream with a lower level seven years before. Another report described a case of eczema of the scalp and face after using a shampoo containing 2% ZPT for a short period.

Ref.: B4, B23

Ingredient based data

From ECHA's website (http://echa.europa.eu/), the SCCS is aware that skin irritation studies have been performed with ZPT. The studies are not available for evaluation. A total of four skin irritation studies have been described in MAK (2012):

6 male New Zealand rabbits were exposed occlusively for 4 hrs to 0.5 g moistened ZPT. Skin reactions were assessed after 0.5, 1, 24, 48 and 72 hrs. After 0.5- 1 hrs, slight erythema of grade 1 (of 4) was observed in three animals and oedema (grade 2 of 4) was observed in two animals. After 24 hrs, the erythema had disappeared and oedema was reduced to a score of 1. After 48 hrs, the oedema also disappeared. From this study, ZPT was considered mildly irritating (below classification level) (Cerven, 1991, taken from MAK (2012)).

The dermal irritancy of a 20 % suspension of ZPT was examined in three animal models (rabbits, guinea pigs and mice). In open patch tests involving five daily applications, ZPT was slightly irritant and induced a marginal epidermal hyperplasia and increased hair growth.
(Lansdown, 1991; cited from MAK (2012)).

In a Buehler-Test performed on 10 male guinea pigs, occlusive 6-hr treatment with 0.4 ml of a 48 % dispersion did not cause skin irritation. (Newcomb, 1996; cited from MAK (2012)).

MAK (2012) also lists the publication Collum and Winek, 1967 in section "skin irritation". However, this study gives no information on skin irritation. It describes the influence of the vehicle on dermal penetration/dermal toxicity of pyrithiones.

In some human repeat insult patch test studies (described in section 3.3.3 – skin sensitisation), slight irritation was observed in some volunteers.

SCCS comment on skin irritation
Skin irritation studies performed with ZPT were not available for evaluation. However, from product based data evaluated in SCCNFP/0671/03, from the description of skin irritation studies performed with ZPT and from human HRIPT tests it can be inferred that ZPT is – at least - a mild skin irritant.

3.4.2.2. Mucous membrane irritation

Product derived data taken from SCCNFP/0671/03

The eye irritation potential of ZPT has been evaluated in a number of product types: Instillation of a soap solution containing 0.25% ZPT to rabbit eyes produced slight transient irritation with the peak effect occurring during the first 4 hours and disappearing completely in 2-4 days.
In another study, undiluted and diluted solutions of shampoo with or without ZPT (2%) were tested. Undiluted solutions produced extensive damage to the eyes of rabbits which were characterised by opalescence of the entire cornea, severe iritis and marked conjunctivitis. In all cases, rinsing was particularly effective in alleviating the condition with very slight to moderate conjunctivitis being observed. In all rinsed cases, damage had cleared by the third day whereas in unrinsed eyes the condition had not cleared by day 42. Dilution of these test solutions to 10% also reduced the ocular irritation and in all cases the condition was cleared by day 7. Again, rinsing was effective in alleviating the condition. No significant differences were observed between the control and the test animals in this study. Repetition of the above study in monkeys with no rinsing produced superficial damage to the corneal epithelium and/or slight conjunctival irritation when the 2% ZPT shampoo was instilled undiluted. Instillation of the shampoo formulation diluted to 10% (0.2% ZPT) resulted in no ocular irritation. Conclusion: The irritation potential of shampoo in rabbit eyes was not increased by the incorporation of ZPT.

Ref.: B61

Ingredient based data

From ECHA's website (http://echa.europa.eu/), the SCCS is aware that eye irritation studies have been performed with ZPT. The studies are not available for evaluation. Two eye irritations performed with ZPT are described in MAK (2012) and HSE (2003):

In an eye irritation study 0.1 ml of powdered zinc pyrithione (95.6 %) was instilled into the conjunctival sac of 6 New Zealand White rabbits, with the other eye serving as a control. Observations were carried out at 1, 24, 48 and 72 h. The mean scores (24, 48 and 72h) were 3 for corneal opacity and conjunctival redness, 4 for conjunctival chemosis and 1.2 for iridial effects (only 4 animals being scored due to excessive discharge making iris scoring difficult in 2 animals). From this study, the substance was considered as a severe eye irritant.

(Olin, 1991b, cited from MAK (2012)).

In a further eye irritation study 0.1 ml of a 48% aqueous dispersion of zinc pyrithione was instilled into the conjunctival sac of 6 New Zealand White rabbits, with the other eye serving as a control. Treated eyes were rinsed 24 h post instillation. Observations were carried out at 24, 48 and 72 h. The mean scores (24, 48 and 72h) were 2.5 for corneal opacity and conjunctival redness, 3 for conjunctival chemosis and 1 for iridial effects.
(HSE (2003), MAK (2012)).

SCCS comment
In SCCNFP/0671/03 only product based information was evaluated. It was concluded that the irritation potential of shampoo in rabbit eyes was not increased by the incorporation of ZPT. Pure ZPT has also been investigated in eye irritation tests, but the studies are not available for evaluation. HSE (2003) concludes that ZPT is a severe eye irritant, MAK (2012) states that ZPT is corrosive to the eye. SCCS notes, that classification as Eye Damage 1; H318 (causes serious eye damage) according to CLP is suggested by the registrant(s) under REACH (ECHA website http://echa.europa.eu).

3.4.3. Skin sensitisation

Animal data

Taken from SCCNFP/0671/03

ZPT was evaluated for its potential to induce contact hypersensitivity to guinea pigs. Using the procedure of Buehler (1965) to detect contact hypersensitivity, 40 animals were exposed to a 50% aqueous slurry of ZPT. No reactions indicative of contact hypersensitivity were seen in any of the animals at challenge.

Ref.: B25

A 0.1% solution of the ZPT (1 % ZPT) soap was injected intracutaneously into depilated guinea pig skin at an initial dose of 0.05 ml and nine subsequent doses of 0.1 ml on alternate weekdays. A single challenge dose of 0.05 ml was injected two weeks later. There was no evidence of sensitisation.

Ref.: B61

New animal data

Guideline:	US EPA 81-6 (complies with OECD 406)
Species/strain/sex:	Guinea pigs, Hartley Albino, male
Group size:	10 dosed animals, 5 controls
Test substance:	Zinc Omadine 48% Suspension
Batch:	I.D.#5RC-088-024ZP
Purity:	reference is given to another document
Dose levels:	0.4 ml of the suspension for induction and challenge
Exposure:	Induction: 6 hrs occlusive/once per week/3 weeks
Positive control:	historical control data
GLP statement:	Yes
Date:	1996

Test substance was applied dermally and held under occlusion for 6 hrs, then test material was removed. The procedure was performed once/week for three weeks. 14 days after the last induction, test and control animals were challenged via the same procedure at naïve sites. Challenged sites were examined and scored at 24, 48 and 72 hrs after challenge

application. Erythema was absent during the induction and challenge phase. Under the conditions of this test, a 48 % suspension of ZPT was not sensitising.

Ref.: E17

Guideline:	OECD 406 (1992), EC B.6
Species/strain/sex:	Guinea pigs / Dunkin Hartley / female
Group size:	2 x 10 animals (test groups), 2 x 5 animals (control group)
Test substance:	Zinc Pyrion
Batch:	A1040010
Purity:	97.9 %
Vehicle:	white petrolatum
Dose levels:	25 % for 1st and 2nd induction, 10 % for challenge
Exposure:	epicutaneous, occlusive
Positive control:	Hexyl Cinnamic Aldehyde (Comparison with previously performed study)
GLP statement:	Yes
Date:	2001

In a preliminary test, 3 females were administered intra-dermally with Freund`s adjuvant. The test substance (50 % w/w) was applied epicutaneously to the sites of the intradermal injections. 7 days later 4 concentrations of the test substance (50 %, 25 %, 10 % and 1 %) were administered epicutaneously to the flanks of the animals. The duration of the exposure was 24 hours. Skin reactions were severe erythema at 50 %, very slight erythema at 25 % and no reaction at 10 % and 1 %.
For induction on day 0, animals received intradermal injections of Freud's complete adjuvans and immediately after that, epicutaneous applications of the test substance to the same site. The second induction (after 7 days) consisted in epicutaneous application of test substance to the site of intradermal injections. Challenge exposure consisted in epicutaneous application of the test substance to the left flank and of vehicle to the right flanks. Control animals received vehicle only.
After the challenge exposure, 2/20 animals of the test substance group had positive skin reactions 24 h after the end of the exposures. No adverse skin reactions were observed in the control animals. Therefore 2/20 animals of the test substance group (10 %) were regarded as sensitised, which is below the threshold of 30 % for classification as a sensitising substance.

SCCS comment
The SCCS concurs with the conclusions drawn by the study authors.

Ref.: E28

In a further Buehler test performed with ZPT of 97% purity (pure test substance, moistened with mineral oil was used) in 10 test and 5 control male Hartley guinea pigs ZPT was not sensitising. HSE states that this study had been adequately conducted (HSE (2003)).

An earlier study performed according to an unusual protocol in guinea pigs using a suspension of 50 % ZPT in mineral oil was considered as not adequate for evaluation because of insufficient documentation and methodological deficits (MAK (2012)).

Human data

Taken from SCCNFP/0671/03

The work by the Danish Contact Dermatitis Group was described in which ZPT (1%) was added to the European Standard Patch Test series. 1652 consecutive dermatitis patients were tested. Only three positive reactions were found. The authors state that in only one of these was the ZPT reaction interpreted with certainty as being of present relevance. They also point out the wide use of ZPT in "shampoos, hair creams and cosmetics". Bearing this in mind, and recalling that all the subjects tested had known skin problems, this is a remarkably low incidence of reactions, and underlines the very low risk from ZPT in the sensitisation area.

Ref.: B4

A multi-centre investigation was conducted in France in order to evaluate the risk of sensitisation by a number of preservatives. 465 subjects were tested. They were suffering from an eczema for which the anecdotal circumstances pointed to an allergy to cosmetics, medicine, industrial products or clothing accessories. Only two patients (0.4%) gave positive patch tests to ZPT.

Ref.: B38*

Two separate closed-patch test studies on human volunteers were conducted. A 1% aqueous solution of shampoo containing 2% ZPT was used. The test solution was placed on the upper arm of the subjects and occluded. Nine serial applications were made on alternate weekdays for three weeks, followed by challenge two weeks later. Challenge patches with the same concentration of test material were placed on both the original site of insult and on an alternate site on the opposite arm to distinguish between skin fatigue and sensitisation. Reactions were scored at both 48 and 96 hours. One subject gave papular reaction at 48 hours, which was scored negative at 96 hours. Unfortunately no follow-up was done with individual ingredients, so it is impossible to determine whether indeed the subject was sensitised, and if so, what the offending material was. The remaining subjects gave only a transient erythematous response indicative of irritation.

Ref.: B61

Cream and lotion shampoo products were tested in two separate HRIPT's. Both studies were conducted according to a modified Draize procedure, in which 0.25% shampoo was patched. No sensitisation was detected in the 82 subjects exposed to the cream nor in the 78 subjects exposed to the lotion. The only responses noted were transient primary irritation in some subjects.

Ref.: B26, B65

A hair dressing cream containing 0.5% ZPT was used to patch test more than 100 women for five months. A minimum of 80% of the subjects were patched weekly for 20 consecutive weeks. Patches were left in place for 48 hours and sites graded 72 hours after removal. Throughout the entire test program, no instances of any skin reactions were observed. Thus, it was concluded that the hairdressing product possessed an extremely low index of sensitisation in humans.

Ref.: B46*

Marketing experience with a commercially available formulation has conclusively demonstrated that ZPT is, at worst, a very weak sensitiser. Few reports of sensitisation have appeared in the literature.
Ref.: B4, B17, B23, B40

Further data

Further information on skin sensitising studies in humans has become available. The information comprises (1) representative samples of HRIPT study reports provided by the applicant, (2) information from MAK (2012) and (3) information on ECHA's website (http://echa.europa.eu/).

Ad 1)

Study 1:
The test substance was a dilution of shampoo with added perfume. The test report has been audited in compliance with the principles of Good Clinical Practice. Informed consent was obtained from the participants. In a pre-test, the irritating potential was investigated by three semi-occlusive 24-hour exposures of the upper outer forearm on days, 1, 3 and 5 to 5% [w/v] and 10 % [w/v] dilutions of the shampoo with added perfume in deionised water. Based on the outcome of the pretest, a dilution of 10% [w/v] shampoo with added perfume (final concentration of perfume: 1.20 %) was chosen for the main study.
The main study involved 9 semi-occlusive 24 hour induction exposures of the upper outer arm over a period of three weeks in 93 subjects from which 87 completed induction and challenge. After the induction period, there was a 14 day rest, followed by the application of the final induction patch prior to duplicate challenge exposures at both original and virgin sites. For most subjects, skin was assessed approximately 48 or 72 hr after induction applications and 48 and 96 hr after challenge application.
Under the conditions of this test, there was no evidence of skin sensitisation, but slight irritation was seen in some of the volunteers.

Ref.: A13

Study 2:
The test protocol was approved by the North West Ethical Committee and the study was performed in accordance with the principles of Good Clinical Practice. A total of 101 volunteers participated in the study, 93 of them fully completed the study. The substance was applied using 2 x 2 cm Webril pad Semi-occlusive Micropure tape. The test material was initially applied at 10 % [w/v] (aq.) to a Webril square for the first four induction patches. This concentration provoked an unacceptably high level of irritation during this period resulting in a reduction to 5% [w/v] for the remainder of the test. A total of 9 induction patches were applied and scored. 48 and 98 hrs after challenge, readings were undertaken. Irritant response of both dermal and epidermal nature with pronounced edge effects were observed which were in some cases more prominent at the 96-hr after-challenge reading when compared to the 48-hr after-challenge reading. Despite the delayed nature of some of these responses, the investigators concluded that there was no evidence of skin sensitisation on the 93 subjects who completed the test.

Ref.: A14

Study 3:
The study adhered to QAU principles. Information on informed consent/ involvement of an ethical committee is not given. The study panel consisted of 84 volunteers who received occlusive patches with the substance during a three week induction period each Monday, Wednesday and Friday. Each patch was left in place for 24 hrs and then removed. Fourteen days after the last induction application, duplicate challenge patches were applied for 24 hrs. 81 volunteers completed the study (the dropping out of volunteers was unrelated to the test). Mild erythematous reaction was observed at many occasions during the induction phase, but the test material showed no evidence of skin sensitisation.

Ref.: A15

Study 4:

The study was performed according to the principles of good laboratory practice. Informed consent was obtained from the 92 volunteers participating in the study (86 volunteers completed the study). The test material was used in a concentration of 0.15 % [w/v] and applied in 0.4 ml amounts to a $2cm^2$ Webril pad and fixed on the lateral surface of the upper arm. In the induction phase, patches were applied on Monday, Wednesday and Friday for three weeks. Test sites were scored before application of each subsequent patch and on the fourth Monday after the final insult patch. After a 2-week rest, challenge patches were applied to both arms of each subject and results were graded after 48 and 96 hrs. During the induction phase, mild erythematous reaction was observed in many occasions. At challenge, none of the volunteers showed reactions greater than that of a mild erythema. It was concluded that the substance did not produce reactions indicative of skin sensitisation.

Ref.: A16

Study 5:
The study adhered to QAU principles. A total of 96 volunteers took part in the study, 92 of them fully completed the study, the reasons for withdrawal of volunteers were unrelated to the test/test material. A 0.15 % [w/v] dilution in distilled water was used. There were 9 induction applications. Results for sensitising effects were graded 48 and 96 hrs after challenge application. The 0.15 % aqueous dilution of test material caused an acceptable level of irritation during the study. There was no evidence of skin sensitising potential observed in the 92 subjects who fully completed the study.

Ref.: A17

Study 6:
The study adhered to QAU principles. The test substance was used in a concentration of 10 % [w/v] in distilled water. The study consisted of nine semi-occluded induction patches over a three week period followed by a 14 – 20 day rest period. The induction patch sites were evaluated at 48 hrs after patch application (72 hrs for weekends). Panellists were challenged on the original and naïve sites and evaluated at 48 and 72 or 96 hrs after patch application. 102 panellists from a total of 117 (informed consent was obtained) completed the study. None of the 102 panellists completing the study exhibited responses during the challenge phase of the study. Thus, it was concluded that there was no clinical evidence indicative of delayed contact hypersensitisation to the substance.

Ref.: A18

SCCS comment on studies 1 – 6 described above
The SCCS does not consider HRIPT studies for determining sensitisation potential to be ethical.

Ad 2)

In a modified Draize test in 10 human volunteers, there was no evidence of skin sensitisation. For induction, either a 3% ZPT suspension in petrolatum or a 1 % solution of ZPT in DMSO was used. Challenge consisted of either a 3% ZPT suspension in petrolatum or of a 0.5 % solution of ZPT in DMSO.

Ref.: D20

In addition, MAK 2012 lists reports on reactions in epicutaneous tests on zinc pyrithione or sodium pyrithione in patients with suspected contact allergy which are presented in table 1 and 2; grading was apparently performed according to the guidelines of the European Society of Contact Dermatitis. With respect to the utilisation of data from sodium pyrithione it should be noted that it is claimed that rather the organic moiety than the Zn ion is

responsible for sensitising effects of ZPT. Therefore, studies using sodium pyrithione are included in the case reports listed in table 1 and 2. Further, worker data from the use of ZPT as a cooling lubricant are also included in the overview.

Table 1: reports on reactions in epicutaneous tests on zinc pyrithione or sodium pyrithione in patients with suspected contact allergy (according to MAK 2012); grading was apparently performed according to the guidelines of the European Society of Contact Dermatitis.

Tested individuals	Test substance, concentration, vehicle	Result	Remarks	Reference
Metal worker with hand eczema	Sodium pyrithione, 0.5% (petrolatum)	1 + and 2 + (after 48 and 96 h)	less than 1% sodium pyrithione in the cooling lubricant concentrate	Le Coz, 2001 (D18)
Patient with facial and scalp eczema*)	zinc pyrithione, 0.05%, 0.2%, 0.5%, 1% (petrolatum)	1 +, 2 +, 3 +, 3 + (at 48 and 72 h)	Shampoo containing 0.5% and 2% zinc pyrithione; no reaction in 20 control persons exposed to 0.1% and 0.5% zinc pyrithione	Goh and Lim, 1984 (B23)
2 patients with scalp eczema	zinc pyrithione, 1% (petrolatum)	2 + / 3 + and - / 2 + (after 48 and 96 h)	zinc pyrithione-containing shampoos, in both patients sensitisation to p-phenylenediamine, and numerous other substances	Gonzalez Perez et al., 1995 (C3)
Patients with dermatitis of the scalp, face, neck and hands	zinc pyrithione, 1% (petrolatum)	2 + (after 48, 72 and 168 h)	Shampoo containing 0.45% zinc pyrithione; patients also have a positive reaction to the shampoo (tested at 2% and 5%)	Hsieh et al., 2010 (D9)
Lathe operator with dermatitis on the back	zinc pyrithione, 1% (petrolatum) and sodium pyrithione, 0.1% (water)	negative; 1+ and 3+ when re-tested after 48 h	coolant with 0.1-1% sodium pyrithione; questionable reaction towards the used coolant and 2 + response to the coolant concentrate (5% in buffer)	Isaksson, 2002 (D11)
Female patient with pustular psoriasis	zinc pyrithione, 1% and 2% (petrolatum)	1 + / 2 + (after 48 and 96 h, respectively)	within 20 days after application of a zinc pyrithione-containing shampoo, pustular	Jo et al., 2005 (D12)

			psoriasis with Koebner phenomenon occurred in the patient with stable psoriasis	
2 patients (tested only in one case) with eczema on the scalp, face and upper body / neck, arms and hands*)	zinc pyrithione, 1% (water), (tested only in one case)	1 + (after 48 and 96 h)	zinc pyrithione-containing anti-dandruff preparations; in the second case no testing of zinc pyrithione, but positive patch test with zinc pyrithione-containing product and no response to zinc pyrithione-free formulation	Muston et al., 1979 (B40)
Female patient with dermatitis of the scalp	Zinc pyrithione (no further information on solvent)	2 + (after 48 h)	zinc pyrithione-containing shampoo, at rechallenge: Koebner phenomenon with the exacerbation of psoriasis	Nielsen and Menné, 1997 (C4)
Patient with dermatitis of the scalp, face and neck	zinc pyrithione, 0.2% and 0.5% (petrolatum)	1 + and 3 + (72 h)	shampoo containing 2 % zinc pyrithione	Nigam et al., 1988 (C5)
Female patient with dermatitis on forehead, neck and hands	zinc pyrithione, 1% (petrolatum)	2 + (after 48 and 96 h)	eczema after treatment of pityriasis capitis, with a shampoo containing 1% zinc pyrithione, no reaction in 14 control subjects at 1% Zinc pyrithione	Pereira et al., 1995 (C1)
Patient with dermatitis of the scalp, face and neck	zinc pyrithione, 5% (petrolatum)	2 + / 3 + (after 48 and 96 h)	no response to 5% zinc pyrithione in 10 control subjects	Yates and Finn, 1980 (D23)

Table 2: Results of patch testing in larger populations

Tested individuals	Test substance, concentration, vehicle	Result	Remarks	Reference
1652 consecutive patients*)	zinc pyrithione, 1% (petrolatum)	1+, 2+ and 3+	zinc pyrithione-containing shampoo; 2+ and 3+ result questionable	Brandrup and Menné, 1985 (C2; B4)

183 metal workers	sodium pyrithione, 0.1% (water)	2 of 183 positive (1 +, after 72 h)	Individuals overlap with Geier et al., 2006	Geier et al., 2004 (D5)
135 metal workers	sodium pyrithione, 0.1% (water)	1 of 135 positive (after 72 h)	Individuals overlap with Geier et al., 2006	Geier et al., 2006 (D6)
181 metal workers	sodium pyrithione, 0.1% (water)	0 of 181 positive		Gruvberger et al. 2003 (D7)
465 patients*)	zinc pyrithione 1% (petrolatum)	2 of 465 positive	no information on the clinical relevance	Meynadier et al., 1982 (B38*)

*) data already considered in section "Taken from SCCNFP/0671/03"

Ad 3) A human patch test is described at ECHA's website (http://echa.europa.eu/). The study is not available for evaluation.

Conclusion on sensitising potential
ZPT is not sensitising in animal studies. Concerning human data, ZPT (or better: the PT moiety) has a low potential to induce contact hypersensitivity when tested per se or as part of a cosmetic formulation. However, in some human HRIPT studies, evaluation was partly hindered by the erythematous reactions observed.

3.4.4. Dermal / percutaneous absorption

3.4.4.1. *In vitro* dermal absorption

Guideline:	OECD 428
Species:	split-thickness skin from rats and humans
Rat skin:	Dorsal skin from Crl:CD(SD) rats
Human skin:	2 abdomen; 1 abdomen/breast; 1 abdomen/arms/upper back/ 1 abdomen/upper arms) from patients aged 23 to 66 years
Test substance a):	[^{14}C] ZPT (Perkin Elmer, Batch 3620165) (radiolabel at position 2+6 of pyridine rings) Radiochemical purity 99.5%, specific activity 3.09 mCi/mmol
Test substance b):	Zinc Pyrithione (Charles River, Batch 0504322141) Purity: 99.4 %
Vehicle:	Carboxymethylcellullose, Darvan, water
Dose levels:	48% and 1 % in rat skin, 1% in human skin (48% was tested previously for human skin)
Receptor fluid:	tissue culture medium containing bovine serum albumin (BSA, *ca* 5%, w/v), glucose (*ca* 1%, w/v), streptomycin (*ca* 0.1 mg/mL) and penicillin G (*ca* 100 units/mL)
Exposure:	8 hrs
GLP statement:	Yes
Date:	2010

Test preparations were applied, at an application volume of 10 µL/cm^2, to human and rat split-thickness skin membranes mounted onto flow-through diffusion cells. Receptor fluid was pumped underneath the skin at a flow rate of *ca* 1.5 mL/h. The skin surface temperature was maintained at *ca* 32°C throughout the experiment. A tritiated water barrier integrity test was performed and any human or rat skin sample exhibiting absorption

greater than 0.6% of the applied dose was excluded from subsequent absorption measurements.
Percutaneous absorption was assessed by collecting receptor fluid in hourly fractions from 0 to 8 h post application and then in 2-hourly fractions from 8 to 24 h post application. At 8 h post application, exposure was terminated by washing the skin surface. At 24 h post application skin was removed, dried and the stratum corneum was removed with 20 successive tape strips. The remaining skin was divided into exposed and unexposed skin. The exposed human skin samples were heat separated to give epidermis and dermis. All the skin samples were solubilised with Solvable® tissue solubiliser. All samples were analysed by liquid scintillation counting.
A summary of the mean results, following topical application of Test Preparation 1 to rat skin and Test Preparation 2 to human and rat skin, is provided in the tables.

Test Preparation	1	
Target Zinc Pyrithione Concentration	48% (w/w)	
Zinc Pyrithione Concentration in Test Preparation by Radioactivity	47.45% (w/w)	
Application Rate of Test Preparation	10 µL/cm^2	
Application Rate of Test Item	5550 µg equiv./cm^2	
Species	Rat	
Distribution	% Applied Dose	µg equiv./cm^2
Dislodgeable Dose 8 h	91.47	5077.47
Total Dislodgeable Dose	91.72	5091.03
Unabsorbed Dose	93.08	5166.78
Absorbed Dose	0.13	7.01
Dermal Delivery	2.26	125.65
Potentially Absorbable Dose	3.14	174.59
Mass Balance	95.34	5292.43

Test Preparation	2			
Target Zinc Pyrithione Concentration	1% (w/w)			
Zinc Pyrithione Concentration in Test Preparation by Radioactivity	1.07% (w/w)			
Application Rate of Test Preparation	10 µL/cm^2			
Application Rate of Test Item	101 µg equiv./cm^2			
Species	Human		Rat	
Distribution	% Applied Dose	µg equiv./cm^2	% Applied Dose	µg equiv./cm^2
Dislodgeable Dose (8 h)	94.52	95.29	87.26	87.98
Total Dislodgeable Dose	94.77	95.55	87.63	88.36
Unabsorbed Dose	95.98	96.76	90.35	91.10
Absorbed Dose	0.02	0.02	1.12	1.13
Dermal Delivery	0.76	0.77	7.75	7.81
Potentially Absorbable Dose	1.31	1.32	9.28	9.36
Mass Balance	96.74	97.53	98.10	98.91

Dislodgeable dose (8 h) = skin wash + tissue swab (8 h) + pipette tips
Total unabsorbed dose = dislodgeable dose (8 h) tissue swab (24 h) + stratum corneum + unexposed skin + cell wash
Absorbed dose = cumulative receptor fluid + receptor rinse
Dermal delivery = exposed skin (dermis and epidermis for human skin) + absorbed dose
Potentially absorbable dose = dermal delivery + stratum corneum (tape strips 6 to 20)
Mass balance = unabsorbed dose + dermal delivery

For Zinc Pyrithione in Test Preparation 1 (48%, w/w) applied to rat split-thickness skin (10 skin samples from 5 different animals were used), the majority of the applied dose (91.47 ± 2.74 %) was removed by washing. At 24 h post application, the total dislodgeable dose was 91.72 ± 2.82% of the applied dose. The stratum corneum retained 1.35 ± 0.74% of the applied dose; 0.46 ± 0.37% was removed with the first 5 tape strips. The total unabsorbed dose was 93.08 ± 2.8 % of the applied dose. The absorbed dose, dermal delivery and potentially absorbable dose were 0.13 ± 0.13 % (7.01 ± 7.45 µg equiv./cm^2), 2.26 ± 0.83 % (125.65 ± 45.87 µg equiv./cm^2) and 3.14 ± 1.14 % (174.59 ± 63.18 µg equiv./cm^2) of the applied dose, respectively. The mass balance was complete with 95.34 ± 2.46 % of the applied dose recovered.
For Zinc Pyrithione in Test Preparation 2 (1 %, w/w) applied to human split-thickness skin (9 skin samples from 5 different donors were used for calculation of means), the majority of the applied dose (94.52 ± 2.08 %) was removed by washing at 8 h post application. At 24 h post application, the total dislodgeable dose was 94.77 ± 2.19 % of the applied dose. The stratum corneum retained 1.18 ± 0.99 % of the applied dose; 0.63 ± 0.6% was removed with the first 5 tape strips. The total unabsorbed dose was 95.98 ± 2.59 % of the applied dose. The absorbed dose, dermal delivery and potentially absorbable dose were 0.02 ± 0.01 % (0.02 ± 0.01 µg equiv./cm^2), 0.76 ± 1.14 % (0.77 ± 1.15 µg equiv./cm^2) and 1.31 ± 1.32 % (1.32 ± 1.33 µg equiv./cm^2) of the applied dose, respectively. The mass balance was complete with 96.74 ± 2.58 % of the applied dose recovered.
For Zinc Pyrithione in Test Preparation 2 (1 %, w/w) applied to rat split-thickness skin (8 samples from 5 different animals were used for calculation of means), the majority of the applied dose (87.26 ± 3.94 %) was removed by washing at 8 h post application. At 24 h post application, the total dislodgeable dose was 87.63 ± 3.99 % of the applied dose. The stratum corneum retained 2.52 ± 1.62 % of the applied dose; 0.99 ± 0.6 % was removed with the first 5 tape strips. The total unabsorbed dose was 90.35 ± 3.46 % of the applied dose. The absorbed dose, dermal delivery and potentially absorbable dose were 1.12 ± 1.06% (1.13 ± 1.07 µg equiv./cm^2), 7.75 ± 5.26% (7.81 ± 5.31 µg equiv./cm^2) and 9.28 ± 5.6% (9.36 ± 5.65 µg equiv./cm^2) of the applied dose, respectively.

Ref.: E5

SCCS comment
Test concentrations of 48 % and 1 % ZPT were used in this study whereas the applicant applies for the use of ZPT at a concentration of 2 %.
As variability is high, the mean ± 2 SD could be used for MoS calculation of solutions using 1 % ZPT.

The assignment of human samples to donors and skin region is unclear.

ZPT is known to be localised in the hair follicles following application (see SCCNFP/0671/03). The above model does not take account of this penetration.

3.4.4.2. *In vivo* dermal absorption

Taken from SCCNFP/0671/03

Based on the submitted *in vivo* animal data it was concluded that percutaneous absorption of ZPT varies from approximately 0.03 to 3.4% (see study descriptions in SCCNFP/0671/03).

Further data

Animal studies

By using Albino rabbits of both sexes which received daily occlusive doses of 400 mg/kg ZPT as a 10 % suspension in a 2 % solution of methylcellulose in either water or DMSO for 14 days, it could be demonstrated that DMSO enhances percutaneous absorption of ZPT with the production of toxic signs and, in most cases, death.

Ref.: D2

Guideline:	OECD 427
Species/strain/sex:	Rat, Sprague-Dawley, male
Group size:	4 animals/group / 3 groups per test preparation
Test substance a):	[^{14}C] ZPT (Perkin Elmer, Batch 3620165) (radiolabel at position 2+6 of pyridine rings) Radiochemical purity 99.5%, specific activity 3.09 mCi/mmol
Test substance b):	Zinc Pyrithione (Charles River, Batch 0504322141) Purity: 99.4 %
Vehicle:	Carboxymethylcellullose, Darvan, water
Dose levels:	48% and 1 %
Exposure:	dermal, protected, 8 hrs
GLP statement:	Yes
Date:	2010

For each test preparation three groups of four male Sprague Dawley rats were tested. The three groups were killed at 24, 48 and 168 hrs post-dose. Following dose administration (dosed area: ca. 10 cm^2, dose volume: 100 µl), the application site was covered with a protective dressing. After an 8 hr exposure period, dressing was removed, sites were washed and a new dressing was applied. Urine and faeces were quantitatively collected for the periods predose, 0-8 and 8-24 hrs, then daily up to termination. At termination, exposed skin was tape-stripped to remove stratum corneum, the exposed area was dissected and the remaining carcass and gastrointestinal tract were retained. Samples were analysed for radioactivity by scintillation counting. The mass balance was complete for [^{14}C]-Zinc Pyrithione for both test preparations and across all time points with a value of *ca* 95% for both. Maximal absorbable doses were composed of absorbed dose (amounts in urine, faeces, cage wash, GI tract and carcass), exposed skin and material associated with stratum corneum of tape strips 6-20. For the 48 % solution, mean and maximal potentially absorbable dose were 0.19 % and 0.27 %. For the 1 % solution, mean and maximal potentially absorbable dose were 0.85 % and 1.13 %.

