# Ethanol, 2-[(4-amino-2-methyl-5-nitrophenyl)amino]-: Human health tier II assessment

**03 July 2015**

## CAS Number: 82576-75-8


- Preface
- Chemical Identity
- Import, Manufacture and Use
- Restrictions
- Existing Work Health and Safety Controls
- Health Hazard Information
- Risk Characterisation
- NICNAS Recommendation
- References


## Preface

This assessment was carried out by staff of the National Industrial Chemicals Notification and Assessment Scheme (NICNAS) using the Inventory Multi-tiered Assessment and Prioritisation (IMAP) framework.

The IMAP framework addresses the human health and environmental impacts of previously unassessed industrial chemicals listed on the Australian Inventory of Chemical Substances (the Inventory).

The framework was developed with significant input from stakeholders and provides a more rapid, flexible and transparent approach for the assessment of chemicals listed on the Inventory.

Stage One of the implementation of this framework, which lasted four years from 1 July 2012, examined 3000 chemicals meeting characteristics identified by stakeholders as needing priority assessment. This included chemicals for which NICNAS already held exposure information, chemicals identified as a concern or for which regulatory action had been taken overseas, and chemicals detected in international studies analysing chemicals present in babies' umbilical cord blood.

Stage Two of IMAP began in July 2016. We are continuing to assess chemicals on the Inventory, including chemicals identified as a concern for which action has been taken overseas and chemicals that can be rapidly identified and assessed by using Stage One information. We are also continuing to publish information for chemicals on the Inventory that pose a low risk to human health or the environment or both. This work provides efficiencies and enables us to identify higher risk chemicals requiring assessment.

The IMAP framework is a science and risk-based model designed to align the assessment effort with the human health and environmental impacts of chemicals. It has three tiers of assessment, with the assessment effort increasing with each tier. The Tier I assessment is a high throughput approach using tabulated electronic data. The Tier II assessment is an evaluation of risk on a substance-by-substance or chemical category-by-category basis. Tier III assessments are conducted to address specific concerns that could not be resolved during the Tier II assessment.

These assessments are carried out by staff employed by the Australian Government Department of Health and the Australian Government Department of the Environment and Energy. The human health and environment risk assessments are conducted and published separately, using information available at the time, and may be undertaken at different tiers.

This chemical or group of chemicals are being assessed at Tier II because the Tier I assessment indicated that it needed further investigation.

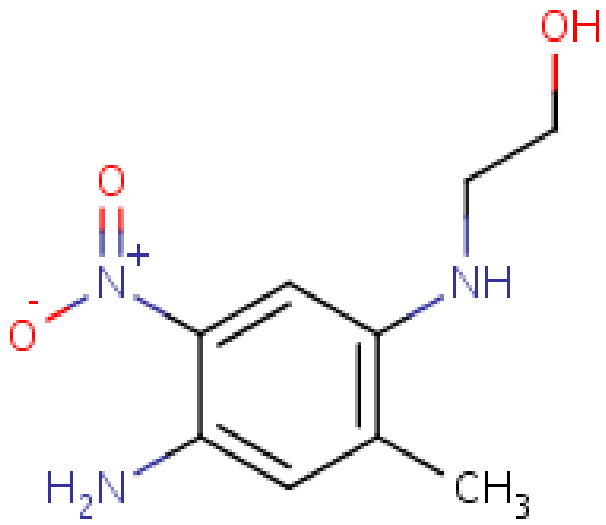
For more detail on this program please visit:www.nicnas.gov.au

**Disclaimer**



Acronyms & Abbreviations

## Chemical Identity

| | |
|---|---|
| Synonyms | HC Violet 1<br>Imexine FAA<br>1-amino-3-methyl-4(2-hydroxyethyl)amino-6-nitrobenzene |
| Structural Formula |  |
| Molecular Formula | C9H13N3O3 |
| Molecular Weight (g/mol) | 211.2 |
| Appearance and Odour (where available) | bright anthracitic crystalline powder |
| SMILES | c1(N)c(N(=O)=O)cc(NCCO)c(C)c1 |

## Import, Manufacture and Use

### Australian

The chemical is on the 'List of chemicals used as dyes in permanent and semi-permanent hair dyes in Australia' (NICNAS, 2007), with reported cosmetic use in semi-permanent hair dye preparations.

### International

The following international uses have been identified through Galleria Chemica; the European Commission Cosmetic Ingredients and Substances (CosIng) database; the United States (US) Personal Care Product Council International Nomenclature of Cosmetic Ingredients (INCI) Dictionary; and the US Environmental Protection Agency's Aggregated Computer Toxicology Resource (ACToR).

