European Union Risk Assessment Report

TRIS (NONYLPHENYL) PHOSPHITE

CAS-No.: 26523-78-4
EINECS-No.: 247-759-6

RISK ASSESSMENT

Cataloguing data can be found at the end of this publication

Luxembourg: Office for Official Publications of the European Communities, 2002
ISBN (insert number here)

Printed in Italy

TRIS(NONYLPHENYL) PHOSPHITE

CAS No.: 26523-78-4
EINECS No.: 247-759-6

RISK ASSESSMENT

Draft of February 2007

France

Rapporteur for the risk assessment of tris(nonylphenyl) phosphite is the Ministry of Ecology and Sustainable Development as well as the Ministry of Employment and Social Affairs in co-operation with the Ministry of Public Health. Responsible for the risk evaluation and subsequently for the contents of this report is the rapporteur.
The scientific work on this report has been prepared by :

Environment risk assessment

National Institute for Industrial Environment and Risks (INERIS)
Direction of chronic risks
Parc Technologique Alata
BP n°2
60550 Verneuil-en-Halatte

Human health risk assessment

Effect assessment, exposure and risk characterisation for consumers

National Institute for Industrial Environment and Risks (INERIS)
Direction of chronic risks
Parc Technologique Alata
BP n°2
60550 Verneuil-en-Halatte

Exposure and risk characterisation for workers

Institut National de recherche et Sécurité (INRS)
Département Risques Chimiques et Biologiques
30, rue Olivier Noyer
75680 Paris cedex 14

Date of Last Literature Search :	**[please insert year]**
Review of report by MS Technical Experts finalised:	**[please insert month and year]**
Final report:	**[please year]**

Foreword

We are pleased to present this Risk Assessment Report which is the result of in-depth work carried out by experts in one Member State, working in co-operation with their counterparts in the other Member States, the Commission Services, Industry and public interest groups.

The Risk Assessment was carried out in accordance with Council Regulation (EEC) 793/93[1] on the evaluation and control of the risks of "existing" substances. "Existing" substances are chemical substances in use within the European Community before September 1981 and listed in the European Inventory of Existing Commercial Chemical Substances. Regulation 793/93 provides a systematic framework for the evaluation of the risks to human health and the environment of these substances if they are produced or imported into the Community in volumes above 10 tonnes per year.

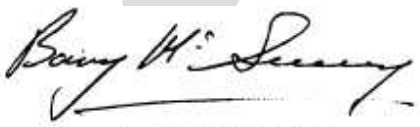
There are four overall stages in the Regulation for reducing the risks: data collection, priority setting, risk assessment and risk reduction. Data provided by Industry are used by Member States and the Commission services to determine the priority of the substances which need to be assessed. For each substance on a priority list, a Member State volunteers to act as "Rapporteur", undertaking the in-depth Risk Assessment and recommending a strategy to limit the risks of exposure to the substance, if necessary.

The methods for carrying out an in-depth Risk Assessment at Community level are laid down in Commission Regulation (EC) 1488/94[2], which is supported by a technical guidance document[3]. Normally, the "Rapporteur" and individual companies producing, importing and/or using the chemicals work closely together to develop a draft Risk Assessment Report, which is then presented at a Meeting of Member State technical experts for endorsement. The Risk Assessment Report is then peer-reviewed by the Scientific Committee on Toxicity, Ecotoxicity and the Environment (CSTEE) which gives its opinion to the European Commission on the quality of the risk assessment.

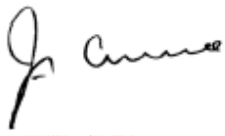
If a Risk Assessment Report concludes that measures to reduce the risks of exposure to the substances are needed, beyond any measures which may already be in place, the next step in the process is for the "Rapporteur" to develop a proposal for a strategy to limit those risks.

The Risk Assessment Report is also presented to the Organisation for Economic Co-operation and Development as a contribution to the Chapter 19, Agenda 21 goals for evaluating chemicals, agreed at the United Nations Conference on Environment and Development, held in Rio de Janeiro in 1992.

This Risk Assessment improves our knowledge about the risks to human health and the environment from exposure to chemicals. We hope you will agree that the results of this in-depth study and intensive co-operation will make a worthwhile contribution to the Community objective of reducing the overall risks from exposure to chemicals.



Barry Mc Sweeney
Director-General
Joint Research Centre



J. Currie
Director-General
Environment, Nuclear Safety and Civil Protection

[1] O.J. No L 084 , 05/04/199 p.0001 – 0075
[2] O.J. No L 161, 29/06/1994 p. 0003 – 0011
[3] Technical Guidance Document, Part I – V, ISBN 92-827-801 [1234]

0 OVERALL RESULTS OF THE RISK ASSESSMENT

CAS Number: 26523-78-4
EINECS Number: 247-759-6
IUPAC Name: Phenol, nonyl-, phosphite (3:1)

Environment

To be updated

Human health effects assessment

Risk assessment of human exposed via the environment was not discussed and will be updated following the update of environment risk assessment.

( ) **(i)** There is a need for further information and/or testing.

(X) **(ii)** There is at present no need for further information and/or testing and no need for risk reduction measures beyond those, which are being applied already.

This conclusion applies to the assessment of the risk to human health through consumer exposure.

(X) (iii) There is a need for specific measures to limit the risks.

This conclusion applies to the assessment of the risk to human health through worker exposure. It is reached because of concerns for sensitisation as a consequence of dermal exposure arising during manufacture of the substance, manufacture of products or use of preparations containing TNPP.

CONTENTS

TABLES

1 GENERAL SUBSTANCE INFORMATION

Parts of this section will be updated in the next version of the environmental risk assessment.

1.1 IDENTIFICATION OF THE SUBSTANCE

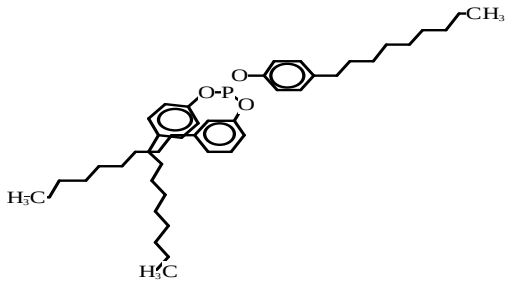
CAS No: 26523-78-4

EINECS No: 247-759-6

IUPAC Name: Phenol, nonyl-, phosphite (3:1)

Molecular formula: $C_{45}H_{69}O_3P$

Structural Formula:



Molecular weight: 689 $g.mol^{-1}$

Synonyms and tradenames: Alkanox TNPP, Lowinox TNPP, Irgafos TNPP, Tris(monononylphenyl)phosphite, Tri(nonylphenyl)phosphite, Weston 399, Weston TNPP, Irgastab CH 55, Naugard TNPP, Polygard, Polygard HR, Polygard LC, TNPP, Trisnonylphenylphosphit.

In this assessment, the name Tris(nonylphenyl)phosphite (TNPP) will be used for the substance as this is the most common name.

1.2 PURITY/IMPURITIES, ADDITIVES

1.2.1 Purity

The purity of TNPP is reported as ca. 95 – 100% w/w.

The following impurities may be found in TNPP :

- Nonylphenol (CAS 25154-52-3) < 5% w/w,
- Phenol (CAS 108-95-2) < 1% w/w,
- Di(nonylphenyl)phenylphosphite (CAS 25417-08-7) 0.05% w/w,
- Chlorine (CAS 7782-50-5) 0.005% w/w.

1.2.2 Additives

1,1',1''-nitrilotripropan-2-ol (CAS No: 122-20-3) is an additive that may be found in TNPP in the proportion of 0.5 to 1% w/w.

1.3 PHYSICO-CHEMICAL PROPERTIES

1.3.1 Physical state (at ntp)

TNPP is a viscous liquid at room temperature.

1.3.2 Melting point

Instead of a melting point, a pour point of 6°C ± 3°C was determined (Reimer&Associates, 2001e). A melting point could not be observed using the differential scanning calorimetric (DSC) method because an endothermic event was not observed in the heat flow *vs* temperature plot. The pour point (the lowest temperature at which the test substance is first observed to flow on warming) is an appropriate measurement for viscous liquid substances. The test was conducted according to ASTM Method D97, as recommended in the OECD 102 guideline.

1.3.3 Boiling point

The boiling point was reported as >303°C (Reimer&Associates, 2001f). The test method was based on OECD 103 guideline. Bubbling was observed for the first 1 to 2 seconds of heating, and then stopped. This was probably due to the boiling of a minor component (<0.1%) present in the test substance. Consequently a new study was undertaken to assess the true boiling point. The TNPP producers have determined that TNPP will begin to degrade before boiling. According to a Thermal Gravimetric Analysis (TGA) of TNPP, the phosphite has an onset of degradation at 322°C under nitrogen.

1.3.4 Relative density

The relative density has been quoted at 0.98 $g.cm^{-3}$ at 20°C (Crompton, 2003).

1.3.5 Vapour pressure

A vapour pressure was estimated using structure activity relationships models developed by the U.S. Environmental Protection Agency and Syracuse Research Corporation (EPIWIN, v. 3.10, US EPA and Syracuse Research Corporation, 2001). The vapour pressure was estimated to 5.10^{-12} Pa (Staples, 2001).

Another value of 0.047 Pa at 20°C was extrapolated from results obtained by isoteniscope (method ASTM D2879) at temperatures ranging from 125 to 375 °C (Phoenix_Chemical_Laboratory, 1997). These measured values are displayed in Table 1-1.

Table 1-1: Vapour pressure data for TNPP

Temperature °C	Vapour Pressure (Pa)
125	22.7
150	65.3
175	160
200	373
225	747
250	1533
275	2800
300	4666
325	8133
350	15330
375	65330

A strong relation between the temperature (1/T) and the vapour pressure is found. Excluding the last value measured at 375°C, the plot of the above results gives a linear regression with a good reliability (see figure below). Vapour pressures of respectively 0.039 Pa and 0.058 Pa at 20°C and 25°C could be derived from this equation. These results are consistent with the extrapolated value of 0.047 Pa at 20°C found in the study summary in the IUCLID file.

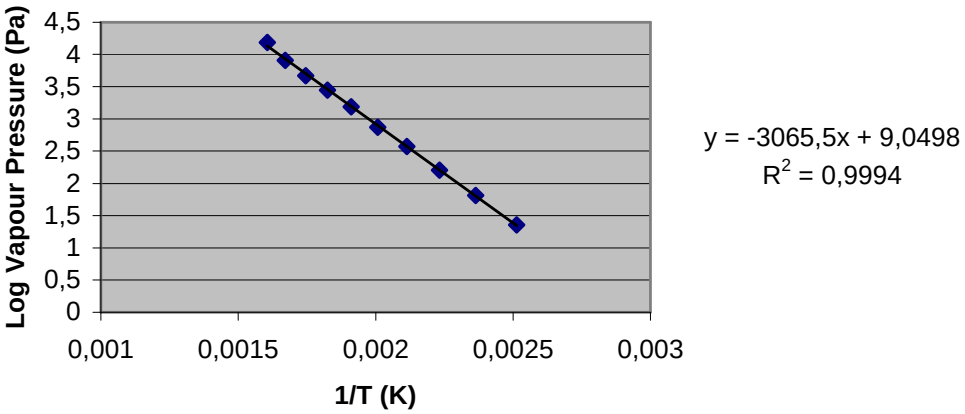


Figure 1-1: Linear regression between the temperature (1/T) and the measured vapour pressures

The isoteniscope method is recommended for the measurement of vapour pressures between 10^2 and 10^5 Pa. The extrapolated value is three orders of magnitude below this range. Consequently, the value of 0.058 Pa at 25°C would need to be confirmed by another vapour pressure result.

The modified Watson correlation method was also used for the estimation of TNPP vapour pressure. According to this method the vapour pressure can be calculated using the boiling point value in the following equation:

Equation 1-1: Calculation of the vapour pressure according to the modified Watson correlation method

$$\ln P_{V_P} = \frac{\Delta H_{VB}}{\Delta Z_b R T_b}\left[1 - \frac{(3-2\frac{T}{T_b})^m}{T/T_b} - 2m(3-2\frac{T}{T_b})^{m-1}\ln\frac{T}{T_b}\right], \text{ with } \frac{\Delta H_{VB}}{T_b} = K_F(8.75 + R\ln T_b)$$

The parameters were chosen as follow:

Table 1-2: Parameters used for the vapour pressure calculation according to the modified Watson correlation method

Parameter	Value	Remark
K_F	1.06	Default
T_b	322°C or 595.15 K	Boiling point, see 1.3.3
ΔZ_b	0.97	Default estimation
T	25°C or 298.15 K	Chosen temperature for subsequent modelling stages
m	0.19	Default value for liquids
R	1.9859 $cal.mol^{-1}.K^{-1}$	Gas constant (8.314 $Pa.m^3.mol^{-1}.K^{-1}$)

The calculation gives a vapour pressure of $3.86.10^{-7}$ atm corresponding to 0.039 Pa. The main drawback of this estimation lies in the fact that the method employed only used the boiling point as experimental data entry and the only data available is the temperature of degradation of the substance: 322°C.

On one hand, the extrapolated value of 0.058 Pa has been calculated based on vapour pressures measured by the isoteniscope method which is not recommended for this range of vapour pressure. On the other hand, the vapour pressure has been calculated with the modified Watson correlation method using the temperature of degradation of the substance as a boiling point. Both methods give similar results.

Finally, the value of 0.058 Pa at 25°C, extrapolated from measured vapour pressures at higher temperatures and confirmed by an estimation method, will be used in the risk assessment.

1.3.6 n-octanol / water partition coefficient

The n-octanol-water partition coefficient was estimated using structure activity relationships models developed by the U.S. Environmental Protection Agency and Syracuse Research Corporation (EPIWIN, US EPA and Syracuse Research Corporation, 2001). The log P_{ow} was estimated to 20.05 (US EPA and Syracuse Research Corporation, 2001).

According to Reimer & Associates, 2001c, it was not appropriate to conduct the partition coefficient measurement because the solubility of TNPP in water was too low (see section 1.3.7) and TNPP was also found to be hydrolytically unstable. The n-octanol / water partition coefficient was therefore calculated using the software from Advanced Chemistry Development Inc. ("ACD/LogP DB"). The result of the calculation was found to be 21.6 ± 0.6 (Reimer&Associates, 2001c).

The annex of the OECD guideline 117 presents some K_{ow} calculation methods that can be used to "provide an estimate when experimental methods cannot be applied". However there are some limitations to the use of such methods. First, the reliability of calculation methods decreases as the complexity of the compound under study increases. Here, TNPP could be classified as a rather complex molecule with a high molecular weight and several functional groups. The domain of application of K_{ow} calculation methods is characterised in terms of chemical structures. For example, some calculation programs cannot be applied to the estimation of Kow for phosphorus compounds including phosphites. Second, the domains of the models is also restricted by the log K_{ow} range of their applicability. In general, clear estimates can be expected

in the region of log K_{ow} 0-5. Some programs have shown good estimates for compounds with log K_{ow} > 5 but estimates for log K_{ow} around 10 or above should be considered rather as qualitative than quantitative information (TGD, Part III, Chapter 4, E.C., 2003).

Considering the high hydrophobic potential of TNPP which contains 27 aliphatic and 18 aromatic carbons, a high log Kow value could be expected for this compound. However, in the absence of other data, the highest recommended value of 8 will be used in EUSES model (E.C., 2004a).

1.3.7 Water solubility

A water solubility was estimated using structure activity relationships models developed by the U.S. Environmental Protection Agency and Syracuse Research Corporation (EPIWIN, US EPA and Syracuse Research Corporation, 2001). The water solubility was estimated to $1.3.10^{-15}$ $mg.L^{-1}$ (Staples, 2001).

Experimental water solubility was determined by (Reimer&Associates, 2001a). The flask method based on OECD Guideline 105 was used. TNPP was not detected in the saturated aqueous test solution. Therefore it is concluded that the water solubility of TNPP is below the detection limit of the substance. This detection limit was estimated to be 0.6 $mg.L^{-1}$, the lowest TNPP concentration that produced a signal that is reliably distinguished from the background signal as determined from chromatograms of TNPP solutions. Therefore, the water solubility of TNPP is < 0.6 $mg.L^{-1}$ at 24°C.

It was not possible to determine a more accurate result. As a matter of fact, no saturated solution could be obtained during the experiment because of the rapid hydrolysis of TNPP in water. The experimental procedure consisted in mixing TNPP in deionized water for at least 24 hours. Half life of TNPP being estimated to 13 hours, it can be supposed that all the TNPP is degraded during the first day.

In this risk assessment report, when a water solubility result is needed for environmental modelling purposes, the value of 0.6 $mg.L^{-1}$ will be retained as a worst case estimation. However, it will be also considered that TNPP is almost instantly degraded into nonylphenol and phosphorous acid when released into aquatic compartments.

1.3.8 Flash point

Values of 183°C (internal reference, Great Lakes Chemical, Italia, Milan) and 195°C (Ciba MSDS) were reported using closed cup methods.

Besides, a value of 207°C was reported using the Pensky-Martin apparatus (closed cup) (Pittsburgh_Testing_Laboratory, 1978). This last value will be retained in this risk assessment because the analytical report was available.

1.3.9 Autoflammability

In a MSDS by Uniroyal, a value of 268°C was quoted. Moreover, using the Setchkin method, a result of 440°C was found (United States Testing Company, 1990).

1.3.10 Explosivity

No result could be found in the literature on any explosion limit. However, on the basis of its chemical structure, TNPP is not expected to have explosive properties.

1.3.11 Oxidising properties

No oxidising property was reported for TNPP (internal reference, Great Lakes Chemical, Milan, Italia).

1.3.12 Viscosity

In a product information sheet, a value of 6000 cps at 25°C is quoted (Crompton, 2003). Other values are also presented in this document showing that the viscosity goes from 15000 cps at 15°C to 18 cps at 120°C. The value at 25°C will be retained for the risk assessment.

1.3.13 Henry's Law constant

The Henry's law constant was estimated using structure activity relationships models developed by the U.S. Environmental Protection Agency and Syracuse Research Corporation (EPIWIN, v. 3.10, sub-model HENRYWIN, US EPA and Syracuse Research Corporation, 2001). At 25°C, a value of 66.1 $Pa.m^3.mol^{-1}$ was calculated (US EPA and Syracuse Research Corporation, 2001).

The Henry's law constant can also be estimated from the ratio of the vapour pressure to the water solubility (E.C., 2003):

$$HENRY = \frac{VP \cdot MOLW}{SOL}$$

Using a vapour pressure of 0.058 Pa, a molecular weight of 689 $g.mol^{-1}$ and a water solubility of 0.6 $mg.L^{-1}$ the Henry's Law constant is equal to 66.6 $Pa.m^3.mol^{-1}$.

This result is coherent with the QSAR calculation above so the value of 66.6 $Pa.m^3.mol^{-1}$ will be retained in this risk assessment.

1.3.14 Summary of physico-chemical properties

The physico-chemical properties of TNPP used in this risk assessment are summarised in the following table:

Table 1-3: Physical and chemical properties of the TNPP

Property	Value	Comments
Physical state at ntp	Viscous liquid	
Molecular weight	689 $g.mol^{-1}$	
Melting Point	6°C ± 3°C	Instead of a melting point, a pour point (more appropriate to viscous liquids) was determined
Boiling Point	322°C	Degradation
Relative density	0.98 $g.cm^{-3}$	
Vapour pressure	0.058 Pa at 25°C	extrapolated from results obtained by isoteniscope (method ASTM D2879)
Partition coefficient	Log Kow = 21.6 Log Kow = 8 (EUSES)	Calculated with software ACD/LogP DB
Water solubility	<0.6 $mg.L^{-1}$	A saturated solution was not obtained and the water solubility result corresponds to the detection limit of the analytical method.
Flash point	207°C	Pensky Martin apparatus (closed cup)
Autoflammability	440°C	Setchkin method
Oxidising properties	No oxidising property	
Henry's law constant	66.6 $Pa.m^3.mol^{-1}$	TGD calculation

1.4 CLASSIFICATION

1.4.1 Current classification

TNPP chemical is not classified under Annex I of Directive 67/547 EEC.

1.4.2 Proposed classification

Human health effects (adopted classification)

Classification was finalised in the Commission working group on the Classification and Labelling of Dangerous Substances in November 2005 (human health) :

Symbol : Xi
R-phrase : R43 : May cause sensitization by skin contact.

Environmental effects

To be updated
.

2 GENERAL INFORMATION ON EXPOSURE

This section will be updated in the next version of the environmental risk assessment.