SCCS comment
The test was performed with 48 % and 1 % ZPT solutions, whereas the applicant applies for approval of a concentration 2% ZPT in cosmetic products.

Ref.: E6

Two further *in vivo* dermal absorption studies are mentioned on ECHA's website (http://echa.europa.eu/). The studies are not available for evaluation.

Further, in submission II, the applicant provided data on the *in vivo* dermal toxicokinetics of ZPT in female CD rats. In order to develop a physiologically-based pharmacokinetic model for ZPT, pharmacokinetics of ZPT was investigated in two phases in the rat. A detailed study description is given in section 3.3.9 Toxicokinetics and PBPK modelling.
Phase 1 of the study was exploratory in nature and therefore not conducted in strict accordance with GLP regulations (and it did not adhere to OECD TGs 427 or 417). The conclusion of this part of the study was: "Dermal exposure to ZPT at levels of 10 mg/kg or

greater resulted in plasma pyrithione concentrations that were measurable (>0.5 ng/mL). A lag time to reach these concentrations of about 8−12 hr from exposure initiation was observed. Decreases in both hind-limb muscle mass and muscle tone were observed in animals exposed daily for 10 days to 100 mg/kg ZPT. The bioavailability of dermally applied ZPT was estimated to be about 3−8%. Since concentrations of pyrithione in plasma were high enough to measure in only a few samples and only at the later time points, these estimates should be considered only to be crude estimates of bioavailability."

Ref.: A19

Phase 2 of the dermal exposure studies included three dose levels (10, 30, and 100 mg/kg) and a vehicle control. Bioavailabilities of the dermal doses were calculated from pyrithione concentrations in plasma (0−24 h) using data from the pilot study for the intravenous dose. These calculated bioavailabilities were **2.3, 8.6, and 0.3%**, respectively, for the **10, 30, and 100** mg/kg/day exposures.

SCCS comment
In principle, the study was performed in line with OECD TGs 427 and 417, although TG adherence was not explicitly mentioned. The study was performed according to GLP. The applicant states that the calculated bioavailabilities should be used with caution: the AUC(0−24h) values for dermal administration do not capture the total AUC, and use of the AUC(0-∞) values are fraught with the difficulties for parameters based on k_{el} as a consequence of high variabilities in various TK parameters. In a manuscript on a dermal PBPK model for ZPT, different absorption percentages were reported for apparently the same study (see also sections 3.4.9.3. and 3.4.9.4).

Ref.: A19; E8

Human studies

Taken from SCCNFP/0671/03

A clinical pharmacokinetic study has investigated deposition, absorption and excretion of ^{14}C radiolabelled ZPT resulting from the use of a ZPT containing shampoo alone (1 % ZPT) and in combination with a ZPT containing leave on hair tonic (0.1% ZPT). This study demonstrated that systemic loading of ZPT was increased significantly less than could be expected from the corresponding skin deposition in those subjects using the shampoo/tonic combination compared with those using the shampoo alone. Additionally, absorption of ZPT in patients with compromised scalps was not found to be statistically different to normal scalps patients.
Deposition, absorption and excretion parameters were measured in 20 volunteers (10 patients using ZPT containing shampoo alone (Group A) and 10 using the ZPT containing shampoo and ZPT containing tonic combination (Group B)) over a 4 day treatment period. Each treatment group was comprised of 5 patients with healthy scalps and 5 patients with compromised scalps with either severe dandruff or seborrheic dermatitis. All patients used 10 g of shampoo per day and those in Group B also used 4 g of tonic per day during the 4 day treatment period.
Measurements of ZPT deposition and excretion were made by analysis of clipped hair, tape stripping areas of the scalp and hands and urinalysis respectively. Previously, preclinical studies have demonstrated that ≥ 90% of absorbed ZPT is excreted in the urine within 24 hours, thus for the purposes of this study the level of ZPT excreted was taken to represent the level of ZPT absorbed (i.e. systemic dose).
Mean ^{14}C-ZPT Systemic Load results are shown in table 3.

Table 3: Mean ^{14}C-ZPT Skin Deposition Measurements (Scalp and Hands)

Day	1 % ^{14}C-ZPT Shampoo		1 % ^{14}C-ZPT Shampoo + 0.1 % ^{14}C-ZPT Tonic	
	LSM systemic load (µg/kg/d)[#]	SEM	LSM systemic load (µg/kg/d)[#]	SEM
1	1.02	0.14	1.39	0.14
2	2.54	0.33	3.31	0.33
3	2.73	0.32	3.32	0.32
4	2.76	0.35	3.43	0.35
5	1.96	0.32	2.29	0.32

LSM = Least Squares Mean
SEM = Standard Error of the Mean
[#] Average body weight per group used for calculation of µg/kg/d values

Analysis of the dermal deposition data indicated:
- ZPT deposition on the hands between the treatment groups A and B was not significantly different throughout the study except on day 2
- ZPT deposition on the scalp between the treatment groups A and B was statistically different with deposition in Group B being significantly higher than group A
- ZPT deposition on the hands was determined to be approximately half the level deposited on the scalp
- ZPT deposition on the hair between the treatment groups A and B was statistically different with deposition on hair in Group A being half the level of Group B.

Analysis of individual subject absorption data indicated that individuals with compromised scalps demonstrated no greater absorption than individuals with normal scalps. Analysis of the urinary excretion curves indicates that steady state conditions were reached within the 4 day treatment period of this study. Statistical analysis indicated that the amount of ZPT excreted in the urine (indicative of systemic exposure) was significantly higher in the shampoo + tonic group (B) compared with the shampoo only group (A) throughout the study.

However the increase was less than what would have been expected from the increase in skin deposition. This suggests that a rate limiting mechanism exists for the absorption of ZPT across the skin.

SCCS comment

From the data it can be seen that systemic exposure to ZPT increases with increasing amount of ZPT applied. Systemic exposure loads up to 3.43 µg/kg/d were obtained from the study.

In a follow-up study to the above mentioned study (which is therein declared as study # CRB9907-083), it is stated that deposition in the study described above was higher than the deposition in the later study. It was discussed that this could - apart from interstudy variation - be due to the fact that deposition is lower when tonic is applied to dry hair. In the first study, tonic was applied to wet hair only, whereas in the subsequent study, only the first application of tonic was on wet hair.

The SCCS notes, that this study has been utilised to establish a dermal PBPK model for ZPT in humans (see section 3.4.9.4). From the PBPK model, a dermal absorption of 0.5 % was calculated.

Study provided in submission II

As the new submission intends to increase of the authorised concentration of ZPT from 1.0% to 2.0% in rinse-off antidandruff hair care products, a new clinical study has been performed to determine the deposition, systemic absorption and excretion of ^{14}C-radiolabeled ZPT resulting from the repeated application of treatment regimens including 2% ZPT containing antidandruff shampoo in a 7-day randomised, parallel group comparison study in which a 4-day treatment period was followed by a 3-day wash-out period. The study adhered to GLP and Quality assurance principle: informed consent was obtained from the participants.
A total of thirty (30) male and female subjects between the ages of 18 to 65 were enrolled. To qualify for enrollment, the subjects had to have mild to severe dandruff or seborrheic dermatitis of the scalp with a total adherent scalp flaking score (ASFS) of $\geq$12.

During the 4-day treatment, subjects washed their own hair with 10 g of the shampoo containing ^{14}C-ZPT. The shampoo was applied only one time per day and was rinsed from the hair. All of the water used for rinsing the shampoo lather from the subjects' hair and hands was collected in a single container for analysis of ^{14}C-radioactivity. The treatment regimens are given in table 6.

Table 4: Treatment regimens of the clinical dermal absorption study provided in submission II

Treatment Regimen	Test Products
A	Shampoo with 1% ^{14}C-ZPT + Leave-on Tonic with 0.1% ^{14}C-ZPT (2 applications)
B	Shampoo with 2% ^{14}C-ZPT + Leave-on Tonic with 0.1% ^{14}C-ZPT (2 applications) + Leave-on Tonic with 0.25% ^{14}C-ZPT (1 application)
C	Shampoo with 2% ^{14}C-ZPT + Leave-on Tonic with 0.25% ^{14}C-ZPT (3 applications)

Immediately following the rinsing of the shampoo, subjects applied the 1st – 4g dose of the ^{14}C-ZPT containing leave-on tonic to their wet hair. Following application, the subjects' hair was blow-dried. Subjects assigned to Treatment Regimen A applied 2 doses of the leave-on tonic. The 1st dose was applied immediately after the shampoo application and the 2nd dose was applied 8 hrs later. Subjects assigned to treatment regimens B and C applied 3 doses of the tonic. The 1st dose was also applied immediately after shampoo application. The 2nd and 3rd doses were applied at 4 to 6 hour intervals after the 1st dose. These doses were applied to dry hair. To measure the deposition of the ^{14}C-ZPT on the scalp and hands, tape-stripping of the scalp and/or fingertips was done on days 1, 2, 3 and 4 after the daily treatment products were applied; on day 5 (24 hrs after the 4th application of the treatment regimen) and, on day 7 of the wash-out period (after the 3rd hair wash with a regular shampoo). In addition to tape stripping, hair specimens were clipped from the subjects' scalps at the same time that the tape-stripping was done to measure the deposition of ^{14}C-ZPT on the hair. To measure the systemic absorption and excretion of ^{14}C-

ZPT, 24 hr urine specimens were collected during the entire study period. Subjects were dismissed on day 8 upon completion of the 24 hr urine collection on day 7.

Results: The measured scalp deposition did not increase with repeated application. The scalp deposition rate was generally highest in treatment regimen C (max. 1.92 µg/cm^2), followed by B (max. 1.39 µg/cm^2), then A (max. 0.51 µg/cm^2). The total mass of ZPT deposited on the hands was typically about 25% - 50% of that on the scalp. Scalp + Hands deposition as a percentage of the applied quantity was less than 1% in all treatment groups (max. 0.52% for A, 0.63% for B and 0.88% for C).

The quantity of ZPT deposited on the hair was typically about 5-10 times higher than the amount deposited on the scalp, demonstrating that most of the ZPT deposited is not on the skin. Hair deposition ranged from approximately 2%-4% in treatment regimens A and B and about 4%-6% in treatment regimen C.

Mean values from excretion measurements calculated on day 4 were used to approximate daily ZPT absorption and excretion during longer term consumer use. The 24 hr excretion measurements were normalized to body weight for each subject to provide estimates of daily internal exposure (systemic load). Systemic loads were significantly higher for Treatment Regimens C (4.66 µg/kg/day) and B (4.38 µg/kg/day) compared to A (2.82 µg/kg/day).

There was no significant difference by gender in scalp deposition, absorption, mass excreted or systemic load of ZPT during the 4-day treatment period.

Ref.: A20, A21

SCCS comments

(1) Calculated recoveries were lower than expected (61 % to 86 %). The rinse water measurements produced values lower than expected, which resulted in generally low calculated recovery of the applied dose. This was attributed to lack of sufficient stirring of the rinse water during sampling. A regression analysis was conducted to evaluate the correlations between the percent of the applied quantity recovered and the quantity in the rinse water, total quantity on the skin and total quantity in the urine. The mean recovery was highly correlated with the rinse water measurements, but not to skin deposition or urinary excretion. Thus, the low recovery does not raise questions about the validity of the deposition data.

(2) The applicant states that "urinary ZPT excretion reached an apparent steady state for all three treatment groups during the 4-day treatment period. This was demonstrated by the lack of significant difference between the mass excreted on day 4 vs. day 3 in all treatment groups. Therefore, the mean values calculated on day 4 were used to approximate daily ZPT absorption and excretion during longer term consumer use."
SCCS compared the systemic loads obtained from the study provided in submission I (using a shampoo containing 1% ZPT and a tonic containing 0.1 % ZPT) with systemic loads from the study provided in submission II. A graphic overview is given in figure 1. From this figure it can be seen that indeed, as the applicant states, apparent steady states could be inferred from the data. However, the respective levels of the individual "apparent" steady states increase with increasing amounts of ZPT applied. Therefore, it cannot be excluded that higher systemic exposure loads than the maximal levels derived from the studies submitted might be obtained in consumers, if ZPT-containing products were applied frequently and in considerable amounts. This is further supported by the results of a dermal toxicokinetic study in female rats, which suggest that maximal absorption of ZPT and therefore pyrithione exposure was achieved at 30 mg/kg (see section 3.3.9).

(3) The applicant uses a 4-day treatment regimen to extrapolate to long-term consumer use. SCCS considers this as inappropriate.

(4) In order to cope with the uncertainties addressed under points (2) and (3) the SCCS will take the value of systemic exposure load from treatment C (4.66 µg/kg/day) + 1 standard deviation (SD) (0.59 µg/kg/d) for risk characterisation which is 5.25 µg/kg/d.

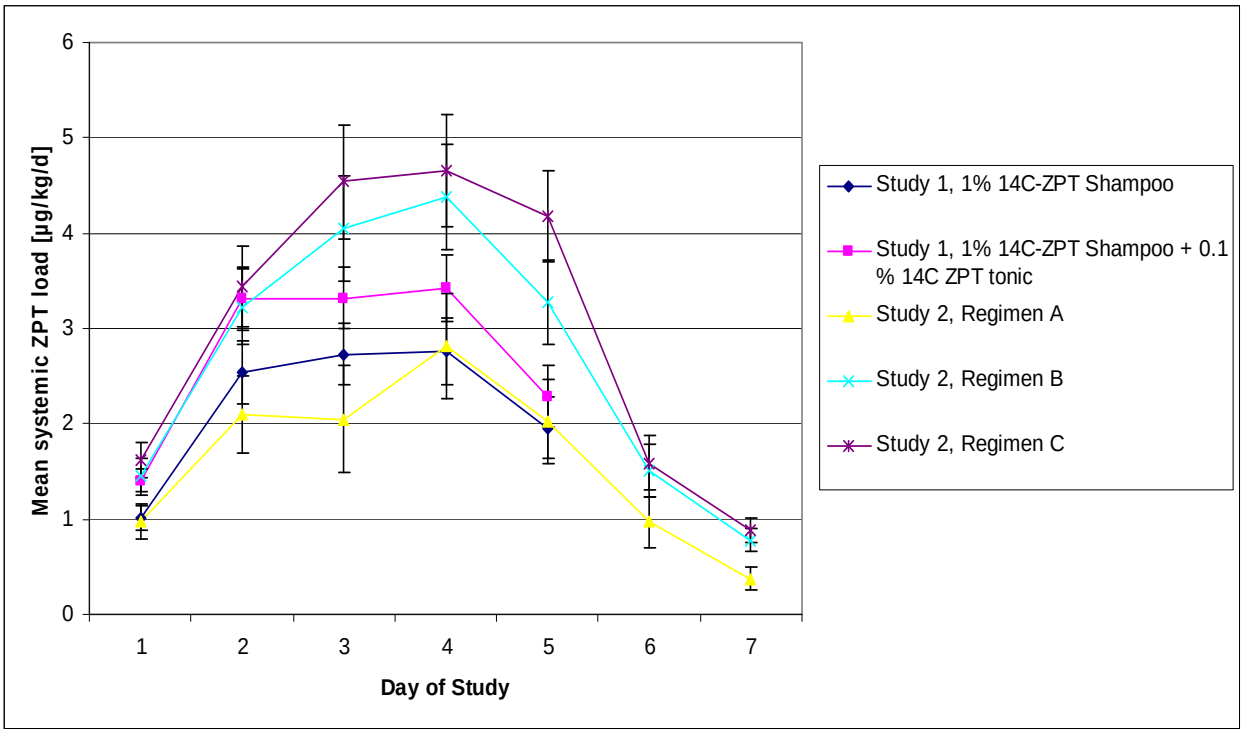


Figure 1: comparison of systemic ZPT loads obtained from two clinical studies performed with shampoo and lotion containing ZPT as provided in submission I and II. Regimen A, B and C of study 2 were as follows: Regimen A: Shampoo with 1% ^{14}C-ZPT + Leave-on Tonic with 0.1% ^{14}C-ZPT (2 applications); Regimen B: Shampoo with 2% ^{14}C-ZPT + Leave-on Tonic with 0.1% ^{14}C-ZPT (2 applications) + Leave-on Tonic with 0.25% ^{14}C-ZPT (1 application); Regimen C: Shampoo with 2% ^{14}C-ZPT + Leave-on Tonic with 0.25% ^{14}C-ZPT (3 applications).

From the daily urinary excretion data given in the study report, SCCS calculated cumulative amounts excreted for Regimen A, B and C. Cumulative excretion amounted to 450, 885.5 and 931.06 mg for regimen A, B, and C, respectively, during the observation period. The cumulative amount excreted during the observation period pointed to 0.22%, 0.16 % and 0.196 % of the applied amounts for regimen A, B, and C, respectively.

Table 5: cumulative amounts of pyrithion-derived radioactivity from a human clinical study using 2% ZPT formulations and a rat dermal toxicokinetic study.

Day	Cumulative Excretion [µg] Human Study 2, Regimen A	Cumulative Excretion [µg] Human Study 2, Regimen B	Cumulative Excretion [µg] Human Study 2, Regimen C
1	78.83	111.68	149.29
2	251.25	364.11	455.06
3	492.83	674.14	850.84
4	721.29	1010.21	1259.11
5	893.63	1256.32	1624.03
6	976.18	1368.91	1758.81
7	1007.82	1429.13	1833.41

Totalapplied quantity [mg]	450	885.5	931.06
% excreted during observation period	0.22%	0.16%	0.196%

Summary dermal absorption:
From the animal studies available in submission I, it was concluded in SCCNFP 0671/03 that percutaneous absorption of ZPT varies from approximately 0.03 to 3.4%. Further studies on dermal absorption of ZPT have been performed thereafter.

In an in vitro dermal absorption study performed according to OECD TG 428 using rat and human split-thickness skin and concentrations of 48 % and 1 % ZPT, the following absorption values were obtained:

Sample	Dermal delivery (dermis, epidermis, receptor fluid, receptor rinse)		Potentially absorbable dose (dermal delivery and stratum corneum tape strips 6-20)	
	[%]	[µg/equiv./cm^2]	[%]	[µg/equiv./cm^2]
Rat, 48 % solution	2.26 ± 0.83	125.65 ± 45.87	3.14 ± 1.14	174.59 ± 63.18
Rat, 1 % solution	7.75 ± 5.26	7.81 ± 5.31	9.28 ± 5.60	9.36 ± 5.65
Human, 1 % solution	0.76 ± 1.14	0.77 ± 1.15	1.31 ± 1.32	1.32 ± 1.33

From an in vivo toxicokinetic study performed in rats with 1 % and 48 % ZPT, mean and maximal potentially absorbable dose were 0.19 % and 0.27 % for the 48 % solution. For the 1 % solution, mean and maximal potentially absorbable dose were 0.85 % and 1.13 %.
From dermal toxicokinetic studies (not fully OECD compliant) performed in female CD rats in order to build up a PBPK model for ZPT, absorption percentages of 2.3, 8.6 and 0.3 were derived after repeated dermal administration of 10, 30 and 100 mg/kg ZPT. However, these values are associated with uncertainties (see section 3.4.9.3). The SCCS is aware that different absorption percentages were reported from apparently the same study in a submitted manuscript on a dermal PBPK model for ZPT.
Dermal absorption was also assessed in two clinical studies in humans. One study investigated the systemic absorption of a shampoo containing 1 % ZPT (with or without combination with a leave-on tonic containing 0.1 % ZPT) in a 4-day treatment regimen. In

this study, a systemic load of ZPT up to 3.43 µg/kg/d was derived. When using a dermal PBPK model, an absorption percentage of 0.5 % was calculated for a 1% ZPT solution. However, no further background information is given for that value. In the second clinical study, the systemic absorption of a shampoo containing 2 % ZPT (either in combination with leave-on tonics containing 0.1 and 0.25 % ZPT or with a leave-on tonic containing 0.25% ZPT only) was investigated in a 4-day treatment regimen. Systemic exposure loads up to 4.66 µg/kg/d were derived. As in the new submission, the applicant applies for an extension of the authorised concentration from 1.0% to 2.0% in rinse-off anti-dandruff hair care products, a systemic exposure load of 4.66 µg/kg/d can be taken for risk characterisation. In order to cope for prolonged application (more than 4 days) and consequently probably higher systemic exposure loads, 1 SD will be added to the systemic exposure load of 4.66 µg/kg/d yielding 5.25 µg/kg/d.
From the daily urinary excretion data given in the study report, SCCS calculated cumulative amounts excreted for Regimen A, B and C. Cumulative excretion amounted to 450, 885.5 and 931.06 mg for regimen A, B, and C, respectively, during the observation period. The cumulative amount excreted during the observation period pointed to 0.22%, 0.16 % and 0.196 % of the applied amounts for regimen A, B, and C, respectively.

3.4.5. Repeated dose toxicity

General Remark on Repeated Dose toxicity
The repeated dose toxicity of ZPT has been investigated in several studies using the oral (gavage and diet), dermal and inhalation route. Further studies are available for sodium pyrithione from which a read across to ZPT may be performed.

3.4.5.1. Repeated Dose oral / dermal / inhalation toxicity (of different duration)

Oral studies

Summary taken from SCCNFP/0671/03

Concerning ZPT:
No significant effects have been noted other than the hind-limb weakness or paralysis which occurred in rats and rabbits within 8 to 14 days when ZPT was administered in the diet at levels from 165 ppm to 330 ppm (8-16 mg/kg/day). Doses greater than 330 ppm required longer periods of administration before paralysis occurred. At levels of 1000 ppm or greater, the animals usually died without developing paralysis. With respect to paralysis, a NOEL of 0.5 mg/kg/day (500 µg/kg/d) has been determined in two year feeding studies in rats. At this dose no toxic effects were observed over the two-year study period.

Concerning shampoo formulations containing ZPT:
Orally administered ZPT in a shampoo formulation produces a reversible paralysis in rats and rabbits within one to two weeks at levels of 10 mg/kg/day. A dose level of 10 mg/kg/day of ZPT in shampoo was used in a monkey gavage study which lasted 16 weeks. No adverse effects were observed. Because of the emetic potential of shampoos, oral dosing of dogs has not produced any toxic symptoms, including ocular effects.

New / additional data
Further repeat dose studies performed in rats or monkeys are given on ECHA's website (http://echa.europa.eu/). Two studies are available for evaluation.

Guideline:	no
Species/strain/sex:	Monkey / Rhesus / both sexes
Group size:	3 per sex at 2.0 and 8.0 mg/kg/d; 4 males, 2 females at 0.5 mg/kg/d

Controls:	3 male and 3 female animals (no further information)
Test substance:	Zinc Omadine Win 9546
Batch:	97-P-158, RN No. 7-72-3
Purity:	not given
Vehicle:	1 % gum tragacanth
Dose levels:	0.5, 2.0, 8.0 mg/kg/d
Exposure:	oral, gavage, 3 months
GLP statement:	no
Date:	1973

Zinc pyrithione, suspended in 1% gum tragacanth, was administered by gavage to Rhesus monkeys at 0, 0.5, 2 or 8 mg/kg/d for 90 d. Clinical chemistry, haematology and urinalysis were performed before substance administration, after 1 month and at study termination. At necropsy, a comprehensive range of tissues was taken for macro- and histopathological examination, including sciatic nerve samples. However, no skeletal muscle samples were examined. No clinical signs of toxicity were reported. No toxicologically significant findings were noted following haematological, clinical chemistry and urinalysis. Electrocardiograms of the high dose group were essentially normal.
No abnormal findings were noted following gross and histopathological examination.
Relative kidney weights in the intermediate and high dose were reduced compared to control, which was attributed to high kidney weights in the control animals. Relative uterus weight was reduced in a dose-related manner at 2.0 and 8.0 mg/kg/d and was associated with apparent immaturity of the uteri.
From this study, a LOAEL of 2.0 mg/kg/d and a NOAEL of 0.5 mg/kg/d can be derived.

Ref.: E9

Guideline:	MITI Guideline (Japan), similar to EC B.7
Species/strain/sex:	Monkey / Cynomolgus / both sexes
Group size:	4 per sex /dose at 5.5 and 11.0 mg/kg/d, 6 per sex /dose at 22.0 mg/kg/d
Controls:	6 male, 6 female
Test substance:	Zinc Omadine ® powder
Batch:	9204084481
Purity:	96.3 %
Vehicle:	
Dose levels:	0, 5.5, 11.0, 22.0 mg/kg/d
Exposure:	oral, gelatine capsules, once daily for 28 days
GLP statement:	yes
Date:	1992

Stability statement: it was confirmed by the sponsor that the test article would be stable for 1 year at room temperature. Dose levels were selected based on the results of a preliminary study. 2 additional animals in the control and high dose group were used for a 2-week recovery period. Animals received ZPT in gelatine capsules once daily for a period of 28 days. Animals were sacrificed after that period (except recovery animals). Blood and urine samples were taken at week 1, at study termination and after the recovery period. At the same time points, animals were weighed and ophthalmic investigations were performed.
One female at the highest dose group died on day 10, among the remaining animals of this dose group, the following clinical signs were observed: decreased activity, decreased appetite, sporadic vomiting and diarrhea. These signs resolved during the recovery period. No abnormal clinical signs were observed in the lower dose groups. Decreased body weights were noted in four males and two females of the highest dose.
Urinalysis revealed an increase in ketone bodies in four males and four females of the highest dose group: pH was reduced in two males.

In the haematologic examination, a decrease in erythrocyte count, haematocrit, haemoglobin concentration and MCHC as well as an increase in MCV were observed in the highest dose group in week 4; the effects resolved during the recovery period.
In the two highest dose groups, a decrease in lymphocytes and a decrease in stab.form neutrophilic leucocytes were observed.
On gross pathologic examination (apart from the female animal that died on day 10) no test-article related abnormalities were found.
The relative organ weights of the adrenal glands and the liver of the high-dosed females were increased. 3 high dose males also had increased relative liver weights. No significant organ weight changes were observed at the lower doses. On histopathologic examination, no abnormalities were observed in the surviving animals.
Based on decreased lymphocyte counts in male animals observed at 11 mg/kg bw/d an NOAEL of 5.5 mg/kg/d is derived from this study.

Ref.: E10

In addition to repeat dose studies performed with ZPT, HSE (2003) derives a NOEL of 0.5 mg/kg/d based on hind limb muscle atrophy from an oral 90-day study in Sprague-Dawley rats performed with **sodium pyrithione,** which is described in HSE (2003), therein cited as Unpublished, 1988. This study is not available for evaluation by the SCCS. A read across from sodium pyrithione to ZPT is considered appropriate based on the following reasoning: data on metabolism of ZPT (see section "toxicokinetics" and see also SCCNFP/0671/03) demonstrate that Zn is cleaved from the molecule after uptake and that ADME of the metal ion and the pyrithione moiety is different. Studies performed in pigs using NaPT and ZPT pointed to a common metabolic pathway (references B.68, B69, B.70). Further, both Zn^{2+} and Na^{+} ions are not considered to be neurotoxic. Thus, it can be assumed that neurotoxic effects observed after ZPT exposure are due to the pyrithione moiety. It can thus be concluded that results from other pyrithione-liberating salts might support the findings obtained with ZPT.

Dermal studies

Taken from SCCNFP/0671/03

Larson (1957) conducted a 90-day percutaneous toxicity study with ZPT (2 ml of water per gram of 50% wettable ZPT powder) using albino rabbits. Doses of 125, 250, 500, 1000 and 2000 mg/kg were applied daily (5 days per week) to groups of three or four animals for 13 weeks. The animals were harnessed during application and remained so until the material dried, at which time the animals were washed. None of the rabbits receiving 1000 or 2000 mg/kg survived the 90-day test period, the longest survival being 21 days. Four of twelve animals dosed at the lower levels survived, and those were necropsied at that time. Focal necrosis of, either the brain or spinal cord in three of the four surviving animals [sic]. There were no histological changes in other organs.
Nelson et al (1965) also conducted studies on the toxicity of ZPT applied topically to rabbits. The material was left in contact with the skin during the periods between application and no effort was made to preclude ingestion of the ZPT. Dosage was daily, and ranged from 50 to 480 mg/kg. After 7 to 15 daily treatments, the rabbits developed hind-limb weakness and diarrhoea. With continued treatment, weakness of both forelegs and death occurred. Treatment was discontinued in three instances coinciding with the onset of severe quadraparesis in two animals and mild quadraparesis in the third rabbit. All three animals recovered. Histopathological observation after autopsy of severely paralysed rabbits revealed no significant structural alterations in the brains, spinal cords, peripheral nerves, muscles, and abdominal or thoracic viscera in 8 of 12 rabbits. In the other four rabbits

various alterations in CNS tissue were seen that the author associated with a protozoan infection.
In work conducted at Food and Drug Research Laboratories, cited by Snyder et al. (1965), two groups of six rabbits received daily topical applications, five days a week, of 5 ml of a 20% aqueous paste of a commercial soap, with or without 1 % ZPT (on a soap basis). This amounts to 10 mg/kg/day. The animals were kept in stocks for six hours after treatment, at which time the skin was washed and dried, and the animals were returned to their cages. There was a total of 65 applications, equivalent to 50 mg/kg/week. No difference was noted in skin effects or in gross or microscopic pathology between the test group and the control group treated with soap alone. No effects on the eye were seen in either group. Histopathologic study of the brains of both groups revealed changes frequently seen in laboratory rabbits that are thought to be associated with the protozoan organism Encephalitozoon cuniculi. This condition is spontaneous, is seen with great frequency, and is mild chronic in nature. There were no compound-related lesions.
In addition to the rabbit studies, two sub-chronic mouse percutaneous toxicity studies were conducted and are summarised below. A six-week study by Dobbs and Nixon (1973) was conducted to determine dose levels of ZPT for an eighteen-month dermal carcinogenicity bioassay study that would not cause systemic toxicity from oral ingestion during grooming. Ten female mice were used per test material and dose level; five were group-housed, and five were housed individually. Application of 0.1 ml of undiluted test material was made five times per week to a 2 x 2 cm clipped area of the interscapular skin for six weeks. Test materials were ZPT at 0.08, 0.4 and 2.0% in a 1 % surfactant (triethanolamine lauryl sulfate)/ 0.5% thickener (Methocel) aqueous slurry, and a vehicle control. These levels correspond to approximately 0.28, 1.4, and 7.0 mg/kg/day. Four animals, two group-housed and two single-housed, treated with the mixture containing 2% ZPT were necropsied after six weeks of treatment, and no gross abnormalities were observed. None of the treatments produced local or systemic effects after a total of 30 applications (six weeks). The study was terminated at this point.
Another mouse study to determine maximum tolerable cutaneous doses of ZPT in a 1% surfactant/0.5% thickener vehicle was conducted by Gargus (1974). Groups of 30 mice (15 male and 15 female), individually housed, were treated topically three time weekly for four weeks with 0.1 ml doses of 0.4, 2.0 and 10.0% (10, 50 and 250 mg/kg/application) ZPT. The high dose group had the 10% concentration applied for one week, and since no toxicity was observed, a 20% concentration (500 mg/kg/application) was substituted for an additional three weeks. After four weeks no skin irritations or other toxicity was observed for animals treated with 0.4 and 2.0% concentration of ZPT. Animals treated with 20% ZPT showed thickening of the skin and erythema. No hind-limb paralysis as observed in rats or rabbits was seen in any of the groups.