The chemical has reported cosmetic use as a hair dye substance in oxidative and non-oxidative hair dye products.

## Restrictions

### Australian

No known restrictions have been identified.

### International

The chemical is listed on the following (Galleria Chemica):

- The European Union (EU) Regulation (EC) No 1223/2009 Annex III, Part 1—List of substances which cosmetic products must not contain except subject to the restrictions and conditions laid down: '(a) the maximum authorised concentration in the finished cosmetic products as a hair dye substances in non-oxidative hair dye products is 0.5 %; and (b) after mixing with hydrogen peroxide under oxidative conditions the maximum concentration applied to hair must not exceed 0.25 %';
- New Zealand Cosmetic Products Group Standard—Schedule 5: Components cosmetic products must not contain except subject to the restrictions and conditions laid down;
- The Association of Southeast Asian Nations (ASEAN) Cosmetic Directive Annex III- Part 1—List of Substances which cosmetic products must not contain except subject to restrictions and conditions laid down;

The Scientific Committee on Consumer Products (SCCP) was of the opinion that ‘the use of HC Violet n° 1 itself as a semipermanent hair dye at a maximum concentration of 0.28 % and as an oxidative hair dye at a maximum concentration of 0.25% in the finished cosmetic product (after mixing with hydrogen peroxide) does not pose a risk to the health of the consumer, apart from its sensitising potential’. It was also noted that the HC Violet 1 should not be used in combination with nitrosating substances. The nitrosamine content should be <50 ppb (SCCP, 2006).

## Existing Work Health and Safety Controls

### Hazard Classification

The chemical is not listed on the Hazardous Substances Information System (HSIS) (Safe Work Australia).

### Exposure Standards

Australian

No specific exposure standards are available.

### International

No specific exposure standards are available.

# Health Hazard Information

## Toxicokinetics

No data are available.

## Acute Toxicity

### Oral

The chemical has low acute oral toxicity based on results from animal tests following oral exposure. The median lethal dose (LD50) was >2000 mg/kg bw in rats.

In an acute oral toxicity study conducted according to Organisation for Economic Co-operation and Development (OECD) Test Guideline (TG) 401, five female Sprague Dawley (SD) rats were administered (gavage) a single oral dose of 2000 mg/kg bw of the chemical. Clinical observations were made for a period of 14 days. No mortality was reported. All animals showed hypoactivity, piloerection and dyspnoea on day one. No other treatment related effects were noted. Necropsy examination revealed no abnormalities. The maximum non-lethal dose was >2000 mg/kg bw (SCCP, 2006).

In another acute oral toxicity study conducted according to OECD TG 401, the chemical was administered (gavage) to SD rats (10). Observations were made daily up to 14 days. All animals showed dark brown staining of the fur but no other treatment related abnormalities were seen. The acute oral LD50 was >2000 mg/kg bw (SCCP, 2006).

### Dermal

No data are available.

### Inhalation

No data are available.

## Corrosion / Irritation

### Skin Irritation

The chemical is reported to slightly irritate skin in animal studies. The effects were not sufficient to warrant hazard classification.

In a skin irritation study in three New Zealand White rabbits, 500 mg of Imexine FAA (trade name of the test chemical; purity 98.5 %) moistened with 0.5 mL water was applied to an approximately 6 $cm^2$ area of abraded and intact skin under a patch. The patches were removed 24 hours after application. Observations were made from 1-48 hours after removal of the patches. All

treated skin sites were stained purple due to the test chemical. Very slight erythema and haemorrhage of the dermal capillaries was noted at intact and abraded skin sites 25 hours after application. Three intact and two abraded skin sites showed very slight oedema 25 hours after application. Very slight erythema and desquamation were noted in one intact and one abraded skin site after 72-hours observation. One intact skin site had desquamation (SCCP, 2006).

In another skin irritation study conducted according to OECD TG 404, 500 mg of Imexine FAA (trade name of the test chemical; purity 98.5 %), moistened with 0.5 mL water, was applied to three male New Zealand White rabbits to approximately 6 $cm^2$ area of the skin under a patch . The patches were removed four hours after the application and observations were made at 1, 24, 48, and 72 hours. All treated sites were stained red due to the test chemical and the evaluation of erythema was obscured. One animal showed severe oedema at one hour and a slight oedema was seen in two animals after one hour but not after 24 hours. The colouration of the skin diminished and reactions were not noted at 48 and 72 hours (SCCP, 2006).

### Eye Irritation

The chemical is reported to be a slight eye irritant in animal studies. The effects were not sufficient to warrant hazard classification.