2.1 PRODUCTION

TNPP is produced all over the world: Unites States, Europe, India, Korea, Russia, China, etc. (Chemical Information Services, 2002). Three facilities are currently producing TNPP in Europe. On the other hand, the major source of TNPP to Europe is from the United States.

2.1.1 Production process

The manufacturing processes used to produce TNPP are reasonably similar in the various plants in the US and Europe. Figure 2-1 is providing an overview of a typical production process.

TNPP production is carried out in a closed system where nonylphenol (NP) and phosphorus trichloride (PCl_3) are added to the reactor (ca. 3 :1) and held at greater than 110°C to ensure all the PCl_3 is consumed. The HCl by-product is vented to an absorber. The HCL by-product can be filtered and stored for sale or use in other processes. Excess nonylphenol is stripped from the product. The stripped nonylphenol can be recycled. The product TNPP in the reactor after stripping is pumped to a storage tank for packaging and sale. The product may be packaged into drums, isotaners, rail cars, or tank trunks.

Environmental release and exposure

The process is fully automated (computer controlled) in a closed system. The reactor is operated under 3-5 lbs (1.4 – 2.3 kg) of pressure. The vacuum pump vent is the only potential process release to the atmosphere, and it is passed through a carbon filter. The storage tank is kept under nitrogen preventing release to the atmosphere. Nitrogen is also used during transfer and packaging.

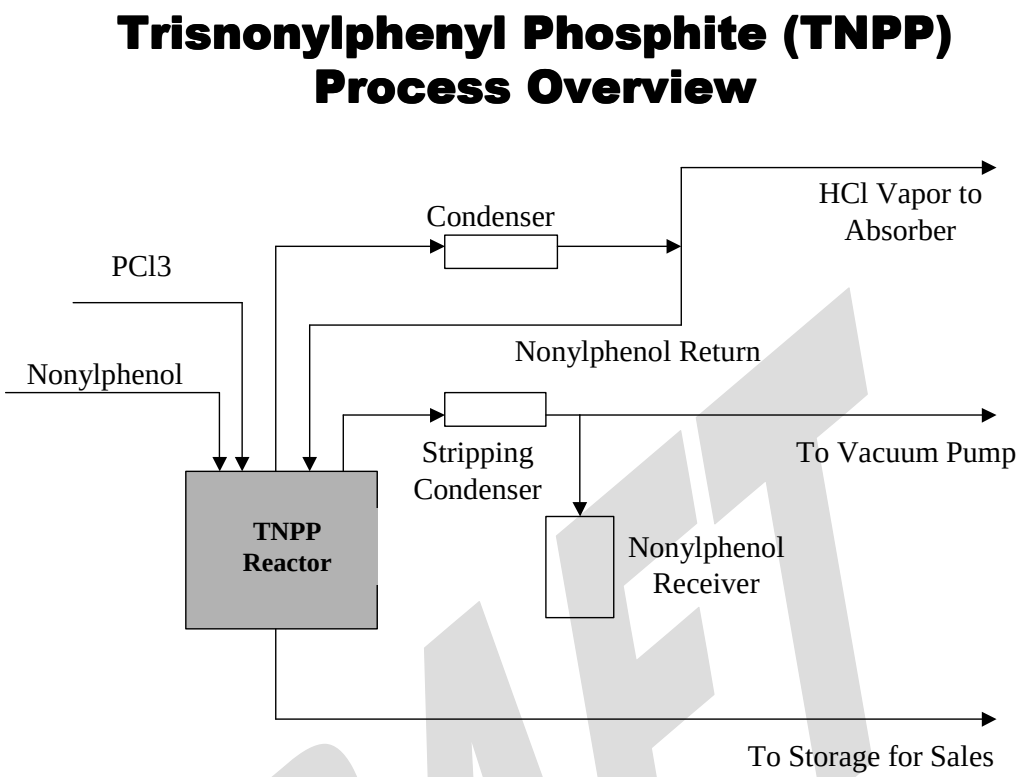


Figure 2-1: Process overview of tris(nonylphenyl)phosphite (TNPP) production

2.1.2 Production capacity

European and North American TNPP producers are organised under the Alkylphenols and Ethoxylates Research Council (APERC), a not-for-profit trade association, whose members have commercial interest in nonylphenol, octylphenol, and derivatives produced from these compounds. Information on production and imports of TNPP in Europe were provided by APERC TNPP Consortium. Hardly any individual volume was provided for each producer/importer.

Three facilities are currently producing TNPP in Europe. A fourth facility ceased TNPP production in 2001. Between 1990 and 1997, the production + import volumes were around 5,000 – 10,000 t/year.

Information is available on the combined estimate of TNPP produced within Europe and imported into Europe over the last three years:

- 1999 – approximately 5,565 tonnes
- 2000 – approximately 5,700 tonnes
- 2001 – approximately 6,800 tonnes

As this information is provided by the APERC TNPP Consortium, it cannot be excluded that these volumes do not take into account shipments of product from producers in other parts of the world than Europe and North America. However, according to the APERC TNPP Consortium, the quantity of TNPP from non-TNPP Consortium companies are not expected to be significant.

European production plants have also reported their production volumes for the year 2001. Imported volume for the same year is also available. Consequently, a total volume in Europe of 8,000 t. calculated with all 2001 data will be used in this report.

2.2 USES

2.2.1 Introduction

TNPP is used as a stabiliser in the processing of various plastic and rubber products. They are used with hindered phenolic antioxidants in plastic food packaging. In the stabilisation process, TNPP is gradually oxidised and nonylphenol is released (Building Research Establishment Ltd., 2001).

TNPP is also used as a secondary antioxidant in polymer formulations (Ullmann, 1985).

About 25 to 35 facilities are processing TNPP in Europe. Their consumption ranges from a few tonnes to over 400 tonnes/year.

An estimate of the breakdown of TNPP uses was developed based on an informal survey of North American and European manufacturers. Quantitative breakdown of TNPP uses are given in Table 2.1. The information pertains to sales of TNPP in 1999. It is expected that the breakdown of uses from the 1999 sales statistics is typical for the current year. Corresponding volumes are calculated using the total tonnage of 8,000 t.

Table 2-1: Typical quantitative breakdown of TNPP Uses

	Percentage of tonnage	Volume (tonnes)	Industrial Category / Use Category
Polyvinylchloride (PVC) film	35%	2,800	IC 11 / UC 49
Polyolefins linear low density polyethylene (LLDPE)	15%	1,200	IC 11 / UC 49
High density polyethylene (HDPE)	10%	800	IC 11 / UC 49
Rubber	37%	2,960	IC 11 / UC 49
Other/Unknown	3%	240	IC 55 / UC 0
TOTAL	100%	8,000	

In the SPIN Database (Substances in Preparations in Nordic Countries), the following industrial uses are described:

Table 2-2: Industrial uses of TNPP in the Nordic Countries (in Tonnes)

	1999[1]	2000[2]	2001[3]
Manufacture of chemicals and chemical products	156	27	< 0.1
Manufacture of rubber and plastic products	38	105	n. i.
Manufacture of furniture; manufacturing n.e.c.	n. i.	0.4	0.1
Manufacture of fabricated metal products, except machinery and equipment	n. i.	0.2	0.1
Construction	n. i.	0.2	0.1
Manufacture of wood and products of wood and cork, except furniture; manufacture of articles of straw and plaiting materials	< 0.1	< 0.1	0.1
Total	194	132.8	0.4

n. i.: not indicated

[1]: Information was available for Sweden only

[2]: Information was available for Sweden, Denmark and Norway

[3]: Information was available for Denmark and Norway.

TNPP is also mentioned in the following industrial categories: publishing, printing and reproduction of recorded media / sale, maintenance and repair of motor vehicles and motorcycles; retail sale of automotive fuel / manufacture of other transport equipment n.e.c. However, the volumes used in such industries could be considered as negligible (> 0.1 t/y in each country).

Besides, the following use pattern is described in the SPIN database:

Table 2-3: Use pattern of TNPP in the Nordic Countries (in Tonnes)

	1999[1]	2000[2]	2001[3]
Stabilizers	118	120	n.i.
Intermediates	-	1	n. i.
Others	1	1	n. i.
Adhesives, binding agents	n. i.	0.5	< 0.1
Paints, lacquers and varnishes	< 0.1	0.3	< 0.1
Fillers	< 0.1	> 0.1	0.2
Total	119	122.8	0.2

n.i.: not indicated

[1]: Information was available for Sweden only

[2]: Information was available for Sweden, Denmark and Norway

[3]: Information was available for Denmark and Norway.

TNPP is also mentioned in the following use categories: lubricants and additives / reprographic agents. However, the volumes used in such applications could be considered as negligible (> 0.1 t/y in each country).

From these tables, it could be stated that TNPP is mainly used as a stabiliser for the manufacture of rubbers and plastic products. The breakdown of TNPP uses described in Table will be used in this risk assessment.

2.2.1.1 Industrial use

Formulation and processing steps are necessary to manufacture plastic and rubber products. Formulation could be defined as the stage where TNPP is combined in a process of blending and mixing into a polymer or into another material while during the processing step, the TNPP containing material is formed. It is not known to what extent formulation and processing may occur at the same site. In the rubber industry, these two steps can often not be viewed separately (E.C., 2003, Emission Scenario Document for IC 15: others: rubber industry).

Therefore, as a worst assumption, formulation and processing stages will be assumed to occur at one site for every uses.

Without any specific information, it could be considered that TNPP is used for polymer processing, in the sub-category “processing of thermoplastics” as a processing aid. This categorisation will be used in the risk assessment for the determination of the default releases factors.

Besides, for plastic and rubber products, stages of private use and recovery may be considered. However, no specific information is available on the possible releases of TNPP during these stages.

All calculations will be performed using EUSES default parameters and, when available, emission factors issued from the emission scenario document on plastics additives (OECD, 2004).

2.2.1.2 Production of Polyvinylchloride (PVC) film

PVC containing TNPP may be used in many products like shower curtains, floorings and wall coverings.

2.2.1.3 Production of Polyolefins linear low density polyethylene (LLDPE)

LLDPE films containing TNPP are used for the manufacture of bags and food packaging. Many national regulations are covering the use of TNPP in food contact materials (Table 2-4

Table 2-4: Global food contact regulations specific to TNPP

Country	Regulation
USA	Food and Drug Administration (FDA) – 21 CFR Part 178.2010
Japan	Self-restrictive Requirements on Food-Contact Articles Japan, Hygienic Olefin and Styrene Plastics Association (JHOSPA) (March 1996), Section A4-2, maximum 1.2%
European Union	Plastics Directive 2002/72/EC, pm/ref. No. 74400, specific migration limit 30 mg/kg
Germany	BfR Recommendation VI, maximum 2.0% total of all stabilisers BGA: maximum 6% in plastics
Netherlands	Food Packaging and Utensils Decree of 01.10.1979 as amended Chapter 1
France	Brochure 1227 (Avril 1990) maximum 1.0%
Italy	Min. Decree of 21.03.1973 maximum 0.3% Min. Decree of 0.04.1985
Spain	Royal Decree 125/1982 of 30.04.1982 Resolution of 4.11.1982
Belgium	Royal Decree of 11.05.1992, specific migration limit 30 mg/kg
United Kingdom	BIBRA/BBF Code of Practice (1991) Rec. No. C.159, maximum 1.0%

2.2.1.4 Production of High density polyethylene (HDPE)

HDPE containing TNPP is used in the manufacture of many products like blow-molded plastic drums or outer wrapping (film) of cigarette boxes or tea boxes.

2.2.1.5 Production of rubber

Rubber containing TNPP are used for example in tires and shoes soles.

2.2.2 Other applications

TNPP is used in other applications than plastic and rubber productions. Using the information provided in the SPIN database, it could be supposed that these other applications include the use of TNPP in publishing, printing and reproduction activities, in the manufacture of products of wood, of fabricated metal products, of furniture and in the construction activities. However, no more specific information is available.

2.2.2.1 Use of end-products

Shower curtains, flooring and wall coverings, bags and food packaging, blow-molded plastic drums, outer wrapping films, tires and shoes soles are examples of plastic and rubber end-products containing TNPP. For all these products, both private and professional end-uses may happen. As a worst case, private use will be considered for all uses in the EUSES program (E.C., 2004b). However, it could be expected that TNPP or NP releases due to the use of end-products are negligible.

2.2.2.2 Recovery and disposal

No information on recovery has been submitted. In view of the end-products containing TNPP that are manufactured, it could be assumed that products containing TNPP may be either recycled into new products, disposed in landfill or incinerated. Therefore, this stage could be considered in the EUSES calculation (E.C., 2004a). However, no default value is actually available for this stage in version 2.0 of the software.

2.3 TRENDS

Releases of TNPP and or NP (nonylphenol) to the environment occur during production, transport, storage, formulation and processing of plastic and rubber products. In addition, releases may also take place through the uses of the end-products. Finally, waste disposal of the end-products may also release TNPP or NP into the environment.

The different industry categories (IC), use categories (UC) and main categories (MC) used in the EUSES calculations are described in Table 2-5

Table 2-5: Industrial Categories (IC), Use Categories (UC) and Main categories (MC) used in EUSES calculations

Life cycle stages		IC	UC	MC	A-Table	B-Table
Production		11	49	I b	A 1.1	B 1.4
PVC films (2,800 t)	**Formulation**	11	49	III	A 2.1	B 2.3
	Processing	11	49	II	A 3.11	B 3.9
LLDPE films (1,200 t)	**Formulation**	11	49	III	A 2.1	B 2.3
	Processing	11	49	II	A 3.11	B 3.9
HDPE films (800 t)	**Formulation**	11	49	III	A 2.1	B 2.3
	Processing	11	49	II	A 3.11	B 3.9
Rubber (2,960 t)	**Formulation**	11	49	III	A 2.1	B 2.3
	Processing	11	49	II	A 3.11	B 3.9
Others (200 t)	**Formulation**	15	55	III	A 2.1	B 2.3
	Processing	15	55	II	A 3.16	B 3.14

For tonnage input in the B tables, regional tonnage of TNPP was set to 700 t for the uses for PVC, LLDPE and rubber (maximum reported consumption range for TNPP processing facilities). For the uses in HDPE and other uses, the regional tonnage was respectively set to 800 t and 240 t.

A default fraction of TNPP in formulation is suggested in TGD (E.C., 2003) Emission Scenario Document for rubber Industry: up to 1.5 % (wt) for processing aids used as stabilisers. However, TNPP manufacturers have submitted better approximations of this value, for different formulated products (Personal communication from TNPP consortium, 1st April 2004):

- PVC film 0.8-1.5 %
- Polyolefins 0.1-0.2 %
- Rubber 0.4-1.0 %

As a worst case, the upper limit of these intervals will be used for the exposure assessment. Then, as a worst case too, fractions of the main source and number of days are derived from Tables B using the tonnage as such for each use.

3 ENVIRONMENT

This part was not provided as it will be updated.

4 HUMAN HEALTH

4.1 HUMAN HEALTH (TOXICITY)

4.1.1 Exposure assessment

4.1.1.1 General introduction

TNPP is a viscous liquid with a very low pressure (0.058 Pa at 25°C). It is produced by two companies in Europe. It is used as a stabilizer in plastic (PVC, LLDPE, HDPE) and rubber for the manufacture of many products like :

- shower curtains, floorings, wall coverings (PVC)
- bags for food packaging (LLDPE)
- drums, outer wrapping of cigarette or tea boxes (HDPE)
- tyre and shoe soles (rubber).

According to the TNPP manufacturers, the maximum amount of TNPP in the polymers is 1.5% (0.8-1.5 % in PVC film, 0.1-0.2 % in polyolefins, 0.4-1.0 % in rubber).

Data extracted by INRS in 2004 from the French product register SEPIA showed that 16 preparations out of the 48 000 registered between 1984 and 2004 contained TNPP. Theses preparations are mainly resins or resin based adhesives. TNPP is always present at a low concentration (< 1 %). Use of TNPP in preparations seems to be a very minor use.

Humans may be exposed to TNPP at workplace, via consumer products and indirectly via the environment. The highest potential exposure is likely to occur during occupational exposure.

Workers are primarily exposed via inhalation and dermal routes. For consumers, the oral route via food contact materials is the most likely.

4.1.1.2 Occupational exposure

Definitions and sources

In this document, unless otherwise stated, the term exposure is used to denote external personal exposure, assessed without taking into account the attenuating effect of any personal protective equipment (PPE) which might have been worn. This definition permits the effects of controls, other than PPE, to be assessed and avoids the considerable uncertainty associated with attempting to precisely quantify the attenuation of exposure brought about by the proper use of PPE. Furthermore, inappropriate use of gloves may even increase dermal uptake.

The estimates generated in this exposure assessment are considered to be worst-case estimates, as they describe high-end or maximum exposures in feasible but not unrealistic situations. They are not intended to account for extreme or unusual use scenarios. The majority of exposures are expected to be well below these estimates.

Since no measured exposure data are available, the assessment of inhalation and dermal exposure is based on model estimates according to the EASE model (Estimation and Assessment of Substance Exposure, version 2). This model is a general purpose predictive model for workplace exposure assessments. It is an electronic, knowledge based, expert system which is used where measured exposure data is limited or not available. It is in widespread use across the European Union for the occupational exposure assessment of new and existing substances.

All models are based upon assumptions. Their outputs are at best approximate and may be wrong. EASE is only intended to give generalised exposure data; it predicts inhalation exposure as ranges for concentrations for continuous exposure at the process under consideration. Dermal exposure is provided by EASE as the quantity of a product adhering to the skin due to a task.

Core exposure information submitted by industry for this assessment was very limited. Therefore most of the parameters chosen to characterise exposure are assumptions based on the knowledge of the assessor about the general circumstances of exposure in the relevant activities. The outcome should be regarded then as a very rough exposure assessment. Given the toxicity profile of TNPP, there is no need to allocate additional effort to generating a detailed assessment of workplace exposure at this initial stage. If there are reasonable grounds for concern for human health at a later stage, the assessment will have to be refined.

Routes of exposure and relevant scenarios

The major occupational routes of exposure to TNPP are inhalation and skin contact. Assuming proper hygiene measures are applied, oral exposure would normally not occur in the workplace.

Exposure may occur during manufacture of TNPP and during handling and further processing in the polymer industry. The following scenarios are regarded as relevant :

- scenario 1 : manufacture of TNPP
- scenario 2 : manufacture of products containing TNPP
- scenario 3 : use of preparations containing TNPP

The stabiliser TNPP is physically bound within the polymer matrix and therefore it could migrate to the surface especially at high temperatures. Release of TNPP from plastic or rubber end products may be a potential way of exposure but due to the very low vapour pressure (0.058 Pa at 25°C) and the small percentage of the stabiliser in the polymers (<1.5 %) exposure to TNPP during subsequent use of products is likely to be negligible.

The number of persons exposed to TNPP is not known.

There are no occupational exposure limits for TNPP.

In the present assessment, inhalation exposure expressed in parts per million (ppm) are converted to mg/m^3 using the following approximation : mg TNPP/m^3 = ppm x 28.6.

4.1.1.2.1 Scenario 1 : Manufacture of TNPP

TNPP is manufactured in three sites in Europe. According the TNPP consortium (North American and European producers), the manufacture is carried out in a closed system where nonylphenol and phosphorus trichloride are added to the reactor and held at 110°C to ensure all phosphorus trichloride is consumed. After the nonylphenol excess has been removed, TNPP is pumped to a storage tank for packaging. The process is fully automated and normally controlled by 1 or 2 operators.

For the large-scale chemical industry, high standards of control at the workplaces are assumed to be practised even if the containment is breached, e.g. during filling. Exposure may occur during coupling and uncoupling of transfer lines, drumming, cleaning, maintenance, repair works and sampling. It is assumed that such activities are performed with local exhaust ventilation (LEV).

Inhalation exposure

The EASE model estimates for production an inhalation exposure in the range of 0-0.1 ppm (closed system, full containment).

If the system is breached in some activities (like maintenance, sampling, cleaning, filling), concentrations are still in the range of 0-0.1 ppm (non-dispersive use, very low tendency to become airborne).

A full shift exposure level of 0.1 ppm (2.86 mg/m^3) is taken as representing a reasonable worst case.

Dermal exposure

Due to the enclosure of the process and control measures taken to minimize skin contact (for example, during transfer to tankers), dermal exposure at the plant is incidental and therefore likely to be low. The main source of potential exposure is during maintenance activities.