From the Snyder et al (1965) study, one can conclude that a dose level of 10 mg/kg/day of ZPT applied topically to rabbits is a no-effect level. Interpretation of the rabbit studies by Larson (1957) and Nelson et al (1965) summarised above, is complicated by several factors. Depending on what Larson (1957) used to wash the animals, some material probably remained on the skin and was subsequently ingested. The Nelson et al (1965) study, and the uncertainty regarding ingestion in these studies make any conclusions tentative, at best. That ingestion of ZPT occurred in these studies can be supported by examining dose levels used in teratology studies conducted by Nolen et al (1975, 1979). He reported that application of 25 to 100 mg/kg/day of ZPT to the backs of rabbits produced no adverse effects when ingestion was meticulously prevented. Therefore, 100 mg/kg/day instead of 10 mg/kg/day is a better estimate of a no-effect level for topical administration to rabbits. The no-effect level in mice for percutaneous toxicity is approximately 100 mg/kg/day, and the effect level (local irritation) is approximately 200 mg/kg/day.

New / Further Data

Guideline:	US EPA 82-3 (similar to OECD TG 411)
Species/strain/sex:	Rat / Sprague-Dawley / both sexes
Group size:	15 per sex /dose
Controls:	15 males, 15 females
Test substance:	Zinc Omadine ® FPS (contains Zinc 2-Pyridinethiol-n-oxide (CAS 13463-41-7; sodium naphthalenesulfonic acid (CAS 9084-06-4-9) and water (CAS 7732-18-5)
Batch:	33-22902781
Purity:	52.2 % active ingredient
Vehicle:	deionized water
Dose levels:	0, 20, 100, 1000 mg/kg/d
Exposure:	dermal, 5 days per week over 13 weeks, semi-occlusive
GLP statement:	yes
Date:	1992

Dose levels were selected based on results from a preliminary range-finding study. Animals were treated by dermal application to intact skin over 13 weeks, 5 times per week. Animals were observed twice daily for mortality and signs of toxicity. Body weight and food consumption were checked weekly. Dermal observations for irritation were conducted daily before the applications. Haematology, serum biochemistry, organ weights (brain, kidneys, liver, testes), macroscopical and microscopical evaluations were performed at the end of the study; ophthalmoscopy was performed before treatment and during the last week of the study.
Test material suspensions were applied at a constant dosing volume of 2 ml/kg to clipped skin areas of approx. 3 x 5 cm. The test article remained in place for 6 hrs by a semi-occlusive tape and was then removed by washing.
All rats survived to scheduled sacrifice, there were signs of irritation such as red foci and desquamation in one male and one female animal at the highest dose. Mean body weights of the 1000 mg/kg males were lower compared to controls during the first four weeks of the study, thereafter they were higher than in controls. In females, mean body weights at 1000 mg/kg/d were lower (17.4% at study termination) than control with statistically significant difference from week 2; correspondingly, food consumption was lower in high dose females.
No test-item related ophthalmoscopic abnormalities were observed. In animals receiving 1000 mg/kg/d, statistically significantly increased leucocyte counts were found in males, statistically significantly depressed erythrocyte counts and haematocrit levels were found in females. Further, statistically significantly increased cholesterol levels were determind in high dose females. No toxicologically significant macroscopical or histopathological findings were observed.
Based on a decreased body weight in top dose females and on haematological changes at the top dose, a NOAEL of 100 mg/kg bw/d is derived from this study.

SCCS comments
An average of 91 % of the initial concentrations of the active ingredient was found after a 10 day storage at room temperature.
Some animals chewed or removed the tape. Data was collected in order to explain probably occurring unexplained toxicity. Oral exposure cannot fully be excluded from this study.

Ref.: E26

ECHA mentions a further dermal toxicity study performed in mice. This study is not available for evaluation.

MAK (2012) describes two dermal repeat dose studies which address mechanistic aspects.

After dermal application of 100 mg ZPT for 10 days to Sprague-Dawley rats, all of 5 treated animals showed reduced amplitude of the evoked compound muscle action potential (CMAP), 4 animals showed signs of a reduced muscle tone.

SCCS comment
Apparently the same study was described shortly in an Addendum to the proposal for Annex III listing of zinc pyrithione provided by Procter and Gamble in January 2002. Therein the following was stated: after 10 day repeated dermal administration of 100 mg/kg/d ZPT to female rats (strain not given) the muscle evoked potential in the hindlimb (measured as M-wave amplitude) was significantly reduced when compared to untreated controls (22.95 ± 11.61 mV in treated animals versus 46.56 ± 5.95 mV in controls).

In a 28-day dermal neurotoxicity study groups of 5 Sprague-Dawley rats received daily dermal doses of 0, 50, 150 and 200 mg ZPT/kg/d (male animals) or 0, 10, 25, 50, 75 and 100 mg ZPT/kg/d (female animals). The vehicle was 0.1 % triethanolamine-lauryl sulfate, the treatment site was protected by a fixed convex piece of plastic shielding. Low muscle tone was observed at 150 and 200 mg/kg/d in male animals beginning on day 8 and day 11 that continued throughout the study duration. Hindlimb and forelimb grip strength as well as muscle tone and body weight were decreased in male animals of the two highest doses on days 14 and 28. No significant changes in plasma, RBC or brain cholinesterase was observed at any dose tested for any time point measured. Decreases in the electrophysiological measurements measured as the maximum amplitude were observed in males at 150 mg/kg/d. In female animals low muscle tone was observed at 50, 75 and 100 mg/kg/d beginning on day 8 in the 100 mg/kg/d group, on day 15 in the 75 mg/kg/d group and on days 22-28 in the 50 mg/kg/d group. On day 14 grip strength was reduced in the 75 and 100 mg/kg/d group and on day 28 grip strength was reduced in the three highest dose groups. No consistent decreases or dose dependent changes were apparent in plasma, RBC or brain cholinesterase at any dose tested. Decreases in the electrophysiological values measured as the maximum amplitude were observed in the 50 and 75 mg/kg/d group (electrophysiological measurements not taken at the 100 mg/kg/d dose level).

Ref.: D1 Arch Chemicals 2003 (Link given in MAK (2012))

SCCS comment
This study demonstrated that hindlimb effects occur after short-term repeated dermal exposure of rats. NOAELs of 50 and 25 mg/kg bw/d were obtained for male and female animals, respectively.

A further subchronic toxicity study is mentioned on ECHA's website (http://echa.europa.eu/). The study is not available for evaluation. A NOAEL of 100 mg/kg/d was derived from that study.

SCCS comment to studies with dermal administration of ZPT
The fact that hindlimb weakness after dermal administration was not observed in some of the studies might be due to the dosing regimen and the vehicle used. In oral studies it could be demonstrated that in contrast to continuous administration, effects were less pronounced or not observable, when there were discontinuities in the dosing (e.g. 5 days per week). Further, when using water or DMSO as vehicle, toxic effects were more pronounced with DMSO as vehicle.

Inhalation studies

Guideline:	OPPTS 870.3465
Species/strain/sex:	Rat / Sprague-Dawley / both sexes
Group size:	20 per sex /dose

Controls:	20 males, 20 females exposed to air only
Test substance:	Zinc Omadine ® > 95% a.i. white powder
Batch:	Zinc Omadine ® Master Log # 74-01B10Cu ZPT Sample 0108244691
Purity:	98.3 % a.i.
Dose levels:	target concentrations: 2, 5 and 12 mg/m^3 Actual dose levels achieved: 2, 6 and 13.5 mg/m^3
Exposure:	inhalation, nose only, 6hrs/d, 5 days/week for 21 days
GLP statement:	yes
Date:	2004/2005

The dose levels were based on a preliminary range-finding study. Animals were nose-only exposed to aerosols generated from the fine powder of the substance. Animals were observed daily for signs of toxicity and mortality. 10 animals from each sex/dose were sacrificed on day 5 (interim sacrifice groups). Body weights were recorded on day 0, 7 and 14, an ophthalmologic examination was performed on day 21. A functional observational battery (FOB) was conducted on days 9 and 16. Blood for haematology was taken from each animal at the end of the study. Animals from the interim group were euthanised on day 5, all other animals on day 21 and subjected to gross necropsy.
Results:
Animals were not acclimatized to test tubes prior to dosing. Therefore some effects might be attributed to the stress produced in the animals. There was one death in the mid dose and 4 deaths in the high dose group. Clinical signs observed were wet fur around the muzzle, gasping, respiratory gurgles, swelling around the eyes, hypothermia and intermittent tip-toe walking. There was no difference between treated and control animals in FOB. Grip strength was not affected by dosing. Concerning clinical chemistry data in plasma and haematology data, there appeared to be no dose-related differences between groups.
Except for lung, there were no differences in organ weights between control and treated animals. Lung weights at the 5-day and 21-day sacrifice were increased for all exposure groups except for the 12.0 mg/m^3 treated males at day 5 and the 2.0 mg/m^3 treated males at day 21.
Histopathological examination revealed the following compound-related effects: hyperplasia of alveolar marcrophages; inflammation of the nasal mucosa and interstitium around the bronchioles and vessels of the lungs; inflammation of the larynx which appeared ulcerative in a few animals; mucous cell hypertrophy of the nasal and bronchial mucosa; squamous metaplasia of the nasal mucosa, larynx and trachea; and smooth muscle hypertrophy of the alveolar ducts. The severity of these effects increased modestly with increasing dose and is consistent with irritation as a result of exposure to the dust of ZPT, a known irritant to mucosal membranes.
Of non-respiratory tissues, effects in thymus and lymph nodes (lymphoid depletion and the lymph nodes and thymus in some animals) were observed.
Based on histopathological data in tissues of the respiratory tract and non-respiratory tissues, a LOAEC of 2 mg/m^3 is derived from this study.

Ref.: E4

Guideline:	OPPTS 870.3465 with some deviations
Species/strain/sex:	Rat / Sprague-Dawley (Crl:CD) / both sexes
Group size:	15 per sex /dose*)
Controls:	15 males, 15 females exposed to filtered air only*)
Test substance:	Zinc Omadine ®
Batch:	0108244691
Purity:	98.3 %
Dose levels:	target concentrations: 0.5, 1.5 and 5 mg/m^3 Mean actual concentrations: 0.52, 1.5 and 5.1 mg/m^3 (males) Mean actual concentrations: 0.5, 1.5 and 5.1 mg/m^3 (males)

Exposure: inhalation, nose only, 6hrs/d, 5 days/week for 4 weeks
GLP statement: yes
Date: 2009

*) On the day following the 5th, 10th and 20th exposures, 5 animals/sex/dose were euthanised and subjected to necropsy and bronchoalveolar lavage.
Exposure concentrations were based on known toxicological information and on a 5-day range-finder study. A special emphasis was given to the evaluation of pulmonary effects, including assessment of bronchoalveolar lavage fluid (BALF) parameters and microscopic examination of the lung following 1, 2, and 4 weeks of exposure.
One female of the highest dose group was found dead on study day 15. Test substance-related clinical observations were noted for the 5.0 mg/m^3 group females.
Clinical observations included thin body condition in 2 additional females and impaired use of the hindlimbs in 1 female on study day 24. One female was also noted as hypothermic (body and extremities cool to the touch) on study day 26.
Test substance-related effects on body weights were noted in the 1.5 and 5.0 mg/m^3 group males and females. Body weight changes in the 5.0 mg/m^3 group were considered to be adverse.
During study days 0 to 4, lower mean food consumption, compared to the control group, was noted in the 5.0 mg/m^3 group males and females and persisted in the 5.0 mg/m^3 females throughout the study. Exposure to the test substance at levels of 0.5 mg/m^3 or higher resulted in an increase in the proportion of eosinophils in the bronchoalveolar lavage fluid (BALF) of rats on study days 5, 12, and 26. Minimal to mild increases in the proportions of neutrophils and lymphocytes were also considered to be test substance-related at levels of 0.5 mg/m^3 and higher. BALF findings also included higher incidence and severity of cell lysis at 0.5 mg/m^3 or higher, and occasional erythrophagocytosis and mucous at 1.5 and 5.0 mg/m^3, and rarely at 0.5 mg/m^3 in females. Higher BALF lactate dehydrogenase and total protein levels were observed for males and females at all exposure levels on study days 5 and 12. Higher total protein levels were also observed for 1.5 mg/m^3 group females on study day 12 and higher LDH and total protein levels were also observed for the 5.0 mg/m^3 group males and females on study day 26.
ZPT exposure was associated with test substance-related and adverse necropsy and microscopic findings including lower final body weight in the 5.0 mg/m^3 group males, higher lung weights in both sexes of all groups, lower thymus weights in the 5.0 mg/m^3 group animals, and broncho-interstitial pneumonitis and smooth muscle hypertrophy in the lungs of both sexes at all exposure levels following 1, 2, and 4 weeks of exposure. The pneumonitis was characterised primarily by perivasculitis with an eosinophilic component, and less pronounced increase in mucus production within bronchioles, subacute inflammation of lung parenchyma, and increased numbers of alveolar macrophages. There was no apparent difference in the incidence or severity of broncho-interstitial pneumonitis in the lungs between the necropsies on study days 5, 12, or 26. However, effects on both lung weights and BALF percent eosinophils in the 1.5 and 5.0 mg/m^3 groups were less pronounced following 4 weeks of exposure than following 1 or 2 weeks of exposure. For broncho-interstitial pneumonitis in the lung, as well as lung weights and BALF percent eosinophils, effects appeared to be more pronounced in females than in males. Smooth muscle hypertrophy of the alveolar ducts in the lungs, which was observed in all test substance-exposed groups as early as the study day 5 necropsy, increased in incidence and severity with increasing exposure concentration, and with increasing time on the study.
Test substance-related enlargement of the mediastinal lymph nodes was observed in 1 male and 1 female in the 1.5 mg/m^3 group and in 1 female in the 5.0 mg/m^3 group (study day 12) and enlargement of the bronchial lymph nodes was observed in 3 females in the 5.0 mg/m^3 groups (study days 5 or 12). For these gross lesions, mild to moderate lymphoid hyperplasia was observed microscopically.
Based on local (portal of entry) effects that were observed at exposure levels of 0.5 mg/m^3 or higher, no NOAEC could be derived. The LOAEC for local effects is 0.5 mg/m^3. Based on systemic endpoints including survival, clinical observations, body weights, food

consumption, organ weights, and microscopic examination of selected tissues (brain, liver, kidneys, stomach, and skeletal muscle), authors considered the NOAEC for systemic toxicity as 1.5 mg/m^3.

Ref.: E14

ECHA's website (http://echa.europa.eu/) mentions a 90-d whole-body study performed in Sprague-Dawley. From this study, a NOAEL of 0.5 g/m³ air was derived. The study is not available for evaluation.

A summary on repeat dose toxicity is given in section 3.4.5.2.

3.4.5.2. Chronic (> 12 months) toxicity

Taken from SCCNFP/0671/03

A two year feeding study was conducted by Larson (1958). Young Wistar rats in groups of ten males and ten females were fed diets containing ZPT at levels of 0, 2, 5, 10, 25 and 50 ppm. These levels correspond to approximately 0, 0.1, 0.25, 0.5, 1.25 and 2.5 mg/kg/day for adult animals. At the start of the study the corresponding levels were 0, 0.2, 0.5, 1.0, 2.5 and 5.0 mg/kg/day for the young rats. Survival in males was not adversely affected by ingestion of the compound, but the highest level caused hind-limb paralysis in some animals. None of the females on the 50 ppm diet lived beyond 80 weeks, and death was commonly preceded by paralysis. Mortality was also increased at 25 ppm, and paralysis occurred in some animals prior to death. In females, growth depression was marked at 50 ppm. Dietary concentrations of 2, 5 and 10 ppm appeared to have an accelerating effect on weight gain in both sexes, and males showed a comparable stimulation at 25 ppm. The no-effect level for males and females was 10 ppm (0.5 mg/kg/day). The only unusual finding upon termination of the study was an increase in neutrophil versus lymphocyte counts in males on the 50 ppm diet. Ratios of organ weights to body weights did not differ significantly among the surviving groups at termination.
Histopathologic examinations did not reveal any lesions that appeared to be attributable to the administration of ZPT. These observations included careful attention to retina, optic nerve, cerebral cortex, and other parts of the central and peripheral nervous systems. There were no significant differences in the rate of frequency of neoplasms between any of the groups.
From this study, an oral NOAEL of 0.5 mg/kg/d (500 µg/kg/d) was derived.

Further data
Two oral chronic studies performed with ZPT are mentioned at ECHA's website (http://echa.europa.eu/), one is apparently the study by Larson (1958) already evaluated for SCCNFP 0671/03. According to the Batch number given, the second study must have been performed with NaPT although not specifically stated on ECHA's website.

Data on metabolism of ZPT (see section "toxicokinetics" and see also SCCNFP/0671/03) demonstrate that Zn is cleaved from the molecule after uptake and that ADME of the metal ion and the pyrithione moiety is different. Studies performed in pigs using NaPT and ZPT pointed to a common metabolic pathway (references B.68, B69. B.70). Further, both Zn^{2+} and Na^{+} ions are not considered to be neurotoxic. Thus, it can be assumed that neurotoxic effects observed after ZPT exposures are due to the pyrithione moiety. It can thus be concluded that results from other pyrithione-liberating salts might support the findings obtained with ZPT. In this respect chronic studies performed with sodium pyrithione can be used as supporting studies.

Oral

Guideline:	US-EPA 83-2 (comparable to OECD TG 453)
Species/strain/sex:	Rat / Crl:CD-1 (ICR) BR (VAF Plus)
Group size:	70 per sex /dose
Controls:	70 males, 70 females exposed to distilled water only
Test substance:	Sodium Omadine ™ 41.2 % aqueous solution
Batch:	8508-P-166H
Purity:	
Dose levels:	0.5, 1.5, 5.0 (3.5)*) mg/kg/d
Exposure:	oral (gavage), once daily, 2 years
GLP statement:	yes
Date:	study completed 1991

*) the highest dose level was reduced to 3.5 mg/kg/d due to low body weight

Animals received daily oral gavage administrations of the test item at the indicated dose levels. Animals were observed daily. Bodyweight and food consumption were recorded weekly for the first 16 weeks and every 4th week thereafter. Ophthalmoscopic examinations were performed on all animals before the study and at all surviving high dose animals at the end of the study. Clinical laboratory investigations wee performed on ten animals during weeks 27, 53, 79 and 103. At necropsy, a wide range of tissues were taken and preserved. Histopathological examinations were performed on liver, kidneys and lungs from all animals and on selected further tissues for control and high dose animals.
Results:
121 males and 90 females died or were killed in extremis, deaths were considered not related to treatment by the authors. Clinical signs observed were hindlimb muscle atrophy in high dose animals. Body weight was reduced in high dose females. Relative lung weights were increased in mid and high dose males. Haematology revealed reductions in red blood cell count and haematocrit in high dose females. Concerning clinical chemistry and urinalysis, there were no toxicologically relevant treatment-related findings.
Non-neoplastic findings observed at the high dose were degeneration of muscle fibres, degeneration of spinal cord and sciatic nerve fibres and peripheral retinal atrophy at the high dose. The effects were also observed to a lesser degree at 1.5 mg/kg bw/d.
The incidence of neoplastic findings was not influenced by substance treatment.
Under the conditions of this study, NaPT was not carcinogenic to rats. Based on the non-neoplastic findings observed, a NOAEL of 0.5 mg/kg bw/d can be derived from this study.

Ref.: E11

Guideline:	OECD TG 453
Species/strain/sex:	Rat / Sprague-Dawley
Group size:	56 per sex /dose for the carcinogenic study (main group) 12 per sex for the chronic study at 0.5 and 1.3 mg/kg (satellite) 20 per sex for the chronic study at the highest dose (satellite)
Controls:	56 males and females for the carcinogenic study (main group) 12 males and females for the chronic study (satellite)
Test substance:	Natrium Pyrion 40% LSG
Batch:	99072150
Purity:	
Dose levels:	0.5, 1.5, 4.0 (2.8 in males; 2.1 in females)*) mg/kg/d
Exposure:	main groups: oral (gavage), 7days/week/ at least 104 weeks Satellite groups: oral (gavage), 7days/week/at least 52 weeks
GLP statement:	yes
Date:	2000 – 2002; report: 2004

*) the high dose was reduced during the course of the study due to severe reaction to treatment

Main group animals were checked daily for clinical signs, animals were palpated weekly. Behavioural examination was performed in satellite animals once weekly. At the end of the study, sensory reactivity and grip strength were measured in satellite animals and motor activity was assessed. Body weight was determined weekly for the first 13 weeks, thereafter once every 4 weeks with deviations (more frequent control at certain occasions).
Ophthalmoscopy (in all satellite and 10 /sex/dose main group animals) was performed prior to commencement and in week 50 and 102 for satellite and main groups, respectively. Blood and urine samples were taken at distinct time points during the study.
At necropsy, weights of selected organs were determined, selected tissues were fixed and examined histopathologically.

Results:

Chronic study:
Signs of toxicity (e.g. impaired limbs, motility impairment, ataxia, reduced body weights) were observed in animals at the mid and high dose. At microscopic examination, treatment-related changes were seen in the skeletal muscle of male and female animals in the high and mid dose groups at the end of the study. Dose-dependent changes in sciatic nerve were also observed in high-dose animals. From the results of the chronic part of the study, a NOAEL of 0.5 mg/kg/d was established.

Carcinogenic study:
Signs of toxicity such as ataxia, decrease muscle tone and emaciation were observed in animals of both sexes. Lower body weight was noted in low and high dose males and in mid- and high dose females. Low dose males were sacrificed on week 97 due to high mortality compared to controls, mortality in mid- and high dose females was higher compared to controls. However, there were no indications of significant different incidences of predominant pathology between control and treated animals.
There were no incidences of neoplasia with NaPT up to 2.8 mg/kg/day.
There were treatment related degenerative changes of the sciatic nerve and skeletal muscle in all treatment groups and gastric reactive change at 2.8 mg/kg/day in male rats only.

SCCS comments
(1) Individual animal data were not presented in this study; the annexes and tables were not provided.
(2) As there were treatment-related degenerative changes of sciatic nerve and skeletal muscle described in all treatment groups from the carcinogenic part of the study and as individual data are not available for deeper analysis, SCCS considers 0.5 mg /kg bw/d as LOAEL for NaPT.

Ref.: E7

Dermal

Guideline:	US-EPA 83-2
Species/strain/sex:	Mouse / Crl:CD-1 (ICR) BR (VAF Plus)
Group size:	50 per sex /dose
Controls:	50 males, 50 females exposed to distilled water only
Test substance:	Sodium Omadine ™ 41.2 % aqueous solution
Batch:	8508-P-166H
Purity:	

Dose levels: 5, 15, 40 mg/kg/d
Exposure: dermal, once daily, 80 weeks
GLP statement: yes
Date: study completed 1991

Dose levels were selected based on the results of a dermal 13-week toxicity study performed in rats. Animals received daily dermal applications of the test item at the indicated dose levels on clipped areas of dorsal skin. Bodyweight and food consumption were recorded weekly for the first 16 weeks and every 4th week thereafter. From week 27 onwards, animals were palpated weekly, during week 52 and 80, blood samples were taken from some animals. After 80 weeks of treatment, surviving animals were necropsied, weights of several organs were recorded and a wide range of tissues were preserved. Tissues from all descendents and control and high dose animals were examined microscopically. Lungs, livers, kidneys, treated skin and gross lesions from animals of the other groups were also examined.
Results:
28 male and 44 female animals died or were killed in extremis. Deaths were considered not treatment-related. There were no treatment-related clinical signs. Palpable masses were rare and only 4 were confirmed as tumours. Body weight was not affected by treatment, leucocyte differential counts were not different between treated and control groups.
Kidney weights of all treated females were slightly reduced dose-dependently. In view of the absence of pathologic findings in kidneys, this was considered as not of toxicological relevance. At the site of treatment, increased incidences of epidermal hyperplasia were observed in both sexes at 15 and 40 mg/kg/d. The effect was statistically significant at the highest dose. No treatment-related effects on sceletal muscle or sciatic nerve were observed in that study.
Dermal administration of NaPT had no effect on tumour incidence. Thus, based on the results of this study, NaPT induced slight histopathologic changes at the treatment site but did not influence tumour formation.

SCCS comment
No information of effects/findings in skeletal muscles and/or sciatic nerves can be obtained from that study. For local effects at the treatment site, a NOEL of 5 mg/kg/d can be obtained from that study.

Ref.: E12

Summary on repeat dose toxicity and chronic toxicity

Oral:
Several oral repeat-dose studies of different durations have been performed with ZPT. In addition, one sub-chronic and two chronic oral studies performed with sodium pyrithione (NaPT) can be considered adequate to assess repeat-dose effects of ZPT.
A NOAEL of 500 µg/kg/d obtained from a chronic oral study (Larson, 1958) performed with ZPT based on paralysis/hind-limb weakness has been derived in SCCNFP 0671/03.
The SCCS is aware that HSE (2003) considered the Larson 1958 study as inadequate due to insufficiently large group sizes to ensure statistical power. However, a 90-day oral study performed with sodium pyrithione (not available to the SCCS) which was considered adequate by HSE, also lead to a NOAEL of 500 µg/kg/d, supporting the outcome of the Larsson study.
Two oral chronic studies performed with NaPT have been provided by the applicant. In a combined chronic toxicity/carcinogenicity study the dose of 500 µg/kg bw/d is considered as LOAEL by the SCCS.

Dermal:
Several dermal repeat-dose studies have been performed with ZPT. Interpretation of the findings is partly hampered by the fact that grooming was not always prevented and that intermittent exposure regimens (causing recovery) have been applied. From a 28-day dermal neurotoxicity study in which grooming was prevented, NOAELs of 25 and 50 mg/kg bw/d were derived in female and male animals, respectively based on reduced electrophysiological parameters and muscle tone. In a two-year dermal chronic study performed with NaPT, a local NOEL of 5 mg/kg bw/d was derived.

Inhalation:
Three inhalation studies of different durations have been performed, two of them are available for evaluation. In a 21-day nose only study performed in Sprague-Dawley rats, a LOAEC of 2 mg/m^3 is derived based on histopathological data in tissues of the respiratory tract and non-respiratory tissues. In a 28-day nose only study performed in Sprague-Dawley rats, no NOAEC could be derived for local effects in the lung and an NOAEC of 1.5 mg/m^3 was derived for systemic effects.
In a 90-day study, animals were whole-body exposed and oral intake cannot be excluded.

3.4.6. Mutagenicity / Genotoxicity

Based on an Ames test, an *in vitro* CHO/HGPRT gene mutation assay, an *in vivo* mouse bone marrow micronucleus test and an UDS-Assay (references B58 and B60), SCCNFP/0671/03 concluded that ZPT has shown no mutagenic effect in any of the *in vitro* and *in vivo* studies conducted.
Since publication of SCCNFP/0671/03, further *in vitro* and *in vivo* studies on mutagenicity/genotoxicity of ZPT have been performed. However, the data are only partly available for evaluation and as far as available described in the following two sections.

3.4.6.1 Mutagenicity / Genotoxicity *in vitro*

Guideline:	US-EPA FIFRA Subdivision F Guideline 84-2 (corresponding to OECD TG 476) 40 CFR Part 158
Test system:	E. coli tester strain WP2 uvrA
Test substance:	Zinc Omadine
Batch:	9212093071
Purity:	97 %
Dose levels:	0.1 – 5000 µg/plate (initial mutagenicity assay) 0.05 – 150 µg/plate (independent repeat assay)
Solvent:	DMSO
Positive controls:	2-aminoanthracene; methyl methanesulfonate
GLP:	Yes
Study period:	07-02-2000 / 24-02-2000

Zinc Omadine was tested in the bacterial reverse mutation assay using E. coli tester strain WP2 uvrA in the presence and absence of Aroclor induced rat liver S9. Concentrations were based on the results of a previous study. Based on this precedent study, a preliminary toxicity study was not performed. Two independent assays were performed via the plate incorporation methodology. In the initial assay, precipitate was observed at concentrations ≥ 1000 µg/plate, toxicity was observed at concentrations ≥ 100 µg/plate. In the independent repeat assay, no precipitate was observed but toxicity was observed at concentations ≥ 50 µg/plate. No positive response was observed in the mutagenicity assays in the presence and absence of S9. Positive controls showed the expected results.

Ref.: E27

Guideline:	US EPA 84-2 (corresponding to OECD TG 476)
Test system:	CHO-K_1-BH_4 cells
Test substance:	2-Mercaptopyridine-N-Oxide Zinc; 48 % aqueous dispersion
Batch:	9RC-290-109 ZP
Purity:	not given
Dose levels:	Initial Assay: without S9: 0.25 – 2.0 µg/ml with S9: 5.0 – 30 µg/ml Confirmatory Assay: without S9 0.25 – 2.2 µg/ml With S9: 2.5 – 30 µg/ml
Solvent:	Ethanol for test substance, DMSO for positive controls
Positive controls:	Ethylmethanesulfonate, Benzo[a]pyrene
Treatment:	5 hrs
GLP:	Yes
Study period:	26-04-1990 / 06-09-1990

Dose levels were selected based on preliminary toxicity tests. S9 from livers from Aroclor-1254 induced male Sprague-Dawley rats was used as metabolic system. CHO cells were exposed to solvent or to concentrations of test substance as indicated in the presence or absence of S9 mix. Cytotoxicities were measured by cloning efficiencies relative to solvent controls.
In the initial experiment, the mutant frequency was statistically significantly increased compared to solvent control at 2.0 µg/ml (without S9). With S9, no mutant frequency was significantly increased compared to control. Positive controls yielded the expected response.
In the confirmatory experiment without S9 mix, mutant frequencies in test-article treated samples were not significantly elevated compared to control. In the presence of S9 mix, mutant frequencies at 20 and 2.5 µg/l were significantly elevated over control values.
As the increases in mutant frequencies were only slight compared to controls, as there was no dose-relationship and as there was no reproduction between the two experiments, the three observed increases in mutant frequenies are not considered biologically relevant. The authors conclude that ZPT is negative in this CHO/HPRGT assay.

Ref.: E13

SCCS comment
Only short treatment was performed and not 24h treatment. In each of two experiments with 5h treatment increased mutant frequency was observed, in one experiment without S9-mix and in the second experiment with S9-mix.

Guideline:	Japanese Guideline
Test system:	Blood lymphocytes from healthy non-smoking volunteers
Test substance:	Zinc Omadine ®
Batch:	9204084481
Purity:	96.3 %
Dose levels:	Main Test, Direct Method 0.6 – 1.2 µg/ml (24 hr -t) 0.2 – 1.0 µg/ml (48 hr) Main Test with metabolic activation: 6.0 – 12.0 µg/ml Main Test without metabolic activation: 4.0 – 12 µg/ml
Treatment:	3 h
Solvent:	DMSO
Negatice Control:	DMSO
Positive controls:	Mitomycin C (without S9); Cyclophosphamide (with S9)

GLP: Yes
Study period: 07-10-1992 / 28-11-1992

Two preliminary dose-finding experiments were performed. As metabolizing system, liver S9 from phenobarbital- and 5,6-benzoflavone induced Sprague-Dawley rats was used. Cytotoxicity was evaluated by using the mitotic index, the mitotic index of the negative control groups was set at 100 %.
After initial incubation periods of 24 and 48 hrs, cells were incubated for 3 hrs with test concentration as indicated, washed thereafter and further cultivated. After 69-hr incubation time in total, cells were incubated with 0.1 µg/ml colcemid and slides were prepared therafter.
Slides were then examined for chromosomal aberrations. No treatment-related increases in cells with structural abnormalities or polyploidies were observed after ZPT treatment, whereas positive controls yielded the expected responses.
Based on the results of this study, ZPT can be considered as non clastogenic to human lymphocytes under the conditions applied.