In an eye irritation study, 73 mg of the chemical (corresponding to a volume of 0.1 mL) was instilled into the conjunctival sac of one eye of three New Zealand White rabbits. Observations were made for up to seven days following treatment. One treated eye showed pale violet staining of the iris one hour after treatment. One animal showed conjunctival irritation, with redness, chemosis and discharge one hour after treatment. Two animals had minimal conjunctival irritation, with redness and chemosis, with or without discharge. All treatment related effects were reversed by day two (SCCP, 2006).

In another eye irritation study conducted according to OECD TG 405, 0.1 mL volume of 1 % chemical dissolved in a 0.5 % suspension of carboxymethylcellulose was instilled into the left conjunctival sac of eye of three male New Zealand White rabbits. Observations were made for up to 72 hours following administration. Two animals showed very slight redness of the conjunctival on days one and two. No other treatment related effects were noted (SCCP, 2006).

## Sensitisation

### Skin Sensitisation

The chemical is considered to be a skin sensitiser based on the positive results seen in local lymph node assays (EC3 is 0.9 %). The available information supports classification for the chemical as skin sensitiser (refer to **Recommendation** section).

In a local lymph node assay (LLNA) conducted according to OECD TG 429, 25 µL of the chemical at 1, 2.5, 5, 10 or 25 % (w/v) in dimethyl formamide (DMF) was applied to the dorsal surface of both ear lobes of female CBA/J mice (five females/group) once daily for three consecutive days. The chemical produced stimulation indices (SIs) of 4.08, 6.11, 9.55, 9.11, 5.64 and 9.79, respectively. The estimated concentration needed to produce a three-fold increase in lymphocyte proliferation (EC3) was calculated to be between 1 and 5 %, indicating strong skin sensitisation potential (SCCS, 2006).

In another LLNA study conducted according to OECD TG 429, 25 µl of the chemical at 0.1, 0.25, 0.5, 1 or 2.5 % (w/v) in DMF was a applied to the dorsal surface of both ear lobes of female CBA/J mice (five females/group) once daily for three consecutive days. The chemical produced SIs of 1.20, 1.67, 1.30, 3.40, 3.83 or 11.05, respectively. The estimated concentration needed to produce a three-fold increase in lymphocyte proliferation (EC3) was calculated to be 0.9 %, indicating strong skin sensitisation potential (SCCS, 2006).

## Repeated Dose Toxicity

### Oral

The limited available data suggest that the chemical does not cause severe effects following repeated oral exposure.

In a repeated dose oral toxicity study, the chemical (in a suspension of 0.5 % aqueous carboxymethyl cellulose) was administered (gavage) to Sprague Dawley derived Crl:CD (SD)BR rats (10/sex) at 50, 150 or 500 mg/kg bw/day for 13 weeks. All treated animals had purple fur and tail staining from the first week of treatment and hair loss and scab formation were seen in all animals from day 48 onwards. Significant dose-related increases in mean absolute and relative liver weights were noted in males of all treatment groups. The females showed slight but significant decreases in absolute liver weight in low dose group and dose-related increases in the mid and high dose groups. Relative kidney weights were slightly increased (6 %) in high dose males, while females showed a significant and dose-related increase in relative kidney weights (7 %, 11 % and 18 %, respectively). Livers of male animals in the high dose groups showed moderate congestion (SCCP, 2006).

Although the authors concluded that the no-observed toxic effect level (NOAEL) in this study was 50 mg/kg bw/day, the SCCP considered the dose of 50 mg/kg bw/day as the lowest observed adverse effect level (LOAEL) due to the change in the kidney weight in this group (SCCP, 2006).

### Dermal

No data are available.

### Inhalation

No data are available.

## Genotoxicity

The available data from in vitro and in vivo studies indicate that the chemical does not have mutagenic or clastogenic potential.

***In vitro studies***

Two independent bacterial gene mutation assays were conducted according to OECD TG 471 using five *Salmonella typhimurium* strains (TA98, TA 100, TA 102, TA1535 and TA 1537).

Concentrations of 1.6, 8, 40, 200, 1000 or 5000 µg/plate of the chemical were used for experiment 1 and 156.25, 312.5, 625, 1250, 2500 or 5000 µg/plate for experiment 2. Both experiments were carried out with and without metabolic activation. A twofold increase of the revertant numbers was seen only in TA98 strains at 1000 and 625 µg/plate of the chemical. The chemical was concluded to be slightly mutagenic in strain TA98 only (SCCP, 2006).