The EASE model estimates a dermal exposure in the range of 0-0.1 mg/cm^2/day (non dispersive use with direct handling and incidental contact). Assuming exposed skin surface area is 420 cm^2, maximum external dermal exposure would be 0-42 mg/day.

4.1.1.2.2 Scenario 2 : manufacture of products containing TNPP

According to the TNPP producers, there are 21 facilities in the EU that formulate TNPP (based on 1999 sales information).

Stabilizers are usually added to polymers during the production of powder or pellets (compounding, master batching) which are then transformed into shaped articles (semi-finished or finished products) by different process : extruding, calendaring, injection moulding.

Inhalation exposure

Highest exposure would normally occur during the transfer of thesubstance to the mixer at ambient temperature and during mixing or transforming at high temperatures. Temperatures

are assumed to be between 100 and 200°C. Exposure to dust during the handling of polymer powders or pellets could also be a source of exposure.

Activities at ambient temperature (transfer of the substance)

The EASE model estimates an inhalation exposure in the range of 0-0.1 ppm (non dispersive, very low tendency to become airborne).

The task is assumed to typically take 2 hours (and is probably not carried out daily), therefore a full shift exposure level of 0.025 ppm (0.72 mg/m^3) is taken as a maximum.

Mixing or transforming at high temperatures

It is assumed that these processes are mainly performed using closed systems but the cooling of worked articles, pellets or sheets could be done in open systems. Taking into account the vapour pressure of TNPP at 200°C is about 373 Pa (category "low tendency to become airborne" regarding the volatility), the EASE model estimates an inhalation exposure in the range of 0.5-1 ppm (non dispersive use, with LEV) or 10-20 ppm (non dispersive use, direct handling with dilution ventilation). Lower exposure levels are obtained when it is assumed a use process consisting of inclusion into the polymer matrix : they become in the range of 0.5-1 ppm (non dispersive use, with LEV) or 3-5 ppm (non dispersive use, direct handling with dilution ventilation).

The model overestimates exposure levels, particularly because of non-consideration of the content of TNPP in the polymer. A simple approach based on a reduction of the highest estimated exposure (20 ppm) by a factor equivalent to the TNPP concentration (1.5 %) leads to a maximum exposure of 0.3 ppm (8.58 mg/m^3). Duration and frequency of exposure are assumed to be full shift and daily.

Exposure to dust

Measurements during loading of powder are available but it is not appropriate to consider the results as analogous/surrogate. Studies were generally performed with inorganic powder whereas the dust for this scenario comes from plastic or rubber. Therefore the EASE model is used to for estimation of the exposure. EASE estimates dust exposure in the range of 2-5 mg/m^3 (dry manipulation with LEV) or 5-50 mg/m^3 (dry manipulation, without LEV). Considering a concentration of TNPP in the polymer of 1.5%, the inhalation exposure to dust amounts to 0.03-0.075 mg/m^3 with LEV and 0.075-0.75 mg/m^3 without LEV. As the task (bag emptying) is assumed to typically take 2 hours, a maximum inhalation exposure of 0.19 mg/m^3, full shift, is predicted.

Conclusion

An inhalation exposure level of 8.58 mg/m^3 (related to mixing or transforming activities at high temperatures) will be considered as the worst case exposure during manufacture of products containing TNPP.

Dermal exposure

The highest dermal exposure may occur during transfer of the substance. Afterwards TNPP is enclosed in the polymer matrix and exposure is negligible.

The EASE model estimates a dermal exposure in the range of 0.1-1 mg/cm^2/day (non dispersive use with direct handling and intermittent contact). Assuming exposed skin surface

area is 420 cm^2 (palms of hands), maximum external dermal exposure would be 42-420 mg/day.

4.1.1.2.3 Scenario 3 : use of preparations containing TNPP

TNPP may be a component of resin based preparations. It is a minor use of TNPP and when it is present, its concentration in the formulation is very low (<1 %). For complete assessment, the use of adhesives containing TNPP is considered in this scenario.

Inhalation exposure

Activities at ambient temperature

Taking into account the low vapour pressure of TNPP and the low concentration in the preparations, inhalation exposure is likely to be negligible.

Activities at high temperatures (e.g. use of hotmelt adhesives)

Taking into account the vapour pressure of TNPP at 200°C is about 373 Pa (low tendency to become airborne), the EASE model estimates an inhalation exposure in the range of 0.5-1 ppm (non dispersive use, with LEV) or 10-20 ppm (non dispersive use, direct handling with dilution ventilation).

The model overestimates exposure levels, particularly because of non-consideration of the content of TNPP in the preparation. A simple approach based on a reduction of the highest estimated exposure (20 ppm) by a factor equivalent to the TNPP concentration (1 %) leads to a maximum exposure of 0.20 ppm (5.72 mg/m^3). Duration and frequency of exposure are assumed to be full shift and daily.

Dermal exposure

The EASE model estimates a dermal exposure in the range of 0.1-1 mg/cm^2/day (non dispersive use with direct handling and intermittent contact). Taking into account the concentration of TNPP in the preparation (1%) and assuming exposed skin surface area is 420 cm^2 (palms of hands), maximum external dermal exposure would be 0.42-4.2 mg/day.

4.1.1.2.4 Occupational exposure summary

Table 4-1: Summary of reasonable worst case exposures

Scenario	8-hour TWA inhalation (mg/m^3)	Dermal (mg/day)
1 - Manufacture	2.86	0-42
2 – Manufacture of products	8.58	42 - 420
3 – Use of preparations	5.72	0.42 - 4.2

4.1.1.3 Consumer exposure

4.1.1.3.1 Introduction

Trisnonylphenylphosphite (TNPP) is used as an antioxidant to stabilise polymers against degradation by ultraviolet light. TNPP is used in plastics such as bathroom curtains, tyres, shoe soles and food packaging. In order to assess the consumer exposure, it is necessary to identify the potential ways of human exposure resulted from inhalation, ingestion or skin contact. First, there is no consumer exposure due to tyres. Besides skin contact may happen with bathroom curtains and shoe soles, but remain short or rather occasional. Moreover TNPP is considered to be a slight irritant to the skin. Thus dermal exposure is not considered to be significant. Furthermore, it seems useful to mention that TNPP might be used in plastic medical devices. Such PVC disposable devices, used for blood transfusion, hemodialysis and peritoneal dialysis for example, are particularly studied as a human potential exposure to phtalates, DEHP (diethyhexylphthalate) in particular (FDA, 2001). On the one hand, the studies conclude that such an exposure has more benefits than adverse health effects as far as public health is concerned (INSPQ, 2004). One the other hand, the results from the alkylphenol work group (AWG, 1998) indicate that no detectable amount of TNPP was shown to migrate from PVC-films in any test conditions. That's why risk assessment for the potential exposure to TNPP from plastic medical devices is not performed. Finally consumer exposure due to food-contact materials is the only source of potential exposure which may be important and that worth being particularly studied. It can also be mentioned as preliminary note that a maximum migration TNPP value into food has been proposed by the European Health & Consumer Protection Directorate-General (DG-SANCO, 2003). Since TNPP doesn't need to be classified on the basis of acute toxicity (see chapter 4.1.2.2), only chronic toxicity due to repeated oral exposure from food-contact materials will be characterised.

TNPP may be partly hydrolysed (acid hydrolysis) and gives nonylphenol. In order to be protective, the TNPP amount in food-contact plastics will be overestimated in so far as the TNPP degradation won't be considered.

4.1.1.3.2 Potential exposure from migration of TNPP from food contact materials

TNPP use and amount in plastics

According Howe *et al.* (Howe, 2001), TNPP is used in three types of food-contact polymers : PVC-films, polyolefins (linear low density polyethylene – LLDPE or ethylene vinyl acetate copolypers – EVA) and rubber. Previously a distinction was often made between "rubber" and "rubber modified polystyrene". Actually a distinction doesn't seem to be needed and the two categories will be considered as one "rubber" category, i.e. high impact polystyrene – HIPS. Various grades of TNPP are used in these polymers as shown in Table 4-2.

Table 4-2: Use of trisnonylphenylphosphite in food-packaging polymers

Polymer type containing TNPP	TNPP concentration (mass %)
PVC Film	0.8 – 1.5 %
LLDPE	0.1 – 0.2 %
EVA	0.05%
Rubber / HIPS	0.4 – 1.0 %

Calculated exposure

The dietary consumption of TNPP depends on :

- the potential level in food;
- the fraction of an individual's diet likely to contact food materials containing TNPP;
- the total weight of food daily consumed by an individual.

The dietary exposure is calculated using the American Food and Drug Administration (FDA) model which uses consumption and food-type distribution factors. Since no European data are available, numerical values for the different factors are those from FDA, derived from simulated food-contact use (with food-stimulating solvents) (FDA, 2002).

When the accurate different food items in contact with the packaging are well identified, the daily dietary concentration in food is the following:

$$\text{Dietary concentration} = P \times \Sigma_i (CF_i \times M_i)$$

M_i is the migration value of TNPP in the considered food-type. This migration value M_i is given by the experimental exposure of the food-contact material to a given food simulant (under the time and temperature exposure) representing the food-type. This food-type is classified as aqueous (aq), acidic (ac), alcoholic (al) or fatty (f).

The consumption factor CF_i represents the fraction of the food-type (which is daily consumed) likely to contact the food packaging.

On the opposite, when food items can not be precisely distinguished, the whole diet is divided into aqueous (aq), acidic (ac), alcoholic (al) or fatty (f) food items. The daily dietary concentration in food is thus the following:

$$\text{Dietary concentration} = P \times CF \times (M_{aq} \times f_{aq} + M_{ac} \times f_{ac} + M_{al} \times f_{al} + M_f \times f_f)$$

M_{type} is the migration value of TNPP in the considered *food-type*. This migration value M_{type} is given by the experimental exposure of the food-contact material to a given food simulant (under the time and temperature exposure) representing the food-type, which can be aqueous (aq), acidic (ac), alcoholic (al) or fatty (f). "10% ethanol" or "90% ethanol" indicate thus food simulants.

The distribution factor f_{type} is the fraction of food of each *type* that will contact the material.

The consumption factor CF represents the fraction of the whole daily diet likely to contact the food packaging.

In both equations, P is the percentage of the considered food-packaging containing TNPP.

The estimated daily intake (EDI) for each type of packaging is determined by multiplying the total weight of food consumed by an individual per day (default 3000 g/day, solids and liquids) by the dietary concentration of TNPP in the studied packaging.

Finally the total estimated daily intake (TEDI) represents the sum of each EDI respectively due to PVC films, LLDPE, EVA and HIPS :

$$TEDI = EDI_{PVC} + EDI_{LLDPE} + EDI_{EVA} + EDI_{HIPS}$$

Estimated daily intake due to PVC

All the food-contact PVC-films contain TNPP. However, the results from the alkylphenol work group (AWG, 1998) indicate that no detectable amount of TNPP was shown to migrate from PVC-films in any test conditions. PVC dietary concentration $\equiv 0$ and $EDI_{PVC} \equiv 0$

Estimated daily intake due to LLDPE

50% of food-contact LLDPE contains TNPP. Since each food type is precisely identified (frozen food, bag-in-box items, …) with their accurate CFs, the dietary concentration is : Dietary concentration = $P \times \Sigma_i (CF_i \times M_i)$.

Table 4-3: Potential TNPP exposure from LLDPE

Application		P	CF	M (ppm)	Exposure (ppb)
Film	Produce	0.5	0.04	ND [a]	-
	Frozen	0.5	0.001	ND [a]	-
	Meat/Poultry	0.5	0.002	1.53 [b]	1.53
	Dry	0.5	0.01	ND [a]	-
	Bag-in-box	0.5	0.006	ND [a]	-
	Snack	0.5	0.002	1.53 [b]	1.53
Films/Coatings		0.5	0.0002	1.53 [b]	0.15
Lids/Tubs	Aqueous	0.5	0.0027	ND [c]	-
	Fatty	0.5	0.0013	2.64 [d]	1.72
LLDPE dietary concentration					5.0 ppb
EDI_{LLDPE}					0.015 mg/day

ND [a]: "10% ethanol" data from films have to be used for film uses involving produce, frozen, dry, and bag-in-box. The results shown in Table 12 from (AWG, 1998) indicate that no detectable amount of TNPP was shown to migrate to 10% ethanol under any of the test conditions used.

[b]: "95% ethanol" data from films have to be used for film uses involving meat, poultry, and snack, and for films/coatings applications (Table 13, AWG, 1998). The maximum value obtained (migration after 4-day test) is used. 4-day and 10-day migration results are not significantly different.

ND [c]:"10% ethanol" data from plaques (condition of use E = 40°C for 10 days) have to be used for the aqueous lids/tubs applications. No TNPP was shown to migrate (Table 12, AWG, 1998).

[d]: "95% ethanol" data from plaques (condition of use E = 40°C for 10 days) have to be used for the fatty lids/tubs applications. The maximum value obtained (migration after 4-day test) is used (Table 13, AWG, 1998).

Estimated daily intake due to EVA

25% of food-contact EVA contains TNPP.

According to the alkylphenol work group, "10% ethanol" data from LLDPE films have to be used for the aqueous food applications and "95% ethanol" data from LLDPE films for the fatty food applications. As far as the aqueous applications are concerned, no TNPP was shown to migrate (Table 12, AWG, 1998). For the fatty applications, the maximum value obtained (migration after 4-day test) is used i.e. 1.53 ppm (4-day and 10-day migration results are not significantly different) (Table 13, AWG, 1998). Since the use level of TNPP in EVA films (500 ppm) is lower than the 1200 ppm TNPP level in the films use for the migration tests and since migration is directly proportional to the concentration, a factor of 42% is used (500/1200). That's why the TNPP migration level that is used is 0.64 ppm.

$$\text{EVA dietary concentration} = P \times CF \times M_f \times f_f = 0.25 \times 0.04 \times 0.64 \times 0.45 = 2.9 \text{ ppb}$$

$$EDI_{EVA} = 0.0087 \text{ mg/day}$$

Estimated daily intake due to HIPS

All the food-contact HIPS-packaging contain TNPP. Since each food type is identified (yoghurts, cheese, …) with their accurate CFs, the dietary concentration is : Dietary concentration = $P \times \Sigma_i (CF_i \times M_i)$.

Since the use level of TNPP in HIPS films (10000 ppm max. value) is higher than the 1200 ppm TNPP level in the films use for the migration tests and since migration is directly proportional to the concentration, a factor of 833% is used (10000/1200). M values in Table 4-4 take into account this 833% factor.

Table 4-4: Potential TNPP exposure from HIPS

Application			P	CF	M (ppm)	Exposure (ppb)
Packaging	Yoghurt cups (aq)		1	0.0036	ND [a]	-
	Cheese/Cream (aq)		1	0.0036	ND [a]	-
	Aseptic/Blow moulded (aq)		1	0.0009	ND [a]	-
Disposable	Fatty	4°C	1	0.0001	0.48 [b]	0.048
		24°C	1	0.0001	0.48 [b]	0.048
		54°C	1	0.0003	11.3 [c]	3.4
	Aqueous	4°C	1	0.0108	ND [a]	-
		24°C	1	0.0188	ND [a]	-
		54°C	1	0.0016	ND [a]	-
	Alcoholic		1	0.0015	ND [a]	-
HIPS dietary concentrations						3.5 ppb
EDI_{HIPS}						0.010 mg/day

ND [a]: 10% ethanol" data from LLDPE plaques have to be used for the aqueous and alcoholic applications. The results shown in Table 12 from (AWG, 1998) indicate that no detectable amount of TNPP was shown to migrate to 10% ethanol under any of the test conditions used.

[b]: 95% ethanol" data from LLDPE plaques have to be used for the fatty applications according to the alkylphenol work group (AWG, 1998). Moreover, the disposable uses involve contact with food for 1 or 2 hour max. The closest time frame to this length of time used in the testing is 2 hours. Thus, the 2-hour data have been considered for the HIPS disposables. Finally for the applications at 4°C and 24°C, results in test condition of use E (= 40°C for 10 days) are used.

[c]: id [b] But for the applications at 54°C, an interpolated M value corresponding to the value between 40°C and 100 °C (condition of use B = 100°C for 2 h, followed by 40°C for 238 h) is used considering that there is a linear relationship between migration and the inverse of temperature (in Kelvin). Thus, the migration value (before correction with the 833% factor) is 1.36 ppm (AWG, 1998).

4.1.1.3.3 Consumer exposure summary

The overall potential dietary exposure, or total estimated daily intake (TEDI), to TNPP from the use in food-contact packaging is the sum of the above EDI values:

$$TEDI = EDI_{PVC} + EDI_{LLDPE} + EDI_{EVA} + EDI_{HIPS}$$

$$TEDI = 0 + 0.015 + 0.0087 + 0.010$$

$$TEDI = 0.0337 \text{ mg/day}$$

4.1.1.4 Indirect exposure via the environment

This section was not provided as it will be updated in the next version of the environmental risk assessment.

4.1.2 Effects assessment : hazard identification and dose (concentration)-response (effect) assessment

4.1.2.1 Toxicokinetics, metabolism and distribution

No specific toxicocinetic study was conducted with trisnonylphenyl phosphite (TNPP).

However qualitative information can be derived from the physico-chemical properties of the substance. Considering the relatively high molecular weight of the molecule (MW = 689 $g.mol^{-1}$), its extremely low water solubility and a very high Log P_{ow}, the absorption of TNPP by the gastro-intestinal tract is expected to be limited.

The vapor pressure of the liquid substance (physical state at 20°C and 101,3 kPa) is very low . Therefore, inhalative exposure can be anticipated only as liquid aerosol.

The molecular weight (> 500) of TNPP, its water solubility (< 1 mg/l) and its Log Pow (> 6) are in favour of a very limited absorption following dermal exposure.

Based on the physico-chemical properties , default values were chosen for oral, dermal and inhalative absorption :

Oral absorption: as indicated above, the absorption of TNPP by the gastro-intestinal tract is expected to be limited. However no quantitative value is available, then as a worst case assumption for oral route, a default value of 50% is chosen.

Dermal absorption: a default factor of 10% is used as MW>500 and Log P_{ow} is higher than 4.

Inhalative exposure: absorption mechanisms via mucous membranes are expected to be the same by oral and inhalation route, thus a default value of 50% is chosen as a worst case assumption.

4.1.2.2 Acute toxicity

Only data on animals are available.

4.1.2.2.1 Oral

The acute oral toxicity of TNPP has been investigated in three animal studies of different quality.

● The most informative study related to acute oral toxicity was conducted by the Food and Drug Research Laboratories for Naugatuck Chemical Corporation in 1957. The report was scientifically acceptable, although the study was not conducted in compliance with the Good Laboratory Practice (GLP) and the international standardisation of testing methods.

Five groups of ten adult albino rats (5 males and 5 females) were given graded doses of a 50 per cent solution of TNPP (purity not specified) in cottonseed oil. The doses (8.19 - 11.32 - 16.38 - 22.62 and 32.72 gram/kg bw.) were administered by stomach tube.

Following dosage, the rats were observed for appearance, behaviour, bodyweight and mortality for a 14-day period. Rats that died, as well as survivors sacrificed at the end of the experiment, were examined for evidence of gross pathology.

All rats showed evidence of abdominal pain and catharsis after dosage. Highest doses (11.32 gram/kg and above) resulted in urinary incontinence and prostration. Gross pathological findings included hemorrhagic lesions in the gastric mucosa and/or duodenum in a few rats that died, and hemorrhagic lungs. According to the authors, the incidence and severity of the abnormalities at the former site being greater at the higher dose levels. However this assertion couldn't be checked because no table of results related to the gross pathological findings was available.

Mortality resulted at scattered intervals over the first five days Table 4-5 . However, growth of the survivors was essentially normal. The Lethal Dose 50 (LD_{50}) was computed according to the method of Miller and Tainter (1944) and was calculated to be 19.5 +/- 3.3 gram/kg bw.Table 4-5: Acute oral toxicity of TNPP

Dose (g/kg)	8.19	11.32	16.38	22.62	32.72
Number of death	0	3	4	7	7

● Another study was conducted by Hill Top Research in 1965. The purpose of this study was to evaluate and compare the acute oral toxicity of four samples of chemicals in rats (two samples of TNPP and two samples of another chemical). This study was not conducted in compliance with the GLP and the international standardisation of testing methods. The samples were received from Argus Chemical Corporation. For the purposes of this study, the purity of each sample was considered to be 100% and no correction was made for possible impurities. The results were incompletely provided as the page of results related to the second sample of TNPP is missing and all the paper files for this study were since discarded.