Ref.: E18

SCCS comment
Only short 3h treatment was used and not treatment for 1.5-2 cell cycle periods as recommended by recent guideline.

An Ames test with Salmonella strains TA98, TA 100, TA 1535 and TA 1537 using ZPT concentrations from 0.03 – 33 µg/plate was negative in the absence or presence of metabolic activation (rat liver S9-mix from Aroclor 1245-treated rats); no information on GLP- and guideline-adherence (Zeiger et al., 1987, taken from MAK 2012).

An Ames test with Salmonella strains TA98, TA 100, TA 1535 and TA 1538 used aqueous 48 % ZPT concentrations from 0.03 – 5.0 µg/plate in the absence of S9 and concentrations from 10 – 333 µg/plate in the presence of rat liver S9 mix. Cytotoxicity was observed from 3.3 µg/plate without metabolic activation and from 333 µg/plate with activation. No mutagenicity was observed (MAK, 2012).

In an in vitro Comet Assay using keratinocytes from human epidermis and human melanocytes from epidermis, DNA strand-breaks were induced after treatment with 100 or 500 nM ZPT. ZPT treatment at 500 nM induced comets with average tail moments that were increased approximately 3-fold over untreated controls within 1 hr of exposure. Significant comet formation was even observed at 100 nM, a dose that does not impair the viability of the cells. In a modified protocol using bacterial formamidopyrimidine-glycosylase, no oxidative DNA-damage was induced after treatment with ZPT.

Reference: D15 (Lamore et al., 2010).

ECHA's website ((http://echa.europa.eu/) mentions further *in vitro* assays which are not available for evaluation:

- a GLP-compliant Ames test performed according to OECD TG 471 with Salmonella strains TA98, TA100, TA1535 and TA1537. ZPT was considered negative in the absence and presence of metabolic activation (S9 mix not further specified) under the test conditions used.

- a GLP compliant mammalian cell gene mutation assay performed according to OECD TG 476 using Chinese hamster lung fibroblasts with and without metabolic activation. The test substance was considered not to induce gene mutations in this system.

- a GLP-compliant *in vitro* mammalian chromosome aberration test in Chinese hamster lung fibroblasts (V79) performed according to OECD TG 473. It was concluded that ZPT induced chromosomal aberrations under the test conditions used.

- a GLP-compliant mammalian cell gene mutation assay performed according to OECD TG 476 using Chinese hamster ovary cells. ZPT was considered negative in the absence or presence of metabolic activation (S9) under the test conditions applied.

- a GLP-compliant bacterial reverse mutation assay performed according to EPA OPP 84-2 using Salmonella strains TA98, TA100, TA1535, TA1537 (with and without metabolic activation; microsomal enzymes from Aroclor induced rat liver as metabolic system). ZPT was considered negative under the conditions applied.

- a GLP-compliant DNA damage and repair assay (unscheduled DNA synthesis) in mammalian cells *in vitro* according to US EPA 84-4 using rat hepatocytes. No conclusions were drawn from this study.

Overall SCCS comment on in vitro genotoxicity/mutagenicity
Results on *hprt* gene mutation test on CHO cells are inconclusive. In each of two experiments positive response was found – in one experiment without S9-mix and in the second experiment with S9-mix. Also only short treatment was used and not 24h treatment as it was also case in *in vitro* micronucleus test on human lymphocytes (no longer treatment for period of 1.5-2 cell cycles was used). The comet assay data on human keratinocytes indicates genotoxicity and there was also a positive effect found in in vitro mammalian chromosome aberration test in V79 cells (ECHA's website (http://echa.europa.eu/)).

3.4.6.2 Mutagenicity/Genotoxicity *in vivo*

Guideline:	Japanese Guideline (similar to EC method B.10 and OECD 473)
Test system:	Blood lymphocytes from Cynomolgus Monkeys enrolled in an oral 28d repeat-dose study
Dose groups:	2 per sex per dose groups
Test substance:	Zinc Omadine ®
Batch:	9204084481
Purity:	96.3 %
Dose levels:	5.5, 11.0 and 22.0 mg/kg/d
Exposure:	oral (gelatine capsules), 28 d
Control:	empty gelatine capsules
Negatice Control:	DMSO
GLP:	Yes
Study period:	07-08-1992 / 28-11-1992

Lymphocytes were obtained from the animals on the day after the end of the exposure period. Lymphocyte cultures were prepared and incubated for 66 hrs. Thereafter, colcemid was added and chromosomes were prepared after further incubation for 6 hrs.
Slides were prepared from the cells and inspected for chromosomal aberrations (100 metaphases per slide were selected for examination) and polyploidy cells. No significant increases in chromosomal aberrations and polyploidy were determined in this study. Under the conditions of this test, ZPT did not induce structural aberrations and polyploidy.

The SCCS considers this test of limited value as no positive control was included.

Ref.: E19

Among the "full study reports" submitted after the commenting period, there was an abstract on a micronucleus assay. As the full study report is not available, no firm conclusions can be drawn from that abstract with respect to genotoxicity:
"Male and female ICR mice were exposed to 11, 22 or 44 mg/kg of 2-Mercaptopyridine-N-Oxide Zinc which was administered in a constant volume of 10 ml/kg as a single IP injection. The high dose level was calculated to be 80 % of the LD_{50}. One of 15 male mice dosed with 44 mg/kg diesd prior to its scheduled sacrifice time and was replaced with an animal from the 44 mg/kg replacement group. No female mice died as a result of treatment. Bone marrow cells, collected 24, 48 and 72 hours after treatment, were examined microscopically for micronucleated polychromatic erythrocytes. No change in the ration of polychromatic erythrocytes to total erythrocytes was observed in male or female mice in the test article treated groups, suggesting that the test article did not induce bone marrow toxicity. No significant increases in micronucleated polychromatic erythrocytes were observed at 24, 48 or 72 hours after dose administration in males or females. The results of the assay indicate that under conditions described in this report, 2-Mercaptopyridine-N-Oxide Zinc did not induce a significant increase in micronucleated polychromatic erythrocytes in male or female ICR mice. 2-Mercaptopyridine-N-Oxide Zinc was concluded to be negative in the mouse micronucleus assay."

Ref.: E20

ECHA's website (http://echa.europa.eu/) mentions two further *in vivo* genotoxicity assays which are not available for evaluation:

- a GLP-compliant *in vivo* mammalian erythrocyte micronucleus test performed according to OECD TG 474 in male and female Crl:NMRI BR mice. It was concluded that ZPT did not produce relevant increases of the numbers of micronuclei in polychromatic erythrocytes after oral gavage of doses up to 1300 mg/kg.

- a GLP-compliant micronucleus assay performed according to EPA OPP 84-2 using Sprague-Dawley mice[*)] and intraperitoneal administration. ZPT was concluded to be negative in this assay. (Comment: this study appears to be identical with a study described in HSE (2003), therein cited as unpublished, 1990c. HSE concluded that ZPT did not increase micronucleus formation and considered this study as adequate).
[*)] the SCCS is not aware that such a mouse strain exists; the mouse strain was not specified in HSE (2003)

Summary on Genotoxicity
From the studies available for SCCNFP/0671/03, it was concluded that ZPT is not mutagenic. Since then, further *in vitro* and *in vivo* genotoxicity/mutagenicity studies have been performed, not all of them are available for evaluation. *In vitro* studies are incomplete and in case of *hprt* gene mutation results are inconclusive with signs of potential mutagenicity that deserve further investigation. *In vivo* micronucleus test only discriminates mutagenic compounds with chromosomal aberration/clastogenic or aneugenic effect and does not detect gene mutation inducing compounds.
Therefore, no firm conclusion with respect to genotoxicity/mutagenicity can be drawn by the SCCS. The SCCS, however, is aware that HSE (2003) and MAK (2012) considered ZPT as non-genotoxic and non-mutagenic.
With respect to the development of cancer, the ambiguous database on genotoxicity might be acceptable as no carcinogenic effect has been observed in chronic oral studies up to dose levels of 2.5 mg/kg bw/d ZPT and 3.5 mg/kg bw/d NaPT (based on systemic findings, higher doses of ZPT/NaPT could not be tested in chronic oral studies). Also with respect to

developmental or reprotoxic effects, the ambiguous database might be acceptable as ZPT and NaPT cannot be considered as reproductive or developmental toxicants.

3.4.7. Carcinogenicity

Oral:
See section 3.3.5.3

Dermal:

Taken from SCCNFP/0671/03

Since the highest dose level of 0.1 ml of 10% ZPT was well tolerated in mouse pilot studies it was deemed suitable for use in the dermal carcinogenicity bioassay conducted by Patterson and Gargus (1979). ICR Swiss mice (730 animals) were selected at random and assigned to the following groups. Each received the noted treatment on a 6 cm^2 clipped area. No attempt was made to control ingestion of the applied material. The two dose levels represent approximately 20 and 100 mg/kg/day.

Table 6: Treatment regimen of the dermal carcinogenicity bioassay by Patterson and Gargus (1979).

Group	Treatment	Number of mice	
		male	female
1	Negative controls, no treatment	139	141
2	Vehicle controls, 0.1 ml	75	75
3	Low dose, 0.1 ml vehicle (2 mg ZPT/application)	75	75
4	High dose, 0.1 ml vehicle (10 mg ZPT/application)	75	75

All animals were housed in individual hanging wire-mesh cages. Individual body weights were recorded initially and at monthly intervals. Observations were made daily for mortality and at each treatment period for evidence of systemic effect and skin lesions in the area of treatment. Treatment was continued for 18 months, at which time it was discontinued, and the mice were maintained until each group mortality reached 75%. The group was then necropsied.
Only the males in the high-dose group experienced a mortality rate of 75% prior to the end of the study. For this group the average survival time was 409 days as compared to 492 days for the untreated males, which suggests a relationship to ZPT toxicity. However, neither gross examination of the animals and tissues, revealed any consistent lesion that would explain the reduced life span. The skin of all three treated groups exhibited changes consistent with exposure to a low-level chemical irritant.
There were no significant differences in the types or incidence of tumours or abnormal tissue masses between any of the groups that could be related to the administration of ZPT.
The chronic study summarised above reveals no evidence of a carcinogenic response when ZPT was applied topically (up to 100 mg/kg/d) in lifetime studies using mice and rats.

New data:
A chronic (80 week) dermal study performed with NaPT in the mouse is described in section chronic (>12 months) toxicity.

Conclusion on carcinogenicity
From chronic oral and dermal studies available in submission I, SCCNFP 0671/03 concluded: "no evidence of a carcinogenic response was seen when ZPT was applied topically (up to 100 mg/kg/d) or given orally (up to 5 mg/kg/d) in lifetime studies using mice and rats."
Since that, further chronic (lifetime) studies performed with ZPT and sodium pyrithione (from which read across to ZPT is considered adequate) using the oral and dermal uptake pathway have become available.
From the studies performed by the oral or dermal route with either ZPT or NaPT there was no evidence for a carcinogenic potential up to dermal doses of 100 mg/kg bw day and up to oral doses of 2.5 mg/kg bw/d ZPT and 3.5 mg/kg bw/d NaPT (based on systemic findings, higher doses of ZPT/NaPT could not be tested in chronic oral studies).
Carcinogenicity of ZPT has not been investigated by the inhalation route.

3.4.8. Reproductive toxicity

3.4.8.1. Fertility

In GLP- and guideline-preceding study, fertility effects of ZPT after dermal administration were investigated in Sprague-Dawley rats. Animals were treated with a 48% ZPT slurry at doses of 1.2, 7.5 and 15.0 mg/kg/d for 8 weeks (grooming not prevented) or only on days 6-15 of gestation (grooming prevented). In animals receiving 8-week treatment, either treated males were mated with untreated females of untreated males were mated with treated females. ZPT dosed rats from the 8-week treatment scheme did not differ significantly from controls in either growth or reproductive characteristics except a statistically significantly lower lactation index in females of the highest dose group. No toxic signs such as paralysis and no test-related histopathology was seen in the males. Further, neither reproduction nor neonatal viability was affected after topical administration of ZPT on GD 6—15. A NOAEL of 7.5 mg/kg/d can be derived from this dermal study.

Reference: B43

SCCS comment
This study was already available for SCCNFP 0671/03, but therein it was discussed together with the developmental/teratogenic studies.

3.4.8.2. Two generation reproduction toxicity

Two two generation reproduction toxicity studies performed with **sodium pyrithione**, from which read-across to ZPT is considered appropriate (see section 3.3.5.3) have been provided.

Guideline:	not stated
Species/strain/sex:	Rat / Sprague-Dawley (OFA-SD (IOPS-Caw))
Group size:	20 F0 pregnant females and 20 male and female F1 animals / dose
Controls:	20 pregnant animals, vehicle control
Test substance:	Sodium Omadine, 40.4 % aqueous solution
Batch:	8508-P-166H
Purity:	not given
Vehicle:	sterile water
Dose levels:	0.5, 1.5, 3.5/4.5*) mg/kg/d

Exposure: oral, once daily from GD 15 to day 21 post partum
GLP statement: yes
Date: 1989 – 1990; report: 1990

*) the highest dose level was reduced from 4.5 to 3.5 mg/kg/d during the first week of lactation.

Groups of male and female Sprague Dawley rats received aqueous suspensions of 0, 0.5, 1.5 and 3.5 (the latter dose was reduced from 4.5 mg/kg/d due to toxicity) mg/kg/d sodium pyrithione by gavage. Parental animals were treated for 11 weeks, and then mated. Dosing of females continued through mating, gestation and lactation. In parental females at the highest dose, body weight was decreased by approximately 10 % at the end of gestation and during lactation. A single female of this dose group was killed *in extremis* due to hind limb impairment. No further mortalities or clinical signs of toxicity were noted in parental animals. At necropsy of parental animals, atrophy of hind limb skeletal muscle (27/50) was found at the top dose, characterised by a reduction and variation in the diameter of muscle fibres. In male parental animals, copulation and fertility indices were both decreased by 26 % at the highest dose group.
In F1 pups there were no effects on gestation success or duration, number of pups born, live births, indices of viability and lactation, cumulative survival or sex ratio. No effects on pup weight were observed. No pup abnormalities were found following necropsy. No effects on development of ear opening, righting reflex or eye opening were observed. However, at the highest dose a reduced incidence of startle response at 15 d was observed (by 10 %).
In the P1 generation, 2 females of the highest dose group were killed *in extremis* due to hind limb impairment. No further mortalities or clinical signs of toxicity were noted. No adverse effects on food consumption or body weight were noted. No adverse effects on the indices of copulation or fertility were observed in the P1 (F0) animals. At necropsy of P1 (F0) animals, abnormal findings among P1 (F0) animals were confined to atrophy of skeletal muscle (29/50) at the top dose.
There were no effects on the success or duration of gestation, number of live births, viability, lactation and the cumulative survival score in the F2 generation. In F2 pups no effects on pup weight were observed. No abnormalities were found following necropsy. No effects on development of ear opening, righting reflex or eye opening were observed. However, in the top dose a reduced incidence of startle response (by 10 %), an indicator of delayed development, was observed on day 15.
From this study, a NOAEL of 1.5 mg/kg/d was established for parental toxicity based on hind limb impairment and skeletal muscle atrophy. An NOAEL of 3.5 mg/kg/d was derived for fertility effects.

Ref.: E22

Guideline: US EPA 83-4 (Complies with OECD 416)
Species/strain/sex: Rat / Crl:CD® (SD) BR
Group size: 25 per sex / dose
Controls: 25 per sex, 0 mg test substance in vehicle
Test substance: Sodium Omadine / Sodium Pyrithione
Batch: 8508-P-166H
Purity: 41.2 % [w/w] aqueous solution
Vehicle: distilled water
Dose levels: 0.5, 1.5 and 3.5*) mg/kg bw/d
Exposure: oral, gavage, once daily
GLP statement: yes
Date: 1987 – 1988

*) 4.5 mg/kg bw/d in weeks 1, 2 and 3

F0 parental animals received daily oral dosing during the premating and mating phase until the day before necropsy which was shortly after the mating period for males and at weaning of the F1 generation for females. After mating, pregnant females were allowed to litter and to rear their offspring until weaning. After weaning, 25 female and male F1 animals were selected for the production of a second generation. The selected F1 animals were also dosed once daily during the premating and mating period until necropsy at the same levels as the F0 generation. Clinical condition, bodyweight, food consumption, fertility and mating performance, macroscopic abnormalities at necropsy and microscopic examination of selected organs were recorded for the parental animals. Litter size, clinical condition, growth and development to weaning and macroscopic abnormalities at necropsy were recorded for the offspring.
Results:
One F0 female and two F1 females at 3.5 mg/kg bw/d were killed because of poor clinical condition due to hindlimb paralysis or impairment of hindlimb movement.
Body weight gain for both F0 and F1 animals at 3.5 mg/kg bw/d was reduced. The number of F1 and F2 pups born at 3.5 mg/kg bw/d was slightly less than the controls. Also, for both F1 and F2 pups, a slight retardation of development was seen at 3.5 mg/kg bw/d. There were no treatment-related clinical signs or necropsy findings. Pup survival was not affected by treatment.
Conclusions:
Oral administration of NaPT at 3.5 mg/kg bw/d elicited toxic changes in parental animals and offspring from two generations: in parental animals, a reduction in body weight gain, atrophy of hind limb muscles, hindlimb paralysis, impaired movement and adverse effects on F0 generation fertility and mating performance were observed. In the offspring, there was evidence of a slight retardation of development during lactation. At 1.5 mg/kg bw/d toxic changes in the parental animals were limited to atrophy of the hindlimb muscles in a few females.
From the results of this study, a parental NOAEL of 1.5 mg/kg bw/d can be derived. For the offspring, NOAELs of 1.5 and 0.5 mg/kg bw/d can be derived for male and female animals, respectively.

Ref.: E21

A further two-2-generation study is mentioned on ECHA's website (http://echa.europa.eu/). This study is not available for evaluation:

- a 2-generation study performed according to EPA OPPTS 870.3800 in male and female Sprague-Dawley rats receiving 0, 0.7, 1.4, and 2.8 mg/kg/d aqueous substance by gavage. For parental animals, NOAELs of 1.4 and 0.7 mg/kg/d were derived for male and female animals, respectively. For F1 animals NOAELs of 1.4 and 0.7 mg/kg/d were derived for male and female animals, respectively.

3.4.8.3. Teratogenicity

Taken from SCCNFP/0671/03

Several teratology/reproduction studies have been conducted using rats and rabbits, in which ZPT was either applied topically or given orally. Topical application (with ingestion during grooming) of levels up to 15 mg/kg/day did not adversely affect reproduction in rats. When pregnant rats were gavaged with 15 mg/kg/day of ZPT, there was an increase in the incidence of forked and fused ribs in the neonates. A dose level of 2.5 mg/kg/day given orally is a no-effect level for teratogenicity/embryotoxicity. No material toxicity was observed in these studies.
Teratology data are summarised in the table below, followed by a brief description of each of the studies.

Table 7: Summary of Teratogenicity Studies performed with ZPT

Species	Route of administration	Dose levels (mg/kg)	Teratology findings	Ref.
Rat	Oral	7.5 15.0	7.5 – none 15.0 – increased incidence of fused or forked ribs	B24
Rat	Oral	7.5 15.0	7.5 – none 15.0 – increased incidence of fused or forked ribs	B43
Rat	Topical with ingestion of applied material	2.5 7.5 15.0	2.5 - none 7.5 - none 15.0 - none	B43
Rat	Topical	2.5 7.5 15.0	2.5 - none 7.5 - none 15.0 - none	B43
Rabbit	Oral	5.0 10.0 20.0	5.0 - fatal to 6/15 dams, no teratogenic effects 10.0 – fatal to 10/15 dams, no teratogenic effects 20.0 – fatal to 15/15 dams	B43
Rabbit	Oral	1.0 2.5 5.0	1.0 – none 2.5 – none 5.0 - none	B43
Rabbit	Topical	25.0 50.0 100.0	25.0 - none 50.0 – none 100.0 - none	B43

Oral, Ingredient based data

In the teratology study conducted by Haley et al (1971) groups of 19, 16, and 20 albino rats were administered dose levels of 0, 7.5 or 15.0 mg/kg/day, respectively, of ZPT for the sixth through the fifteenth day of gestation. The material was dosed as a solution in corn oil by oral intubation. All animals were allowed food and water ad libitum. Maternal body weights were depressed in the groups given ZPT. The mean weight gain per animal for the

control group was 132 mg, for the group receiving 7.5 mg/kg, 69 mg; and for the group receiving 15.0 mg/kg, 89 mg. An increased incidence of skeletal abnormalities, particularly fused and forked ribs, was seen in the foetuses in the higher dose group, but not in the group receiving 7.5 mg/kg/day.

Nolen and Dierckman (1979) in a series of studies evaluated the embryotoxic/teratogenic effects of ZPT in rats and rabbits. Initially they confirmed the report by Haley et al. (1971) in that pregnant rats dosed orally with 15 mg/kg/day produced litters with an increased incidence of skeletal abnormalities. They also confirmed that 7.5 mg/kg day did not cause a statistically significant increase in the number of foetal abnormalities. ZPT administered orally to pregnant rabbits from day 6 through day 18 of pregnancy was lethal to 6/15 at the 5 mg/kg level, 10/15 at the 19 mg/kg level and 15/15 at the 20 mg/kg level. The surviving animals lost weight and had significantly higher incidences of embryonic resorption.

However, there was no evidence of teratogenicity. In another experiment, rabbits were dosed orally with 1, 2.5, or 5 mg/kg of ZPT. Animals receiving 5 mg/kg lost a significant amount of weight, but none died. In addition, the number of resorptions was significantly increased compared to the water control. Dams dosed with 2.5 mg/kg ZPT gained less weight than controls and had a higher incidence of resorptions, neither observation being statistically significant, while the data from dams treated with 1 mg/kg were similar to those of the control. None of the ZPT doses had any adverse effects on foetal development. Thus, 2.5 mg/kg/day is a no-effect level for orally administered ZPT in teratology studies.

Dermal, Ingredient based data
No deleterious effects were seen in rabbits treated topically with 25, 50, or 100 mg ZPT/kg/d with oral ingestion controlled by a leather harness, and as in the other rabbit studies, no teratogenic effects were observed in this study (Nolen and Dierckman (1979)).

In a separate study using rats, Nolen and Dierckman (1979) topically applied a 48% aqueous slurry of ZPT to three groups of 10 animals at levels of 2.5, 7.5, and 15 mg/kg/day. The animals were dosed from eight weeks before mating until day 15 of gestation. No attempt was made to control ingestion during grooming in that the material was not washed off, and additional applications were made over the previous treatment. Three other groups of rats were treated at the same dose levels, but ingestion of the ZPT was prevented by means of plastic domes glued to their backs. The animals in this portion of the study were treated from day 6 through day 15 of gestation. In the rat reproduction portion of the study, ZPT produced no adverse effects on growth, pathology, or conception in the parents, or on viability, post-weaning growth, or pathology in the neonates. Ingestion of the material during grooming did cause hind-limb paralysis in 2/10 dams dosed at 7.5 mg/kg/day and in 5/10 dams dosed at 15 mg/kg day. There was no evidence of any adverse effects in any group in either the dams or foetuses when ingestion was prevented by the plastic dome. No teratogenic or adverse effects on reproduction have been seen when the material is applied topically to rats and rabbits at levels up to 15 and 100 mg ZPT/kg/day respectively (highest doses tested).

Product based data
Teratology studies have been conducted using rabbits and pigs in which ZPT was incorporated into a product base and applied topically. Jordan and Borzelleca (1975) studied the effects of 1%, 2% or 6% ZPT applied as part of a shampoo formulation to the backs of pregnant Yorkshire swine at levels equivalent to 50, 100, or 300 mg/kg/day. Product was applied from day 12 through day 36 of gestation. There was no evidence of embryotoxicity nor teratogenic effects in the foetuses.
In a study reported by Wedig et al (1976), Yorkshire pigs were again used as the test species. In this test, a 50% (w/v) suspension of ZPT in Aquafor Cream (commercial product) was applied to a 380 cm^2 area of the backs of the animals. Eight dose sites were used in rotation to prevent irritation at the dosing area. An untreated group and a group

treated with Aquafor Cream without ZPT were used as controls. Dose levels of ZPT were 30, 100, and 400 mg/kg/day. Material was left on the skin for eight hrs/day from the eighth through the thirty-second day of gestation. During the treatment period each animal was individually confined in such a manner as to prevent oral ingestion of the test materials. Slight erythema was observed in some animals during the dose administration period, but all lesions were reversible and were not apparent at sacrifice (day 100). No signs of systemic toxicity were observed in any of the animals. Maternal body weights were not depressed by administration of ZPT. No evidence of teratogenic effects was observed in the foetuses from the ZPT-treated animals either grossly following examination of internal organs or upon skeletal examination.

Nolen et al (1975) have reported the results of a percutaneous teratology study of ZPT in rabbits. A lotion shampoo containing 2% ZPT was applied for two hours each day at either 1 or 2.5 g/kg (20 or 50 mg/kg of ZPT) from the seventh to the eighteenth day of gestation to groups of 15 rabbits. A third and fourth group of animals received either no treatment or the shampoo base without ZPT. These were control groups. Oral ingestion was prevented by harnessing the animals and cleaning the cages. ZPT had no effect on maternal weight gains and was not teratogenic under these conditions.
No teratogenic effects were seen in rabbits topically treated from the seventh to the eighteenth day of gestation with shampoo containing up to 50 mg/kg ZPT. Neither were effects seen in pigs topically treated from the eighth to the thirty sixth day of gestation with a shampoo containing up to 300 mg/kg ZPT.

Further data

Guideline:	US-EPA 83-3
Species/strain/sex:	Rat / Charles River Crl:CD® VAF/Plus®
Group size:	30 per dose
Controls:	30 animals, vehicle
Test substance:	Zinc Omadine ®, 48 % aqueous solution
Batch:	33-22902781
Purity:	not given
Vehicle:	deionised water
Dose levels:	0.75, 3.0 and 15.0 mg/kg/d
Exposure:	oral, gavage, once daily on GD 6 - 15
GLP statement:	yes
Date:	1992, report 1993

Dose levels were selected based on a precedent study. Animals received the test substance at 0.75, 3.0 and 15.0 mg/kg/d on GD 6-15 and were sacrificed on GD 20. Animals were observed twice daily; clinical signs and toxicity were recorded once daily from GD 6 - GD 20. Body weights and food consumption were recorded on selected time points. On GD 20, surviving animals were killed and maternal and fetal tissues were investigated.
Results
Maternal toxicity (significant inhibition of weight gain during treatment and a higher incidence of increased salivation post-dose relative to the control group) was present at 3.0 and 15.0 mg/kg bw/d with a higher degree in high dose animals. Body weight was decreased in the high dose group by 10 % from day 12 until study termination. Body weight gain and food consumption were also decreased by 30 % and 15 % respectively in the high dose group, throughout the study. Clinical signs reported were dilated pupils in high dose animals and a dose-related increase in the incidence of excessive salivation in mid- and high dose groups.
Developmental toxicity was present in the mid- and high dose groups (less pronounced in the mid dose group). At the high dose level, skeletal malformations and developmental malformations were observed for the majority of litters. Fetal survival (increased

postimplantation loss) and mean fetal weight were also adversely affected at the high dose. At 3.0 mg/kg bw/d, a slight increase in the incidence of fetal malformations and a less pronounced increase in mean postimplantation loss compared to controls were observed.
However, as fetal effects occur at dosages where maternal toxicity is observed, the test article is not considered as a developmental toxicant.
A NOAEL of 0.75 mg/kg/d is derived from the study for maternal and developmental toxicity.

Ref.: E23

Guideline:	US-EPA 83-3 (Comparable to OECD TG 414)
Species/strain/sex:	New Zealand White Rabbits
Group size:	20 females/dose
Controls:	20 female animals receiving vehicle
Test substance:	Zinc Omadine ®, 48 % aqueous solution
Batch:	33-22902781
Purity:	52.2 %
Vehicle:	deionised water
Dose levels:	0.5, 2.0, 4.0, 8.0 and 12.0 mg/kg bw/d
Exposure:	oral, gavage, once daily on GD 6 - 18
GLP statement:	yes
Date:	1993

Dose levels were selected based on a precedent study. Animals received the test substance at 0.5, 2.0, 4.0, 8.0 and 12.0 mg/kg/d on GD 6-18 and were sacrificed on GD 29. Animals were observed twice daily; clinical signs and toxicity were recorded once daily from GD 6-GD 29. Body weights and food consumption were recorded on selected time points. On GD 20, surviving animals were killed and maternal and fetal tissues were investigated.
Results:
One animal of the mid dose died, probably due to a dosing error. Body weight and food consumption were decreased during the dosing period.
At study termination, post implantation losses of 12 % were observed in controls and low dose animals, which increased with increasing dose. The number of dams with viable fetuses was decreased at the highest dose. The incidence of fetuses with abnormalities was increased at the highest dose. The cases of fetal abnormalities were different in type and there was no dose-relationship.
A very high incidence of post-implantation losses was observed at the highest dose, which is considered treatment-related, but might be seen as a consequence of maternal toxicity.
A NOAEL of 0.5 mg/kg bw/d for maternal and developmental toxicity can be derived from this study.

Ref.: E24

Guideline:	OPPTS 870.3700
Species/strain/sex:	Rat / Crl:CD® (SD)IGS BR VAF/Plus®/ female
Group size:	24 at 10, 15 and 30 mg/kg bw/d; 25 at 60.0 mg/kg bw/d
Controls:	23 animals, vehicle
Test substance:	Zinc Pyridinethione (ZPT) – an off-white to tan powder
Batch:	0108244691
Purity:	95 – 99 %
Vehicle:	reverse osmosis membrane deinonized water
Dose levels:	10.0, 15.0, 30.0 and 60.0 mg/kg/d
Exposure:	dermal, ingestion prevented, once daily on GD 0 – 20, 6hrs/d
GLP statement:	yes

Date: 2004 - 2005

Test formulations were applied to shaved areas of the backs of rats considered pregnant, the area was secured with tape and Elizabethan collars were placed around the necks of the animals. After 6-hr exposure periods, tapes and collars were removed and the area was washed. Skin sites were examined before the applications. Rats were examined for viability, general appearance, clinical observations, abortions, premature deliveries and deaths. A hindlimb neurological evaluation was performed on GDs 8, 12, 16 and 20. Body weights were recorded weekly, food consumption on GDs 0, 3, 6, 9, 12, 15, 18 and 20. On GD 21, animals were sacrificed, Caesarean sections were performed and examinations on maternal animals and fetuses were performed.
Results:
Two deaths occurred and were considered not to be related to the test item. Body weights and body weight gains were reduced or significantly reduced at 30 and 60 mg/kg/d when compared with controls. Uterine weights in the highest dose group were significantly reduced. Pregnancy occurred in 23 to 24 animals per dose group. Fetal body weights were significantly reduced in the 60 mg/kg/d group, which was also maternally toxic. In this dose group there was an increased number of fetuses with incomplete ossification of the sternal centra and/or with wavy ribs. Further, there were significant reductions in the ossification site averages for the caudal vertebrae, forelimb phalanges, metacarpals, hindlimb phalanges and metatarsals.
There was an increased number of rats with limited use of hindlimbs in the 30 mg/kg/d group and an (significantly) increased number of rats with erythema grade 1, flaking grade 1, limited or no use of hindlimbs, low muscle tone, shuffling gait, dehydration, ungroomed coat, low carriage, chromodacryorrhea, emaciation, chromorhinorrhea and hunched posture in the 60 mg/kg/d dose group.
Based on the results of this study, a maternal (systemic) NOAEL of 15 mg/kg bw/d based on limited use of hindlimbs and a developmental NOAEL of 30 mg/kg bw /d were derived. ZPT was not considered as a selective developmental toxicant.