Two independent mammalian cell gene mutation assays (HPRT locus) were conducted according to OECD TG 476 using L5178Y *tk* $^{+/-}$ mouse lymphoma cells. The chemical was tested up to a maximum concentration of 1000 µg/mL in the absence and presence of a rat liver metabolic activation system. Negative results were reported (SCCP, 2006).

A micronucleus test in accordance with the recommendations of the International Workshop on Genotoxicity Testing (IWGT) using human peripheral blood lymphocytes from two healthy female donors was also conducted. The chemical was tested up to a maximum concentrations of 1720 µg/mL, with and without metabolic activation. The chemical did not cause an increase in the induction of micronuclei in cultured human peripheral blood lymphocytes (SCCP, 2006).

***In vivo studies***

In a mouse micronucleus test, the chemical was administered (gavage) to CD-1 mice (five animals/sex/dose) at concentrations of 500, 1000 or 2000 mg/kg bw as a single dose. Bone marrow cells were collected at 24 or 48 hours (for the 2000 mg/kg dose group only) after administration of the chemical and the polychromatic erythrocytes (PCE) were examined. As no increase in micronucleated PCEs was found, the chemical was concluded to be non-mutagenic in this study (SCCP, 2006).

Another micronucleus test was also conducted according to OECD TG 474 in Crl:CD(SD)BR rats (five animals/sex/dose). The chemical was administered (gavage) at concentrations of 500, 1000 or 2000 mg/kg bw as a single dose. Bone marrow cells were collected at 24 or 48 hours (for the negative control and 2000 mg/kg) after administration of the chemical and the PCEs were examined. As no increase in micronucleated PCEs was found, the chemical was non-mutagenic in this study (SCCP, 2006).

### Carcinogenicity

No data are available.

However, the lack of genotoxicity in vivo and negative bacterial reverse mutation assay results indicate that the likelihood of carcinogenic effects is low.

### Reproductive and Developmental Toxicity

Limited information indicated that the chemical does not show specific reproductive or developmental toxicity.

In a reproductive and developmental toxicity study conducted in accordance with OECD TG 414, three groups of pregnant female SD rats (24/dose) were administered (gavage) 0, 50, 200 or 800 mg/kg bw/day of the chemical on gestation days (GD) 6–15. On day 20 of the pregnancy, all females were euthanised and foetuses were removed by Caesarean section. Maternal toxicity characterised by a reduction in body weight gains and food consumption was noted only in the highest dose group (800 mg/kg bw/day). All treated groups had red coloured urine and red staining of the tail and fur, but no other treatment related toxicity was observed in dams or foetuses.

The NOAEL for embryo/foetotoxicity is 800 mg/kg bw/day and the NOAEL for maternal toxicity is 200 mg/kg bw/day (SCCP, 2006).

## Risk Characterisation

### Critical Health Effects

The critical health effect for risk characterisation includes a local effect (skin sensitisation).

### Public Risk Characterisation

The chemical is reported to be used in semi-permanent hair dye preparations in Australia (NICNAS, 2007).

The EU, New Zealand and ASEAN have restricted the use of the chemical in cosmetics with the following restriction: '(a) the maximum authorised concentration in the finished cosmetic products as a hair dye substances in non-oxidative hair dye products is 0.5 %; and (b) after mixing with hydrogen peroxide under oxidative conditions the maximum concentration applied to hair must not exceed 0.25 %' (see **Regulatory 1: international**). The SCCP also stated that the chemical should also not be used in combination with nitrosating substances and the nitrosamine content should be <50 ppb (SCCP, 2006).

Currently, there are no restrictions in Australia on using this chemical in cosmetics products. In the absence of any regulatory controls, the characterised critical health effects, particularly skin sensitisation, have the potential to pose an unreasonable risk to the public given the identified uses.

### Occupational Risk Characterisation

During product formulation, dermal and ocular exposure may occur, particularly where manual or open processes are used. These could include transfer and blending activities, quality control analysis, and cleaning and maintaining equipment. Worker exposure to the chemical at lower concentrations could also occur while using formulated products containing the chemical. The level and route of exposure will vary depending on the method of application and work practices employed.

Given the critical local health effects, the chemical could pose an unreasonable risk to workers unless adequate control measures to minimise dermal exposure are implemented. The chemical should be appropriately classified and labelled to ensure that a person conducting a business or undertaking (PCBU) at a workplace (such as an employer) has adequate information to determine the appropriate controls.

The data available support an amendment to the hazard classification in the Hazardous Substances Information System (HSIS) (Safe Work Australia) (refer to **Recommendation** section.

# NICNAS Recommendation

Further risk management is required. Sufficient information is available to recommend that risks to public health and safety from potential use of the chemical in cosmetics products (hair dye preparations) be managed through changes to the Poisons Standard, and risks for workplace health and safety be managed through changes to classification and labelling.