Graded doses up to 10 ml/kg of TNPP (equivalent to 9.8 g/kg, based on a relative density of 0.98 $g.cm^{-3}$ at 20°C) were administered orally by stomach tube to six groups of five male albino rats, Holtzman strain. Each TNPP sample was administered as a 10% or 50% volume/volume solution in corn (Mazola) oil at dosage levels of 0.215 - 0.464 - 1.00 - 2.15 - 4.64 and 10.0 ml/kg bw (equivalent to 0.21 – 0.45 – 0.98 – 2.11 – 4.54 and 9.80 g/kg bw). Larger doses could not be administered without exceeding the capacity of the rat stomach.

All animals were observed closely for gross signs of toxicity and mortality at frequent intervals during the day of dosage, and at least once daily thereafter for a total of 14 days. Gross autopsies were performed on the animals that died. At the end of the 14-day observation period, the surviving rats were weighed, sacrificed and gross autopsies were performed.

For sample one, there was no mortality at any dosage level tested. The acute oral LD_{50} of TNPP for male albino rats was therefore established to be greater than 10.0 ml/kg bw (9.8 g/kg bw).

All the rats exhibited normal appearance and behaviour during the observation period. The average body weight gain for each group of rats was within the normal range of values for rats of the sex, age and strain used in this study. At gross autopsy, the organs of all animals appeared grossly within normal limits.

There was no information related to sample two, except for the summary that mentioned that LD_{50} was greater than 10.0 ml/kg bw too (9.80 g/kg bw).

- A study conducted by Majlathova (1981) was related to the evaluation of aralkyl phenylphosphite antioxidants by an acute peroral experiment on mice and rats and by epicutaneous and conjunctival test on rabbits. The publication available related to this study was written in Slovakian but it contained an abstract written in English by the authors. The abstract states that the starting LD_{50} concentration of the TNPP product tested was greater than 10 grams/kg bw but that the storage (time) makes it become more toxic. Yet 5 gram/kg bw do not affect the health condition of the animals.

4.1.2.2.2 Inhalation

No study was found.

4.1.2.2.3 Dermal

● Acute dermal toxicity was studied in a recent and well-conducted study, following OECD guideline 402 (Tay, 2001a).

TNPP (purity not indicated) was evaluated for its potential to produce systemic toxicity or death after a single topical 24-hour application to the skin of albino rabbits at a dose of 2000 mg/kg (limit test). Five male and five female New Zealand White rabbits were used for the test. The test substance was introduced under gauze patches, two single layers thick, and applied directly to the skin of the body surface (approximately 10%) of each of ten animals. At the completion of the exposure period, the skin was gently wiped to remove any test substance still remaining. The animals were observed frequently during the first day, and then a careful clinical examination was made at least once a day. The animals were also observed for signs of erythema and oedema after the exposure period according to the Draize scale for scoring skin reactions. Animals were weighed at day 0 (prior to dose administration), day 7 and day 14. A gross necropsy was performed on all animals whether found dead or sacrificed at the end of the study, on the 14^{th} day.

All animals gained weight during the post treatment-period, except one male rabbit (weight loss = 0.02 kg). In all rabbits, no other sign of systemic toxicity was evident during the course of the study and no animal died. At necropsy, there was no abnormality or lesion noted. No erythema or oedema was observed at any of the test sites.

The LD_{50} in rabbits was then found to be greater than 2000 mg/kg bw.

● Another recent dermal acute toxicity study, performed in rats by Ciba-Geigy (1992), confirms these results. The study was conducted in compliance GLP and following the OECD Test Guideline 402.

The purity of the test article was > 94%.

Ten young adult albino rats of both sexes (5 males and 5 females) were exposed to the dose of 2000 mg/kg bw. The test article was evenly dispersed on the back of the rat (at least 10% of the body surface was shaved with an electric clipper). After 24h under semi-occlusive conditions the dressing was removed and the skin was cleaned with lukewarm water.

Animals were weighed at day 0 (immediately before application), day 7 and day 14. Mortality and symptoms were observed daily for 14 days and the animals were submitted to a gross necropsy at the end of the observation period.

All animals gained weight during the post treatment-period. No mortality occurred in this study. Piloerection and hunched posture were seen, being common symptoms in acute dermal tests. The animals recovered within 2 days. At necropsy, no deviation from normal morphology was found.

The LD_{50} in rats of both sexes was then found to be greater than 2000 mg/kg bw.

● The Food and Drug Research Laboratories (1961) indicate that toxicological screening tests, conducted in these laboratories, demonstrated that 24-hour dermal applications in massive doses were not lethal to the rabbits. However, the report for these screening tests was not available.

4.1.2.2.4 Other

An acute intraperitoneal toxicity study was conducted in rat (Ciba-Geigy, 1983).

This study was not conducted in compliance with GLP and the international standardisation of testing methods. The sample provided by the sponsor was Irgafos TNPP (trade name) but no information was provided about its purity.

In this study, 5 male and 5 female albino rats were administrated a single dose of 1000 mg/kg bw, by intraperitoneal injection. The vehicle used was distilled water containing 0.5% carboxymethylcellulose and 0.1% polysorbate 80. Animals were weighed at day 1, 7 and 14. Mortality and symptoms were observed daily for 14 days or until all symptoms have disappeared. The animals were submitted to a gross necropsy at the end of the observation period.

Dyspnoea, exophtalmus, ruffled fur and curved body position were seen, being common symptoms in acute tests. Animals recovered within 12 days. No mortality occurred during the study. At autopsy, peritoneal adhesions in the liver and spleen area were found in 8/10 animals.

The LD_{50} in rats of both sexes was then found to be greater than 1000 mg/kg bw.

4.1.2.2.5 Summary of acute toxicity

No human data is available. In animals, TNPP has a very low acute toxicity by the oral route, with a LD_{50} value of about 19.5 +/- 3.3 gram/kg bw for the rat. Hemorrhagic lesions in the gastro-intestinal tract and the lungs are seen in some animals, following the administration of a lethal dose. This value was used for the risk assessment. The other studies couldn't be used in the risk assessment due to shortcomings or unavailable study reports. Furthermore a LD_{50} could not be derived from these studies as no mortality was observed at doses up to the highest doses tested (about 10 g/kg). Nevertheless, these results are in accordance with the value of 19.5 g/kg bw derived from the study from Naugatuck (1957).

The acute toxicity of TNPP by the dermal route seems to be very low too, with a LD_{50} greater than 2000 mg/kg in rabbits. No data is available on the acute inhalation toxicity, although the non-corrosive and non-irritant nature of TNPP (see section 4.1.2.3.1 on skin irritation) may suggest that toxicity would not be enhanced following exposure by this route.

By intraperitoneal route, the LD_{50} was found to be > 1000 mg/kg in rats.

Classification and labelling :

According to the criteria of the European Union, this chemical does not need to be classified on the basis of its acute toxicity.

4.1.2.3 Irritation

Only animal data are available.

4.1.2.3.1 Skin

● Acute dermal irritation and corrosion were studied in a recent and well-conducted study, following OECD guideline 404 (Tay, 2001b).

The degree of purity of the test substance was 99.3%.

TNPP was evaluated for its potential to produce skin irritation and/or corrosion after a single topical application for four hours to the intact skin of New Zealand White rabbits.

One male and two females were used for the test. A dose of 0.5 ml liquid test substance was applied to a small area (approximately 6 cm^2) of skin. The test report does note indicate if the test substance was applied as pure or not, but the hypothesis of a pure substance application is the most probable. Animals were observed for signs of erythema and oedema at 60 minutes and then at 24, 48 and 72 hours after patch removal. Observations were scored according to the Draize scale for scoring skin reactions. Daily clinical observations included all toxicological and pharmacological signs. Animals were weighed at the end of the observation period.

All of the test animals exhibited a gain in body weight during the study. No overt sign of toxicity was evident in any of the animals during the course of the study. Very slight erythema was observed in three out of three rabbits following a 4-hour exposure. By the 24-hour observation point, the irritation was reversed, with no sign of erythema present at the 24-, 48- and 72-hour observations for all three rabbits. No oedema was observed at any of the observation points (see Table 4-6).

Table 4-6: Skin mean reaction scores (4-hours exposure)

Time after exposure	60 minutes		24 Hours		48 Hours		72 Hours	
	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema
	1	0	0	0	0	0	0	0

TNPP was considered to be a very slight irritant to the skin.

● Ciba-Geigy conducted a skin irritation study in rabbits in 1981. The study was not conducted in compliance with GLP and the international standardisation of testing methods, however, the procedure used is based on the Proposed Guidelines of the United States Environmental Agency (US EPA) : "Primary dermal irritation study" (1978).

The sample provided by the sponsor was Irgafos TNPP (trade name) but no information was provided about its purity.

The test was performed on 3 male and 3 female adult New Zealand White rabbits. Before treatment, the entire back and the flank of the rabbits were shaved with electric clipper and immediately before treatment, the shaven skin on one side was slightly scarified. Gauze

patches, laden with 0.5 ml of the test material, were applied to the prepared abraded and intact skin of the rabbits. After 24h under occlusive conditions the dressings were removed and the skin reaction was appraised upon removal and during an observation period of 7 days. The grading system for skin irritation was similar to the Draize scale for scoring skin reactions.

The study report states that in 3/6 animals, the application sites showed necrosis, but it did not give any further information on this effect. In 5/6 animals the erythemas extended beyond the treated areas. Erythema and edema of intact skin were reversed within 7 days, except in abraded skin in 2/6 animals for which erythema was still moderate to severe. The calculated primary irritation index was 2.5 : TNPP was found to cause moderate irritation when applied to intact and abraded rabbit skin (see Table 4-7).

Table 4-7: Skin mean reaction scores (24-hours exposure)

Time after exposure	Erythema		Edema	
	Intact skin	Abraded skin	Intact skin	Abraded skin
24 hours	2.0	2.7	1.0	1.4
72 hours	0.7	1.7	0.0	0.7
7 days	0.2	1.2	0.0	0.3

- In New Zealand rabbits, the acute dermal toxicity study (OECD 402) conducted by Tay (2001a) (see section 4.1.2.2.3 dermal acute toxicity), a single topical 24-hour application of pure substance to the skin exhibited no signs of erythema and oedema at any of the test sites after the exposure period according to the Draize scale for scoring skin reactions.

- The Food and Drug Research Laboratories (1961) indicate that toxicological screening tests conducted in these laboratories demonstrated that TNPP was a primary irritant when applied repeatedly to the skin of rabbits, but that the effects were reversible. The report for these screening tests was not available.

4.1.2.3.2 Eye

- Acute eye irritation and corrosion were studied in a recent and well-conducted study, following OECD guideline 405 (Tay, 2001c).

The degree of purity of the test substance was 99.3%.

TNPP was evaluated for its potential to produce an irritating effect on the ocular tissue of New Zealand White rabbits. Two males and two females were used for the test.

Both eyes of each rabbit were examined for macroscopic findings and were scored before and after treatment. The grading system for ocular irritation was the one presented in OCDE test guideline 405. The eyes of the animals were screened with fluorescein stain before dosing. The left eye of each animal was treated with 0,1 ml of the test substance, the right eye remaining untreated and thus, served as a control. The eyes of the test animals were not washed prior to 24 hours following instillation of the test substance. Following the 24-hour observation, the treated eyes were not rinsed. The initial procedure was performed on one

rabbit, using a 10% dilution of the substance. As no severe effect was observed, the test was performed on three rabbits with the undiluted test substance.

Eyes were examined at 1, 24, 48 and 72 hours after treatment using the scale of grades for ocular lesions. After recording the observations at 24 hours, the eyes of all rabbits were examined with the aid of fluorescein to further characterise corneal opacity. Animals were also observed daily for clinical manifestations and were weighed at the end of the observation period.

All the animals exhibited an increase in body weight during the course of the study. No overt sign of toxicity other than the ocular effects was evident during the course of the study in any of the animals.

Eye Scores :

- No corneal opacity was observed in any of the treated eyes, at any of the observation periods.
- No fluorescein staining was observed in any of the treated eyes, at all observation points.
- The iris response was normal in all treated eyes.
- Three out of three treated eyes exhibited slight conjunctival redness and chemosis at the 1-hour observation point (grading score of 1). They persisted in 2 of 3 animals for 24 hours and were resolved by the 48-hour time point. In the other animal, all signs of irritation were resolved by the 24-hour observation point.

TNPP was considered to be a slight irritant to the ocular tissue of New Zealand White rabbits.

● Ciba-Geigy conducted an eye irritation study in rabbits in 1981. The study was not conducted in compliance with GLP and the international standardisation of testing methods, however, the procedure used is based on the Proposed Guidelines of the US EPA : "Primary eye irritation study" (1978).

The sample provided by the sponsor was Irgafos TNPP (trade name) but there is no information about its purity.

The test was performed on 3 male and 3 female New Zealand White rabbits. 0.1 ml of the test material was inserted into the conjunctival sac of the left eye of the rabbits. In 3 of the 6 rabbits approximately 30 seconds after treatment, the treated eye was flushed with 10 ml of physiological saline. The eye irritation was appraised with a slit-lamp on day 1, 2, 3, 4 and 7 and was scored for each individual rabbit. The grading system for ocular irritation was similar to the one presented in OCDE test guideline 405.

No corneal opacity was observed in any of the treated eyes, and the iris response was normal in all treated eyes at any of the observation periods. Slight redness and chemosis were the only observable effects. They were completely reversible in rabbits with rinsed eye within 7 days. The test material was found to cause minimal irritation when applied to the rabbit eye mucosa, whether the eyes were rinsed or unrinsed (seeTable 4-8).

Table 4-8: Rabbit eye irritation scores (conjunctiva mean reaction scores)

	24 Hours		48 Hours		72 Hours		4 days		7 days	
Eye treatment	Not rinsed	Rinsed	Not rinsed	Rinsed	Not rinsed	Rinsed	Not rinsed	Rinsed	Not rinsed	Rinsed
Redness	1	1	0.3	0.7	0.7	0	1	0.3	0.7	0
Chemosis	1	0.7	0.3	0.7	0.3	0.3	0.3	0.3	0	0

4.1.2.3.3 Summary of irritation

No information is available from human studies. Based on the available data on rabbits, it can be assumed that TNPP is a very slight to moderate irritant to the skin, varying according to tests conditions used : TNPP was a very slight irritant when administered to intact skin for a 4-hours exposure, whereas a 24-hour exposure on intact and abraded skin under occlusive conditions elicited more severe irritation properties. The two available studies indicate that TNPP is a slight irritant to the eye. In each case, the effects were generally reversed within a few days.

Classification and labelling :

According to the cutaneous and eye irritation test methods cited in Annex V, similar to OCDE guideline 404 and 405, TNPP should not be classified as an irritant to skin and eye.

4.1.2.4 Corrosivity

The results from the study of Tay (2001b) indicate that after a 4-hour exposure under semi-occlusive conditions TNPP is not corrosive on intact skin (OECD 404 conditions). However, the study conditions of another study (Ciba-Geigy, 1981) elicit corrosive properties of TNPP. These were harsh conditions (24h exposure under occlusive conditions on abraded and non-abraded skin), furthermore the study report indicates no further details on necrosis observed (was necrosis observed on intact or abraded skin? After what time of application the necrosis was observed?). Based on exposure conditions adopted by OECD guideline for classification, the results of the study of Tay were used in the risk assessment.

Classification and labelling :

TNPP should not be classified as corrosive to skin or eye according to the criteria of the European Union.

4.1.2.5 Sensitisation

Only animal data are available.

4.1.2.5.1 Skin sensitisation

- A Buehler Sensitisation Test was performed, following OECD guideline 406 and GLP (Tay, 2001d).

The degree of purity of the test substance was 99.3%.

TNPP was evaluated for its potential to produce allergenic skin reactions following epicutaneous application to albino guinea pigs. 19 males and 19 females were used for the test. Test animals were distributed into the following groups :

Experimental : 10 males/ 10 females

Negative controls : 5 males/ 5 females

Positive controls : 3 males/ 2 females

Preliminary Irritation : 1 male/ 2 females

- Preliminary irritation : four different concentrations were applied to the skin for 6 hours : 100%, 50%, 25% and 10% of the test substance diluted with 0.9% sodium chloride for injection. The test substance was determined to be a non-irritant, therefore, it was used neat for the induction and the challenge phase.

- Induction phase : closed patches were applied directly to the skin and removed after 6 hours of exposure. The test substance was applied once per week for 3 consecutive weeks (days 0, 7 and 14) on one side of the animals. The positive control substance (dinitrochlorobenzene, DNCB) was applied in the same manner. Naive animals, i.e., untreated during the induction phase, served as a negative control group.

- Challenge phase : The challenge test was performed on virgin skin sites of test and naive animals in the same way as the 6-hour test of the induction phase (closed patches).

All animals gained weight during the course of the study. No abnormal clinical observations were evident in any of the animals during the course of the study.

All animals showed no sign of erythema or oedema at the 24 and 48-hour observation points for the challenge phase. No reactions were observed in the negative control group and 100% reactivity was observed in the positive control group at challenge.

● A Maximisation Test was also performed, following OECD guideline 406 and GLP (Ciba-Geigy, 1992).

The degree of purity of the test substance was > 94%.

The test was performed on 10 male and 10 female guinea pigs in the test group and 5 male and 5 female in the control group.

The sensitivity of the strain is checked every six months with a known sensitiser, such as 2,4-dinitrochlorobenzene, paraphenylene-diamine or potassium-dichromate.

Control group : The control group was treated with adjuvant and the vehicle during the induction period. During the challenge period, the group was treated with the vehicle as well as with the test article to check the maximum subirritant concentration of the test article in adjuvant treated animals.

- Induction (weeks 1 and 2) : it was a 2-stage operation. First, intradermal injection into the neck region (adjuvant/saline mixture, 5% of test article in Oleum arachidis (well tolerated dose) and test article in the adjuvant/saline mixture). Second, one week later, closed patch exposure of 10% TNPP in vaseline (concentration leading to erythema reactions) over the injection sites for 48 hours.

- Rest period : during weeks 3 and 4, no treatment was performed.

- Challenge (week 5) : the animals were tested on the flank with 1% TNPP in vaseline (subirritant concentration) and the vehicle alone (occluded administration for 24 hours).

24 and 48h after removing the dressings, the challenge reactions were graded according to the Draize scoring scale. The body weight was recorded at start and end of the test.

All the animals gained weight during the study. No positive reaction was observed in control animals. In the test group, there were 12/20 and 15/20 positive animals respectively 24h and 48h after occlusive epidermal application (showing erythema scores of 1 to 2). Therefore, TNPP is classified as a strong sensitiser in albino guinea pig according to the grading of Magnusson and Kligman.

● In the study of Majlathova (1981), related to the evaluation of aralkyl phenylphosphite antioxidants by an acute peroral experiment on mice and rats and by epicutaneous and conjunctival test on rabbits, the abstract of the publication states that acute toxic contact dermatitis developed in rabbits after epicutaneous administration of Polygard TNPP exposed to time. The changes healed spontaneously ad integrum within three days after drug withdrawal.

4.1.2.5.2 Summary of sensitisation

No human data is available. The results of the Buehler sensitisation test and of the Maximisation test, both conducted on guinea pig and following OECD TG 406, are not in accordance.

Adjuvant-type tests are likely to be more accurate in predicting a probable skin sensitising effect of a substance in humans than those methods not employing Freunds Complete Adjuvant (FCA), and are thus the preferred methods. Then, the results of the Guinea-Pig Maximisation test will be used for the risk assessment, as this test is considered to be more sensitive than the Buehler test.

No information on respiratory tract sensitisation is available.

Classification and labelling :

TNPP needs to be classified as a skin sensitiser according to the criteria of the European Union (Xi, R43).

4.1.2.6 Repeated-dose toxicity

4.1.2.6.1 Animal data

There is no data for the inhalation and the dermal route.

Four studies provide an assessment of the oral repeated dose toxicity of TNPP.

Three of them were conducted between 1957 and 1961 by the Food and Drug Research Laboratories for the Naugatuck Chemical Company and the last one is a recent reproductive/developmental toxicity screening test conducted by Tyl et al. in 2002.

The Food and Drug Research Laboratories conducted a 90-day study in rats in 1957 and two two-year studies, one in rats and the other one in dogs, in 1961.

Theses studies were not GLP and did not follow specific EU or international guideline. However all of the documents provided were acceptable, well-documented study reports, which met basic scientific principles.