Ref.: E1

Conclusion on Reproductive toxicity
In SCCNFP 0671/03 the following conclusions were drawn with respect to Reproductive toxicity of ZPT:
- 2.5 mg/kg/d administered orally to rats is a no effect level for teratological effects
- no reproductive effects have been observed when ZPT was applied topically to rats and rabbits at levels up to 15 and 100 mg ZPT/kg/d respectively (highest doses tested) and ingestion of the test material was controlled.
- no reproductive or teratogenic effects have been observed in rabbits and pigs following topical application of shampoo formulations containing 50 and 400 mg ZPT/kg/d respectively.

Since that, further generation studies have been performed with ZPT and NaPT as well as developmental toxicity studies with ZPT. One 2 generation study which is mentioned at ECHA's website is not available for evaluation by SCCS. However, based on the overall picture given by the available data (and by the summary of the non-available study), the conclusions of other scientific bodies can be supported: the SCCS is aware that HSE (2003) did not identify any potential concern to humans regarding adverse effects on fertility. Further, both MAK (2012) and HSE (2003) concluded that adverse effects on development were most likely attributable to maternal toxicity.

3.4.9. Toxicokinetics and PBPK modeling

3.4.9.1 Summary of toxicokinetics taken from SCCNFP/0671/03

- percutaneous absorption of ZPT varies from approximately 0.03 to 3.4%
- the distribution of radioactivity in tissues after oral administration of labelled ZPT showed that the radioactivity rapidly disappeared from the blood, and the primary route of excretion was via the urine. The residual radioactivity was low (4.5% of dose), ZPT was distributed throughout the body, and was not concentrated in any particular tissue.
- all animal species investigated (rat, rabbit, dog, and monkey) biotransformed ZPT in qualitatively similar ways. This similarity with regard to ZPT metabolism suggests that human metabolism is likely to be similar. This has been confirmed by Wedig et al. (1984).

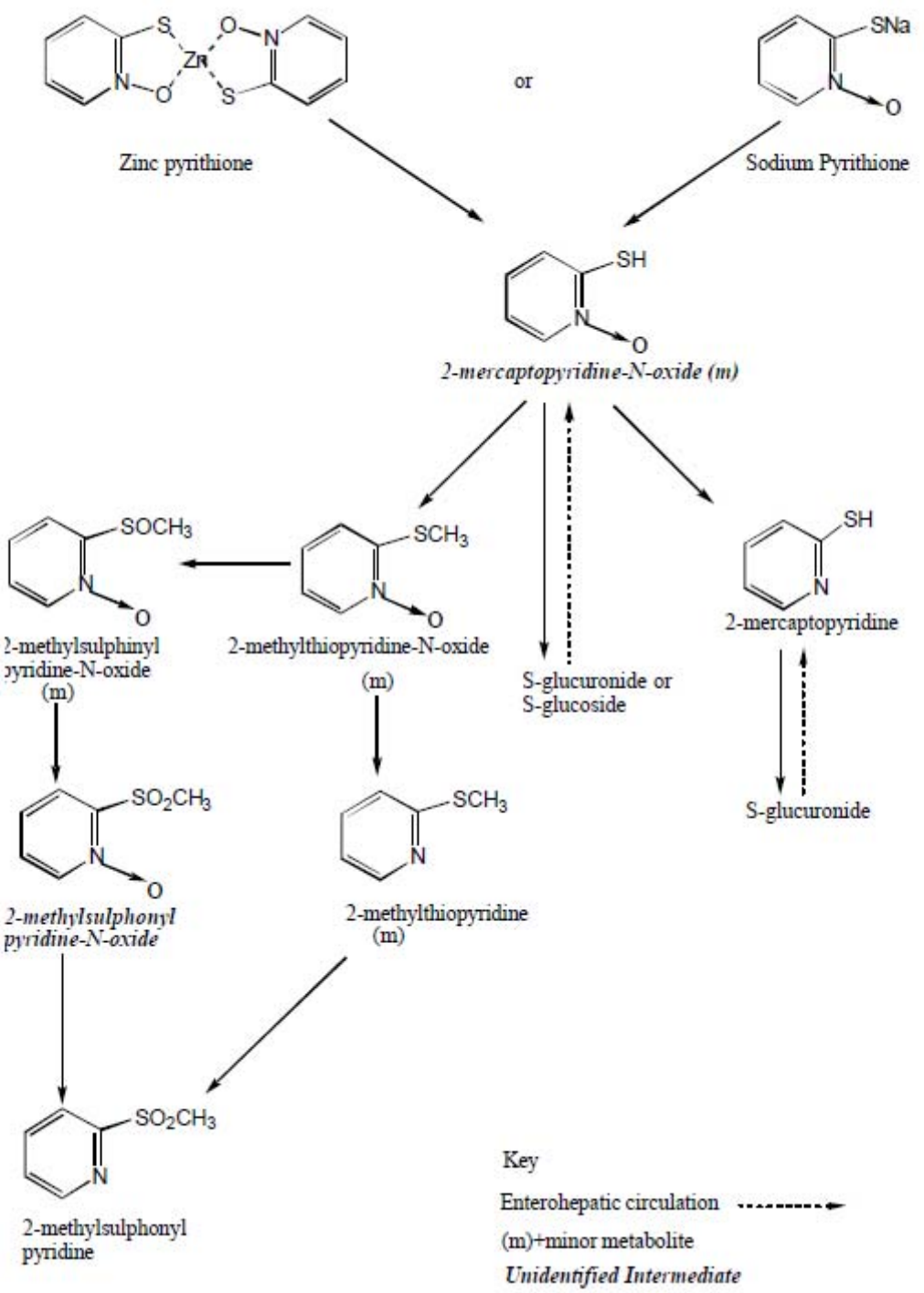
3.4.9.2. General information on Metabolism and Toxicokinetics of ZPT

The metabolism and toxicokinetics of ZPT have been well investigated in different species. An overview on toxicokinetic studies performed with ZPT and NaPT in animals and humans is given in HSE (2003) where ADME studies are summarised as follows and a metabolic scheme is given:

"Studies have been presented in experimental animals to address the toxicokinetics of zinc pyrithione and sodium pyrithione following oral administration, and zinc pyrithione only following dermal administration. Studies are also presented to address the toxicokinetics of zinc pyrithione and the related compound sodium pyrithione in a number of experimental animal species following i.v. administration. No specific information was identified to address the toxicokinetics of zinc pyrithione following single inhalation exposure or repeated exposure by any route. However, from the information available it was possible to conduct an adequate assessment of the toxicokinetics of zinc pyrithione. No further toxicokinetic information is required at the present time.
Following oral dosing, absorption of ^{14}C labelled zinc pyrithione and sodium pyrithione was high, around 75-90% of the administered radiolabel. Studies performed with ^{65}ZnP indicate that following oral administration, zinc pyrithione disassociates to liberate Zn and the pyrithione moiety which are then absorbed independently. Dermal absorption of zinc pyrithione was found to be minimal. Short term tissue retention of radiolabel was confined to erythrocytes. The metabolic profiles of orally administered zinc pyrithione and i.v. administered sodium pyrithione were extensive, and qualitatively similar in rats, rabbits, dogs and monkeys. In all species, independent of route or pyrithione salt, a common terminal metabolite, 2-methylsulphonylpyridine was identified, which was also identified in workers involved in pyrithione manufacture. The identification of a common terminal metabolite suggests a common metabolic pathway. Data from studies performed in the pig with both zinc pyrithione and sodium pyrithione support the suggestion of a common metabolic pathway. Excretion of zinc pyrithione and sodium pyrithione was found to be rapid, principally via the urine, with faecal excretion being a minor route. It was found that biliary excretion and enterohepatic circulation are both important in the rat.
As zinc pyrithione is rapidly metabolised, the potential for bioaccumulation of the parent molecule is low. However, it was found that 2-methylsulphonylpyridine levels rise with time after dosing (up to 72 hours), although insufficient information was available to indicate bioaccumulation potential. As the zinc pyrithione metabolites are anticipated to be water soluble, then distribution of these metabolites is expected to be extensive. Thus, exposure of the developing embryo/fetus to zinc pyrithione metabolites is likely. However, given the very high lipid content of breast milk, it is thought unlikely that postnatal exposure could occur via this route.
Given the detection of 2-methylsulphonyl pyridine in human plasma, it can be concluded that the metabolic pathways are qualitatively similar across a range of species. However, as

the molecular weight cut off for biliary excretion in humans is higher than in rats, this route of excretion is likely to be of diminished importance, in humans."

Figure 2: metabolic scheme for ZPT (according to HSE (2003))



3.4.9.3. New data on metabolism and toxicokinetics of ZPT

(1) Toxicokinetics of ZPT after single (i.v.) and repeat dose oral (gavage, diet) and dermal administration (Experiments performed in a first phase to assist in the parameter selections for the main repeat dose studies described below)

Ref.: A22

The experiments consisted of a series of single dose experiments involving two animals per experiment (pilot studies) and several repeat dose studies involving five animals per experiment (satellite studies). Experiments of this first phase were exploratory in nature and therefore not conducted in strict accord with GLP regulations. Further, data obtained after i.v. administration in the first phase was used to calculate bioavailability in the main study.
The following major results were obtained from the first phase:
• Urine is the primary route of excretion of radiolabeled compounds derived from ZPT following administration of ZPT to female rats by intravenous injection, dermal exposure, oral gavage or in fortified feed.
• The estimated elimination half-life of pyrithione, the bioactive, organic moiety of ZPT, after intravenous administration of ZPT is about 2 hrs.
• Dietary exposure to feed containing 1 or 10 ppm of ZPT (actual consumption of 0.03−0.4 mg/kg), produced pyrithione concentrations that were either near or below the lower limit of quantitation of 0.5 ng/mL. Exposure to feed containing 50 or 250 ppm of ZPT (actual consumption of 0.7−2.7 mg/kg) produced measurable pyrithione concentrations in plasma for ca. 12 h. Repeated exposure for 10 days to feed containing 250 ppm of ZPT resulted in weight loss and decreased hind-limb muscle mass.
• Dermal exposure to ZPT at levels of 10 mg/kg or greater resulted in plasma pyrithione concentrations that were measurable (>0.5 ng/mL). A lag time to reach these concentrations of about 8−12 hrs from exposure initiation was observed. Decreases in both hind-limb muscle mass and muscle tone were observed in animals exposed daily for 10 days to 100 mg/kg ZPT.
• Bioavailabilities of ZPT administered either by oral gavage or dietary via ZPT-fortified feed were greater than 70%. The bioavailability of dermally applied ZPT was estimated to be about 3−8%. Since concentrations of pyrithione in plasma were high enough to measure in only a few samples and only at the later time points, the estimates of dermal bioavailability should be considered only to be crude estimates of bioavailability.
• ZPT-derived compounds do not appear to be concentrated in any tissue/organ examined.
• Concentrations of total radiolabel in blood were higher and much more persistent in blood than concentrations of pyrithione in plasma. This is consistent with previous studies that identified pyrithione metabolites as the main constituents in serum.

2) Toxicokinetics of ZPT – main repeat dose studies

Oral gavage study:

Guideline:	not mentioned, but experiments were in accordance with OECD TG 417
Species/strain/sex:	Rat (CD), female
Group size:	designed to obtain data from 6 animals per treatment group[*)]
Test substance:	a) non-labelled: ZPT powder from Arch Chem lot #0108244691, Master Log I.D. # 74-01B10CuZPT; purity 98,3 % b) radiolabelled ZPT from Perkin Elmer, lot #3547-059, specific activity 11.0 mCi/mmol, radiochemical purity 97.3 %
Dose levels:	1.25, 3 and 10 mg/kg/d (3 treatment groups per dose)

Controls:	vehicle control (0 mg/kg/d) (3 treatment groups)
Vehicle:	suspensions of ZPT in appropriate volumes of 0.1% aqueous triethanolamine lauryl sulfate
Exposure:	gavage, volume: 5 ml/kg Treatment groups A and B: 10 daily doses; treatment group C: 3 daily doses
GLP statement:	Yes
Date:	2005- 2007

*) this could not always be achieved, therefore the study plan was amended and sometimes only data from 5 animals were available

Treatment group A consisted of animals that had a jugular cannula installed to facilitate the collection of serial blood samples. These animals were dosed daily for 10 days (9 days for the highest oral gavage group), and 8−9 serial blood samples were taken over 24 hr following the first dose and following the last dose of ZPT. Additional blood samples were taken 48 and 96 hr following the last dose of ZPT, at which time the animals were sacrificed. Blood volume in these animals was maintained by injections of plasma taken from donor animals. Serial blood samples were not taken from groups B and C. Animals of group B were sacrificed 24 hr following nine daily doses of ZPT while the animals in group C were sacrificed 24 hr following three daily doses of ZPT. Animals were housed individually in glass metabolism chambers equipped for separate collection of urine and faeces. Excreta were collected daily from animals in all treatment groups, necropsies were performed at sacrifice. Biological samples were assayed for radioactivity by liquid scintillation spectrometry. Pyrithione (PT) was measured in plasma samples using LC/MS. Several functional assessments were conducted prior to sacrifice to ascertain the extent of hind-limb weakness in the animals. These measurements consisted of "muscle mass" and "muscle tone". Necropsies were performed on each animal following sacrifice, with specific tissues being collected for radiochemical analysis. Pharmacokinetic analyses were conducted using WinNonlin.

Results

Concentrations of PT in plasma reached maximal levels within 0.4−0.9 hrs of ZPT administration. PT concentrations then declined with mean half-lives that ranged from 2.6 to 6.9 hrs with no difference between half-lives of the first dose and the last dose at any dose level. Estimates of PT half-life in plasma were considerably shorter than those for total radioactivity in blood (40−60 h). Maximal concentrations of PT increased in a dose-related manner following both the first and the last dose of ZPT.

Systemic exposures (expressed as AUC) also increased in a dose-related manner following the first and the last dose of ZPT as administered dose increased from 1.25 to 10 mg/kg. Ratios of study day 10 (study day 9 for the highest dose group) to study day 1 for both Cmax and AUC were less than 3 at all doses, suggesting that accumulation of pyrithione was minimal after 9−10 days of oral administration to rats.

The majorities of the oral gavage doses were excreted in urine at all dose levels with more than half of this occurring within 24 hrs of dosing. Overall, urine accounted for about 73% of the administered dose, and excretion in urine (as a percentage of dose) was independent of dose level or length of dosing.

Excretion of total radioactivity in faeces was much lower, accounting for 5−10% of the dose. For animals sacrificed 96 hrs after their final dose of ZPT, only about 1% of the total dose remained in the carcass at sacrifice. Much higher percentages of the dose (up to 17% of the highest dose) were still in the carcasses of animals sacrificed 24 hrs following three daily doses of ZPT. Highest concentrations of radioactivity were found in livers, followed by kidneys.

In general, animals administered the vehicle control and the two lower doses maintained or slightly increased body weight throughout the study. Animals at the highest dose lost approximately 15% of their study day 1 body weight at the end of nine days of dosing. Muscle mass and muscle tone decreased with the length of dosing and with increasing dose

levels of ZPT. Reductions of both muscle mass and muscle tone were also found in animals in the vehicle control Treatment Group, in which serial blood samples were drawn.

Oral dietary study:

Guideline:	not mentioned, but experiments were in accordance with OECD TG 417
Species/strain/sex:	Rat (CD), female
Group size:	designed to obtain data from 6 animals per treatment group*)
Test substance:	a) non-labelled: ZPT powder from Arch Chem lot #0108244691, Master Log I.D. # 74-01B10CuZPT; purity 98,3 % b) radiolabelled ZPT from Perkin Elmer, lot #3547-059, specific activity 11.0 mCi/mmol, radiochemical purity 97.3 %
Dose level:	250 ppm [^{14}C] ZPT in feed (3 treatment groups)
Controls:	vehicle control (feed meal without ZPT) (3 treatment groups)
Vehicle:	suspensions of ZPT in appropriate volumes of 0.1% aqueous triethanolamine lauryl sulfate
Exposure:	oral via diet; treatment groups A and B: 10 daily doses; treatment group C: 3 daily doses
GLP statement:	Yes
Date:	2005- 2007

*) this could not always be achieved, therefore the study plan was amended and sometimes only data from 5 animals were available

Dietary studies involved daily doses of ZPT in feed meal (target concentration 250 ppm) or of feed meal alone (vehicle control) for periods up to 10 days. Animals were placed in one of three treatment groups (A, B and C) for the ZPT feed and vehicle control, respectively. Each animal in the A treatment groups had a cannula implanted into a jugular vein for serial blood sampling. Serial blood samples were removed from these animals at specified times following access to dosed feed on study day 1 and study day 10. Animals in Treatment Groups B and C were sacrificed at intermediate time points. Group C animals were sacrificed on study day 4 and group B animals were sacrificed on study day 10, 24 hrs following their final exposure to ZPT-fortified feed. Excreta were collected daily from animals sacrificed at the intermediate time points.
Blood, urine, faeces, and tissue samples were analysed for total radioactivity, 12 urine samples and 12 faeces samples, were chosen at random from animals in the vehicle control treatment groups for analysis. Tissues from animals administered ZPT and from two animals in each of the vehicle control treatment groups were analysed. Pyrithione was measured in plasma, prepared from selected blood samples.
Several functional assessments were conducted prior to sacrifice to ascertain the extent of hind-limb weakness in the animals. These measurements consisted of "muscle mass" and "muscle tone". Necropsies were performed on each animal following sacrifice, with specific tissues being collected for radiochemical analysis. Pharmacokinetic analyses were conducted using WinNonlin.

Results
Animals given ZPT-fortified feed for 9−10 days lost an average of 19% of their study day 1 body weights. Concentrations of PT reached maximal levels within 2.4−4.1 hrs of introduction of ZPT-fortified feed. Following C_{max}, plasma levels of PT declined with a half-life that ranged from 3.7 (study day 1) to 11.0 hrs (study day 10). Both PT C_{max} and AUC increased significantly from study day 1 to study day 10 as would be expected for increased ZPT intake on study day 10. When C_{max} and AUC were normalised for daily dose, neither parameter showed large changes from study day 1 to study day 10. Thus the data suggest that accumulation of pyrithione was minimal after 10 days of ZPT administration to rats via dosed feed.

Excretion in urine accounted for 84−92% of the ingested ZPT. Animals exposed to 250 ppm ZPT in their diets for 9−10 days exhibited slightly to greatly reduced muscle mass and moderate to no muscle tone.

Dermal study:

Guideline:	not mentioned, but experiments were in accordance with OECD TG 417
Species/strain/sex:	Rat (CD), female
Group size:	designed to obtain data from 6 animals per treatment group
Test substance:	a) non-labelled: ZPT powder from Arch Chem lot #0108244691, Master Log I.D. # 74-01B10CuZPT; purity 98,3 % b) radiolabelled ZPT from Perkin Elmer, lot #3547-059, specific activity 11.0 mCi/mmol, radiochemical purity 97.3 %
Dose level:	10, 30 and 100 mg/kg bw/d ZPT (3 treatment groups)
Controls:	vehicle control (0 ppm) (3 treatment groups)
Vehicle:	suspensions of ZPT in appropriate volumes of 0.1% aqueous triethanolamine lauryl sulfate
Exposure:	dermal; treatment groups PK and 10-day intermediate sacrifice group: 10 daily doses; 4-day intermediate sacrifice group: 3 daily doses
GLP statement:	Yes
Date:	2005- 2007

A dose area of 12 cm^2 was clipped free of fur in order to apply the dose to bare skin; rats were fitted with rodent jackets in order to prevent grooming, an appliance was created to surround the application site. Calculated daily target doses of 10, 30 and 100 mg/kg bw/d based on the actual body weight were applied via syringe and a non-occlusive cover was attached to the top of the appliance which was then secured by a metal screen in order to prevent removal of the appliance. Six hours after application of the dermal doses, the cover was removed and treatment sites were washed. Since the dose sites were uncovered except for the 6-h period each day when the animals were being exposed dermally to ZPT, oral ingestion of any small amounts of ZPT remaining on the surface of the skin during the interval between dermal exposures cannot be ruled out.
The animals of the PK group were dosed daily for 10 days and 8 serial blood samples were taken over 24 hrs following the beginning of the first dermal exposure, on study days 2, 3, 4 und 5 and up to 96 hrs following the last 6 hr exposure to ZPT. 96 hrs following the last exposure to ZPT the animals were sacrificed. Blood volume in these animals was maintained by injections of plasma taken from donor animals. Serial blood samples were not taken from the other two exposure groups. Animals of the 10-day intermediate sacrifice group were sacrificed 24 hrs following nine daily doses of ZPT while the animals of the 4-day intermediate sacrifice dose were sacrificed 24 hrs following three daily doses of ZPT. Animals were housed individually in glass metabolism chambers equipped for separate collection of urine and faeces. Excreta were collected daily from animals in all treatment groups, necropsies were performed at sacrifice. Biological samples were assayed for radioactivity by liquid scintillation spectrometry. Pyrithione (PT) was measured in plasma samples using LC/MS. Several functional assessments were conducted prior to sacrifice to ascertain the extent of hind-limb weakness in the animals. These measurements consisted of "muscle mass" and "muscle tone". Necropsies were performed on each animal following sacrifice, with specific tissues being collected for radiochemical analysis.
Approximately 24 hrs following application of the last dose, the dose sites were washed again (2% soap in water followed by deionized water) and dried. The dose site was then left uncovered for the remainder of the study. Unabsorbed dose (total radioactivity) recovered from the collections of the protective appliances, cloth coverings, and gauzes were measured. Necropsies were performed on each animal following sacrifice, with specific

tissues being collected for radiochemical analysis. Pharmacokinetic analyses were conducted using WinNonlin.

Results
Radioactivity was only slowly absorbed following dermal exposure. Concentrations of pyrithione reached maximal levels after 12 −26 hrs of administration of dermal ZPT doses. On study day 1, pyrithione concentrations were too low to measure at the time points shorter than 8 hrs; thus, the lag time required for transiting skin is longer than the 6-hr exposure period. High variability in study day 1 pyrithione Cmax and AUC point to variability in the penetration of ZPT. Study day 10 AUC increased in a roughly proportional manner between rats receiving 10 and 30 mg/kg daily. However, between 30 and 100 mg/kg dose groups, study day 10 AUCs were similar. This suggests that maximal absorption of ZPT and therefore pyrithione exposure was achieved at 30 mg/kg.
Because of variability caused by apparent delayed absorption of ZPT estimates of tmax and parameters based on on k_{el} ($t_{1/2}$, AUC(0-∞), Vd/F and CL/F) should be used with caution.
Bioavailabilities of the dermal doses can be calculated from pyrithione concentrations in plasma (0−24 hr) using data from the pilot study for the intravenous dose (concentrations of pyrithione in plasma following intravenous administration of ZPT were measurable for less than 24 hrs). These calculated bioavailabilities are 2.3, 8.6, and 0.3%, respectively, for the 10, 30, and 100 mg/kg/day exposures. However, the AUC (0−24hrs) values for dermal administration do not capture the total AUC, and use of the AUC (0-∞) values are fraught with the difficulties described above for parameters based on kel. Only small proportions of the administered doses were excreted in urine (1−7% of the total dose for animals kept 4 days following the last dose of [^{14}C]ZPT), and even smaller proportions were excreted in faeces. The potential exists that even these small proportions are inflated due to contamination by residual dose present in the dose site.
Animals exposed to the lowest dose of ZPT excreted the largest percentages of the dose in urine and faeces. The lowest percentages of the dose were excreted by animals exposed to the highest doses of ZPT. As opposed to urinary excretion following oral gavage administration of ZPT, the rate of urinary excretion of total radioactivity following dermal administration did not decrease during the four days following the last dermal exposure. However, based on high variabilities in AUC and C_{max} values, no conclusion on probable accumulation after repeat dose dermal administration can be made from that study.
For animals examined 96 hrs after their final dose of ZPT, about 1−3% of the total dose remained in the carcass at sacrifice, over half of which was in the dose site skin. Somewhat larger percentages of the dose (up to 9% of the lowest dose) were still in the carcasses of animals sacrificed 24 hrs following their last dermal exposure to ZPT. The largest contributor to the residual radioactivity, in general, was the dose site skin and the second highest concentrations were in the liver.
In general, body weights decreased during the first few days of the study for animals in all dermal treatment groups, including the vehicle control groups. Weights then remained relatively constant during the period of daily dermal exposure. Hind-limb muscle mass and muscle tone both decreased with increasing exposure to ZPT. Reductions in muscle mass and muscle tone were already observed at the lowest dose testes, which could therefore be considered as LOAEL. For the animals exposed to 100 mg/kg of ZPT for 10 days, muscle mass was greatly reduced with low to no muscle tone.

SCCS comment
The SCCS notes that different absorption values have been reported in the manuscript on the dermal PBPK model which has been submitted after the commenting round (table 4 of the manuscript). Apparently the same study yielded observed absorption values of 9.0, 5.2 and 1.3 % for dermal doses of 10, 30 and 10 mg/kg bw/d ZPT in contrast to 2.3, 8.6, and 0.3% for dermal doses of 10, 30 and 10 mg/kg bw/d ZPT as described above. As it is also mentioned above, that values should be taken with caution, an explanation of the difference in dermal absorption values should be provided.

Inhalation Study

Guideline:	not mentioned, but mainly in accordance with OECD TG 417
Species/strain/sex:	Rat / Sprague-Dawley (Crl:CD) / female
Group size:	5 per sex /dose*)
Controls:	5 females exposed to filtered air only*)
Test substance:	a) non labelled: Zinc Omadine ®
Batch:	0108244691
Purity:	98.3 %
	b) Zinc pyrithione, [pyridine ring-2,6-^{14}C]-CUSC70360000MC; Lot 3620139
	60.0 mCi (2.22 GBq; 9.81 mCi/mmol (362.97 MBq/mmol)
Dose levels:	target concentrations: 0.5 and 1.5 mg/m^3
	Mean actual concentrations: 0.51, 1.6 mg/m^3
	(corresponding to calculated doses of 0.033 and 0.102 mg)
Exposure:	inhalation, nose only, 6hrs/d
	0, 4 or 9 exposures to non-labelled material, followed by 1 exposure to Radioactive labelled material
GLP statement:	yes
Date:	2009/2010

The objective of the study was to determine the toxicokinetic profile of [^{14}C]-ZPT in rats following a single nose-only inhalation exposure following 0, 4, or 9 prior nose-only inhalation exposures to non-labelled ZPT (1, 5, and 10 total exposures).
The test substance was administered as 0, 4, or 9 6-hr nose-only inhalation exposures to non-labelled ZPT, followed by one 6-hr exposure to the radio-labelled ZPT on exposure days 1, 5, or 10. After the respective exposures animals were placed in metabolism cages for collection of urine and faeces. Plasma was obtained from 0.25, 0.5, and 1-hr post-exposure timepoints. All animals were euthanised at 24 hours post-exposure and blood, lung, liver, kidney, spleen, brain, stomach, gastrointestinal (GI) tract, and carcass were collected and analysed using liquid scintillation counting.
Results:
Mean Plasma concentrations are given in table 8:

Table 8: mean plasma concentrations of [^{14}C]-ZPT derived radioactivity after inhalation exposure.

Dose [mg/m^3]	1 exposure		5 exposures		10 exposures	
	C_{max} [µg/g ±SD]	t_{max} [hr]	C_{max} [µg/g ±SD]	t_{max} [hr]	C_{max} [µg/g ±SD]	t_{max} [hr]
0.5	0.027 ± 0.008	0.5	0.035 ± 0.013	1	0.029 ± 0.012	1
1.5	0.233 ± 0.149	0.5	0.124 ± 0.036	1	0.110 ± 0.050	0.5

Among the tissues examined, the greatest amount was found in the liver, followed by the GI tract, kidney, stomach, lung, brain, and spleen (as a percent of the calculated dose).
The mean carcass concentration corresponded to approximately 33 % and 41 % of the calculated dose at the low and high dose concentration, respectively.
For the low dose, urinary excretion of ^{14}C-ZPT-derived radioactivity accounted for approximately 43 %, 58 %, and 53 % of the total dose in animals receiving 1, 5, or 10

exposures, respectively. At the high dose, urinary excretion accounted for 76 %, 48 %, and 44 % of the dose in animals receiving 1, 5, or 10 exposures, respectively. Total fecal excretion of ^{14}C-ZPT-derived radioactivity accounted for <6 % of the total dose regardless of exposure concentration or the total number of exposures. Mean recovery of total radioactivity in the excreta, tissues, and carcass was approximately 93 % of the mean calculated dose at 0.5 mg/m^3 and 106% of the mean calculated dose at 1.5 mg/m^3.
The study demonstrates, that appreciable amounts of ZPT become systemically available when administered by the inhalation pathway.

Ref.: E29

3.4.9.4. PBPK modelling

Toxicokinetic data has been used to build up physiologically-based-pharmacokinetic (PBPK) models for ZPT.

3.4.9.4.1 Oral model
At first a preliminary model has been developed based on data mainly obtained from rabbit studies. The development of this model and its informative value has been described and discussed in the document, "Feasibility Study for a Physiologically-based Pharmacokinetics Model of Zinc Pyrithione (August 2003) which was provided by the applicant (Ref. A23).
Afterwards, a preliminary rat model was developed by using data obtained from Wedig et al., 1978 (Ref.: A26), from which a first rat model was successfully parameterized and optimized (no data have been provided for the preliminary and the first rat model). The first rat model failed to predict the multi-dose kinetics of pyrithione carbon (PTC) as observed in the experiments described above (Ref. A22 / Annex I of submission II) and could not simulate the kinetics of pyrithione, as parameters for PT metabolism were not developed for the first rat model. Therefore, in addition to data already used for the older models, data from the experiments described above (Ref. A22 / Annex I of submission II) were used to parameterize and calibrate the second rat model (Ref. A24 / Annex I of submission II).

Data used
Data from reports compiled in a Procter and Gamble document "Zinc pyrithione Studies of Absorption, Distribution, Metabolism, and Excretion" prepared by J.F. Nash, 2004 (the document is not available to SCCS, extracted data were given in tabulated form in Annex I of submission II) formed the basis for the second rat model. Data on tissue masses, tissue volumes and blood and plasma flows were from ILSI (1994). Data on tissue:plasma partition coefficients and on plasma – red blood cell mass transfers mainly based on Wedig et al., 1978 (Ref. A26).
Data used for model calibration were (1) plasma PT concentrations for the first 24 hours following the first dose of ZPT (1.25 mg/kg) from Ref. A22 /Annex II of submission II; (2) blood PTC concentrations for the first 24 hours following the first dose of ZPT (1.25 mg/kg) from Ref. A22 /Annex II of submission II; (3) relative abundance of 2-(methyl)sulfonyl pyridine (MSP) and S-glutathione (SG) in blood at 1, 4, and 16 hours following a single oral dose of ZPT (1 mg/kg), from Gibson and Turan (1978) (this study is an internal Procter and Gamble report. Data utilized from this study were given in tabulated form in Ref. A24); and (4) cumulative urinary and fecal excretion (% of dose) of ^{35}S (pyrithione sulfur, PTS) up to 72 hours after a single oral dose of ZPT (25 mg/kg) from Ziller et al. (1977) (Ref. B74). Data used for calibration, evaluation and parameter estimation were from Ref. A22 /Annex II of submission II, from the Gibson and Turan (1978) study, from Ref. B69 and from Ref. B74.
All data were compiled in tabular form. Models were constructed and implemented in Advanced Continuous Simulation Language (acslXtreme, v. 2.0.1.6). Parameter estimation was performed in acslXtreme Optimum using the Nelder-Mead and/or conjugate gradient algorithms, set to minimize relative error for each parameter. No attempt was made to weigh observations for measurement error or uncertainty. Goodness of fit was judged by

visual inspection of model predictions compared to observations. Univariate sensitivity analysis consisted of running the model after perturbing values for single parameters by a factor of 0.01, in the up and down directions.