● In the 90-day range-finding feeding test with rats (strain not specified) (Food and Drug Research Laboratories, 1957), groups of five male and five female rats were exposed to TNPP (purity not specified) via incorporation in the diet, at doses of 0.2, 1.0 and 5.0% of the daily feeding ration, designed to provide each day about 200, 1000 and 5000 mg/kg bw of TNPP.

The rats were inspected daily for appearance and behaviour. Bodyweight and food intake were recorded weekly for 12 weeks and the efficiency of food utilisation (EFU) was calculated. Haematological (haemoglobin, hematocrit and white cell count) and chemical examinations (blood sugar and blood non-protein nitrogen) were made on the blood of two male and two female rats per group at the 12-week period. All rats that died and all survivors (sacrificed at the end of the 90-day period) were examined at autopsy for evidence of gross pathology. The liver weights were determined for all survivors.

Up to the dose of 1000 mg/kg/d, no adverse effect of the ingestion of this chemical was observed.

In the group exposed to a dose of 5000 mg/kg/d, two females died (on the 35th and 49th day respectively). The principal abnormalities seen at autopsy suggest that the deaths were at least in part due to pulmonary pathology (fibrinous exudate in thorax and hemorrhagic lungs).

In this same group, growth and food efficiency were depressed to a highly significant ($p=0.001$) degree in both sexes. These were the only results which were given a statistically significance. The report didn't indicate which statistical test was used, however, it is reasonable to assume that the statistics presented are intergroup comparisons using the Student's t-test.

The haematological observations disclosed no evidence of abnormalities at any dose level. The blood sugar levels were slightly increased at the dose of 200 mg/kg/d and slightly depressed at the 5000 mg/kg/d dose. The blood non-protein nitrogen levels were slightly elevated in rats of both sexes at the 5000 mg/kg/d dose and in the male at the 1000 mg/kg/d dose. However, all values for both of these biochemical components were within normal limits for the rat.

Pathological changes were observed in the lung and the kidney. Pulmonary lesions were lesions commonly seen in laboratory rats. In the kidney, no change was observed in the test groups up to the 1000 mg/kg/d dose level but at the dose of 5000 mg/kg/d, 8 of the 9 animals examined showed evidence of acute and chronic pyelonephritis with foci of calcification. Chronic pyelonephritis typically represents an inflammatory process that begins in the renal pelvis area (either primarily or as an extension from the urinary bladder) and often spreads upwards into the kidney. There can be many contributory factors associated with pyelonephritis, thus it may not be treatment-related.

However, based on pathological changes observed in the kidney at the dose level of 5000 mg/kg/d, this study identifies a NOAEL (No Observed Adverse Effect) of about 1000 mg/kg bw/d (1% TNPP in the diet) and a LOAEL (Low Observed Adverse Effect) of about 5000 mg/kg bw/d (5% TNPP in the diet) for a 90-day exposure of rats.

Chronic ingestion studies on TNPP (purity not specified) were conducted in rats and dogs (strains not specified) over a 2-year period. The test material was incorporated in nutritionally adequate rations for the respective species at levels of 1000, 3300 and 10 000 ppm in the diet and comparison was made with control groups receiving the basal diet without the additive.

● The rat experiment (daily intake corresponding to 50, 167 and 500 mg/kg bw), combined a chronic and a reproductive toxicity study. Weanling rats were distributed into four groups of 25 males and 25 females each. Observations were made of behaviour and appearance, growth, food intake and efficiency. At 12 weeks and at approximately half-yearly intervals, clinical examinations were made in ten rats of each sex in the control and highest test level groups and in five of each sex at the lower levels. These include erythrocyte and leukocyte counts, blood haemoglobin, hematocrit, sugar and non-protein nitrogen determinations, and urine examinations for protein, sugar and sediment. Blood cholesterol levels, prothrombin time and various other biochemical determinations were made at several intervals. After the rats were on test for approximately 100 days, reproduction and lactation studies were initiated and mating continued through the lifetime of the females of the F0 generation (leading to a total of six matings). Ten representative rats of each sex from the second litters produced by the females were selected at the time of weaning and placed on the same ration as their respective parents. Observations similar to those described above were carried out through these and two additional descendant generations. Rats that died or were sacrificed when moribund were examined grossly. At the termination of the two-year period of the F0 rats and at various periods after the weaning of the second litter in the descendant generations of rats (72, 36 and 14 weeks respectively for F1, F2 and F3), they were sacrificed and also examined grossly. At the time of autopsy, eight of the major organs were weighed (liver, spleen, heart, testes and ovaries, and the adrenal, thyroid and pituitary glands). Extensive histopathological examinations were carried out in the F0 generation.

At levels up to 167 mg/kg/d, no adverse effect was noted in any generation, with respect to any of the criteria employed.

At the dose of 500 mg/kg/d, there was a slight but statistically significant retardation in growth of the F0 (p=0,05), F2 (p=0,001) and F3 (p=0,05) males and of the F3 females (p=0,001). The weight attained at 12 weeks was approximately 20 g lower than in the other corresponding control groups. The efficiency of food utilisation was slightly depressed in these same groups, with a significant difference from controls in F0 and F2 males (p=0.05) at the highest dose and in F3 females at the 2 highest doses used (p=0.001). In F3 females, the decrease of food utilisation efficiency was dose related.

Haematological examinations were made at five different periods up to the 100th week and they didn't disclose any aberration. In all groups of each generation and at all time intervals, the findings were within normal limits for the rats. Blood sugar levels and blood non-protein nitrogen levels were normal too. At the 10 000 ppm level, serum cholesterol level was elevated in the females of the F2 generation and in the males of the F0 generation. Urinary findings were negative.

The survival data showed few deaths occurring during the first year of the study. Mortality during the second year was significantly higher among the females that were being carried through the reproduction studies than among the males but the effects were not dose-related. As a matter of fact, the survival of the low and middle dosage levels female rats were somewhat better than the control group, which in turn was equal to the 500 mg/kg/d level. Survival of the males at the highest dose level was 68% compared to 76% in the controls and was better than observed with the two lower dose levels.

The table summarising the organ weight of the F0 generation is missing in the study report. However, information could be found in the original study report of the Food and Drug Laboratory Research : 5/25 F0 females were reported to have a very high absolute liver weight (6.08-8.36 g). Furthermore the missing table also indicates an increased absolute kidney weight of F0 males at the dose level of 500 mg/kg/d. No statistical method was applied to the organ weight data.
The authors indicate a possible dose-dependant increase in liver weight of the F0 females receiving 500 mg/kg/d. They suggest that this may be attributable to the stress of frequent pregnancies and lactation with the concomitant increase in food intake and hence, elevation of dosage level.

In the F0 generation, the principal gross abnormalities seen at autopsy were pulmonary (inflammation, congestion, infection, bronchiectasis…), but of a character commonly seen in laboratory-housed rats. They occurred in one-third to one-half of the rats of each group. Other findings were scattered throughout the groups and in various organs, but in no case did there appear to be a dose-relationship. The liver and kidneys were examined in at least 20 rats of each sex per group and 19 additional organs were examined in at least 10 rats of each sex per group. The incidence of positive finding was somewhat greater in the males than in the females ; however, they do not indicate any particular organ to be affected nor any significant differences between the test groups and the controls. Several of the livers that had elevated weights in relation to body weight showed no histopathologic alterations. The tumour incidence at all levels of TNPP was approximately the same as that of the control group and consisted mainly of fibroadenomas and fibromas of mammary origin. The gross findings at autopsy of the rats of the F1, F2 and F3 generations also revealed no dose-related effects, the principal change being in the lungs and the incidence being greater in the older rats. However, in no case was it greater in the test groups than in the controls (Food and Drug Research Laboratories, 1961).

Overall, based on limited observed effects at the highest dose : a slight retardation of growth in males and an elevation of the absolute liver weight in F0 females, a NOAEL of about 167 mg/kg bw/d (3300 ppm of TNPP in the diet) was derived for this study and 500 mg/kg bw/d (10 000 ppm) is considered as a LOAEL.

- In the study with dogs, three males and three females per group were fed with a diet containing 1000, 3300 and 10 000 ppm TNPP. The report didn't indicate the equivalence between the doses expressed in ppm and the doses in mg/kg bw, absorbed by the dogs. No reproduction testing was conducted, but the same observations and clinical tests as in the rats study were carried out in all of the dogs. In addition, particularly in the control and highest dose level groups, analyses were made of blood serum cholesterol levels, alkaline phosphatase, SGO (Serum Glutamooxaloacetate) and SGP (Serum Glutamopyruvate) transaminases, and anticholinesterase activity. A series of neurological reactions were tested in the control and in the highest dose level group at 12 weeks and at frequent periods thereafter (evaluation of behaviour, of patellar, tonic neck and tonic eye reflexes, and of placing, supporting and righting reactions, were made).

When the study had been under way for five month, one dog at the 1000 and another at the 3300 ppm level died with symptoms of encephalytic meningitis. After confirmation of the diagnosis at autopsy, these deaths were considered irrelevant to the study and the animals were replaced.

For all the other dogs, there was no adverse effect on growth, behaviour and appearance, at any dose level, during the course of the study. Neurological reactions were normal.

The haematological data remained in normal ranges throughout the test period for all groups. No abnormal values were found in blood sugar or non-protein nitrogen levels either. Starting at 48 weeks, the blood cholesterol level of one of the 10 000 ppm female dogs was markedly elevated and continued so to the end of the study. At 100 weeks, there were also high cholesterol levels in the females of this level and also in the female controls, but the values were not excessive in the 1000 and 3300 ppm groups. Alkaline phosphates, SGO and SGP transminases and anticholinesterase values fell within normal limits for dogs. Urinary findings were negative.

Microscopic examination of approximately 23 organs and tissues was realised.

A reticulum cell sarcoma was observed in one male dog at the lowest dose. In this same dog, abnormal germinal epithelial cells were observed in testes. However there was no related finding in any of the dogs at the higher levels.

One female dog exposed to the 3300 ppm level exhibited some granulomas in lungs and a myocarditis and another female dog fed with the highest dose level exhibited reticulated cytoplasm in some hepatic cells. One male dog exposed to the 3300 ppm level exhibited some mucinous degeneration in media of aorta. These findings were considered incidental.

One male dog exposed to the highest dose exhibited a chronic inflammation in renal pelvis. This finding may be linked to the renal impact of TNPP observed in male rats in the 90-day study (described above) and in the OECD TG 421 study (described below).

One other finding of possible significance in the gross or histopathological examinations was a slight to moderate degree of hyperplasia of the thyroid (with focal collections of lymphocytes) in two female dogs at the highest dose level group. A very slight hyperplasia of the thyroid (focal collections of lymphocytes) was also observed in one male control dog (Food and Drug Research Laboratories, 1961).

Based on this thyroid change at the highest dose, a NOAEL of 3300 ppm and a LOAEL of 10 000 ppm of TNPP in the diet were derived from this study with dogs (Food and Drug Research Laboratories, 1961).

● A reproductive/developmental toxicity screening test of TNPP with rats, conducted by Tyl *et al*., 2002, also provides some information on repeated dose toxicity. This well-conducted study (also described in chapter 4.1.2.9) meets and enhances the OECD testing guideline 421. The purity of the test material was of 99.98%.

TNPP was administered by oral gavage once daily, seven days per week in CD® (Sprague-Dawley) rats at dose levels of 50, 200 and 1000 mg/kg/day, at a dose volume of 5 ml/kg/day in Mazola® corn oil. Animals were divided in groups of ten per sex per dose. TNPP was administered for two weeks of prebreed exposure (males and females) and two weeks of mating (males and females) for F0 parents. F0 females continued to be dosed for three weeks each of gestation and lactation, as were F1 offspring (ten per sex per treatment group) from weaning through scheduled sacrifice, at approximately 85 days of age. In addition, five F0 males per group from the control and the 1000 mg/kg/day groups were designated as recovery animals and held without dosing for two weeks, after the F0 male dosing period was completed to evaluate recovery from any possible treatment related effects identified in the high dose.

Observations for mortality were made twice daily and the general condition of all animals was checked daily. Clinical examinations, body weights and feed consumption were recorded

regularly. All F0 parental animals in all groups, as well as the recovery males, all retained F1 adults and non selected F1 weanlings in all groups were subjected to a complete gross necropsy, and full histopathology of the organs was performed for five high dose and control F0 and F1 males and females.

Unscheduled deaths occurred in F0 females at 50 and 1000 mg/kg/day (1 and 4 F0 females respectively). The unscheduled deaths of the low dose F0 female during gestation and one of the high dose F0 female during lactation were attributed to dosing errors and were not considered treatment related. Of the three remaining unscheduled F0 females' deaths, all were found on gestation day 22, possibly attributable to dystocia. Dystocia was evident due to the inability of the dams to deliver their pups. Their demise was considered treatment related. However, this effect is linked with reproduction and thus, won't be taken into account for the assessment of repeated dose toxicity.

F0 parental females did not exhibit any other overt adult systemic toxicity at any dose, as evidenced by a lack of statistically significantly different body weights or gross necropsy findings. However, trends towards increased feed consumption in females from the high dose group (except during lactation) were noted. The authors considered this finding as most likely because of the excessive rooting behaviour observed during the dosing period, however, the excess rooting behaviour observed in F0 females at the highest dose tested was observed during gestation and lactation but not during the prebreed and mating period. Gross necropsy and histological findings of F0 parental females exhibited no treatment- or dose-related pattern of incidence or severity at scheduled sacrifice.

There was no unscheduled death for the adult F1 females. There was no significant difference in body weight or weight gain for the F1 females during the post weaning period. Feed consumption values, presumably associated with excessive rooting behaviour, were increased at 1000 mg/kg/day. There were no treatment-related effects for the gross necropsy or histopathological findings.

There was no treatment-related death for the F0 males. Minor systemic toxicity was present at 1000 mg/kg/day, expressed as trend toward decreased body weights and reduced body weight gains. Feed consumption values, presumably associated with excessive rooting behaviour, were increased at 1000 mg/kg/day during mating. Paired kidney weights, both absolute and relative to terminal body and brain weights, were significantly increased at 1000 mg/kg/day. Histological findings included minimal corticomedullary junction mineralisation of the kidney in three males out of five at 1000 mg/kg/day (with no male with this finding at 0 mg/kg/day), which correlated with the increase of kidney weight at this dose. There was no effect on kidney weights in the recovery group.

There was no unscheduled death for the adult F1 males. There was no significant difference in body weight or weight gain during the post weaning period at any dose. Increased feed consumption at 1000 mg/kg/day, considered related to increasing in rooting behaviour was observed. There was no treatment-related effect for gross necropsy findings. However, histologic findings of F1 males included minimal (one male) and moderate (one male) corticomedullary junction mineralisation of the kidney in two males at 1000 mg/kg/day, versus none at 0 mg/kg/day. These findings were considered treatment related.

The renal lesions observed in F0 and F1 males, were characterised by the presence of basophilic deposits of mineral occurring along the corticomedullary junction. These findings were considered treatment related, since this kind of lesion is rarely, if ever, observed in control males (although it is a common finding in control females). The mineralisation

observed within the high-dose F0 and F1 males appeared similar to the mineralisation noted in the female (control and treated) animals. The increased kidney weight data in the males could have been related to the mineralisation.

Corticomedullary mineralisation in the rat is often diet and/or sex related, but in this study, the reason for the presence of corticomedullary mineralisation in the F0 and F1 male rats could not be determined. No evidence of necrosis or other lesions, which could lead to mineralisation within this area, were observed.

A variety of other histopathologic changes were observed in males and females of both generations at both control and dosage levels (e.g., cyst on renal medulla, necrosis of renal tubule epithelium and nephropathy). These changes were typical of the spontaneous microscopic renal pathology that can be observed at this age and in this strain of rat and were not considered treatment related lesions (Tyl *et al*., 2002).

The abnormal rooting behaviour which is reported in rats at the highest dose level could be associated with a neurotoxic activity of the test compound.

Based on the renal lesions in F0 and F1 males and on the abnormal rooting behaviour (males and females) observed at the dose level of 1000 mg/kg/day, the NOAEL derived from this study for repeated dose toxicity was 200 mg/kg/day.

● A study on delayed neurotoxicity was performed in chicken (Van Velsen *et al*., 1980). The study report was in Dutch ; data presented in the report come from a summary in English. Chickens were exposed to a mixture of tris(mono- and dinonylphenylphosphite (Polygard^{R}) and as positive controls two organophosphates, tri-o-cresylphosphate (TOCP) and O-methyl, O-4-bromine-2,5-dichlorophenyl, phenylphosphonothioate (leptofos) known for causing delayed neurotoxicity. Hubbard chickens weighing 1.5 to 2.5 kg were exposed on day 0 to 4 ml olive oil as control, 400 mg leptofos, 500 mg TOCP or 4000 mg Polygard per kg bw by gavage into the gizzard (6 animals per group). Body weights and food intake were determined once a week. Chickens were taken out of the cage every day to observe the gait and appearance. On day 0, 7, 14, 21 and 28, cholinesterase activity in plasma was measured. From day 28 on, autopsy was performed on 6 animals per day by perfusion via heart with 4% formaldehyde and 1% glutaraldehyde in phosphate buffer (3 animals per group) or by abdominal exsanguination (3 animals per group) both after intravenous injection with Nembutal. From all animals, heart, brain, liver, kidney, stomach and gizzard were histopathological investigated. From the perfused animals, the spinal column and peripheral nerves (nervus ischiadicus, nervus tibialis and nervus peronealis with part of the innerved muscles) were dissected.
Body weights were significantly reduced in the leptofos group (to 64%) and TOCP group (to 87%) with no effects in the Polygard group. Food intake was significantly higher in the Polygard groups in the second, third and fourth week, while leptofos induced a lower food intake. Animals in the leptofos and TOCP group showed signs of delayed neurotoxicity. Cholinesterase activity was not changed in any of the groups on all days. TOCP and leptofos showed degenerative changes in the axons and myelin sheath in the spinal cord and peripheral nerves. No degenerated axons were found in the control and Polygard group. Histological investigations of the other organs resulted in no effects caused by the different treatments.
The authors concluded that it is not plausible that Polygard would result in delayed neurotoxicity in humans, based on the lack of clinical and electronmicroscopical detectable symptoms of delayed neurotoxicity after a single exposure to 4000 mg Polygard/kg bw to chickens.

Human data

No human data was available

4.1.2.6.2 Summary and discussion of repeated dose toxicity

For repeated dose toxicity, confidence is gained by the evaluation of several generations in the two-year studies. These studies provide a profile of limited repeated dose toxicity for TNPP.

A 90-day exposure to a dose of 5000 mg/kg/day (5%) of TNPP in rat resulted in the observation of toxic symptoms and of pathological changes in the kidney, but no adverse effect was observed at lower doses. Over a longer period (2-year), ingestion of TNPP at a dose level of 10 000 ppm (corresponding to 500 mg/kg/d in rats) led to a slight retardation of growth in male rats, an increase of the liver weight in F0 female rats and a thyroid change (doubtful relationship to dosage) in dogs. One male dog exposed to 10 000 ppm also exhibited a renal chronic inflammation in pelvis. In these 2-year studies, 3300 ppm of TNPP in the diet (corresponding to 167 mg/kg/d in rats), was derived as a NOAEL, both for rat and dog. In the modified and enhanced OECD TG 421 study with rats, the NOAEL for systemic toxicity was established at 200 mg/kg/day, based on an excessive rooting behaviour in males and females and on a treatment-dependent corticomedullary junction mineralisation of the kidney in males observed at the highest dose level (1000 mg/kg/day). However, microscopic examination was only performed on 5 males and 5 females of the control and the highest dose group, thus, the NOAEL could not be used for the risk assessment.

Based on this lack of information in the study of Tyl *et al.* and on the respective duration of the studies, the NOAEL used for risk assessment for repeated dose toxicity is 3300 ppm (corresponding in rats to 167 mg/kg), derived from the 2-year study in rat (Food and drug research laboratories) and based on the following limited effects: a slight retardation of growth in males and an elevation of the absolute liver weight in F0 females. This NOAEL is rather conservative.

Factors such as hydration, diet, or intratubular pH may alter the mineral balance within kidneys (Montgomery *et al*, 1990 ; Kahn *et al*., 2002). Additionally, compounds with vitamin D activity could promote mineralisation. Compounds such as oestrogen or having estrogenic activity can influence mineralisation as well, however, the high-dose, F0 and F1 females did not show any evidence of increased severity of mineralisation. There are sex-related differences in the renal metabolism and handling of some xenobiotics in the rat kidney which could have also influenced this change. In particular female kidneys present some kind of down regulation to oestrogen-like compounds as they are exposed to a high level of oestrogens in physiological conditions, whereas male kidney which are not exposed to such a high level of oestrogen are more reactive to an oestrogen-like stimulation.

It could be suggested that abnormal rooting behaviour, reported in rats at 1000 mg/kg/day in the study of Tyl *et al*. (2002) could be linked with a neurotoxic activity of the test compound. However, “rooting in bedding” typically postdosing (but also predosing) in a dose-related incidence was observed in every gavage study performed in rats in the laboratory which conducted the study and in many others too. The consensus is that it is an expression of taste aversion, likely the animal’s attempt to get rid of the bad taste in its mouth from the oral gavage dosing. The higher the dose, the more test material, the greater the incidence of rooting; in this study all rooting was observed postdosing. This behavior is therefore

considered indicative of a conditioned adaptive behavior. Furthermore, abnormal behaviour was not observed in the other available studies. An unpublished study carried out by the Dutch National Institute of Public Health and Environment, on delayed neurotoxicity in chickens did not show any evidence of delayed neurotoxicity in chickens for TNPP (Van Velsen *et al*.,1980).

Classification and labelling :

This chemical is not classified according to the criteria of the European Union. R48 should not be applied.

4.1.2.7 Mutagenicity

Only data from *in vitro* test systems are available.

4.1.2.7.1 Studies *in vitro*

Genetic mutations

- A bacterial reverse mutation assay was recently performed by Wagner and Klug, Bioreliance Laboratory (2001). It was conducted according to the GLP and followed the OECD guideline 471. The purity of the test article was of 98 to 99%.

Salmonella typhimurium tester strains TA98, TA100, TA1535 and TA1537 and *Escherichia coli* tester strain WP2 *urv*A were exposed to TNPP with and without S9 activation. All dose levels of test article, negative controls and positive controls were plated in triplicate.

The substance was dissolved in acetone. Acetone alone was used for the negative controls. Positive control experiments were carried out simultaneously with the following substances : 1) all *Salmonella* strains and WP2 *urv*A with S9 : 2-aminoanthracene ; 2) TA98 without S9 : 2-nitrofluorene ; 3) TA100 and TA1535 without S9 : sodium azide ; 4) TA1537 without S9 : 9-amino-acridine ; 5) WP2 *urv*A without S9 : methyl methanesulfonate.

A preliminary toxic assay was used to establish the dose-range over which the test article would be assayed. Vehicle and ten dose levels of the test article were plated, one plate per dose, with overnight cultures of TA98, TA100, TA1535 and TA1537 and WP2 *urv*A on selective minimal agar in the presence and absence of Aroclor-induced rat liver S9.

The mutagenicity assay was then used to determine the mutagenic potential of the test article. A minimum of five dose levels of test article (0, 75, 200, 600, 1800, 5000 µg/plate) along with appropriate vehicle and positive controls were plated with TA98, TA100, TA1535 and TA1537 and WP2 *urv*A in the presence and absence of Aroclor-induced rat liver S9.

In the preliminary toxicity assay, the maximum dose tested was 5000 µg per plate. Precipitate was observed beginning at 1000 µg per plate. No appreciable toxicity was observed. Based on the findings of the toxicity assay, the maximum dose plated in the mutagenicity assay was 5000 µg per plate. Precipitate was observed beginning at 600 µg per plate. No appreciable toxicity was observed and no positive response was observed with any of the tester strains in the presence and in the absence of metabolic activation.

TNPP was concluded to be negative in the Bacterial Reverse Mutation Assay.

● Another bacterial reverse mutation assay was performed by Ciba Geigy in 1990. This Salmonella/mammalian-microsome mutagenicity test was carried out in accordance with GLP and the OECD guideline 471 (May 26, 1983) with the exception of statistical analysis. The purity of the test substance was > 94%.

Salmonella typhimurium tester strains TA98, TA100, TA1535 and TA1537 were exposed to TNPP with and without S9 activation. The activation mixture contained S9 fraction of liver from rats induced with Aroclor and a solution of co-factors.

The substance was dissolved in acetone. Acetone alone was used for the negative controls. Positive control experiments were carried out simultaneously with the following substances : 1) for strain TA 98 : donorubicin-HCl 2) for strain TA 100 : 4-nitroquinoline-N-oxide 3) for strain TA 1535 : sodium azide 4) for strain TA 1537 : 9(5)-amino-acridine hydrochloride monohydrate.

All dose levels of test article, negative controls and positive controls were plated in triplicate and the experiments were repeated in order to confirm the results.

A preliminary toxic assay (9 concentrations ranging from 20 to 5000 µg/0.1ml) was used to establish the dose-range over which the test article would be assayed. From the results obtained, the highest concentration suitable for the mutagenicity test was found to be 5000 µg/0.1 ml. The following concentrations were used with and without microsomal activation : 313, 1250, 2500 and 5000 µg/0.1 ml.

The test substance is considered to be positive in this test system if a reproductible increase of the mean number of revertants per plate above that of the negative control, at any concentration level, by at least a factor of 1.5 and 2 respectively for strain TA 100 and for strains TA 98, TA 1535 and TA 1537, is observed.

In the experiments performed without and with microsomal activation, comparison of the number of histidine-prototrophic mutants in the controls and after treatment with TNPP revealed no marked differences. At the concentrations of 2500 and 5000 µg/0.1 ml, the test substance precipitated in soft agar.

No evidence of the induction of point mutations by TNPP or by its metabolites formed as a result of microsomal activation was detectable in the strains of *Salmonella typhimurium* used in these experiments.

● Other Ames tests were conducted in a large Japanese study (Hachiya, 1987), which aim was the evaluation of chemical genotoxicity by a series of short-term tests (82 substances were subjected to a battery of short-term assays). However, documentation is insufficient for an assessment of the study, as the report is in Japanese version, except for the summary and the table of results.

The results with TNPP were all negative with and without metabolic activation for the *Salmonella typhimurium* strains TA97, TA98, TA100 and TA102 and for *Escherichia coli* strain WP2/pKM102, at concentrations up to 5000 µg/plate.

● An *in vitro* mammalian cell gene mutation test was conducted too, following OECD guideline 476 and in compliance with GLP (San and Clarke, 2001). The purity of the test article was of 98 to 99%.

TNPP was tested in the L5178Y/TK$^{+/-}$ Mouse Lymphoma Mutagenesis Assay. Exposures were for 4 hours in the absence and presence of metabolic activation (Aroclor-induced rat liver S9). The vehicle used was acetone. Methyl methanesulfonate was used as the positive control for the non-activated test system and 7,12-Dimethylbenz(a)anthracene was used as positive control for the S9- activated test system.

Based on the results of the preliminary toxicity assay, the doses chosen for treatment of the mutagenesis assay ranged from 1.0 to 200 µg/ml for both the non-activated and S9-activated cultures. Visible precipitate was present at concentrations of ≥ 100 µg/ml in treatment medium. No visible precipitate was present at concentrations of ≤ 50 µg/ml in treatment medium. The doses chosen for cloning ranged from 5.0 to 100 µg/ml with and without S9 activation. No cloned cultures exhibited mutant frequencies, that were at least 55 mutants per 10^6 clonable cells over that of the solvent control. Toxicity in the cloned cultures was not observed at any dose levels.

TNPP was concluded to be negative in the L5178Y/TK$^{+/-}$ Mouse Lymphoma Mutagenesis Assay.

●An *in vitro* gene mutation test with Chinese hamster cells V79 was also conducted by Ciba-Geigy (1990). The test was carried out in accordance with GLP and the OECD guideline 476. The purity of the test substance was > 94%.

TNPP was dissolved in ethanol. Two negative controls (ethanol) and one positive control (N-nitroso-dimethylamine) were also tested. The study was conducted with and without activation (Aroclor-induced rat liver S9). The cells were treated in the experiments with microsomal activation for 5 hours and in the experiments without microsomal activation for 21 hours. The results of each original experiment were confirmed in a second and independent experiment (confirmatory experiment). Based on the results of a preliminary toxicity assay, the original experiments were performed at the following concentrations with microsomal activation : 0.6, 1.2, 2.4, 4.8, 7.2, 9.6 and 12.0 µg/ml and without microsomal activation : 0.3, 0.6, 1.2, 2.4, 3.6, 4.8 and 6.0 µg/ml. Because the intended toxicity was not obtained in the original experiments, in the confirmatory experiments, the concentrations applied were increased to 0.8, 1.6, 3.2, 6.4, 9.6, 12.8 and 16.0 µg/ml with microsomal activation and to 0.4, 0.8, 1.6, 3.2, 4.8, 6.4 and 8.0 µg/ml without microsomal activation. In all the experiments, comparison of the number of mutant colonies in the controls and in the cultures treated with the various concentrations of the test substance revealed no significant deviation of the mutant frequencies.

In both investigations with and without microsomal activation, criteria for a negative response were reached : a difference in the treated and untreated dishes of at least 20 clones per 10^6 cells plated was not detected and there was no indication of a concentration mutant-frequency relation in any experiment.

TNPP was concluded to be negative in the gene mutation test with Chinese hamster cells V79.

Chromosomal effects

● One mammalian chromosome aberration assay, following OECD guideline 473 and in compliance with the GLP was carried out on Chinese hamster ovary (CHO) cells both in the absence and presence of metabolic activation (Aroclor-induced S9 activation system) (Gudi and Brown, 2001). The purity of the test article was of 98 to 99%.

Acetone was determined to be the solvent of choice, based on the solubility of the test article and compatibility with the target cells. Mitomycin C was used as the positive control in the non-activated test system and cyclophosphamide was used as the positive control for the S9 activated test system.

The preliminary toxicity assay was performed for the purpose of selecting dose levels for the chromosome aberration assay and consisted of an evaluation of test article effect on cell growth. The chromosome aberration assay was performed using standard procedures (Evans, 1976), by exposing duplicate cultures of CHO cells to the test article as well as positive and solvent controls.

Dose levels for the chromosome aberration assay were selected based upon the lowest precipitating dose : the doses chosen ranged from 18.75 to 200 µg/ml for both the non-activated and the S9 activated 4-hour exposure groups and from 6.25 to 150 µg/ml for the non-activated 20-hour continuous exposure group. No toxicity and no statistically significant structural or numerical chromosomal aberrations were observed under any treatment condition in the assay.

TNPP was concluded to be negative for the induction of structural and numerical chromosome aberration in Chinese hamster ovary cells.

● Another mammalian chromosome aberration assay, following OECD guideline 473 and in compliance with GLP was carried out on Chinese hamster ovary cells (cell line CCL 61) both in the absence and presence of metabolic activation (Aroclor-induced S9 activation system) (Ciba-Geigy, 1990). The purity of the test article was > 94%.

TNPP was dissolved in acetone. Acetone was used as negative control. Mitomycin C 0.2 µg/ml, a mutagen not requiring S9 activation and cyclophosphamide 40.0 µg/ml, which requires activation, were used as positive controls.

Based on the results of a preliminary toxicity assay, the 2 experiments of the original study were performed at the following concentrations : 62.5, 125.0 and 250.0 µg/ml (with activation and without activation). Since at the upper concentration of 250.0 µg/ml, only metaphase of inferior quality, insufficient for scoring were present, the concentrations of 31.25, 62.5 and 125.0 µg/ml were selected for the four experiments of the confirmatory study (with and without activation).

The number of cells with specific chromosomal aberrations in the treatment groups showed no marked difference in comparison with the negative control. The incidence of changes observed is within the range of spontaneous aberrations inherent to this particular cell line used.

No evidence of clastogenic effects was obtained in Chinese hamster ovary cell *in vitro* treated with TNPP.

4.1.2.7.2 Studies *in vivo*

No *in vivo* study is available.

4.1.2.7.3 Summary of mutagenicity

In vitro mutagenetic tests did not reveal any genotoxic effect in six well-conducted tests, two Bacterial Reverse Mutation Assays, two *in vitro* Mammalian Cell Gene Mutation Tests, and two *in vitro* Mammalian Chromosome Aberration Tests.

Although neither human data nor *in vivo* tests are available, the available data from *in vitro* tests support the view that TNPP is a non-genotoxic substance.

Classification and labelling :

This chemical is not classifiable as mutagenic according to the criteria of the European Union.

4.1.2.8 Carcinogenicity

There is no carcinogenicity study available, however, two studies that were described in the repeated dose toxicity section (4.1.2.6) were conducted on rats and dogs for 2 years. These studies are not in compliance with international guidelines for the assessment of carcinogenicity because the number of animals used was not sufficient, compared to what is required by the international guidelines. In the rat experiment, there were only 25 rats per sex for each group instead of 50 in the guideline, and not all of them were examined histopathologically for all organs : the liver and kidneys were examined in at least 20 rats of each sex per group and 19 additional organs were examined in at least 10 rats of each sex per group. In the experiment with dogs, 3 dogs per sex per group were used.

In these chronic ingestion studies, TNPP was incorporated in nutritionally adequate rations for the respective species at levels of 1000, 3300 and 10 000 ppm in the diet and comparison was made with control groups receiving the basal diet without the additive.

In the rat experiment, histopathological examinations of the liver and kidney of all except an occasional (autolysed) animal and of the spleen, adrenal, thyroid and pituitary gland, heart, stomach, small and large intestine, pancreas, bladder, gonads, salivary glands, lymph nodes, lungs, bone marrow, muscle, brain and spinal cord in at least half the rats at all levels in F0 generation were conducted. All tumours were examined microscopically in F0 generation.

The tumour incidence (Table 4-9) at all levels of TNPP was approximately the same as that of the control group and consisted mainly of non-malignant tumours (fibroadenomas and fibromas of mammary origin).

In the dog experiment (Table 4-10), a reticulum cell sarcoma was observed in one male dog at the lowest dose. In this same dog, abnormal germinal epithelial cells were observed in testes. However there was no related finding in any of the dogs at the higher levels. (Food and Drug Research Laboratories, 1961).

Table 4-9: Summary of Histopathological Findings in F0 generation rats (Negative findings omitted)

Organ and finding	Dose (mg/kg)							
	0		50		167		500	
	M	F	M	F	M	F	M	F
Tumours :								
Necrotic tumour	1							
Reticulum cell sarcoma	1		2					
Fibroadenoma		6	1	1	1	3		8
Fibroma		1	2	1		1	1	1
Adenoma		1		1		1		1
Linoma			1					
Malignant papillary mesothelioma				1				
Squamous carcinoma				1				
Adenocarcinoma					1			
Spindle cell sarcoma					1			
Liposarcoma					1			
Teratoma						1		
Angiomyoma							1	
Plasma cell tumour involving spleen, liver, BM, LN and soft tissues							1	
Mammary carcinoma								1

[1] : Liver and kidneys examined in 20 or more rats per sex per group ; the remaining 19 tissues were examined in at least 10 rats per sex per group.

Table 4-10: Summary of gross and histopathological findings in dogs (negative findings omitted)

Level	Dog No & sex[4]	Fate[5]	Gross Abnormalities	Histopathological Findings[6]
ppm :				
None	476M	S-2yr		
	482M	"		
	488M	"	Colon : few petechial hemorrhages	Thyroid : very slight hyperplasia and focal collections of lymphocytes
	492M	"		
	474F	"		
	483F	"	Spleen mottled	
1000	479M	S-2yr		
	489M	"		Mucinous degeneration in media of aorta (1 of 2 sections)
	493M	D 655 days	Possible tracheal insufflation of food	Reticulum cell sarcoma in lung, liver, spleen and lymph node. Abnormal germinal epithelial cells in testes.
	472F	S-2yr		
	495F	"		Kidneys : small foci chronic inflammation in medulla and pelvis
3300	477M	S-2yr		
	490M	"	Focal hemorrhages in small intestine	
	494M	"		
	473F	"		Granulomas in lung; myocarditis
	485F	"		
10 000	478M	S-2yr		
	481M	"		
	491M	"		Kidneys : chronic inflammation in pelvis
	475F	"	Hemorrhagic mucosa in intestines	
	486F	"		Thyroid : slight focal hyperplasia collections of lymphocytes.
	487F	"		Moderate diffuse hyperplasia; Liver : reticulated cytoplasm in some hepatic cells

[4] Dogs No. 464 F (3300 ppm) and 571F (1000 ppm) had not completed the 2-year period at the time of this data collection.

[5] S = sacrificed, D = died

[6] Organs examined were liver, kidneys, spleen, aorta, heart, lungs, stomach, small and large intestines, pancreas, gall bladder, urinary bladder, salivary glands, thymus, gonads, adrenal and thyroid glands, lymph nodes, bone marrow, muscle, brain, spinal cord and pituitary gland.

Summary

There are no reliable study available on carcinogenicity, however, on the basis of the information currently available on mutagenicity, TNPP is considered as a non-genotoxic substance, so concerns for cancer caused by a genotoxic mechanism are low.

Considering the potential for carcinogenicity by a non-genotoxic mechanism, no evidence of a significant increase of tumour incidence was found in the 2-year chronic studies carried out on a small sample of rats and dogs.

Although only limited data are available, these data tend to indicate that TNPP is not of concern for a carcinogenic potential.

Classification and labelling :

This chemical is not classifiable as a carcinogen according to the criteria of the European Union.

4.1.2.9 Toxicity to reproduction

Reproduction and developmental toxicity were evaluated in two oral multi-generation studies in rats and in one non-standard study on chick embryos.

Fertility and reproductive toxicity

● The 2-year study on rats (described in the repeated dose toxicity section 4.1.2.6), combines a chronic and a three-generation reproductive toxicity study. This study is not GLP and it does not follow specific international guideline, however, this multi-generation study is well-documented and meets basic scientific principles.

Rats were distributed into four groups of 25 males and 25 females each and fed with a diet containing TNPP at a level designed to be equivalent to approximately 1000, 3300 and 10000 ppm (daily intake corresponding to 50, 167 and 500 mg/kg bw).

After the rats were on test for approximately 100 days, reproduction and lactation studies were initiated and mating continued through the lifetime of the females of the F0 generation. From the second litters born of these dams, young were raised and carried through similar feeding and reproduction studies. Their young, and in turn the descendant of that generation, were carried through similar experiments, making a total of three descendant generations studied. Reproduction and lactation experiments on the F0 generation were carried through six matings. The F1 and F2 generations were carried through only two matings and the F3 generation was not mated.

Records were kept of the date of birth, the number of pups born, their weight and survival during lactation. The criteria employed for evaluating the performance of the rats were a series of indexes for fertility, gestation, viability and lactation.

Reproductive organs (testes and ovaries) were autopsied and weighed.

Growth was normal at all dosage levels in F0, F1 and F2 females. At the dose level of 500 mg/kg/d, there was a slight but statistically significant retardation in growth of the F2 (p=0,001) and F3 (p=0,05) males and of the F3 females (p=0,001), along with a decrease in the efficiency of food utilisation for F2 males (p=0.05) at the highest dose and F3 females at

the 2 highest doses used (p=0.001). In F3 females, the decrease of food utilisation efficiency was dose related.

The authors state that the findings related to reproductive parameters were comparable with observations in rats of the stock colony and were considered normal. No statistical analysis was performed but this seems true for most of the parameters. There was no indication of adverse effect in the F0 generation at any dose level. However, diminution in the number of pups born per litter in the F1 and F2 high dose groups, and a small decrease in the fertility and viability indexes in F2 at this same high dose level exposure were observed (see Table 4-11). (Food and Drug Research Laboratories, 1961).

Based on those results, indicating a possible effect on reproduction at the dose of 500 mg/kg/d, a NOAEL for reproduction of 167 mg/kg bw/day can be derived.