PBPK model Structure

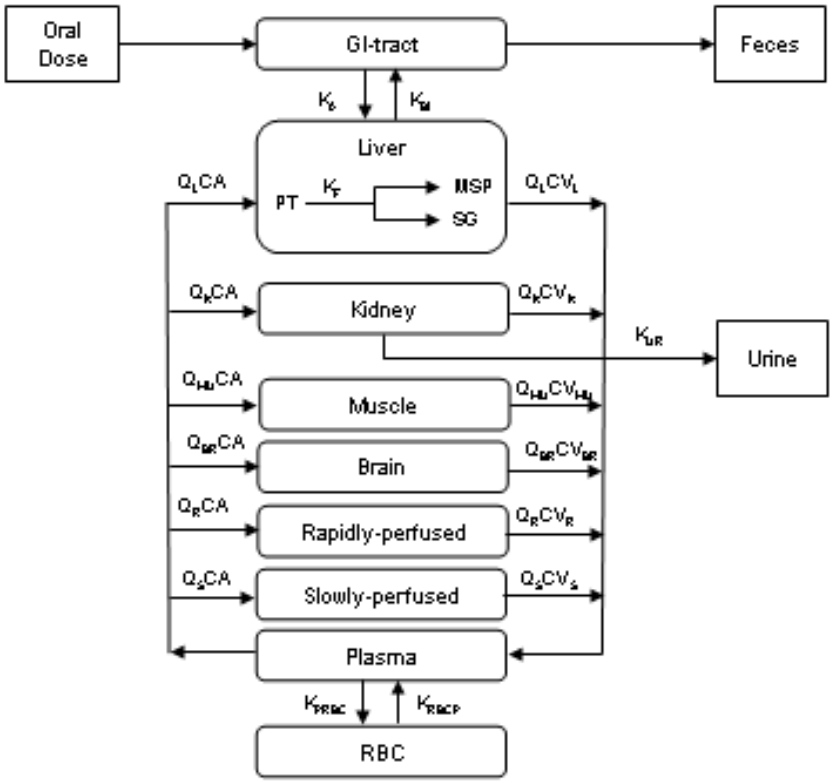
The model structure is depicted in Figure 2. The model simulates the kinetics of absorption, distribution, and excretion of PT and products of its two major metabolic pathways, the 2-(methylsulfonyl)pyridine (MSP)-pathway and the S-glucuronide (SG)-pathway. The central distributing compartment is plasma. Gastrointestinal tract (GI-tract) absorption processes for PT and metabolites are simulated as first-order kinetic processes of unlimited capacity. These are represented with absorption (i.e., bioavailable) fractions and absorption rate constants, with mass flow from the GI-tract to liver, conceptually representing the hepatic portal flow. Seven tissue compartments are represented: kidney, liver, brain, skeletal muscle, other rapidly-perfused tissues (tissues other than liver and kidney that have a relatively high-blood flow/g tissue; e.g., heart, viscera), slowly-perfused tissue (e.g., adipose, skeletal muscle, skin), and red blood cells (RBC). Transfers of PT and metabolites between plasma and tissues other than RBC are assumed to be sufficiently rapid to produce a steady state within the time of blood flow from the arterial to venous vasculature of each tissue (i.e., flow-limited). Transfers between plasma and RBC are represented as first-order processes of unlimited capacity. Metabolism is attributed to the liver compartment and is represented as distinct first-order (unlimited capacity) processes for conversion of PT to MSP and SG. Two excretory pathways are simulated: first-order transfer from kidney to urine, and first-order transfer from liver to GI-tract, the latter representing biliary secretion; both pathways have unlimited capacity. Distinct absorption parameters for PT and metabolites provide a means of simulating reabsorption of chemical secreted in bile. The above specification of the model does not include simulations of binding or sequestration of PTC in tissues. The model is implemented as a series of first-order differential equations.

SCCS comment

The SCCS notes, that skeletal muscle is assigned to slowly-perfused tissue but at the same time as an individual compartment.

The SCCS notes that the applicant has considered this issue. In the dermal model, skeletal muscle has been pulled out as an individual compartment and was considered part of slowly perfused tissues.

Figure 2: Structure of the oral PBPK model for ZPT



Model calibration

In the model calibration phase, sequential optimisations were performed in order to achieve a good representation of the data obtained from the studies submitted in Annex II of submission II (Ref. A22, described in 3.3.9.3). The adequacy of the outcome of the optimisations was assessed by visual inspection of model simulations against the observations from the multi-dose oral phase II study (Ref. A22).

Results

Comparisons between simulated and observed values were performed. It should be noted that partly overlapping data have been used to build up the model and to optimise and check the model.

A comparison between observed and simulated kinetics was performed with respect to the following aspects:

- kinetics of plasma PT
- kinetics of blood pyrithione carbon
- kinetics of urinary and fecal excretion of pyrithione carbon
- kinetics of tissue pyrithione carbon
- kinetics of metabolites in blood

The comparisons demonstrated that the model quite reasonably replicates the observed short-term elimination kinetics of PT and pyrithione carbon following single doses and longer-term temporal patterns of blood concentrations of pyrithione carbon during repeated dosing schedules. The model also accounts for the production and rapid elimination of SG, the major metabolite of PT in urine, as well as production and slower elimination of the minor metabolite, MSP. The latter is the major pyrithione carbon species in blood within several hours following a dose of ZPT.

It was stated that physiological parameters and most chemical parameters that are highly influential in simulation of blood and tissue pyrithione carbon levels are scaled to body weight and could therefore be scaled to represent different species.
In a submitted poster (Ref. A25) it is stated that the rat model has been allometrically scaled to humans.

By using the rat model allometrically scaled to humans, the applicant provided a calculated external human concentration which would correspond to that AUC in the rat, which is obtained from an oral dose of 500 µg/kg/d. This human equivalent external dose was 2170 µg/kg/d (or 2260 µg/kg/d - two slightly differing values are given in reference A 25; in the supplemental submission the applicant gives the value of 2170 µg/kg/d as the human equivalent external dose).
The applicant states in his supplemental submission:
(1) "The predictive validity of the ZPT PBPK model was verified in repeat dose pharmacokinetic studies conducted in female rats. Toxicologically-relevant doses of ^{14}C-ZPT were administered by gavage, in the diet, topically, or intravenously. Radiolabel (PTC from ^{14}C-ZPT) and parent (PT) concentrations were measured in blood and plasma, respectively, over the 14-day study (10-day dosing + 4-day recovery). These experimental observations confirmed the predictive validity of the ZPT PBPK model. Furthermore, these data provided additional support to estimates of the internal dose of ZPT in humans based on measures of cumulative urinary radioactivity following use of shampoos containing 2% ^{14}C-ZPT thereby linking PBPK estimates to the data obtained in human subjects."
(2) "In summary, the PBPK model, verified by the rat pharmacokinetic studies, and human clinical study results provide new evidence supporting the human safety of 2% ZPT used in anti-dandruff shampoos."

SCCS comments on the oral PBPK model for ZPT
No information has been given how the rat model was allometrically scaled to humans (i.e. which parameters have been used to scale the model to humans in terms of e.g. partition coefficients, organ masses and volumes) and whether and to which extent species differences (e.g. with respect to enterohepatic circulation, with respect to metabolic parameters) have been considered when scaling the rat models to humans. Further, no description has been provided how the rat model had been verified by human clinical study results.

Therefore, it cannot be reproduced, whether the human external dose of ZPT equivalents, that would correspond to the rat NOAEL of 500 µg/kg/d (2260 and 2170 µg/kg/d), are sound and reliable values to base MoS calculation on.
Thus, solely from the information available from the SOT 2008 poster (Ref. A25) and the statements in the submission (see citations (1) and (2) above) the human model cannot be used for deriving quantitative threshold values, as there is no transparent information on the human model and its validation.
This is in line with the REACH guidance on information requirements and chemical safety assessment, Chapter R-8, where it is stated: "Furthermore, if a PBPK model is used to extrapolate from animals to humans, the proposed model should be validated by data from humans if these are available, and extrapolations from the model should be within or close to the range of experimental measurements used to validate the model. If there is no validation of the model by data from humans, PBPK models may be used to support an interpretation of toxicodynamic data or toxicological findings rather than as a basis for the derivation of a DN(M)EL."

Ref.: D24

The SCCS acknowledges that PBPK modelling is a straightforward way to perform various types of extrapolation in risk assessment and that it might assist in reducing uncertainties associated with conventional extrapolation procedures. However, when the model is not sufficiently documented, confidence in the model is not given.

This means in case of submission II for ZPT that without more detailed information especially with respect to the oral human model the data cannot be accepted for the assessment of ZPT and for the derivation of a safe exposure level.

The SCCS notes that the applicant has responded after the commenting period that standard scaling factors had been used in order to scale the rat model to the human model. Reference has been given to the parameters listed in the manuscript for the dermal ZPT model. This does not change the fact that the oral model had not been validated against human data.

The SCCS notes that manuscripts concerning oral PBPK modelling of ZPT submitted to the SCCS have not yet been published in a peer-reviewed scientific journal although it was stated that they had been submitted.

The SCCS notes that after the commenting period in 2013 the applicant has submitted a modified manuscript for the oral rat PBPK model, where the allometric scaling to human has been eliminated. In order to address toxicokinetics in humans, the applicant has submitted a manuscript of dermal PBPK modelling of ZPT.

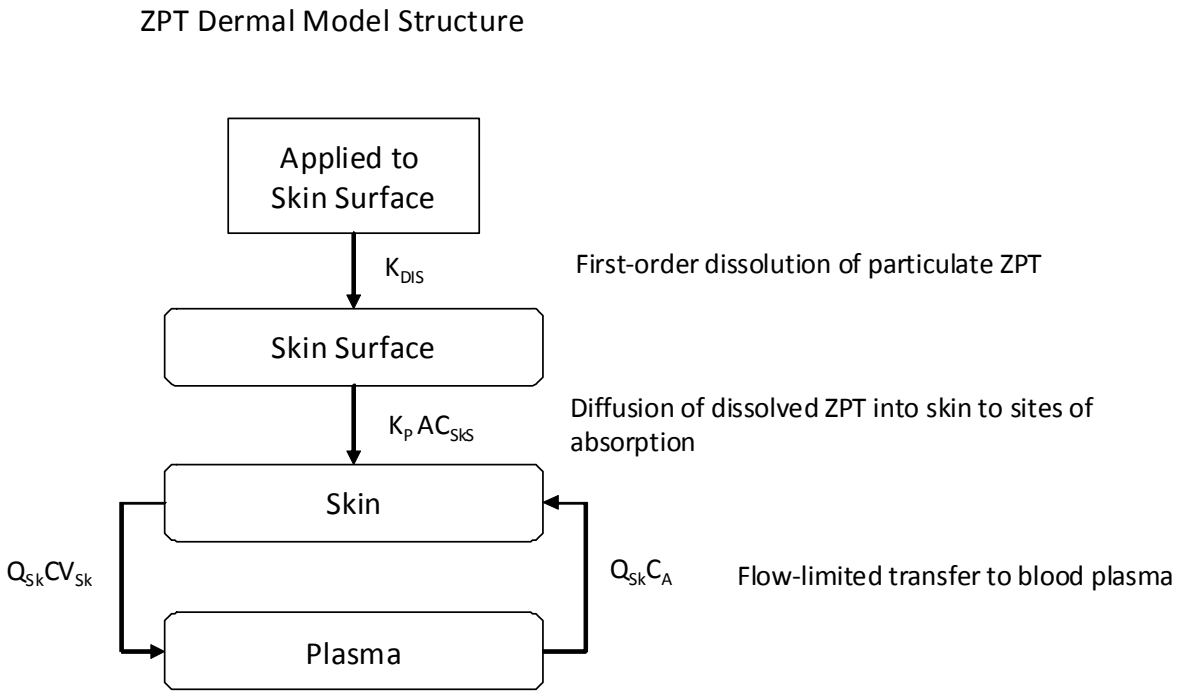
3.4.9.4.2 Dermal model
Data used:
Data from the following studies were used to create the dermal model for ZPT: (1) an in vivo dermal repeat dose study using 0, 10, 30 or 100 mg/kg bw/d ZPT (apparently the same study as that described in section 3.4.9.3); (2) a human repeat-dose study using an antidandruff formulation containing 1 % ZPT and a leave-on tonic containing 0.1 % ZPT (see section 3.4.4.2; Group A applied a 1 % ZPT shampoo formulation on 4 consecutive days, Group B applied a 1% ZPT shampoo formulation, followed by a leave-on tonic containing 0.1 %ZPT), an in vitro skin permeation study using full thickness skin from Sprague-Dawley rats and humans (the study has been submitted after public consultation of SCCS 1512/13 see section 3.4.4.1).
All data used were processed in Excel, data in figures were transcribed into tabular form after digitisation of the figures. Statistical analyses were performed using Statgraphics Centurion XV. Noncompartmental modelling pharmacokinetic analyses were conducted using WinNonlin. The PBPK model was constructed and implemented in Advanced Continuous Simulation Language (acslXtreme). Parameter estimation was performed in acslXtreme Optimum using the Nelder-Mead and/or conjugate gradient algorithms, set to minimize relative error for each parameter. No attempt was made to weigh observations for measurement error or uncertainty. Goodness of fit was judged by visual inspection of model predictions compared to observations.

PBPK model structure
The model structure is depicted in figure 3. The dermal absorption of ZPT is determined by the processes: (1) dissolution of ZPT particulates at the skin surface; (2) fugitive loss of dissolved ZPT (e.g. by skin sloughing, washing, drying); (3) diffusion of dissolved ZPT into skin; (4) exchange of skin ZPT with blood plasma. All the processes involved were described by equations. Dissolution of ZPT particulate was treated as a slow, unidirectional first-order process (KDIS). Dermal penetration of dissolved ZPT from the skin surface is regarded as a bidirectional diffusion process which is governed by partitioning of dissolved ZPT at the skin surface and skin, the dermal permeability coefficient (KPS) and the concentration gradient between skin and skin surface. Fugitive loss of dissolved ZPT from skin surface was treated as a unidirectional first-order process (KFUG). A capacity limitation concerning the absorptive flux was introduced by defining a maximum achievable concentration of bioavailable ZPT (CSSMAX). The skin:skin surface partition coefficient was set equal to the octanol-water partition coefficient, skin:blood partition coefficients for rats and humans were based on the in vitro dermal penetration studies performed. In the absence of

experimentally determined values, values for KPS, KDIS, KFUG and CSSMAX were statistically optimized against the data from the 10 mg/kg bw/d repeated dermal absorption study in rats. Selected combinations of dermal absorption parameters were then optimised to blood ^{14}C data from all three doses of the dermal rat study. Adequacy of the outcome of the optimizations was assessed by visual inspection of model simulations against the observations from the repeat-dose rat study.

Extrapolation to humans
The rat model was allometrically scaled to humans. With respect to scalp doses, simulations were run using the mean ±SE to account for uncertainty in the dose estimate. Optimisations for the dermal permeability coefficient (KPS) and scaling factor for elimination rates (BWSFK), based on the human repeated dose study were explored and found to improve model performance.

Figure 3: Structure of the dermal PBPK model for ZPT



Results:
The rat model replicated the dose-dependency of PTC bioavailability in the study. The model accounted for approximately 98 % of the observed variability in blood PTC. With respect to the AUC of PTC, larger residuals (residual = predicted – observed mean/standard deviation of observed mean) were observed for the highest dose (100 mg/kg/d). The model further reproduced the kinetics of observed urinary excretion of PTC. The model accounted for approximately 85 % of the observed variability in daily urinary PTC excretion. Prediction of PT kinetics in plasma showed less agreement with experimental data (residuals > ± 2), which was explained by the high variability in plasma PT concentration. The dose-dependency of plasma PT, however, was well predicted. Model predictions for PTC in most tissues ranged within a 1 – 3-fold factor of observations. Model predictions for overall absorption (3 – 10 % of the dose) were similar to observations (1 – 9 %). The observed and predicted mass balance for 14[C] in rats following dermal dosing with ZPT is given in table 9 (table 4 from the manuscript) (it should be noted that it is not clear whether the last row should be the dose of 100 mg/kg/d instead of 10, i.e. whether there is a typing error in the table):

Table 9. Observed and Predicted Mass Balance for ^{14}C in Rats Following Dermal Dosing with ZnPT

	Dermal Dose (mg/kg/day)		
Compartment	10	30	10
Urine	6.7 (8.4)	3.4 (2.8)	1.0 (0.84)
Faeces	1.0 (0.72)	0.39 (0.24)	0.15 (0.07)
Systemic Tissues[a]	1.3 (0.61)	1.5 (1.8)	0.12 (2.4)
Absorbed (%)[b]	9.0 (9.7)	5.2 (4.8)	1.3 (3.3)

Observations are shown along with predictions from the model in parentheses. All values are expressed as a percentage of the total applied dose.
[a]All tissues excluding skin at the dosing site.
[b]Excreted + systemic tissues

The human model replicated the observed rate of daily PTC excretion, whereby the performance was better for group A than for group B. Inspection of the simulations revealed that the model is predicting slower elimination kinetics than observed. After optimisation of the allometric scaling factor, better agreement between predicted and calculated values was observed, however, residuals were only modestly improved. As no blood data were available from human studies, the model could not be used to compare predicted with measured plasma data. Calculated AUC values were 0.0093 and 0.032 hr-mg/l for group A and B, respectively with optimized scaling factors. The model predicted absorption of 0.5 % of the dose. The authors discuss that the lower absorption fraction in humans might be due to longer exposure times in the rat study (6 hrs) compared to the 1 minute shampoo exposure applied in humans.

SCCS comments
The SCCS notes that the rat dermal PBPK model quite reasonably reflects internal exposure to PTC whereas internal exposure to PT was predicted with lower performance.

The SCCS notes that observed values for absorption percentages as given in the manuscript on the dermal PBPK model (table 4 from the manuscript, see above) differ from the absorption values given in the study report of the in vivo dermal absorption study described in section 3.4.9.3., although the data base was apparently the same.

The SCCS notes that skeletal muscle was considered as individual, rapidly perfused tissue but was also assigned to slowly perfused tissue.

The SCCS acknowledges that the dermal PBPK model has been allometrically scaled to humans und validated against urinary excretion data from a human study performed with 1 % ZPT. The SCCS notes, that the allometrically scaled human model reasonable reflected the observed urinary excretion when allometric scaling factors were statistically optimized to values of 0.025 (group A) and -0.13 (group B), respectively. These modified factors lack physiological plausibility, and the explanation given by the authors ("one possible explanation for the improved simulation of excretion [remark: by using modified scaling

factors] is that the dominant mechanism for excretion of ^{14}C in urine is renal tubular secretion of SG, which may be more strongly correlated with kidney weight than with body weight." is an unproven hypothesis.

The human dermal model has not been validated for blood PT or blood PTC. Therefore, there is no confidence of the human dermal model in any prediction of the availability of ZPT in blood (i.e. on PTC-AUC) which would be the relevant dose metrics.

Further, the overall confidence in the human dermal model is low as it has not been demonstrated that it reproduces a variety of data from at least more than one experiment and it exhibits further weaknesses when reflecting arguments concerning the credibility of PBPK models which are given in Kohn, 1995.

In this respect, the argumentation given by the applicant in response to SCCS/1512/13 cannot be accepted ("The most important and relevant aspect of the model is shown in the table below. We took the oral rat NOAEL, 500 µg/kg/day, and used the rat PBPK model to predict the corresponding area-under-curve (AUC) of pyrithione (PT) or 14C-PT (PTC, i.e., parent and metabolites) concentration plasma (i.e., internal dose). The predicted plasma AUC (0-∞) values corresponding to the 500 µg/kg/day dose group in the rat are 68.6 hr. · µg/L for PT and 3416 hr. · µg/L for plasma PTC. These values represent the systemic exposure to PT or PTC at the dose corresponding to the NOAEL from the 2 yr oral toxicity study. We then used the PBPK model to predict the "Human Applied Dermal Dose" that would yield the same systemic exposure as the rat oral NOAEL. The resulting ratios of human/rat dose represent the dose equivalence ratio for oral dosing of the rat and dermal dosing of humans. The human dose/rat dose ratio is 0.93 for PT and 2.07 for 14C-PT. Therefore, the PBPK models predict systemic dose equivalence when the human applied dermal dose is approximately equal to or 2-fold higher than the rat oral dose").

The SCCS notes, that the manuscript concerning dermal PBPK modelling of ZPT submitted to the SCCS has not yet been published in a peer-reviewed scientific journal.

As a conclusion, the SCCS acknowledges the efforts undertaken by the applicant in order to build up a PBPK model for dermal administration of ZPT. The SCCS further acknowledges, that the model quite well reflects the observed toxicokinetics of ZPT in the rat. However it is not acceptable for quantitative risk assessment in humans.

Therefore, as experimental data on systemic human exposure from formulations containing 2 % ZPT are available, SCCS gives preference to the experimental human data for risk characterisation.

Ref.: E8

3.4.10. Photo-induced toxicity

3.4.10.1. Phototoxicity / photoirritation and photosensitisation

No data available

3.4.10.2. Phototoxicity / photomutagenicity / photoclastogenicity

No data available

3.4.11. Human data

See section 3.3.3. Skin sensitisation and section 3.3.4.2. *In vivo* dermal absorption

3.4.12. Special investigations

3.4.12.1. Mode of Action and Neurotoxicity studies

Taken from SCCNFP/0671/03

(a) Work by Snyder et al (1977, 1979), Dejesus et al (1978), Chrisman and Ross (1978) and Sahenk and Mendell (1979, 1979a, 1980) has provided considerable information concerning ZPT induced paralysis.
Snyder et al (1977) determined that the hind-limb muscle wasting reported by Dearwester and Johnson (1974) was a disuse atrophy secondary to neurologic effects. Using in situ sciatic nerves Snyder et al (1977) found conduction velocities were normal, but they observed a decrease in the force of muscle contraction. Effects on serum cholinesterase were rejected as a possible cause, since levels were measured and found to be normal. Dejesus et al (1978) in a later study confirmed their finding of normal conduction velocities using the sural nerve, but in addition he found a reduction in the amplitude of the sensory potential and a reduction in duration of the evoked response.
Similar studies by Chrisman and Ross (1978) investigated the clinical and electrophysiologic response to several dose levels of ZPT using rats. The animals were fed diets containing 0, 10, 50, 250, 500, 750 and 1000 ppm ZPT for 12 weeks. At a concentration of 10 ppm in the diet (0.6 mg/kg/day), there were no changes in neurologic signs or electrophysiologic function during the course of the study. However, at 50 ppm in the diet (4.0 mg/kg/day for females and 2.6 mg/kg/d for males) severe neurologic deficits and electrophysiologic abnormalities were noted. Reduction in the electrophysiologic response began after about one week on the diet, and neurologic deficit was grossly apparent about a week later. These changes became progressively more pronounced and were most severe at six weeks. After eight weeks on the diet, the animals began to improve, and some of the rats completely recovered clinically. Animals receiving 250 ppm of ZPT in the diet died prior to termination of the study. All of the rats were severely affected, and no recovery was observed prior to death. Concentration of 500 ppm and greater in the diet produced mild or no neurologic deficit or electrophysiologic changes prior to death.
Milligram/kilogram equivalents have not been provided for dietary concentration of ZPT above 50 ppm because of excessive body weight loss and extreme variability in the amount of chow eaten.
(b) it seems likely that a critical systemic level of ZPT or a metabolite must be attained and maintained for a sufficient period of time to produce paralysis. Gibson (1979) has shown that animals partially recover over the weekend when they are dosed 5 days/wk. Intermittent reduced food consumption or food avoidance on some days could produce a similar effect that in turn affects the systemic level.
(c) During the course of many studies, the animals failed to gain weight, and some even lost weight. This was due to reduced food consumption, either because of progressing paralysis (part of this weight loss was due to a significant decrease in food consumption, since the animals had difficulty reaching their food) or because of reduced palatability of chow with higher levels of ZPT added.

Further data

In vivo studies

Acute in vivo neurotoxicity study

Guideline:	OECD 424
Species/strain/sex:	Rat / Crl:CD® (SD)IGS BR VAF/Plus ®
Group size:	10 per sex / dose
Controls:	10 per sex, 0 mg test substance in vehicle
Test substance:	Zinc Pyridinedione (ZPT) – off-white to tan powder
Batch:	01008244691

Purity:	98.3 %
Vehicle:	reverse osmosis membrane deionized water
Dose levels:	25, 75 and 150 mg/kg bw/d
Exposure:	oral, gavage, one application
GLP statement:	yes
Date:	2004 - 2005

Animals received one oral (gavage) administration at the indicated dose levels. Checks for viability were made twice daily. Clinical observations, general appearance and body weights were recorded before and during the study. FOB evaluations, which included detailed clinical observations, and motor activity evaluations were conducted before the day of dosage, on the day of dosage (DS 1), seven and 14 days after dosage. After FOB evaluations, muscle tone and mass of the calf muscles were assessed.
Surviving rats were sacrificed on DS 16. A gross necropsy of the thoracic, abdominal and pelvic viscera was performed. The calvaria were removed and the head immersed in fixative. After an interval of at least 24 hours, the brain was removed from the skull and weighed.
Five rats per sex per group were selected for neurohistological examination. The tissues from rats selected in the control and high concentration groups were dissected further and appropriate tissues were processed and retained.
Results
Two females at 75 mg/kg and one male and six females at 150 mg/kg did not survive to scheduled sacrifice. The male rat and two of the six female rats in the 150 mg/kg dosage group were injured during dosage administration and the deaths were not considered test substance-related. The number of female rats found dead in the 150 mg/kg dosage group was significantly increased, as compared to the vehicle control group. No test substance-related alterations were noted in the central or peripheral nervous system tissues, eyes with retinas and optic nerves, or skeletal muscle. There were no microscopic changes in the gross lesions observed in this study that were considered to be the result of test substance administration. The changes were typical of those that occur as a result of inadvertent dosing accidents or spontaneously as incidental findings in laboratory rats of this age and strain.

Ref.: E30

A study was performed in order to investigate the electrophysiological correlates of ZPT-induced neuromuscular dysfunction in rats after oral (dietary) intake of ZPT. Male Fischer F344 rats (6-8 animals/group) received diet containing 50 ppm ZPT of unspecified impurity (equivalent to approximately 2.5 mg/kg/d) for 14 d, followed by a 42 d recovery period. Control animals received diet without ZPT. A further group of animals received diet without ZPT but at reduced levels, in order to determine whether reduced food intake could account for the observed neuromuscular deficit. Electrophysiological, observational and direct measurements were used to monitor the progress of changes in neuromuscular function. No toxicologically significant alterations in food consumption were observed during the study; body weights were reduced (by 13%) in animals receiving the reduced diet.
In ZPT-treated animals, mean hind limb strength was decreased by approximately 40 % from day 8 of dosing to day 8 of the recovery period. Forelimb strength was reduced by approximately 25 % from day 12 to day 4 of the recovery period. Electrophysiological changes, consistent with the observed decreases in hind limb function were also noted.
Electrophysiological alterations (M wave changes) remained evident for 42 d of the recovery period, although hind-limb functionality returned after 8 d. There were no apparent changes in nerve conduction velocity following ZPT treatment. There was no apparent neuromuscular deficit in the group receiving reduced amount of food. The data indicate that the earliest neuromuscular deficits after oral ZPT administration are observed in hind limbs. Animals

regained hind-limb function following removal of treatment, although electrophysiological dysfunction remained detectable.

Ref.: D21

2 studies already mentioned in section 3.3.5.1 (data summarized from MAK 2012)

After dermal application of 100 mg ZPT for 10 days to Sprague-Dawley rats, all of 5 treated animals showed reduced amplitude of the evoked compound muscle action potential (CMAP), 4 animals showed signs of a reduced muscle tone.

In a 28-day dermal neurotoxicity study groups of 5 Sprague-Dawley rats received daily dermal doses of 0, 50, 150 and 200 mg ZPT/kg/d (male animals) or 0, 10, 25, 50, 75 and 100 mg ZPT/kg/d (female animals). The vehicle was 0.1 % triethanolamine-lauryl sulphate, the treatment site was protected by a fixed convex piece of plasic shielding. Low muscle tone was observed at 150 and 200 mg/kg/d in male animals beginning on day 8 and day 11 that continued throughout the study duration. Hindlimb and forelimb grip strength as well as muscle tone and body weight were decreased in male animals of the two highest doses on days 14 and 28. No significant changes in plasma, RBC or brain cholinesterase was observed at any dose tested for any time point measured. Decreases in the electrophysiological measurements measured as the maximum amplitude were observed in males at 150 mg/kg/d. In female animals low muscle tone was observed at 50, 75 and 100 mg/kg/d beginning on day 8 in the 100 mg/kg/d group, on day 15 in the 75 mg/kg/d group and on days 22-28 in the 50 mg/kg/d group. On day 14 grip strength was reduced in the 75 and 100 mg/kg/d group and on day 28 grip strength was reduced in the three highest dose groups. No consistent decreases or dose dependent changes were apparent in plasma, RBC or brain cholinesterase at any dose tested. Decreases in the electrophysiological values measured as the maximum amplitude were observed in the 50 and 75 mg/kg/d group (electrophysiological measurements not taken at the 100 mg/kg/d dose level).

A dermal NOAEL of 25 mg/kg bw/d can be derived from the study.

Ref.: D1

In vitro studies

Knox et al. (2004) performed *in vitro* studies using bag cell neurons of a marine snail (Aplysia) in order to investigate the mechanisms underlying the reversible neurotoxicity of pyrithiones by using sodium pyrithione. It could be demonstrated that NaPT caused intracellular Ca^{2+} elevation. Several hypotheses which could build the molecular basis for this calcium entry have been tested. It could be demonstrated that the elevation of intracellular Ca^{2+} results from influx of calcium ions from the external medium, which is not affected by blockage of voltage gated Ca^{2+} channels. Intracellular Ca^{2+} elevation was also unaffected by inhibition of voltage-gated Na^{+} channels or a Na^{+}/K^{+} ATPase inhibitor. It could be demonstrated that the NaPT-induced elevation of Ca^{2+} was attenuated by two potential inhibitors of store-operated Ca^{2+} entry (SKF 96365 and Ni^{2+}) suggesting that NaPT activates a form of calcium influx in these invertebrate neurons. From the studies performed, the authors concluded that pyrithione-evoked Ca^{2+} entry into the cells might also trigger activation of this nonselective cation current and depolarize neurons, thus explaining at least in part the neurotoxic effects of pyrithiones in rodents.