Table 4-11: Comparison of first two matings in three generations of rats

Dose	Generation	Total No. of mating	No. litters born alive	Pups born alive	Pups per litter born	No. litters weaned	Average weight of pups at weaning[7]	F.I.[8]	G.I.[9]	V.I.[10]	L.I.[11]
Mg/kg							**Gm**				
None	**F0**	49	41	328	8.0	34	40.0	98.0	82.9	87.2	96.2
	F1	20	19	216	11.3	19	36.3	95.0	100.0	87.0	89.5
	F2	20	17	151	8.9	16	42.7	90.0	94.5	93.2	87.5
50	**F0**	49	40	354	8.8	36	36.5	91.8	90.0	91.8	88.0
	F1	20	20	213	10.7	20	41.6	100.0	100.0	96.0	90.0
	F2	20	19	159	8.4	16	40.0	95.0	94.5	87.6	81.1
167	**F0**	50	45	415	9.2	41	37.9	94.0	95.7	95.7	87.7
	F1	20	20	212	10.6	20	40.1	100.0	100.0	95.5	94.5
	F2	20	19	151	8.0	12	42.6	95.0	100.0	94.5	71.0
500	**F0**	48	40	337	8.4	37	36.0	100.0	83.3	93.8	87.3
	F1	17	16	113	7.0	13	36.0	100.0	100.0	93.5	96.0
	F2	20	17	122	7.3	13	43.8	85.0	100.0	79.7	89.7

[7] At 21 days
[8] Fertility index = (No. pregnancies / No. matings) X 100
[9] Gestation index = (No. litters born alive / pregnancies) X 100
[10] Viability index = (No. pups at 1d. / No. pups born alive) X 100
[11] Lactation index = (No. pups at 21d. / No. pups at 1d.) X 100

● A study combining a reproductive and a developmental toxicity screening of TNPP was conducted on CD® (Sprague-Dawley) rats (Tyl *et al.*, 2002). This recent, well-conducted study, was performed in compliance with OECD Guideline 421 and GLP. It exceeded the OECD TG 421 study design as follows : enhanced evaluation of toxicity in the F0 generation, including the evaluation of a recovery group of males ; evaluation of developmental landmarks in the F1 generation (time of vaginal opening or preputial separation, normality and length of oestrous cycle) ; and following the F1 offspring to adulthood, with continued exposure and assessment of reproductive structures and functions including potential effect on sperm.

The purity of the test material was of 99,98 %.

TNPP was administered by oral gavage once daily, seven days per week in rats at dose levels of 50, 200 and 1000 mg/kg/day, at a dose volume of 5 ml/kg/day in Mazola® corn oil, ten animals/sex/dose. TNPP was administered for two weeks of prebreed exposure (males and females) and two weeks of mating (males and females) for F0 parents. F0 females continued to be dosed for three weeks each of gestation, lactation, as were F1 from weaning through scheduled sacrifice. On the day of birth (postnatal day or pnd 0), anogenital distance was measured and bodyweight recorded for all live F1 pups in all litters. They were also examined to determine the number of viable and stillborn pups from each litter. Thereafter, litters were evaluated for survival on pnd 4, 7, 14 and at weaning (pnd 21). F1 litters were culled on pnd 4 to yield, as nearly as possible, five males and five females per litter. The culled F1 pups were weighed, euthanized and necropsied with complete external and visceral examinations. For the remaining F1 pups, survival indices were calculated at least weakly through weaning (pnd 21). At weaning, at least one female and one male (whenever possible) from each F1 litter were randomly selected to continue treatment for approximately seven more weeks, with dosing for F1 selected pups begun on pnd 22 until all pups were at least 85 days of age. F1 postweaning observations and procedures for each retained female included examination for patency of vaginal opening (from pnd 22 until acquisition of vaginal opening). Oestrous cyclicity and normality were evaluated by vaginal smears from F1 females taken daily the last three weeks of the postwean exposure period prior to scheduled sacrifice. For each retained F1 male offspring, observations for the cleavage of the balanopreputial gland (preputial separation) began at 35 days of age and continued until acquisition of preputial separation. Andrologic assessment was also performed on the F1 retained males at necropsy. All F0 parental animals and retained F1 adults were subjected to a complete gross necropsy with the following organs : ovaries, uterus with cervix and vagina, prostate, epididymides, testes and seminal vesicles with coagulating glands and their fluids.

In this part of the hazard identification, the results of this study will focus on the effects related to reproduction and development.

Three of ten pregnant F0 females at 1000 mg/kg/day died in late pregnancy (gestation day 22). These deaths may have been related to dystocia, since the dams appeared to be unable to deliver their normal appearing pups. Examination of the F1 pups and the necropsy of 3 dams indicated that the pups were full term and normal in external appearance. Two F0 females respectively exposed to 50 mg/kg/day (during gestation) and 1000 mg/kg/day (during lactation) were also found dead. But these deaths were attributed to dosing errors and were not considered treatment related.

Ovary weights (absolute and relative to terminal body and brain weights) were significantly decreased at 1000 mg/kg/day in F0 but not F1 adult females.

There was no effect of exposure to TNPP on any F0 reproductive indices during the production of F1 offspring. Mating, fertility, pregnancy and gestational indices were equivalent across groups ; gestational length was equivalent across all groups. According to the authors, there was also no difference across groups for the number of total implantation sites per litter, percent post-implantation loss per litter, or number of total, live or dead pups per litter at birth.

There was evidence of F1 offspring toxicity (see Table 4-12) observed postnatally at 1000 mg/kg/day, expressed, according to the authors as reduced litter size on postnatal day (pnd) 4,but not on postnatal day 0. This finding could be linked with maternal toxicity, expressed as a behaviour change towards its litter. However, the assumption of the study report (reduced litter size on pnd 4 but not on pnd 0) is not in accordance with the tables of values provided in the report. The tables indicate that there is a reduction of litter size at the highest dose, compared to the control group and that this reduction is the same at pnd 0 and at pnd 4 tending to indicate a direct offspring toxicity and not an indirect one, linked with a maternal behaviour change.

There was no treatment-related death for the adult F1 males and females. No effect on reproductive parameters, developmental landmarks, F1 oestrous, or F1 andrology was observed. In F1 males, paired epididymides weight, relative to terminal body weights, were significantly decreased at 1000 mg/kg/day.. There was no treatment-related effect for gross necropsy findings in F1 males and females.

Table 4-12: Summary of F1 offspring toxicity

	Trisnonylphenyl Phosphite (mg/kg/day)			
	0	50	200	1000
N° of live litters				
Postnatal Day 0	10	8	10	7
Postnatal Day 4	10	7[a]	10	7
Postnatal Day 7	10	7	10	7
Postnatal Day 14	10	7	10	7
Postnatal Day 21	10	7	10	6[b]
Average number of live pups per litter (pnd 0)	14.9** ± 0.5	12.8 ± 1.6	15.9 ± 0.6	12.0 ± 1.4
Average number of live pups per litter (pnd 4, precull)	14.8** ± 0.5	14.3 ± 0.6	15.6 ± 0.5	12.0* ± 1.4
Average number of live pups per litter (pnd 7, postcull)	9.8 ± 0.1	10.0 ± 0.0	10.0 ± 0.0	9.1 ± 0.9
Average number of live pups per litter (pnd 14, postcull)	9.8 ± 0.1	10.0 ± 0.0	10.0 ± 0.0	9.1 ± 0.9
Average number of live pups per litter (pnd 21, postcull)	9.8 ± 0.1	10.0 ± 0.0	10.0 ± 0.0	10.0 ± 0.0

[a] The entire litter for female 30 was missing and presumed dead on postnatal day 4.

[b] Female 24 was found dead (possible dosing error) on postnatal day 15, therefore, her entire litter had to be euthanized and was not included in any parameters after postnatal day 14.
* $p<0.05$ Dunnett's test
** $p<0.05$ ANOVA test

The decrease in ovary weight (absolute and relative) in F0 females and the decrease in relative paired epididymides weight in F1 males, at 1000 mg/kg/d may be related to an hormonal,

oestrogen-like effect of the substance. Actually, regarding the decreased paired epididymides weight, the difference in the dosing period could be a reasonable explanation for, why this organ weight is decreased in the F1 males and not in the F0 males. The F1 males are dosed during the critical period of reproductive system development thereby enhancing sensitivity to endocrine disrupters compared to the parent generation, which are only dosed during adulthood. Andrology parameters measured at 1000 mg/kg/day did not reveal any change compared to controls.

Based on a slight reduction of the litter size, on a slight decrease in relative paired epididymides weight in F1 males and on signs of maternal toxicity (death on gestation day 22, decrease in ovary weight) at 1000 mg/kg/day, NOAELs for maternal and offspring toxicity of 200 mg/kg/day were derived from this study.

Developmental toxicity

● In this reproductive and a developmental toxicity screening study (Tyl *et al.*, 2002), no effect was observed in developmental landmarks in the F1 generation (time of vaginal opening or preputial separation, normality and length of oestrous cycle) at any dose level.

All F1 pups culled on pnd 4 (see Table 4-12) were subjected to a complete external and visceral examination, including examination of all thoracic and abdominal organs, bisection of kidneys and heart dissection. These examinations did not reveal any developmental effect up to the dose level of 1000 mg/kg/day. The NOAEL for teratogenicity is ≥ 1000 mg/kg/day.

However, it must be underlined that while the OECD 421 study design specifies termination of the study on pnd 4, with external and internal examination of the F1 pups at this time, the modified study design used in this study provides, for continuation of the F1 offspring, with continuing exposure until sexual maturity. Thus, to provide data on the pnd 4 pups, the pups culled to standardise litters on pnd 4 were euthanised and subjected to complete gross necropsy, but this number is very limited since F1 litters were culled on pnd 4 to yield, as nearly as possible, five males and five females per litter. This leads to nearly 2 animals in the highest dose group and 4 in the other groups. The other pups were subjected to a complete gross necropsy at weaning (pnd 21), except for at least one male and one female per litter that were selected to continue treatment for seven more weeks. No malformation was observed at any stage.

● The effects of TNPP on the survival rate of chick embryos were studied. This non-standard study is very shortly reported and is not considered as an adequate developmental toxicity study. The materials employed (purity not indicated) were submitted to the Food and Drug Research Laboratories by Weston Chemical, Inc.

A dose of 5 mg of TNPP in corn oil was injected directly into the yolk sac of fertile hen's eggs. Two replicate runs were conducted, in which groups of 16 fertile White Leghorn eggs were used per group. With each series, there were four comparison groups : (1) untreated eggs, as a control on hatchability, (2) eggs which were drilled and the needle inserted into the yolk sac, without the injection of anything, (3) eggs injected with distilled sterile water and (4) eggs injected with corn oil from the same lot used to prepare the sample solution.

The result was given in terms of percent of survival of the embryos at 5, 10 and 18 days of incubation and in terms of the final number of live chicks which hatched.

It was evident that any manipulation that disrupted the integrity of the egg membrane caused some mortality of the embryos. Mortality was further slightly increased by the injection of either water or plain corn oil. No statistical test was performed but conclusion was made difficult as results appeared to be different between the two runs, one of them showing a very slight effect of

the injection of TNPP and the other one showing a more important effect on embryos' survival. Examination of embryos that died before hatching and of the newly hatched chicks revealed no gross abnormality or malformation peculiar to the test groups (Food and Drug Research Laboratories, 1971).

Summary and discussion

TNPP exposure over four generations did not reveal any significant effect on reproduction up to 500 mg/kg/d, the highest dose tested, except for a possible reduction of litter size, born from F1 and F2 generations at the highest dose. This slight tendency seems to be confirmed by the OECD 421 study in which a slight but significant litter size reduction was observed at the highest dose (1000 mg/kg/day). In this same study, maternal toxicity was observed at the dose of 1000 mg/kg/day. At the dose of 1000 mg/kg/day, a decrease of the ovary weight of F0 females and the decrease of epididymides weight in F1 males suggest an oestrogen-like activity of the test substance. No other significant effects on reproductive toxicity were observed in this study.

Phenomenon of dystocia observed in dams at the highest dose in the study of Tyl (2002) is viewed as maternal toxicity, due from the adjustments of dosing volume on gd 14 and especially on gd 20, resulting in over dosing the dams in late gestation. Actually, the dosing volume of the test chemical was adjusted for each dam based on each new body weight. This means that the dosing volumes for the F0 dams during gestation were adjusted on gd 0, 7, 14, and 20. The pregnant rat CD (SD) females gain approximately 150 g or more during gestation but with the body weight gain from gd 14 to parturition (the "last trimester") of at least 100 g, due almost entirely to the rapid growth of the uterine contents. For gavage studies, test chemical intake (in mg/day) during this period is increased by as much as 30% because of the adjustment for maternal body weight, especially from gd 20 to parturition (gd 22 ± 1). Thus, the dose in mg/kg/day, based on the actual maternal body weight minus the uterine contents, is similarly increased by ~30%. This can result in overdosing the dam (and conceptuses) and is likely the cause of the excessive peri-parturitional maternal toxicity observed.
The risk of increased maternal toxicity in late pregnancy from bolus gavage dosing is due to: (a) the maternal liver (although it is enlarged in late pregnancy in response to the pregnancy and the increased test chemical load) is not enlarged commensurate with the increased test chemical dose; (b) test chemical is likely not equally distributed between maternal and fetal compartments, so the relative maternal burden may be even greater; and (c) gastrointestinal tract motility is reduced in late pregnancy, so there is likely increased absorption of the test chemical from the gut due to longer transit times.

Based on these observations, the NOAELs for reproductive toxicity and for maternal toxicity were 200 mg/kg/day, derived from the OECD 421 study (considered as a key study for risk characterisation as a recent study, following OECD guideline).

No indication of any developmental effect was observed in both of the studies. $NOAEL_{terato}$ is ≥ 1000 mg/kg/day, although these parameters were observed on a very reduced number of animals.

Classification and labelling :

This chemical is not classified as toxic to reproduction (fertility and development) according to the criteria of the European Union.

4.1.3 Risk characterisation

4.1.3.1 General aspects

No human data are available, so this assessment of the hazardous properties of TNPP is based only on animal data.

No studies on Toxicokinetics were conducted so the only information we have are those that can be derived from the physico-chemical properties of the molecule : a relatively high molecular weight (MW = 689g.mol^{-1}), an extremely low water solubility and a high Log P_{ow}. Thus absorption of TNPP is expected to be limited.

From many studies by oral, dermal or intraperitoneal route, it can be assumed that the acute toxicity of TNPP is very low. The derived LD_{50} for oral route is 19.5 +/- 3.3 gram/kg. Acute dermal and intraperitoneal limit tests (at 2000 mg/kg and 1000 mg/kg respectively) did not cause any mortality. No data is available on the acute inhalation toxicity, although the non-corrosive and very slight irritant nature of TNPP may suggest that toxicity would not be enhanced following exposure by this route. It is to note that tests conditions proposed by OECD guidelines do not elicit irritant or corrosive properties of the test substance although stronger conditions may reveal a moderate irritation or a possible corrosive action of TNPP to the skin. TNPP is a slight eye irritant.

According to the maximisation test, TNPP showed a strong grade of skin-sensitising potential in albino guinea pigs. Although a Buehler sensitisation test gave a negative result, the maximisation test is preferred as it is the most sensitive one. No information on respiratory tract sensitisation is available.

The main toxic effect is a renal impact observed in F0 and F1 male rats in a reproductive/developmental toxicity screening test, at the highest dose of 1000 mg/kg/day by oral route (microscopic examination was only performed in control and high dose group animals in this study). Paired kidney weights, both absolute and relative to terminal body and brain weights, were significantly increased at this dose and histological findings included corticomedullary junction mineralisation of the kidney. The renal lesions observed in F0 and F1 males were characterised by the presence of basophilic deposits of mineral occurring along the corticomedullary junction. In a 2-year study on rat, an oral dose of 10 000 ppm (corresponding to 500 mg/kg/day) led to a few effects on growth and liver weight. A NOAEL of 3300 ppm, corresponding to 167 mg/kg/day was derived from this long-term repeated dose toxicity study and can be used in the risk characterisation.
No repeated dose studies with inhalation and dermal application route were available.

Concerning mutagenicity, TNPP was negative in all of the six well-conducted studies available *in vitro* (two Bacterial Reverse Mutation Assays, two *in vitro* Mammalian Cell Gene Mutation Tests, and two *in vitro* Mammalian Chromosome Aberration Tests). No *in vivo* study was available, overall, the evidence indicates that TNPP is not mutagenic.

There are no carcinogenic studies conducted according to international guidelines. However, some information on the carcinogenic potential of TNPP can be derived from other data. On the basis of the information currently available, it is unlikely that TNPP is mutagenic, so concerns

for cancer caused by a genotoxic mechanism are low. Considering the potential for carcinogenicity by a non-genotoxic mechanism, no evidence of an increase of tumour incidence was seen in 2-year repeated dose toxicity studies in rat and dog. There is low concern for carcinogenicity by a non-genotoxic mechanism too.

TNPP exposure over several generations did not reveal any significant effect on reproduction up to the highest dose tested (500 mg/kg/day) in F0 but a slight reduction of litter size born from F1 and F2 generations, which tended to be confirmed by the OECD 421 study in which a slight but significant litter size reduction was observed at the highest dose (1000 mg/kg/day). In this same study, maternal toxicity was observed at the dose of 1000 mg/kg/day. At the dose of 1000 mg/kg/day a decrease of the ovary weight of F0 females and the decrease of epididymides weight in F1 males suggest an oestrogen-like activity of the test substance.

The NOAELs for reproductive toxicity and for maternal toxicity, were derived from the OECD 421 study and were considered to be 200 mg/kg/day.

No indication of any developmental effect was observed in both of the studies. $NOAEL_{terato}$ is ≥ 1000 mg/kg/day, although these parameters were observed on a very reduced number of animals on pnd 4 due to the modified procedure of the study. However, internal and external examination on a larger number of pups, on pnd 21 also gave negative results.

Overall for the risk characterisation :

$NOAEL_{maternal}$ = 200 mg/kg/day

$NOAEL_{repro}$ = 200 mg/kg/day

$NOAEL_{terato}$ ≥ 1000 mg/kg/day

4.1.3.2 Workers

Occupational exposure may occur by inhalation and dermal route during manufacture of TNPP, manufacture of products and use of preparations containing TNPP.

Route-to-route extrapolation and calculation of internal doses

Inhalation and dermal route are the relevant occupational routes whereas all NOAELs are available by oral route only. Therefore route-to-route extrapolation has to be done and corrections should be made for differences in bioavailability as determined by percentages of absorption.

There are no data on the absorption of TNPP for the different routes of exposure. For oral absorption (starting route), a default value of 50 % may be chosen. For inhalation route, a default absorption of 50 % is proposed. For dermal absorption, a default value of 10 % can be used based on a MW>500 and log P_{ow} higher than 4.

Internal doses are presented in table 4.4 via the different routes for each scenario. They are calculated using a human body weight of 70 kg and a ventilation rate of 10 m^3/8 hours.

Table4-13: Calculated internal doses for workers

Scenario	Route of penetration				Combined routes
	Inhalation		Dermal		
	External exposure mg/m^3	Internal dose mg/kg/day	External exposure mg/day	Maximum internal dose mg/kg/day	Internal dose mg/kg/day
1 - TNPP manufacture	2.86	0.20	0 - 42	0.06	0.26

2 - Manufacture of products containing TNPP	8.58	0.61	42 - 420	0.60	1.21
3 - Use of preparations containing TNPP	5.72	0.41	0.42 - 4.2	0.006	0.42

For risk characterisation at the workplace, MOSs should normally be determined for route-specific as well as combined inhalation and dermal exposure. For simplification, only MOSs derived from combined exposure are presented.

4.1.3.2.1 Acute toxicity

Acute dermal toxicity was found to be very low (LD_{50} > 2000 mg/kg). No data is available for acute inhalation toxicity but taking into account the very low acute toxicity by dermal and oral routes and that TNPP is a very slight to moderate irritant, inhalation acute toxicity is likely to be low as well. Acute toxicity is not considered of concern.

Conclusion (ii) for all scenarios

4.1.3.2.2 Irritation

TNPP is considered as a slight skin and eyes irritant and it may be presumed that it does not induce significant respiratory irritation. Therefore irritative effects are not considered of concern.

Conclusion (ii) for all scenarios

4.1.3.2.3 Sensitisation

One study conducted according to Buehler gave a negative response while a positive result was observed in a maximisation test. Thus TNPP is classified as a skin sensitiser.

No human data are available, however, according to the TNPP consortium, no case of sensitisation was observed at existing production sites. There are no data on respiratory sensitisation.

Exposure to TNPP during manufacture of the substance, manufacture of products and use of preparations may lead to concern. Risk reduction measures which should be applied as a result of its classification as the proper use of personal protective equipment can effectively reduce sensitisation at the work place. However, if protective equipment is not used properly and conscientiously and appropriate work procedures are not followed, it is likely that sensitisation might be induced in the worker. Although proper personal protection use and work procedure might be in use in most of the plants handling TNPP, there is no certainty that this is the situation of all plants in the EU. Conclusion iii is drawn in all worker scenarios. This conclusion is mitigated given the non dispersive use of the substance and the lack of reported case of sensitisation.