Ref.: D13

In a follow-up study, the same group investigated whether pyrithione is also capable of inducing influx of Ca^{2+} into mammalian neurons and whether there would be any difference between cells obtained from rats (strain not mentioned) and Rhesus monkeys.
It could be demonstrated that in isolated rat as well as in isolated monkey motor neurons NaPT produced an increase of intracellular Ca^{2+}. The study authors discussed that elevation of intracellular Ca^{2+} can be used as an explanation for the accumulation of tubovesicular profiles within the terminals of rodent motor neurons following exposure to NaPT *in vivo.* In rat motor neurons, the NaPT induced Ca^{2+} entry was unaffected by nefedipine, a blocker of L-type voltage-dependent calcium channels or by tetrodotoxin, which blocks voltage-dependent sodium channels. Ca^{2+} elevation was also unaffected by ouabain, an inhibitor of the plasma membrane Na^{+}/K^{+} ATPase. As has been demonstrated for bag cell neurons of Aplysia in a precedent study, SKF 96365, an antagonist of certain store-operated plasma membrane Ca^{2+} channels, inhibited NaPT induced Ca^{2+} entry into the cells in rat as well as in Rhesus monkey motor neurons, suggesting that PT targets such channels and suggesting that the mechanism of action of NaPT in motor neurons is conserved across species. Despite the qualitative similarities, quantitative differences in NaPT induced intracellular Ca^{2+} elevation were observed between rat and Rhesus monkey motor neurons. In rat motor neurons, the NaPT concentration that produces an increase in intracellular Ca^{2+} corresponding to 50 % of the maximum response (EC_{50}) is 0.31 µM, whereas in Rhesus monkey motor neurons the EC_{50} value for NaPT induced Ca^{2+} elevation is 10 µM, i.e. 30 times higher compared to the rat. This finding might explain the species differences between rats and monkeys observed after ZPT administration *in vivo*.
In addition to NaPT, the authors also investigated the effects of 2-MSP (2-methylsulfonylpyridine, the terminal serum metabolite of NaPT or ZPT) on intracellular Ca^{2+} elevation. In rat as well as monkey motor neurons, this metabolite did not induce Ca^{2+} elevation (which is in line with studies demonstrating that this metabolite fails *in vivo* to produce neurotoxicity in rats).

Ref.: D14

In a study aiming at systematically identifying potassium channel modulators, Xiong et al. discovered by using cell lines stably expressing potassium channels KCNQ2 and KCNQ2/3 that ZPT activates both recombinant and native KCNQ m currents (remark: an M-current is a non-inactivating potassium current found in many neuronal cell types. In each cell type, it is dominant in controlling membrane excitability by being the only sustained current in the range of action potential initiation. It can be modulated by a large array of receptor types, and the modulation can occur either by suppression or enhancement. Modulation of M-current has dramatic effects on neuronal excitability. (Marrion N.V. (1997): Annu. Rev. Physiol. 59, 483 – 504)). The activation of KCNQ potassium channels is reversible and not mediated by Zn^{2+} ions but consistent with ZPT directly binding to KCNQ channels. In the same study it could be demonstrated that ZPT is able to upregulate mutants of KCNQ channels exhibiting reduced currents which are thought to be associated with benign familial neonatal convulsions.

Ref.: D22

Lamore et al. (2010) demonstrated that cultured primary human skin keratinocytes and melanocytes display an exquisite vulnerability to nanomolar concentrations of ZPT resulting in pronounced induction of heat shock response gene expression and impaired genomic integrity. In keratinocytes treated with nanomolar concentrations of ZPT, expression array analysis revealed massive upregulation of genes encoding heat shock proteins (HSPA6, HSPA1A, HSPB5, HMOX1, HSPA1L, and DNAJA1) further confirmed by immunodetection. Moreover, ZPT treatment induced rapid depletion of cellular ATP levels and formation of poly (ADP-ribose) polymers. Consistent with an involvement of poly(ADP-ribose) polymerase (PARP) in ZPT-induced energy crisis, ATP depletion could be antagonised by

pharmacological inhibition of PARP. This result was independently confirmed using PARP-1 knockout mouse embryonic fibroblasts that were resistant to ATP depletion and cytotoxicity resulting from ZPT exposure. In keratinocytes and melanocytes, single-cell gel electrophoresis and flow cytometric detection of γ-H2A.X revealed rapid induction of DNA damage in response to ZPT detectable before general loss of cell viability occurred through caspase-independent pathways. Combined with earlier experimental evidence that documents penetration of ZPT through mammalian skin, the authors point out that their findings raise the possibility that this topical antimicrobial may target and compromise keratinocytes and melanocytes in intact human skin.

Ref.: D15

Lamore et al. (2011) further demonstrated that ZPT causes rapid accumulation of intracellular zinc in primary keratinocytes as observed by quantitative fluorescence microscopy and inductively coupled plasma mass spectrometry (ICP-MS), and that PARP activation, energy crisis, and genomic impairment are all antagonized by zinc chelation. In epidermal reconstructs (EpiDerm™) exposed to topical ZPT (0.1–2% in Vanicream™), ICP-MS demonstrated rapid zinc accumulation, and expression array analysis demonstrated upregulation of stress response genes encoding metallothionein-2A (*MT2A*), heat shock proteins (*HSPA6*, *HSPA1A*, *HSPB5*, *HSPA1L*, *DNAJA1*, *HSPH1*, *HSPD1*, *HSPE1*), antioxidants (*SOD2*, *GSTM3*, *HMOX1*), and the cell cycle inhibitor p21 (*CDKN1A*). Immunohistochemistry analysis of ZPT-treated EpiDerm™ confirmed upregulation of Hsp70 and TUNEL-positivity. Thus, ZPT impairs zinc ion homeostasis and upregulates stress response gene expression in primary keratinocytes and reconstructed human epidermis, activities that may underlie therapeutic and toxicological effects of ZPT.

Ref.: D16

Effects of nM zinc pyrithione on cell stress response pathways, including p53 and stress kinase p38, was demonstrated on primary human skin fibroblasts during 24h of exposure also by Rudolf and Cervinka (2011).

Ref.: E32

3.3.12.2. Ocular Toxicity

Ocular effects of ZPT as observed in Snyder (1965) and Cloyd et al., (1978) after oral administration are species specific and are not considered of relevance for humans: studies performed in various species provide evidence that the tapetum lucidum is the target tissue for the ocular toxicity of ZPT, since the eyes of animals lacking this choroidal structure are not affected. Humans and non-human primates do not possess a tapetum lucidum.

Ref.: B61; D3

Conclusions on Special Investigations
Reversible hind limb paralysis is the most prominent effect observed in rats after repeated oral administration of ZPT. In the first instance dermal administration was considered to not cause hind limb paralysis. However, mechanistic studies and further dermal repeat-dose studies have demonstrated that dermal administration of ZPT caused electrophysiological changes and decreases in hindlimb and forelimb grip strength and muscle tone. An acute inhalation study (described in section 3.4.1.3) demonstrates that neurotoxic (hindlimb) effects are also exerted after uptake by the inhalation route.
The loss of hind limb function is mediated by peripheral axonopathy. Thus, muscle atrophy is considered as a secondary event due to underlying nerve damage.

Species differences have been observed with respect to loss of hind limb function with monkeys being appreciably less sensitive to ZPT induced loss of hind-limb function.
In vitro studies so far contributed to a mechanistic understanding of ZPT-induced neurological effects: Pyrithione stimulates an influx of calcium into both rat and Rhesus monkey motor neuron preparations. This influx is mediated via pyrithione-stimulated Ca^{2+} release-activated Ca^{2+} channels. Although quantitative differences in Ca^{2+} influx were observed between rat and monkey motor neurons *in vitro*, which might be used to explain differences in sensitivities to the neurotoxic effects of ZPT in rats and monkeys, no conclusions with respect to human sensitivity can be drawn from these studies. Furthermore, it has to be kept in mind that in the *in vitro* studies no attempt had been made to relate the *in vitro* pyrithione concentration to *in vivo* blood levels.
Apart from an interaction with Ca^{2+} channels, ZPT is also able to activate KCNQ potassium channels.
In cultured human primary keratinocytes, ZPT caused an upregulation of heat shock proteins and other stress response genes, depletion of cellular ATP levels, formation of poly (ADP-ribose) polymers and impairment of zinc homeostasis. Further, ZPT induced DNA damage in keratinocytes and melanocytes as shown by single cell gel electrophoresis.

3.4.12.3 Market experience

Taken from SCCNFP/0671/03

ZPT has been used as an anti-dandruff active in shampoo formulations at levels of 1.0 and 2.0% since the 1940's. During this time there has been little evidence of any serious adverse effects from this usage, and those few effects that have been recorded are limited to eye and skin irritation as can be expected for surfactant-based formulations.
The effective use of shampoos containing this ingredient involves regular (2-3 times a week at least) usage.
Recent tracking of consumer marketplace complaints continue to indicate that ZPT containing shampoos have a very similar, low problem incidence profile compared to conventional shampoos which do not contain the material.

New information:
In its Comments on SCCS Opinion: Zinc Pyrithione – P81 (Ref. E31) the applicant states that in the past year, a 2% ZPT anti-dandruff shampoo has been sold in the US without any change in post-market health-related consumer comments.

SCCS comment
Consumer surveillance should be continued.

3.4.13. Discussion of Safety evaluation (including calculation of the MoS)

CALCULATION OF THE MARGIN OF SAFETY

Although modern approaches were submitted by the applicant to demonstrate the safety of the substance, these approaches were associated with many uncertainties, even contradictions in such that they have not been used for risk assessment by the SCCS.

3.4.13.1. Discussion of safety evaluation based on oral studies and internal exposure

Systemic exposure dose (4.66 µg/kg/d+ 1 SD) = 5.25 µg/kg/d
Lowest observed adverse effect level (mg/kg) = 500 µg/kg/d
(oral chronic/carcinogenicity study, rat)
Adjusted to NOAEL (application of adjustment factor of 3) = 167 µg/kg/d
100 % bioavailability = 167 µg/kg/d
88% bioavailability = 147 µg/kg/d

Margin of Safety (100 %bioavailability)	**NOAEL / SED**	**= 31.8**
Margin of Safety (88 %bioavailability)	**NOAEL / SED**	**= 28.0**

A reduced MoS is suggested for the safety evaluation of ZPT. Different frameworks (e.g. WHO/IPCS) further split up the MoS of 100 into sub-factors which could be replaced by data-derived subfactors, if such data is available: the MoS of 100 is composed of assessment factors (AF) of 4 (interspecies differences in toxicokinetics) x 2.5 (interspecies differences in toxicodynamics or) x 3.16 (human variability in toxicokinetics) x 3.16 (human variability in toxicodynamics).
Within REACH the interspecies AF consists in the correction for metabolic rate (allometric scaling) and a factor of 2.5 for toxicokinetic (TK) differences not related to metabolic rate (smaller part of 2.5) and toxicodynamic (TD) differences (larger part of 2.5).
With respect to ZPT the interspecies AF could be reduced as information on TK differences not relating to metabolic rate are available (suggestion: 4 for allometric scaling and 2 for differences in toxicodynamics) leading to an overall interspecies AF of 8.

In addition, also the AFs for intraspecies variability in TK and TD could be reduced based on the following grounds:
With respect to the TK part: the value of the systemic exposure dose of 5.25 µg/kg bw/d comprises three layers of conservatism: (1) it results from the concomitant use of rinse-off shampoo in combination with leave-on tonic, (2) it is a value, to which 1 standard deviation has been added in order to cope for more extended periods of application and (3) it results from the upper boundary of internal exposure determined in the human clinical study.
With respect to the TD part: (1) the previously derived NOAEL of 500 µg/kg bw/d has been converted to a LOAEL based on the occurrence of some effects which could not be retraced further due to unavailability of data. (2) ZPT has been used as an anti-dandruff active in shampoo formulations at levels of 1.0 and 2.0% since the 1940's. During this time there has been little evidence of any serious adverse effects from this usage, and those few effects that have been recorded are limited to eye and skin irritation as can be expected for surfactant-based formulations.

Therefore an overall intraspecies AF of 4-5 is considered acceptable.

An interspecies AF of 8 and an intraspecies AF of 4 – 5 would yield an overall MoS of 32 – 40.

Under these aspects, the ZPT could be considered safe which would be in line with calculations based on external exposure as given below (the SCCS does not use this calculation as first and only choice as internal exposure data from a human clinical study were available).

3.4.13.2. Discussion of safety evaluation based on oral studies and external exposure (SCCS NoG 8th version, p. 69)

Lowest observed adverse effect level (mg/kg) = 500 µg/kg/d

(oral chronic/carcinogenicity study, rat)167
Adjusted to NOAEL (application of adjustment factor of 3) = 167 µg/kg/d
100 % bioavailability = 167 µg/kg/d
88% bioavailability = 147 µg/kg/d

A = 1.51 mg/kg bw/d (shampoo) + 0.6 mg/kg bw/d (conditioner) = 2.11 mg/kg/d.

C (%) (concentration of substance in finished cosmetic product) (%) = 2

Dap (%) = 1

Assuming 1 % dermal absorption in humans is supported by a human clinical study using 2 % ZPT shampoo formulations in combination with 0.1 % or 0.25 % ZPT containing leave-on formulations. In this study up to 0.22 % of the applied dose was excreted via urine. Taking into consideration that further amounts could have been excreted at later time points not considered in the test interval or by faecal excretion and also considering some tissue retention, total absorption is most probably not higher than 1 %.

A x C / 100 x Dap = 2.11 mg/kg/d x 2/100 x 0.01 = 0.00042 mg/kg/d
100 % bioavailability 0.167 mg/kg/day ÷ 0.00042 mg/kg/day = 398
88% bioavailability 0.147 mg/kg/day ÷ 0.00042 mg/kg/day = 350

Overall conclusion on MoS calculation:

Based on all above considerations, the substance is considered safe under the use conditions applied for.

Physico-chemical properties
Concerning some physico-chemical properties, diverging values are available from different sources of information.

Toxicity
In addition to acute oral toxicity studies evaluated in SCCNFP/0671/03, further studies have been performed. The data is not available for evaluation. SCCS notes, however, that classification as Acute Tox 3; H301 (toxic if swallowed) according to CLP is suggested by the registrant(s) under REACH (ECHA website (http://echa.europa.eu/).
The acute dermal toxicity of ZPT appears to be higher than 2000 mg/kg.
Acute inhalation studies have been performed with ZPT. According to one of the studies, classification as Acute Tox 3; H331 (toxic if inhaled) according to CLP as suggested by the registrant(s) under REACH (ECHA website (http://echa.europa.eu/) is justified.
Several oral repeat-dose studies of different durations have been performed with ZPT. In addition, one sub-chronic and a chronic oral studies performed with sodium pyrithione can be considered adequate to assess repeat-dose effects of ZPT.
An **oral** NOAEL of 500 µg/kg/d obtained from a chronic oral study (Larson, 1958) performed with ZPT based on paralysis/hind-limb weakness has been derived in SCCNFP 0671/03.
The SCCS is aware that HSE (2003) considered the Larson 1958 study as inadequate due to insufficiently large group sizes to ensure statistical power. However, a 90-day oral study performed with sodium pyrithione (not available to the SCCS) which was considered adequate by HSE, also lead to a NOAEL of 500 µg/kg/d, supporting the outcome of the Larsson study.
Two oral chronic studies performed with NaPT have been provided by the applicant. In a combined chronic toxicity/carcinogenicity study the dose of 500 µg/kg bw/d is considered as LOAEL by the SCCS.

Several **dermal** repeat-dose studies have been performed with ZPT. Interpretation of the findings is partly hampered by the fact that grooming was not always prevented and that intermittent exposure regimens (causing recovery) have been applied. From a 28-day dermal neurotoxicity study in which grooming was prevented, NOAELs of 25 and 50 mg/kg bw/d were derived in female and male animals, respectively based on reduced electrophysiological parameters and muscle tone. In a two year dermal chronic study performed with NaPT, a local NOEL of 5 mg/kg bw/d was derived.
Three inhalation studies of different durations have been performed, two of them are available for evaluation. In a 21-day nose only study performed in Sprague-Dawley rats, a LOAEC of 2 mg/m^3 is derived based on histopathological data in tissues of the respiratory tract and non-respiratory tissues. In a 28-day nose only study performed in Sprague-Dawley rats, no NOAEC could be derived for local effects in the lung and an NOAEC of 1.5 mg/m^3 was derived for systemic effects.
In a 90-day study, animals were whole-body exposed and oral intake cannot be excluded.

Skin/eye irritation and sensitisation
Skin irritation studies performed with ZPT were not available for evaluation. However, from product based data evaluated in SCCNFP/0671/03, from the description of skin irritation studies performed with ZPT and from human HRIPT tests it can be inferred that ZPT is – at least - a mild skin irritant.
ZPT has been investigated in eye irritation tests, but the studies are not available for evaluation. HSE concludes that ZPT is a severe eye irritant, MAK (2012) states, that ZPT is corrosive to the eye. SCCS notes, that classification as Eye Damage 1; H318 (causes serious eye damage) according to CLP is suggested by the registrant(s) under REACH (ECHA website (http://echa.europa.eu/)).
ZPT is not sensitising in animal studies. Concerning human data, ZPT (or better: the PT moiety) has a low potential to induce contact hypersensitivity when tested per se or as part of a cosmetic formulation. However, in some human HRIPT studies, evaluation was partly hindered by the erythematous reactions observed.

Summary dermal absorption:
From the animal studies available in submission I, it was concluded in SCCNFP 0671/03 that percutaneous absorption of ZPT varies from approximately 0.03 to 3.4%. Further studies on dermal absorption of ZPT have been performed thereafter.

In an in vitro dermal absorption study performed according to OECD TG 428 using rat and human split-thickness skin and concentrations of 48 % and 1 % ZPT, the following absorption values were obtained:

Sample	Dermal delivery (dermis, epidermis, receptor fluid, receptor rinse)		Potentially absorbable dose (dermal delivery and stratum corneum tape strips 6-20)	
	[%]	[µg/equiv./cm^2]	[%]	[µg/equiv./cm^2]
Rat, 48 % solution	2.26 ± 0.83	125.65 ± 45.87	3.14 ± 1.14	174.59 ± 63.18
Rat, 1 % solution	7.75 ± 5.26	7.81 ± 5.31	9.28 ± 5.60	9.36 ± 5.65
Human, 1 % solution	0.76 ± 1.14	0.77 ± 1.15	1.31 ± 1.32	1.32 ± 1.33

From an in vitro toxicokinetic study performed in rats with 1 % and 48 % ZPT, mean and maximal potentially absorbable dose were 0.19 % and 0.27 % for the 48 % solution. For the 1 % solution, mean and maximal potentially absorbable dose were 0.85 % and 1.13 %.
From dermal toxicokinetic studies (not fully OECD compliant) performed in female CD rats in order to build up a PBPK model for ZPT, absorption percentages of 2.3, 8.6 and 0.3 were derived after repeated dermal administration of 10, 30 and 100 mg/kg ZPT. However, these values are associated with uncertainties (see section 3.4.9.3). The SCCS is aware that different absorption percentages were reported from apparently the same study in a submitted manuscript on a dermal PBPK model for ZPT.
Dermal absorption was also assessed in two clinical studies in humans. One study investigated the systemic absorption of a shampoo containing 1 % ZPT (with or without combination with a leave-on tonic containing 0.1 % ZPT) in a 4-day treatment regimen. In this study, a systemic load of ZPT up to 3.43 µg/kg/d was derived. When using a dermal PBPK model, an absorption percentage of 0.5 % was calculated for a 1% ZPT solution. However, no further background information is given for that value. In the second clinical study, the systemic absorption of a shampoo containing 2 % ZPT (either in combination with leave-on tonics containing 0.1 and 0.25 %ZPT or with a leave-on tonic containing 0.25% ZPT only) was investigated in a 4-day treatment regimen. Systemic exposure loads up to 4.66 µg/kg/d were derived. As in the new submission the applicant applies for an extension of the authorised concentration from 1.0% to 2.0% in rinse-off anti-dandruff hair care products, a systemic exposure load of 4.66 µg/kg/d can be taken for risk characterisation. In order to cope for prolonged application (more than 4 days) and consequently probably higher systemic exposure loads, 1 SD was added to the systemic exposure load of 4.66 µg/kg/d yielding 5.25 µg/kg/d.
From the daily urinary excretion data given in the study report, SCCS calculated cumulative amounts excreted for Regimen A, B and C. Cumulative excretion amounted to 450, 885.5 and 931.06 mg for regimen A, B, and C, respectively, during the observation period. The cumulative amount excreted during the observation period pointed to 0.22%, 0.16 % and 0.196 % of the applied amounts for regimen A, B, and C, respectively.

Mutagenicity/genotoxicity

From the studies available for SCCNFP/0671/03, it was concluded that ZPT is not mutagenic. Since then, further *in vitro* and *in vivo* genotoxicity/mutagenicity studies have been performed, not all of them are available for evaluation. *In vitro* studies are incomplete and in case of *hprt* gene mutation results are inconclusive with signs of potential mutagenicity that deserve further investigation. *In vivo* micronucleus test only identifies mutagenic compounds with chromosomal aberration/clastogenic or aneugenic effect and does not detect gene mutation inducing compounds.
Therefore, no firm conclusion with respect to genotoxicity/mutagenicity can be drawn by the SCCS. The SCCS, however, is aware that HSE (2003) and MAK (2012) considered ZPT as non-genotoxic and non-mutagenic.
With respect to the development of cancer, the ambiguous database on genotoxicity might be acceptable as no carcinogenic effect has been observed in chronic oral studies up to dose levels of 2.5 mg/kg bw/d ZPT and 3.5 mg/kg bw/d NaPT (based on systemic findings, higher doses of ZPT/NaPT could not be tested in chronic oral studies). Also with respect to developmental or reprotoxic effects, the ambiguous database might be acceptable as ZPT and NaPT cannot be considered as selective reproductive or developmental toxicants.

Carcinogenicity
From chronic oral and dermal studies available in submission I, SCCNFP 0671/03 concluded: "no evidence of a carcinogenic response was seen when ZPT was applied topically (up to 100 mg/kg/d) or given orally (up to 5 mg/kg/d) in lifetime studies using mice and rats."

Since that, further chronic (lifetime) studies performed with ZPT and sodium pyrithione (from which read across to ZPT is considered adequate) using the oral and dermal uptake pathway have become available.
From the studies performed by the oral or dermal route with either ZPT or NaPT there was no evidence for a carcinogenic potential up to dermal doses of 100 mg/kg bw day and up to oral doses of 2.5 mg/kg bw/d ZPT and 3.5 mg/kg bw/d NaPT (based on systemic findings, higher doses of ZPT/NaPT could not be tested in chronic oral studies).
Carcinogenicity of ZPT has not been investigated by the inhalation route.

Reproductive toxicity
In SCCNFP 0671/03 the following conclusions were drawn with respect to Reproductive toxicity of ZPT:
- 2.5 mg/kg/d administered orally to rats is a no effect level for teratological effects
- no reproductive effects have been observed when ZPT was applied topically to rats and rabbits at levels up to 15 and 100 mg ZPT/kg/d respectively (highest doses tested) and ingestion of the test material was controlled.
- no reproductive or teratogenic effects have been observed in rabbits and pigs following topical application of shampoo formulations containing 50 and 400 mg ZPT/kg/d respectively.
Since that, further generation studies have been performed with ZPT and NaPT as well as developmental toxicity studies with ZPT. One 2 generation study which is mentioned on ECHA's website is not available for evaluation by SCCS. However, based on the overall picture given by the available data (and by the summary of the non-available study), the conclusions of other scientific bodies can be supported: the SCCS is aware that HSE (2003) did not identify any potential concern to humans regarding adverse effects on fertility. Further, both MAK (2012) and HSE (2003) concluded that adverse effects on development were most likely attributable to maternal toxicity.

4. Conclusion

Although modern approaches were submitted by the applicant to demonstrate the safety of the substance, these approaches were associated with many uncertainties, even contradictions in such that they have not been used for risk assessment by the SCCS.

Based on the scientific data provided the SCCS considers that zinc pyrithione, when used in a concentration up to 2.0% as an anti-dandruff agent in rinse-off hair care products, is safe for the consumer.

Aggregate exposure to Zinc Pyrithione from non-cosmetic sources has not been considered.

The SCCS recommends thorough post-marketing surveillance of the product.

5. Minority opinion

-

6. References

A References from submission II

Annex

I. FINAL REPORT: Physiologically-based pharmacokinetics model of zinc pyrithione in the rat (June 2006). FINAL REPORT: Feasibility study for a physiologically-based pharmacokinetics model of zinc pyrithione (August 2004)

II. FINAL REPORT: Disposition of zinc pyridinethione (ZPT) in female rats: repeated dose oral and dermal pharmacokinetic studies (2007). RTI Study No. 0208173.000 Final Report

III. CLINICAL STUDY FINAL REPORT: An evaluation of the deposition, absorption, and excretion of 14C resulting from an anti-dandruff treatment regimen consisting of shampoo and a leave-on tonic containing 14C-pyrithione zinc (Study Number CRB 0005-067 HC)

References from submission II

A1. Bruckner, J. V. (2000). Differences in sensitivity of children and adults to chemical toxicity: The NAS panel report. Regulatory Toxicology and Pharmacology 31, 280-285.

A2. Diamond, G.L. and Nash, J.F. (2008): Use of a Physiologically-based Pharmacokinetic (PBPK) Model to Predict Internal Dosimetry of Zink Pyrithione (Zn-PT) in Rats and Humans. Society of Toxicology 2008. (Poster)

A3. Demchuk, E., Ruiz, P., Wilson, J. D., Scinicariello, F., Pohl, H. R., Fay, M., Mumtaz, M. M., Hansen, H., and De Rosa, C. T. (2008). Computational toxicology methods in public health practice. Toxicology Mechanisms and Methods 18, 119-135.

A4. Gibson, W. T., Chamberlain, M., Parsons, J. F., Brunskill, J. E., Leftwich, D., Lock, S., and
A5. Safford, R. J. (1985). The effect and mode of action of zinc pyrithione on cell growth. I. *In vitro* studies. Food Chem Toxicol 23, 93-102.

A6. Krishnan, K., Haddad, S., liveau, M., and Tardif, R. (2002). Physiological modelling and extrapolation of pharmacokinetic interactions from binary to more complex chemical mixtures. Environmental Health Perspectives 110, 989-994.

A7. Mitoma, C., Steeger, T., Rogers, J., Thomas, D., and Wedig, J. H. (1983). Metabolic disposition of pyrithiones. Fundam.Appl.Toxicol 3, 256-263.

A8. Nash, J.F. and Diamond, G.L. (2006): A Physiological-Based Pharmacokinetic Model of Zinc Pyrithione in the Rat. Society of Toxicology 2006. (Poster)

A9. Nash, J.F., Jeffcoat, A.R., Coleman, D., Garner, E., Cooper, V.H. and Skoulis, N.P. (2007): Disposition of Pyrithione and Muscle weakness in the Rat after Repeat Dose Administration. Society of Toxicology 2007 (Poster)

A10. Price, P. S., Conolly, R. B., Chaisson, C. F., Gross, E. A., Young, J. S., Mathis, E. T., and Tedder, D. R. (2003). Modeling Interindividual Variation in Physiological Factors Used in PBPK Models of Humans. Critical Reviews in Toxicology 33, 469-503.

A11. Scientific Committee on Cosmetic Products and Non-Food Products Intended for Consumers. Opinion Concerning Zinc Pyrithione, Colipa No. P81. SCCNFP/671/02. 12-17-2002. 12-17-2002.

A12. Skoulis, N.P., Jeffcoat, A.R., Valentine, J.L., Coleman, D.P. and Nash, J.F. (2007): Disposition of Pyrithione in the Rat after Oral, Dermal and Intravenous Routes of Administration. Society of Toxicology (2007) (Poster)

A13: Procter & Gamble Human Repeat Insult Patch Test, Sponsor's Reference HBE BTS 0602/01, Quintiles Study number PGU/32/V, November 1996

A14: IS Consultancy Limited (issued to Procter & Gamble Technical Centres Ltd.). Report No. 05033 Final, HRIPT ISC No. 134, Project No. 0503, SAF 186-50287, TSAN 10000330, December 1998

A15: CTC International Ltd (issued to Procter & Gamble SA, European Technical Center). CTC Schedule No. PGM/604, CTC Report No. 89/PGN604A/0329. Human Repeat Insult Patch Test CTC 83, RVE BTS 0030, RE-0079, April 1989.

A16: Inveresk Research International (issued to Procter & Gamble Technical Centre) Report No. 5289, HRIPT IRI No. 88, IRI Project No. 240356, RVE BTS 0013, RE0034, Report completed August 1988.

A17: IS Consultancy Limited (issued to Procter and Gamble European Technical Center). Report No. 02232, HRIPT ISC No. 65, Project No. 0223, HBE BTS 0121, RE0199, November 1999.

A18: Hill Top Research Inc. (issued to Procter and Gamble Company). Human Repeat Insult Patch test Report No. 93-3560-73A, Procter and Gamble identification numbers TSIN: BH0031-134, DRD: BCS0122, July 1994.

A19: Health Sciences Science and Engineering Group RTI International, RTI Report No. RTI/0208173 phase 1. July 2001

A20: Biopharmaceutical Analysis Laboratory, Department of Pharmaceutics, Virginia Common Wealth University (issued to Procter and Gamble). Sponsor Study number CRB 0005-067 HC. An evaluation of the Deposition, Absorption, and Excretion Resulting from an Anti-dandruff Treatment Regimen Consisting of Shampoo, conditioner and a Leave on Tonic Combination Containing 14C-Pyrithione Zinc. Investigator report. September 2000.

A21: Biopharmaceutical Analysis Laboratory, Department of Pharmaceutics, Virginia Common Wealth University (issued to Procter and Gamble). Sponsor Study number CRB 0005-067 HC. An evaluation of the Deposition, Absorption, and Excretion Resulting from an Anti-dandruff Treatment Regimen Consisting of Shampoo, conditioner and a Leave on Tonic Combination Containing 14C-Pyrithione Zinc. Final report. 2000

A22: Health Sciences Science and Engineering Group RTI International (issued to Procter amd Gamble). RTI study code number Rt04-919. Disposition of Zinc Pyridinethione (ZPT) in female rats: Repeated Dose Oral and Dermal Pharmacokinetic studies. July 2007.

A23: Syracuse Research Corporation (prepared for Dr. J.F. Nash, Procter and Gamble): Feasibility study for a Physiologically based Pharmacokinetics Model of Zinc Pyrithione, August 2004.

A24: Syracuse Research Corporation (prepared for Dr. J.F. Nash, Procter and Gamble):Physiologically-based Pharmacokinetics Model od Zinc Pyrithione in the Rat. June 2006

A25: Diamond G.L. and Nash, J.F. (2008) Use of a Physiologically-based Pharmacokinetic (PBPK) Model to Predict Internal Dosimetry of Zinc Pyrithione (Zn-PT) in Rats and Humans; Poster at SOT 2008.

A26: Wedig, J.H., Wentworth, R.A., Gallo, M.A., Babish, J.G., and Henion, J.D. (1978): Disposition of zinc pyrithione in the rat. Food Cosmet Toxicol 16, 5553- 561.

B. References from previous submissions:

B1. ADAMS, M.D., WEDIG, J.H., JORDAN, R.L, SMITH, L.W., HENDERSON, R., BORZELLECA, J.F. (1976), "Urinary excretion and metabolism of salts of 2-I-oxide following intravenous administration to female Yorkshire pigs", Toxicol. Appl. Pharmacol., 36 (3), 523-31.