Conclusion (iii) for all scenarios

4.1.3.2.4 Repeated dose toxicity

Comparing the estimated combined internal exposure with the NOAELs of 167 mg/kg/day derived from a 2-year study in rats, the following MOSs can be calculated:

Table 4-14: MOSs for systemic effects by repeated exposure

Scenario	Internal Exposure mg/kg/day	Internal NOAEL mg/kg/day	MOS	Conclusion
1 - Manufacture	0.26	83.5	321	ii
2 – Manufacture of products	1.21	83.5	69	ii
3 – Use of preparations	0.42	83.5	199	ii

The effects observed at the LOAEL in the 2-year study in rats (500 mg/kg/day) are changes on growth and liver weight. The main toxic effect is a renal impact observed in a reproductive/developmental toxicity screening test in rats at 1000 mg/kg/day.

A minimal MOS of 50 can be derived from the following assessment factors:

- 10 for interspecies differences (default value)
- 5 for intraspecies differences (homogeneous population)
- 1 for type of the effect
- 1 for the confidence in the data base.

Compared to the minimal MOS, the MOSs are considered acceptable.

Conclusion (ii) for all scenarios

4.1.3.2.5 Mutagenicity

Available *in vitro* data do not reveal a genotoxic potential. Effects are not anticipated to occur.

Conclusion (ii) for all scenarios

4.1.3.2.6 Carcinogenicity

Data concerning carcinogenicity are not available. Based on results of mutagenicity testing, TNPP is not anticipated to be a genotoxic carcinogen. There is a low concern for carcinogenicity by a non-genotoxic mechanism too.

Conclusion (ii) for all scenarios

4.1.3.2.7 Toxicity to reproduction

Fertility and reproductive toxicity

Comparing the estimated combined internal exposure with the NOAEL of 200 mg/kg/day derived from a reproductive/developmental study in rats, the following MOSs can be calculated :

Table 4-15: MOSs for reproductive effects by repeated exposure

Scenario	Internal Exposure mg/kg/day	Internal NOAEL mg/kg/day	MOS	Conclusion
1 - Manufacture	0.26	100	385	ii
2 – Manufacture of products	1.21	100	87	ii
3 – Use of preparations	0.42	100	238	ii

The adverse effects observed at 1000 mg/kg/day in the reproductive/developmental study in rats are decrease of ovary weight in F0 females, a decrease of epididymes weight in F1 males and a slight litter size reduction. No other significant reproductive effects were observed.

A minimal MOS of 50 can be derived from the following assessment factors:

- 10 for interspecies differences (default value)
- 5 for intraspecies differences (homogeneous population)
- 1 for the type of the effect
- 1 for the confidence in the data base

Compared to the minimal MOS, the MOSs are considered acceptable.

Conclusion (ii) for all scenarios

Developmental effects

No indication of any developmental effect was observed up to the highest dose of 1000 mg/kg/day. Effects are not anticipated to occur.

Conclusion (ii) for all scenarios

4.1.3.3 Consumers

4.1.3.3.1 Introduction

Risk may occur by ingestion of food in contact with plastic containing TNPP. It is the only route of significant exposure for the consumer.

4.1.3.3.2 Risk characterisation due to migration from food contact materials

The total daily intake due to food-contact materials has been estimated to 0,0337 mg/day. For an adult with a bodyweight of 70 kg, the systemic dose resulting from this unique route of ingestion is 0,48 µg/kg/day. Systemic and reproductive effects are observed in animals with repeated dose. With the available NOAELs, the following MOSs can be calculated:

Table 4-16: MOS for systemic effects by repeated exposure

Scenario	Exposure µg/kg/day	NOAEL mg/kg/day	MOS	Concern for risks to human health	Conclusion
Food contact materials	0.48	167	350000	low	ii

Table 4-17: MOS for reproductive effects by repeated exposure

Scenario	Exposure µg/kg/day	NOAEL mg/kg/day	MOS	Concern for risks to human health	Conclusion
Food contact materials	0.48	200	420000	low	ii

4.1.3.3.3 Summary of risk characterisation for consumers

Repeated dose toxicity and reproductive effects are of low concern (**conclusion ii**).

4.1.3.4 Human exposed via the environment

This section was not provided as it will be updated in the next version of the environmental risk assessment.

4.1.3.4.1 Summary of risk characterisation for exposure via the environment

This section was not provided as it will be updated in the next version of the environmental risk assessment

4.2 HUMAN HEALTH (PHYSICO-CHEMICAL PROPERTIES)

Exposure assessment

The exposure assessment, to the extent it is related to physico-chemical properties, has already been discussed. No specific exposure information is available.

Effects assessment : Hazard identification and dose (concentration) - response (effect) assessment

Explosivity

TNPP has no explosive properties.

Flammability

TNPP has a very low degree of flammability (flash point : 207°C).

Oxidising potential

TNPP has no oxidising potential.

Risk characterisation

TNPP has neither explosive nor oxidising properties. The likelihood of an adverse effect deriving from flammability is very low.

Conclusion (ii) for all scenarios

5 RESULTS

5.1 ENVIRONMENT

To be updated

5.2 HEALTH

Risk assessment of human exposed via the environment was not discussed and will be updated following the update of environment risk assessment.

( ) (i) There is a need for further information and/or testing.

(X) (ii) There is at present no need for further information and/or testing and no need for risk reduction measures beyond those, which are being applied already.

This conclusion applies to the assessment of the risk to human health through consumer exposure.

(X) (iii) There is a need for specific measures to limit the risks.

This conclusion applies to the assessment of the risk to human health through worker exposure. It is reached because of concerns for sensitisation as a consequence of dermal exposure arising during manufacture of the substance, manufacture of products or use of preparations containing TNPP.

6 REFERENCES

Health part

AWG (1998) Alkylphenol Work Group, Potential dietary exposure to p-nonylphenol from food contact use of tris(nonylphenyl)phosphite (TNPP)

Chapin RE, Delaney J, Wang Y, Lanning L, Davis B, Collins B, Mintz N, Wolfe G, 1999. The effects of 4-nonylphenol in rats: a multigeneration reproduction study. Toxicol Sci Nov 52:1 80-91

Ciba-Geigy, 1992. Acute dermal toxicity in the rat. Test n° 924059.

Ciba-Geigy, 1983. Acute intraperitoneal LD_{50} in rat. Project n° 831156.

Ciba-Geigy, 1981. Skin irritation in the rabbit after single application of TK 10 417. Project n° 811807.

Ciba-Geigy, 1981. Eye irritation in the rabbit after single application of TK 10 417. Project n° 811806

Ciba-Geigy, 1992. Skin sensitisation test in the Guinea pig, Maximisation test. Test number 924058.

Ciba-Geigy, 1990. Gene mutation test with Chinese hamster cells V79. test number 894549.

Ciba-Geigy, 1990. Salmonella/Mammalian-microsome mutagenicity test. Test number 894547.

Ciba-Geigy, 1990. Chromosome studies on Chinese hamster ovary cell line CCL 61 in vitro. Test number 894548.

Cunny HC, Mayes BA, Rosica KA, Trutter JA, Van Miller JP, 1997. Subchronic toxicity (90-day) study with para-nonylphenol in rats. Regul Toxicol Pharmacol Oct 26:2 172-8

DG Sanco - European Health & Consumer Protection Directorate-General. Provisional list of monomers and additives notified to European Commission as substances which may be used in the manufacture of plastics intended to come into contact with foodstuffs (updated to 25 July 2003). http://europa.eu.int/comm/food/food/chemicalsafety/foodcontact/synoptic

European Union Risk Assessment Report on Nonylphenol ; Priority Setting, 2nd Priority List. R017_0104_env_hh

Evans HJ, 1976. Cytological methods for detecting chemical mutagens. A. Hollaender (Ed.), Chemical Mutagens, Principles and Methods for their Detection, vol. 4. Plenum Press, New York, NY.

FDA (2001) Safety Assessment of Di(2-ethylhexyl)phthalate (DEHP) Released from PVC Medical Devices. http://www.fda.gov/cdrh/ost/dehp-pvc.pdf

FDA (2002) Preparation of Food Contact Notifications and Food Additive Petitions for Food Contact Substances: Chemistry Recommendations, FINAL GUIDANCE, April 2002

http://www.cfsan.fda.gov/~lrd/foodadd.html

Food and Drug Research Laboratories, 1957. Toxicological studies with TNPP. Acute oral DL_{50}. Subacute Feeding Tests. Unpublished report n°72652

Food and Drug Research Laboratories, 1961. Two-year feeding studies of TNPP in rats and dogs. Unpublished report n°79060

Food and Drug Research Laboratories, 1971. The effects of Three Samples on the Survival Rate of Chick Embryos. Unpublished report n° 0582

Green T, 2001. Nonylphenol : Bioavailability in the rat. CTL ReportCTL/024139 ; Submitted to APERC October 24, 2001

Gudi R and Brown C, 2001. *In Vitro* Mammalian Chromosome Aberration test. Unpublished report n° AA37MU.331.BTL, BioReliance, Rockville, MD 20850

Hachiya N, 1987. Evaluation of Chemical Genotoxicity by a series of Short-term Tests. Akita J. Med, 14, 269-292

Han XD, Tu ZG, Gong Y, Shen SN, Wang XY, Kang LN, Hou YY Chen JX. The toxic effects of nynylphenol on the reproductive system of male rats. Reprod. Toxicol. 2004, 19, 215-221

Hill Top Research, 1965. Acute oral administration of nonylated phenyl phosphite (samples #1 and #2), Mark 488, and Mark 492 to rats. Unpublished report

Hossaini A, Dalgaard M, Vinggaard AM, Frandsen M, Larsen J-J. *In utero* reproductive study in rats exposed to nonylphenol. Reprod. Toxicol. 2001, 15, 537-543

Howe *et al.* (2001) Potential dietary exposure to p-nonylphenol from food contact use of tris(nonylphenyl)phosphite (TNPP), Food Additives and Contaminants, 2001, Vol.18(11), 1021 – 1039

INSPQ - Institut National de Santé Publique du Québec, Les phtalates : état des connaissances sur la toxicité et l'exposition de la population générale. (2004) http://www.inspq.qc.ca/pdf/publications/252-Phtalates.pdf

Kahn KNM and Alden CL, 2002. Kidney. In. Handbook of Toxicologic Pathology, 2nd Ed. Academic Press, San Diego, CA, pp.255-336

Majlathova L, Lekarskej F, Komenskeho M, 1981. Evaluation of aralkyl phenylphosphite antioxidants by an acute peroral experiment on mice and rats and by epicutaneous and conjonctival test on rabbits. Bratisl. Lek. Listy ; vol 76, ISS 3, 315-26

Miller LC, Tainter ML, Proc. Soc. Exptl. Biol. And Med., 57, 261 (1944)

Montgomery CA and Seely JC, 1990. Kidney. In. Pathology of the Fischer rat. Academic Press, San Diego, CA, pp. 127-153

Nagao T, Wada K, Marumo H, Yoshimura S, Ono H, 2001. Reproductive effects of nonylphenol in rats after gavage administration : a two-generation study. Reprod Toxicol May-Jun 15:3 293-315

Odum J, Lefevre PA, Tinwell H, Van Miller JP, Joiner RL, Chapin RE, Wallis NT, Ashby J, 2002. Comparison of the developmental and reproductive toxicity of diethylstilbestrol administered to rats in utero, lactationally, preweaning, or postweaning. Toxicol Sci Jul 68:1 147-63

Reimer GJ, 2001. Physical/Chemical Property of Tris-nonylphenol Phospite (TNPP). Hydrolysis as a function of pH (OECD 111). Unpublished report n° 1711234, Reimer Analytical and Associates Inc., Vancouver, BC, V6S 1B6

San RHC and Clarke JJ, 2001. *In Vitro* Mammalian Cell Gene Mutation Test (L5178Y/TK$^{+/-}$ Mouse Lymphoma Assay). Unpublished report n° AA37MU.702.BTL, BioReliance, Rockville, MD 20850

Tyl RW, Hamby BH, Myers CB, Marr MC, 2002. Reproductive/Developmental Toxicity Screening Test of Trisnonylphenyl Phosphite (TNPP) administred via Oral Gavage to CD® (Sprague-Dawley) Rats (Modified OECD 421). Unpublisbed report n° 65C-07895.300, Center for Life Sciences and Toxicology, Research triangle park, NC 27709-2194

Tay CH, 2001a. Tris-nonylphenol (TNPP) : Acute dermal toxicity study - (OECD 402). Unpublished Report n° 01-4176-G1, Toxicon Corporation

Tay CH, 2001b. Tris-nonylphenol (TNPP) : Acute dermal irritation/corrosion - (OECD 404). Unpublished Report n° 01-4176-G2, Toxicon Corporation

Tay CH, 2001c. Tris-nonylphenol (TNPP) : Acute eye irritation/corrosion - (OECD 405). Unpublished Report n° 01-4176-G3, Toxicon Corporation

Tay CH, 2001d. Tris-nonylphenol (TNPP) : Buehler sensitisation test - (OECD 406). Unpublished Report n° 01-4176-G4, Toxicon Corporation

F.L van Velsen, J.A.M.A. Dormans, G.J.A. Speijers, and M.J. van Logten, 1980.Research on delayed neurotoxicity in chickens after treatment with organophosphites. RIVM report 78/80

Wagner VO and Klug ML, 2001. Bacterial Reverse Mutation Assay. Unpublished report n° AA37MU.502.BTL, BioReliance, Rockville, MD 20850

Environment part

Building Research Establishment Ltd. (2001). http://www.bre.co.uk/

Chemical Information Services (2002). http://www.chemicalinfo.com/

Crompton (2003). WESTON TNPP Phosphite, product information, 08-01-2003.

E.C. (2003). Technical Guidance Document on Risk Assessment in support of Commission Directive 93/67/EEC on Risk Assessment for new notified substances, Commission Regulation (EC) N° 1488/94 on Risk Assessment for existing substances, Directive 98/8/EC of the European Parliament and of the Council concerning the placing of biocidal products on the market. Luxembourg, Office for Official Publications of the European Communities.

E.C. (2004a). EUSES 2.0, the European Union System for the Evaluation of Substances.National Institute of Public Health and the Environment (RIVM).

E.C. (2004b). EUSES, the European Union System for the Evaluation of Substances, Version 2.0.National Institute of Public Health and the Environment (RIVM).

OECD (2004). Emission scenario document on plastics additives, Organisation for Economic Co-operation and Development: 125.

Phoenix_Chemical_Laboratory (1997). Vapor pressure by isoteniscope (ASTM D2879). Chicago, May 19, 197, General Electric Specialty Chemicals: 1.

Pittsburgh_Testing_Laboratory (1978). Flash Point. Pittsburgh, July 12, 1978: 1.

Reimer&Associates (2001a). Physical/chemical property of tris-nonylphenyl phosphite (TNPP) [26523-78-4] - Solubility in Water (OECD 105). Pittsfield, August 2001, General Electric Company: 19.

Reimer&Associates (2001c). Physical/chemical property of tris-nonylphenyl phosphite (TNPP) [26523-78-4] - n-Octanol/Water partition coefficient. Pittsfield, August 2001, General Electric Company: 4.

Reimer&Associates (2001e). Physical/chemical property of tris-nonylphenyl phosphite (TNPP) [26523-78-4] - Melting (pour) point (OECD 102). Pittsfield, August 2001, General Electric Company: 10.

Reimer&Associates (2001f). Physical/chemical property of tris-nonylphenyl phosphite (TNPP) [26523-78-4] - Boiling temperature (OECD 103). Pittsfield, August 2001, General Electric Company: 10.

Staples, C. A. (2001). Fugacity modeling to estimate transport between environmental compartments for tris-nonylphenyl phosphite (TNPP) (CAS Reg. No.26523-78-4). Fairfax, November 2001, Assessment Technologies, Inc.: 5.

Ullmann (1985). Ullmann's encyclopedia of industrial chemistry, vol., VCH.

United States Testing Company, I. (1990). Report of Test. Hoboken, New Jersey, February 6, 1990, United States Testing Company, Chemical Services Division: 1.

US EPA and Syracuse Research Corporation (2001). EPI Suite, v.3.10, US EPA.

GLOSSARY

Standard term Abbreviation	Explanation/Remarks and Alternative Abbreviation(s)
Ann.	Annex
AF	assessment factor
BCF	bioconcentration factor
bw	body weight / *Bw, b.w.*
°C	degrees Celsius (centigrade)
CAS	Chemical Abstract System
CEC	Commission of the European Communities
CEN	European Committee for Normalisation
CEPE	European Council of the Paint, Printing Ink and Artists' Colours Industry
d	day(s)
d.wt	dry weight / dw
DG	Directorate General
DT_{50}	period required for 50 percent dissipation (define method of estimation)
DT_{50lab}	period required for 50 percent dissipation under laboratory conditions (define method of estimation)
DT_{90}	period required for 90 percent dissipation (define method of estimation)
$DT_{90field}$	period required for 90 percent dissipation under field conditions (define method of estimation)
EC	European Communities
EC	European Commission
EC_{50}	median effective concentration
EEC	European Economic Community
EINECS	European Inventory of Existing Commercial Chemical Substances
EU	European Union
EUSES	European Union System for the Evaluation of Substances
f_{oc}	Fraction of organic carbon
G	gram(s)

PNEC(s)	Predicted No Effect Concentration(s)
$PNEC_{water}$	Predicted No Effect Concentration in Water
(Q)SAR	Quantitative Structure Activity Relationship
STP	Sewage Treatment Plant
TGD	Technical Guidance Document[12]
UV	Ultraviolet Region of Spectrum
UVCB	Unknown or Variable composition, Complex reaction products or Biological material
v/v	volume per volume ratio
w/w	weight per weight ratio
w	gram weight
GLP	Good Laboratory Practice
h	hour(s)
ha	Hectares / *h*
HPLC	High Pressure Liquid Chromatography
IARC	International Agency for Research on Cancer
C_{50}	median immobilisation concentration or median inhibitory concentration 1 / *explained by a footnote if necessary*
ISO	International Standards Organisation
IUPAC	International Union for Pure Applied Chemistry
kg	kilogram(s)
kPa	kilo Pascals
K_{oc}	organic carbon adsorption coefficient
K_{ow}	octanol-water partition coefficient
Kp	Solids water partition coefficient
l	litre(s)
log	logarithm to the basis 10
$L(E)C_{50}$	Lethal Concentration, Median
LEV	Local Exhaust Ventilation
m	Meter
µg	microgram(s)

[12] Commission of the European Communities, 1996. Technical Guidance Documents in Support of the Commission Directive 93/67/EEC on risk assessment for new substances and the Commission Regulation (EC) No 1488/94 on risk assessment for existing substances. Commission of the European Communities, Brussels, Belgium.
ISBN 92-827-801[1234]

mg	milligram(s)
MAC	Maximum Accessibility Concentration
MOS	Margins Of Safety
NOAEL	No Observed Adverse Effect Level
NOEC	No Observed Effect Concentration
NOEL	No Observed Effect Level
OEL	Occupational Exposure Limit
OECD	Organisation for Economic Co-operation and Development
OJ	Official Journal
pH	potential hydrogen -*logarithm* (to the base 10) of the hydrogen ion concentration $\{H^+\}$
pKa	-*logarithm* (to the base 10) of the acid dissociation constant
pKb	-*logarithm* (to the base 10) of the base dissociation constant
Pa	Pascal unit(s)
PEC	Predicted Environmental Concentration
STP	Sewage Treatment Plant
WWTP	Waste Water Treatment Plant

ANNEX 1

European Commission

EUR .:[click here to insert EUR No.] - European Union Risk Assessment Report
[click here to insert SUBSTANCE NAME, and volume no.]

Editors: B.G. Hansen, S.J. Munn, S. Pakalin, C.J.A. Heidorn, R. Allanou , S. Scheer, G. Pellegrini, S. Vegro, J.De Bruijn, M.Luotamo, K. Vormann, H. Loonen, F. Berthault, A. Naughton, V. Anfossi, L. Praderio.(keep this updated)

Luxembourg: Office for Official Publications of the European Communities

2000 – VIII pp. .:[click here to insert page count, number of pages.]
pp. – 17.0 x 24.0 cm

Environment and quality of life series

ISBN .:[click here to insert ISBN No.]

Price (excluding VAT) in Luxembourg: EUR .:[click here to insert price.]

The report provides the comprehensive risk assessment of the substance Trisnonylphenylphosphite (TNPP). It has been prepared by France in the frame of Council Regulation (EEC) No. 793/93 on the evaluation and control of the risks of existing substances, following the principles for assessment of the risks to man and the environment, laid down in Commission Regulation (EC) No. 1488/94.

Conclusions of risk assessment [click here to insert text]

Conclusions of risk assessment [click here to insert text]