B2. ADAMS, M., JORDAN, R., BORZELLECA, J., (Undated), "Acute toxicity and disposition of sodium omadine, zinc omadine and omadine-MDS following intravenous administration to swine", Medical College of Virginia, report to Olin Chemical Corporation

B3. BLACK, J.G., HOWES, D., RUTHERFORD, T. (1975), "Skin deposition and absorption of zinc pyrithione in laboratory animals", Actualites de Dermocosmetologie, Centre European de Dermocosmetologie, 69 Lyon, VII, 128-142

B4. BRANDRUP, F., MENNE, T. (1985), "Zinc pyrithione (Zinc Omadine) allergy", Contact Dermatitis, 12, (1),50

B5. BUEHLER, E.V. (1965), "Delayed contact hypersensitivity in the guinea pig", Arch. Dermat., 91, 171-5

B6. CALANDRA, J.C. (1972), "Acute oral toxicity study with Zinc omadine in albino rats", Industrial BioTest Laboratories, Report to Olin Corporation

B7. CALVIN, G., LAWHORN, G.T. (1972), "The percutaneous absorption of zinc pyridinethione in monkeys", Procter & Gamble Internal Report

B8. CALVIN, G., LAWHORN, G.T. (1972a), "The effect of ZPT in the diet of Rhesus monkeys", Procter & Gamble Internal Report

B9. CHRISMAN, C.L., ROSS, J.F. (1978), "ZPT dose-response study: electrophysiology and clinical signs", Report to Procter & Gamble

B10. CUMMINS, L.M., KIMURA, E.T. (1971), "Safety evaluation of selenium sulfide antidandruff shampoos", Toxicol. Appl. Pharmacol., 20 (1),89-96

B11. DEARWESTER, D.D., JOHNSON, G.R. (1974), "28 day feeding study on zinc pyridinethione (ZPT)", Procter & Gamble internal report

B12. DEJESUS, C.P.V., TOWFIGHI, J., SNYDER, D.R. (1978), "Sural nerve conduction study in the rat: a new technique for studying experimental neuropathies", Muscle and Nerve, 1 (2), 162-7

B13. DOBBS, J., NIXON, G., (1973), "Pilot mouse painting study", Procter & Gamble intern report

B14. DOYLE, R.L., ELSEA, J.R. (1960), "Emetic dose study on UDB-107 in pigeons", Hill Top Research Institute, Report to Procter & Gamble

B15. DOYLE, R.L., ELSEA, J.R. (1962), "Emetic dose studies on UDB-662, -655, -659 and -685", Hill Top Research Institute, Report to Procter & Gamble

B16. EVANS, P.G.E., SUGDEN, J.K., VAN ABBE, N.J. (1975), "Aspects of the photochemical behaviour of 1-hydroxypryidine-2-thione", Pharm. Acta Helv., 50, 94-8

B17. FISHER A.A. (1976), "Highlights of the First International Symposium on Contact Dermatitis", Cutis, 18,645-62

B18. GARGUS, J.L. (1974), "Repeated dermal applications-mice, (Progress Report 1), Project No. 297147", Hazleton Laboratories, Report to Procter & Gamble

B19. GIBSON, W.B. (1977), "The metabolism and distribution of zinc pyridinethione: VI. The excretion of radioactivity by rats after oral or intravenous administration of (^{14}C) and the tissue distribution of radioactivity in rats after intravenous administration of ^{14}C ZPT', Procter & Gamble Report

B20. GIBSON, W.B., CALVIN G. (1978), "Percutaneous absorption of zinc pyridinethione in monkeys", Toxicol. Appl. Pharm., 43 (3), 425-37

B21. GIBSON, W.B. (1979), "The importance of dose frequency to zinc pyridinethione - induced paralysis", Paper presented at annual meeting of Soc. of Toxicology

B22. GIBSON, W.B., JEFFCOAT, A.R., TURAN, T.S., WENDT, R.H., HUGHES, P.F., TWINE, M.E. (1982), "Zinc pyridinethione: Serum metabolites of zinc pyridinethione in rabbits, rats, monkeys and dogs after oral dosing", Toxicol. Appl. Pharmaol., 62, 237-50

B23. GOH, C.L., LIM, K.B. (1984), "Allergic contact dermatitis to zinc pyrithione", Contact Dermatitis, 11, 120

B24. HALEY, S., PLANK, J.B., WRIGHT, F.L., KEPLINGER, M.L. (1971), "Teratology study with zinc omadine in albino rats", Industrial Bio-Test laboratories, report to Olin Corp.

B25. HILL TOP TOXICOLOGY (1977), "Delayed contact hypersensitivity study in guinea pigs", Report to Procter & Gamble

B26. HINK, G. (1977), "Repeated insult patch test of four test materials", Hill Top Research Inst., Report to Procter & Gamble

B27. HOWES, D., BLACK, J.G. (1975), "Comparative percutaneous absorption of pyrithiones", Toxicology, 5,209-20

B28. HOWLETT, H.C.S., VAN ABBE, N.J. (1975), "The action and fate of sodium pyridinethione when applied topically to the rabbit", Presented at the International Federation of Societies of Cosmetic Chemists VIIIth International Congress

B29. JEFFCOAT, A.R., GIBSON, W.B., RODRIGUEZ, P.A., TURAN, T.S., HUGHES, P.F., TWINE, M.E. (1980), "Zinc pyridinethione: Urinary metobolites of zinc pyridinethione in rabbits, rats, monkeys and dogs after oral dosing", Tox. Appl. Pharmacol., 56 (1), 141-54

B30. JORDAN, R.L., BORZELLECA, J.F. (1975), "Teratogenic studies with zinc omadine in swine", Anat. Rec., 181, 388

B31. KABACOFF, B.L., FAIRCHILD, C.M., BURNETT, C. (1971), "Pyridinethione glucuronide as a metabolite of sodium pyridinethione", Food Cosmet. Toxicol., 9, 519-26

B32. KLAASSEN, C.D. (1976), "Absorption, distribution and excretion of zinc pyridinethione in rabbits", Toxicol. Appl. Pharmacol., 35, (3), 581-7

B33. LARSON, P.S. (1956), "Studies on the acute oral toxicity of zinc pyridinethione, Na pyridinethione, (pure grade) and Na pyridinethione (technical grade) to rats", Report to Olin Corporation

B34. LARSON, P.S. (1956a), "Three-month study on the effect of adding zinc pyridinethione to the diet of rats", Report to Olin Corporation

B35. LARSON, P.S. (1957), "Subacute (90 day) percutaneous toxicity of omadine in the rabbit", Report to Olin Corporation,

B36. LARSON, P.S. (1958), "Toxicologic observations on the effect of adding Zn pyridinethione to the diet of rats for a period of two years", Report to Olin Corporation

B37. MAIBACH, H.I. (1976), "The effect of Head & Shoulders shampoo on the level of pigmentation in human skin", Report to Procter & Gamble

B38. MAJORS, P.A., RUBENKOENIG, H.L. (1960), "The emetic dose of sample UDV - 1994", Hill Top Research Inst., Report to Procter & Gamble

B38* MEYNADIER, J.-M., MEYNADIER, J., COLMAS, A., CASTELAIN, P.-Y,. DUCOMBS, G., CHABEAU, G., LACROIX, M., MARTIN, P., NGANGU, Z. (1982), "Allergie aux Conservateurs", Ann. Dermatol. Venereol. (Paris), 109, 1017-23

B39. MIN, B.H., PAREKH, C., COLBERG, L., McCHESNEY, E.W. (1970), "Experimental studies of sodium pyridinethione (II - urinary excretion following topical application to rats and monkeys)", Fd. Cosmet. Toxicol., 8,161-6

B40 MUSTON, H.L., MESSENGER, A.G., BYRNE, J.P. (1979), "Contact dermatitis from zinc pyridinethione, an anti-dandruff agent", Contact Dermatitis, 5 (4), 276-7

B41. NELSON, J.S., WOOLSEY, R.M., MURPHY, M.Q. (1965), "The effect of pyridinethiol oxide on the Central Nervous System", Excerpta Medica International Congress Series No. 100, Proceedings of the Vth International Congress of Neuropathology (Zurich, 1965), 798-801

B42. NOLEN, G.A., PATRICK, L.F., DIERCKMAN, T.A. (1975), "A percutaneous teratology study of zinc pyridinethione in rabbits", Toxicol. Appl. Pharmacol., 31, 430-3

B43. NOLEN, G.A., DIERCKMAN, T.A. (1979), "Reproduction and teratology studies of zinc pyridinethione administered orally or topically to rats and rabbits", Food Cosmet. Toxicol., 17, 639-49

B44. OKAMOTO, K., ITO, T., HASEGAWA A., (1967), "Percutaneous absorption of zinc bis (2-pyridylthio)1,1'-dioxide and residual amount on skin surface (Zinc omadine, zinc pyrithione)", J. Hyg. Chem., 13, 323-9

B45. OKUMURA, T., HAYASHI, S., TOKIWA, F., HORIN, S. (1975), "Adsorption of zinc pyrithione onto hair and skin", Cosmet. Perfum., 90, 101-4

B46. OPDYKE, D.L., BURNETT, C.M., (1967), "The chemical toxicity of zinc pyrithione", Paper presented to Sept. 1967 meeting of IUPAC Congress (Prague)

B46*.OPDYKE, D.L., BURNETT, C.M., BRAUER, E.W (1967a), Antiseborrheic qualities of ZPT in a cream vehicle. II. Safety evaluation", Food Cosmet. Toxicol., 5 (3), 321-6

B47. PAREKH, C., MIN, B.H., GOLBERG, L. (1970), "Experimental studies of sodium pyridinethione (I: Percutaneous absorption in laboratory animals)", Food Cosmet. Toxicol., 8, 147-60

B48. PARRAN, J.J. (1965), "Deposition on the skin of particles of antimicrobial agents from detergent bases", J. Invest. Derm., 45, (2) 86-88

B49. PATTERSON, D.R., GARGUS, J.L. (1979), "Repeated dermal application - mice", Hazleton Laboratories, Report to Procter & Gamble

B50. PIEPER, K.M. (1972), "Dominant lethal assay for zinc pyridinethione", Procter & Gamble Internal Report

B51. PIEPER, K.M. (1975), "An *in vivo* cytogenetic study on zinc pyridinethione", Procter & Gamble Internal Report

B52. RENO, R.E., BANAS, D.A. (1975), "A four-week subchronic feeding study in rhesus monkeys", Report to Procter & Gamble

B53. RUTHERFORD, T., BLACK, J.G. (1969), "The use of autoradiography to study the localization of germicides in skin", Br. J. Derm., 81 (4), 75-87

B54. SAHENK, Z., MENDELL, J.R. (1979), "Ultrastructural study of zinc pyridinethione - induced peripheral neuropathy", Division of neurology and department of pathology, Ohio State University College of Medicine, Columbus, Ohio, 1-32

B55. SAHENK, Z., MENDELL, J.R. (1979a), "Evidence for the distal axon as the site of axoplasmic transport abnormality in ZPT - induced neuropathy", Neurology, 29 (4), 590

B56. SAHENK, Z., MENDELL, J.R. (1980), "Zinc Pyridinethione", In "Experimental and Clinical Neurotoxicology" Ed. Spencer and Schaumburg, Williams and Wilkins, Baltimore, 578-92

B57. SEGAWA, T., TAKAGI, M. (1970), "Bis (1-hydroxy-2(1 H)-pyridinethionato) zinc (ZPT): Pharmacological properties of bis (1-hydroxy-2(1 H)-pyridinethionato) zinc (ZPT)", Oyo Yakuri, 4 (5), 883-90

B58. SKARE, J.A., WONG, T.K. (1983), "Unscheduled DNA synthesis assay in cultured human fibroblasts (WI-38 Cells) - Zinc Pyridinethione (ZPT)", Internal Procter & Gamble Report

B59. SKARE, J.A., WONG, T.K. (1983a), "Unscheduled DNA synthesis assay in primary cultures of rat hepatocytes - Zinc Pyrithione (ZPT)", Internal Procter & Gamble Report

B60. SKOULIS, N.P., BARBEE, S.J., JACOBSON-KRAM, D., PUTMAN, D.L., SAN, R.H. (1993), "Evaluation of the genotoxic potential of zinc pyrithione in the Salmonella mutagenicity (Ames) assay, CHO/HGPRT gene mutation assay and mouse micronucleus assay", J. Appl. Toxicol., 13, 283-9

B61. SNYDER, F.H., BUEHLER, E.V., WINEK, C.L. (1965), "Safety evaluation of zinc-2-pyridinethiol 1oxide in a shampoo formulation", Toxicol. Appl. Pharmacol., 7, 425-37

B62. SNYDER, D.R., GRALLA, E.J., COLEMAN, G.L., , J.H. (1977), "Preliminary neurological evaluation of generalized weakness in zinc pyrithione-treated rats", Food Cosmet. Toxicol., 15, (1), 43-7

B63. SNYDER, D.R., DEJESUS, C.P.V., TOWFIGHI, J., JACOBY, R.O., WEDIG, J. (1979), "Neurology, microscopic and enzyme - histochemical assessment of ZPT toxicity", Food Cosmet. Toxicol., 17 (6), 651-60

B64. SPIKER, R.C., CIUCHTA, H.P. (1980), "Effects of pyrithiones and surfactants on zinc and enzyme in rabbits", Am Ind. Hyg. Assoc. J., 41, (4), 248-53

B65. STOTTS, J. (1977), "Human sensitization tests of Head & Shoulders cream with RBD9818 perfume (7710-4) and RBD 9819 perfume (7711-3)", Procter & Gamble internal report

B66. WEAVER, J.A., PIEPER, K.M. (1977), "Genetic safety testing of zinc pyridinethione via the host mediated assay", Procter & Gamble Internal Report
B66*. WEDIG, J.H. (1976), "Toxicology summary of zinc omadine", Olin Corporation internal eport, 8/18/76

B67. WEDIG, J.H., KENNEDY, G.L., JENKINS, D.H., HENDERSON, R., KEPLINGER, M.L. (1976), "Teratologic evaluation of dermally applied zinc pyrithione on swine", Toxicol. Appl Pharmacol., 36, 255-9

B68. WEDIG, J.H., MITOMA, C., HOWD, R.A., THOMAS, D.W. (1978), "Identification of metabolites from salts of pyridine-2-thiol-1-oxide following intravenous and dermal administration to swine", Toxicol. Appl. Pharmacol., 43 (2), 373-9

B69. WEDIG, J.H., WENTWORTH, R.A., GALLO, M.A., BABISH, J.G. and HENION J.D. (1978a), "Disposition of ZPT in the rat", Food Cosmet. Toxicol., 16 (6) 553-61

B70. WEDIG, J.H., BARBEE, S.J., MITOMA, C. (1984), "Pyrithione: plasma metabolite in man", Fundam. Appl. Toxicol., 4(3) Pt. 1, 497-8

B71. WINEK, C.L. (1963), "Toxicity of pyridinethiones", Procter & Gamble internal report

B72. WINEK, C.L., BUEHLER, E.V. (1966), "Intravenous toxicity of zinc pyridinethione and several zinc salts", Toxicol. Appl. Pharmacol., 9 (2), 269-73

B73. ZILLER, S.A. (1969), "The effect of 2-pyridinethiol-1-oxide on animals. (III: Pharmacology and Toxicology)", Procter & Gamble internal report

B74. ZILLER, S.A. (1977), "Absorption, excretion and tissue distribution of 2-pyridinethiol-1-oxide", Fd. Cosmet. Toxicol., 15 (1), 49-54

C. SCCNFP references

C1. F. Pereira, C. Fernandes, M. Dias, and M. H. Lacerda. Allergic contact dermatitis from zinc pyrithione. Contact.Dermatitis. 33 (2):131, 1995.

C2. F. Brandrup and T. Menne. Zinc pyrithione (Zinc Omadine) allergy. Contact Dermatitis. 12 (1):50, 1985.

C3. R. Gonzalez Perez, A. Aguirre, J. A. Raton, X. Eizaguirre, and J. L. Diaz Perez. Positive patch tests to zinc pyrithione. Contact.Dermatitis. 32 (2):118-119, 1995.

C4. N. H. Nielsen and T. Menne. Allergic contact dermatitis caused by zinc pyrithione associated with pustular psoriasis. Am.J.Contact Dermat. 8 (3):170-171, 1997.

C5. P. K. Nigam, S. Tyagi, A. K. Saxena, and R. S. Misra. Dermatitis from zinc pyrithione. Contact.Dermatitis. 19 (3):219, 1988.

D. SCCS References

D1. Arch Chemicals (2003) cited Zinc pyrithione. Preliminary results following dermal application for 28-consecutive days. Letter from Arch Chemicals Inc to USEPA. http://yosemite.epa.gov/oppts/epatscat8.nsf/ALLIDS/F9CBE92520181B4285256D51004652BD/$FILE/88030000115.pdf?OpenElement

D2. Collum, W.D. and Winek, C.L. (1967): Percutaneous toxicity of pyridinethiones in a demethylsulfoxide vehicle. J. Pharm. Sci 56, 1673- 1675.

D3. Cloyd, G.G., Wyman, M., Shadduck, J.A., Winrow, M.J., and Johnson, G.R. (1978): Ocular toxicity studies with Zinc Pyridinethione. Tox. Appl. Pharmacol. 45, 771-782.

D4. EC (2007): EC Commission Regulation No. 1451/2007 of 4 December 2007 on the second phase of the 10 year work programme referred to in Article 16(2) of Directive 98/8/EC of the European Parliament and of the Council concerning the placing of biocidal products on the Market.

D5. Geier, J., Lessmann, H., Dickel, H., Frosch, P.J., Koch, P., Becker, D., Jappe, U., Aberer, W., Schnuch, A., Uter, W. (2004): Patch test results with the metalworking fluid series of the German Contact Dermatitis Research Group (DKG). Contact Dermatitis 51, 118–130.

D6. Geier, J., Lessmann, H., Becker, D., Bruze, M., Frosch, P.J., Fuchs, T., Jappe, U., Koch, P., Pföhler, C., Skudlik, C. (2006): Patch testing with components of water-based metalworking fluids: results of a multicentre study with a second series. Contact Dermatitis 55, 322–329.

D7. Gruvberger, B., Isaksson, M., Frick, M., Pontén, A., Bruze, M. (2003): Occupational dermatoses in a metalworking plant. Contact Dermatitis 48, 80–86.

D8. HSE (The Health and Safety Executive) (2003): Advisory committee on pesticides No 208. Evaluation on: Zinc Pyrithione: use as a booster biocide in antifouling products (http://www.pesticides.gov.uk).

D9. Hsieh, C.W., Tu, M.E., Wu, Y.H. (2010): Allergic contact dermatitis induced by zinc pyrithione in shampoo: a case report. Dermatol Sin 28, 163-166.

D10. ILSI (1994): Physiological Parameter Values for PBPK models. International Life Science Institute, Risk Science Institute, December 1994.

D11. Isaksson, M. (2002): Delayed diagnosis of occupational contact dermatitis from sodium pyrithione in a metalworking fluid. Contact Dermatitis 47, 248–249.

D12. Jo, J.H., Jang, H.S., Ko, H.C., Kim, M.B., Oh, C.K., Kwon, Y.W., Kwon, K.S. (2005): Pustular psoriasis and the Kobner phenomenon caused by allergic contact dermatitis from zinc pyrithione-containing shampoo. Contact Dermatitis 52, 142–144.

D13. Knox et al. (2004): Activation of a calcium entry pathway by sodium pyrithione in the bag cell neurons of Aplysia, J. Neurobiol 60, 411-423.

D14. Knox et al., (2008): Comparative effects of sodium pyrithione evoked intracellular calcium elevation in rodent and primate ventral horn motor neurons. Biochem. Biophys. Res. Commun 366, 48-53.

D15. Lamore, S.D., Cabello, C.M., and Wondrak, G.T. (2010): The topical antimicrobial zinc pyrithione is a heat shock response inducer that causes DNA damage and PARP-dependent energy crisis in human skin cells. Cell Stress and Chaperones 15, 309-322.

D16. Lamore, S.D. and Wondrak, G.T. (2011): Zinc pyrithione impairs zinc homeostasis and upregulates stress response gene expression in reconstructed human epidermis. Biometals 24, 875 - 890.

D17. Lansdown, A.B.G. (1991): Interspecies Variations in response to topical application of selected zinc compounds. Food Chem Toxicol 29, 57 – 64.

D18. Le Coz, C.J. (2001): Allergic contact dermatitis from sodium pyrithione in metalworking fluid. Contact Dermatitis 45, 58–59.

D19. MAK (2012) (http://onlinelibrary.wiley.com/doi/10.1002/3527600418.mb1346341d0052/pdf)

D20. Marzulli, F.N. and Maibach, H.I. (1973): Antimicrobials: experimental contact sensitization in man. J. Soc. Cosmet. Chem. 24, 399 – 421.

D21. Ross, J.F. and Lawhorn, G.T. (1990): ZPT-related distal axonopathy: behavioural and electrophysiologic correlates in rats. Neurotoxicology and Teratology 12, 153-159.

D22. Xiong, Q., Sun, H. And Li, M. (2007): Zinc pyrithione-mediated activation of voltage-gated KCNQ potassium channels rescues epileptogenic mutants. Nature Chemical Biology 5, 287-296.

D23. Yates, V.M., Finn, O.A. (1980): Contact allergic sensitivity to zinc pyrithione followed by the photosensitivity dermatitis and actinic reticuloid syndrome. Contact Dermatitis 6, 349–350.

D24. ECHA (update 2012): Guidance on information requirements and chemical safety assessment Chapter R.8: Characterisation of dose [concentration]-response for human health. Available at: http://echa.europa.eu/web/guest/guidance-documents/guidance-on-information-requirements-and-chemical-safety-assessment

E. References obtained/retrieved after the commenting period

E1. Barnett, J.F. (2005): Percutaneous Developmental Toxicity Study of Zinc Pyridinedione (ZPT) in rats. Project ID: AEN00006
Performing Laboratory: CR-DDS Argus Division, 905 Sheehy Drive, Building A, Horsham, Pennsylvania 19044-1241, USA.
Sponsor: Arch Chemicals, Inc., 350 Knotter Drive, Cheshire, Connecticut 06410-0586

E2. Blagden S.M. (1996): Zinc Pyrithione: Acute inhalation toxicity (nose only) study in the rat, Safepharm Laboratories Limited, UK, SPL Project No 369/054 (unpublished); EZPTF 6013-001

E3. Carter, L. (2004): Final Report Acute inhalation toxicity study in rats, Laboratory study number 7866-03. Submitted to Arch. Chem.

E4. Carter, L. (2005): 21-Day Inhalation Toxicity Study with a 5-Day Interim Sacrifice in Rats (OPPTS 870.3465). Conducted by Stillmeadow, Inc., 12852 Park One Drive, Sugar Land, TX 77478. Submitted to Arch Chem

E5. Charles River (Testing facility), Sponsor: Arch Chem (2010a): The in vitro Percutaneous Absorption of Radiolabelled Zinc Pyrithione in an Aqueous Dispersion Through Human and Rat Skin. Test Facility Study No. 788821, Report No. 31307.

E6. Charles River (Testing facility), Sponsor: Arch Chem (2010b): The in vivo Percutaneous Absorption of Radiolabelled Zinc Pyrithione in an Aqueous Dispersion at Two Concentrations in the Rat (OECD 427). Test Facility Study No. 192727, Report No. 31323.

E7. Cicalese, R. and Argentino-Storino, A. (2004): Natrium Pyrion Combined Chronic Toxicity/Carcinogenicity Study in Rats. RTC Study No. 7403, RTC Report No. 7403/T/401/2001
Sponsor: Rütgers Chemicals AG, Germany

E8. Diamond, G.L., Skoulis, N.P., Jeffcoat, A.R., and Nash, J.F. (manuscript dated from 2013): A physiological-based pharmacokinetic model for the antimicrobial zinc pyrithione for predicting dosimetry in humans.

E9. Drobeck, H.P., Gumaer, K.I., Owen, S.D., and Donikian, M.R. (1973): Oral administration of zinc Omadine® (WIN 9546) to rhesus monkeys. Sterling Winthrop Research Institute, Report No. PD. 2-543 (unpublished)

E10. Funato, M. (1992): A repeated dose toxicity study of zinc Omadine powder to Cynomolgus monkeys for 28 days followed by a 2-week recovery period. Shin Nippon Biomedical Laboratories, Ltd., Kagoshima, Japan; Report No. SBL 40-32 (unpublished).

E11. Husband, R.F.A., Newman, A.J. and Lee, P.N. (1991a): 104 Week Oral (gavage) Combined Carcinogenicity and Toxicity Study in the Rat.
Performing Laboratory: Toxicol Laboratories Limited, Bromyard Road, Ledbury, Herefordshire, HR8 1LH, England; Laboratory Project ID OLA/3/90
Sponsor: Olin Corporation

E12. Husband, R.F.A., Newman, A.J. and Lee, P.N. (1991b): Sodium Omadine ^{R} 80 Week Dermal Toxicity Study in the Mouse.
Performing Laboratory: Toxicol Laboratories Limited, Bromyard Road, Ledbury, Herefordshire, HR8 1LH, England; Laboratory Project ID OLA/7/90
Sponsor: Olin Corporation

E13. Jacobson-Kram, D. and Sigler, C.I. (1990) CHO/HGRPT Mutation Assay with Confirmation; Test article 2-Mercaptopyridine-N-Oxide Zinc
Performing Laboratory: Microbiological Associates, Inc., 9900 Blackwell Road, Rockville, Maryland 20850. Laboratory study number T9153.332001
Sponsor: The Olin Corporation, 91 Shelton Avenue, New Haven, CT 06511

E14. Kirkpatrick, D.T. (1990): A 4-Week Inhalation Toxicity Study with Emphasis on Pulmonary Effects of Zinc Omadine in Sprague-Dawley rats. Study number WIL-544008, performed by WIL Research Laboratories, LLC, 1407 George Road, Ashland, OH 44805-8946
Sponsor: Arch Chemicals, Inc., 350 Knotter Drive, Cheshire, CT 06410

E15. Kohn, M.C. (1995): Achieving credibility in risk assessment models. Toxicology Letters 79, 107 – 114.

E16. MAK (1994): Sodium Pyrithione

E17. Newcomb, T. (1996): Zic Omadine 48 % Dispersion, I.D.#5RC-088-024ZP Delayed Contact Dermal Sensitization Test, Unpublished Report by MB Research Laboratories, Inc., MB 96-4868.06

E18. Ohta, K. (1992a): An in vitro Chromosomal Aberration Test of Zinc Omadine® Powder on the Human Peripheral Lymphocyte.
Testing facility: Shin Nippon Biomedical Laboratories Ltd., Miyanoura, Yoshida, Kagoshima 891-13, Japan, Study number SBL 40-36.

E19. Ohta, K. (1992b): An in vivo Chromosomal Aberration Test of Zinc Omadine ® Powder on the Peripheral Lymphocyte of the Cynomolgus Monkeys.
Testing facility: Shin Nippon Biomedical Laboratories Ltd., Miyanoura, Yoshida, Kagoshima 891-13, Japan, Study number SBL 40-34.

E20. Putman, D.L. and Morris, M.J. (1990): Micronucleus Cytogenetic Assay in Mice.Test Article 2-Mercaptopyridine-N-Oxide Zinc.
Performing Laboratory: Microbiological Associates, Inc., 5221 River Road, Bethesda, Maryland 20816, Study Number T9153

E21. Ridgeway, P. and Wood, C.M. (1989): Sodium Omadine. Rat Two Generation Reproduction Toxicity Study.
Performing Laboratory: Toxicol Laboratories Limited, Bromyard Road, Ledbury, Herefordshire. HR8 1LH

E22. Ridgeway, P. (1990): Sodium Omadine Rat Peri and Post Natal Study. Alcon Laboratories Inc. Reference No. ALC/1/89
Testing Facility: Toxicol Laboratories Limited, Bromyard Road, Ledbury, Herefordshire. HR8 1LH England
Sponsor: Alcon Laboratories Inc., 6201 South Freeway, Fort Worth, Texas 76134, USA

E23. Schardein, J.L. (1993a): Developmental Toxicity Study in Rats with Zic Omadine.
Performing Laboratory: International Research and development Corporation, 500 North Main Street, Mattawan, Michigan 49071, Laboratory Project Identification 397-055

E24. Schardein, J.L. (1993b): Developmental Toxicity Study in New Zealand White Rabbits with Zinc Omadine. Performing Laboratory: International Research and Development Corporation, 500 North Main Street, Mattawan, Michigan 49071, Laboratory Project Identification 397-056

E25. Ulrich, C.E. (1991): Acute inhalation Toxicity Evaluation on Zinc Omadine in Rats. International Research and Development Corporation; Laboratory Product Identification 397-051

E26. Ulrich, C.E. (1993): 90-day subchronic dermal toxicity study on zinc Omadine ® in rats. International Research and Development Corporation, Mattawan, MI, USA, Report No. 397-057.

E27. Wagner, V.O. and Klug, M.L. (2000): Bacterial Reverse Mutation Assay with an independent Repeat Assay. Test Article Zinc Omadine.Performing Laboratory: BioReliance, 9630 Medical Center Drive, Rockville, MD, 20850, Study number AA25YE.502.BTL
Sponsor: Arch Chemicals, Inc., 350 Knotter Drive, Cheshire, CT 06410

E.28. Weiss-Fuchs, U. (2002): „Zinc Pyrion": Skin Sensitisation Study (Guinea pig maximisation test) modified according to Maurer and Hess, ARC Seibersdorf research GmbH (unpublished)

E29. Thomas, J.A. (2010): Inhalation of ^{14}C-Zinc Omadine. Study number WIL-544011, WIL Research Laboratories, LLC, 1407 George Road, Ashland, OH 44805-8946
Sponsor: Arch Chemicals, Inc., 350 Knotter Drive, Cheshire, CT 06410

E30. Reference: Barnett, J.F. (2005): Oral (Gavage) Acute Neurotoxicity Study of Zinc Pyridinedione (ZPT) in Male and Female Rats. Laboratory Project ID CR-DDS Argus Division Protocol number: AEN00002. Performing Laboratory: CR-DDS Argus Division, 905 Sheehy Drive, Building A, Horsham, Pennsylvania, 19044-1241 USA.
Sponsor: Arch Chem, Inc. 350, Knotter Drive, Cheshire, Connecticut 06410-0586.

E31: The Procter and Gamble (P&G) Company (2013): Comment on SCCS Opinion: Zinc Pyrithione – P81. Letter dated from 8 August 2013.

E32: Rudolf, E., Cervinka, M. (2011): Stress responses of human dermal fibroblasts exposed to zinc pyrithione.Toxicol Lett. 204, 164-173. doi: 10.1016/j.toxlet.2011.04.028. Epub 2011 Apr 30.

7. List of abbreviations

A:	amount
ADME:	Absorption, distribution, metabolism, excretion
ATP:	Adenosine triphosphate
AUC:	area under the curve
BALF:	broncho-alveolar lavage fluid
Br:	brain
CA:	arterial concentration
C_{max}:	maximal concentration
Cl:	clearance
CLP:	Classification, Labelling, Packaging
DMSO:	demethyl sulphoxide
ECHA:	European Chemicals Agency
F:	fraction
GLP:	good laboratory practice
HRIPT:	Human Repeat Insult Patch Test
HSE:	Health and Safety Executive
ICP-MS:	Inductively coupled plasma mass spectrometry
K:	rate constant
K_{el}:	elimination rate constant
LC/MS:	liquid chromatography/mass spectrometry
MAK:	Maximale Arbeitsplatz Konzentration (maximum workplace concentration)
MMAD:	mass median aerodynamic diameter
MSP:	2-(methyl)sulfonyl pyridine
Mu:	muscle
NaPT:	sodium pyrithione
PBPK:	physiologically-based pharmacokinetic modeling
PK:	pharmacokinetic
PT:	pyrithione
PTC:	pyrithione carbon
Q:	plasma flow
QAU:	Quality assurance unit

R:	rapidly perfused tissue
RBC:	red blood cells
REACH:	Registration, Evaluation, Authorisation and Restriction of Chemicals
S:	slowly perfused tissue
SCCNFP:	Scientific committee on cosmetic products and non-food products
SG:	S-glutathione
T_{max}:	time of maximal concentration
UDS:	unscheduled DNA synthesis
UR:	urine
V:	volume
Vd:	distribution volume
ZPT:	zinc pyrithione