Assessment of the chemical is considered to be sufficient provided that risk management recommendations are implemented and all requirements are met under workplace health and safety and poisons legislation as adopted by the relevant state or territory.

## Regulatory Control

### Public Health

Given the risk characterisation, it is recommended that the chemical should be included in Schedule 6 of the Poisons Standard 2015 - *Standard for the Uniform Scheduling of Medicines and Poisons* (SUSMP) with an appropriate concentration cut-off (exemption) for hair dye use.

Consideration should be given to the following:

- the chemical is a strong skin sensitiser;
- overseas restrictions for use of the chemical in hair dyes where the maximum concentration allowed in the finished cosmetic product in hair dye products is 0.5 % and the maximum use concentration upon application is 0.25 % (after mixing under oxidative conditions);
- the concentration may be based on the lowest EC3 value calculated (0.90 %) for skin sensitisation (see **Skin sensitisation**); and
- the risk could be controlled by including warning statements on the label of hair dye formulations containing the chemical at any concentration.

### Work Health and Safety

The chemical is recommended for classification and labelling under the current approved criteria and adopted GHS as below. This assessment does not consider classification of physical and environmental hazards.

| Hazard | Approved Criteria (HSIS)[a] | GHS Classification (HCIS)[b] |
|---|---|---|
| Sensitisation | May cause sensitisation by skin contact (Xi; R43) | May cause an allergic skin reaction - Cat. 1 (H317) |

[a] Approved Criteria for Classifying Hazardous Substances [NOHSC:1008(2004)].

[b] Globally Harmonized System of Classification and Labelling of Chemicals (GHS) United Nations, 2009. Third Edition.

[*] Existing Hazard Classification. No change recommended to this classification

## Advice for consumers

Products containing the chemical should be used according to the instructions on the label.

## Advice for industry

***Control measures***

Control measures to minimise the risk from dermal and ocular exposure to the chemical should be implemented in accordance with the hierarchy of controls. Approaches to minimise risk include substitution, isolation and engineering controls. Measures required to eliminate, or minimise risk arising from storing, handling and using a hazardous chemical depend on the physical form and the manner in which the chemical is used. Examples of control measures which could minimise the risk include, but are not limited to:

- health monitoring for any worker who is at risk of exposure to the chemical, if valid techniques are available to monitor the effect on the worker's health;
- minimising manual processes and work tasks through automating processes;
- work procedures that minimise splashes and spills;
- regularly cleaning equipment and work areas; and
- using protective equipment that is designed, constructed, and operated to ensure that the worker does not come into contact with the chemical.

Guidance on managing risks from hazardous chemicals are provided in the *Managing risks of hazardous chemicals in the workplace—Code of practice* available on the Safe Work Australia website.

Personal protective equipment should not solely be relied upon to control risk and should only be used when all other reasonably practicable control measures do not eliminate or sufficiently minimise risk. Guidance in selecting personal protective equipment can be obtained from Australian, Australian/New Zealand or other approved standards.

***Obligations under workplace health and safety legislation***

Information in this report should be taken into account to help meet obligations under workplace health and safety legislation as adopted by the relevant state or territory. This includes, but is not limited to:

- ensuring that hazardous chemicals are correctly classified and labelled;
- ensuring that (material) safety data sheets ((M)SDS) containing accurate information about the hazards (relating to both health hazards and physicochemical (physical) hazards) of the chemical are prepared; and
- managing risks arising from storing, handling and using a hazardous chemical.

Your work health and safety regulator should be contacted for information on the work health and safety laws in your jurisdiction.

Information on how to prepare an (M)SDS and how to label containers of hazardous chemicals are provided in relevant codes of practice such as the *Preparation of safety data sheets for hazardous chemicals—Code of practice* and *Labelling of workplace hazardous chemicals—Code of practice*, respectively. These codes of practice are available from the Safe Work Australia website.

A review of the physical hazards of the chemical has not been undertaken as part of this assessment.

# References

CosIng. Cosmetic Ingredients and Substances. Accessed April 2015 at http://ec.europa.eu/consumers/cosmetics/cosing/

Galleria Chemica. Accessed April 2015 at http://jr.chemwatch.net/galeria/

NICNAS 2007. LIST OF CHEMICALS USED AS DYES IN PERMANENT AND SEMI-PERMANENT HAIR DYES IN AUSTRALIA. http://www.nicnas.gov.au/Publications/Information_Sheets/Existing_Chemical_Information_Sheets/Hair_Dyes_list_Feb_07_PDF.pdf

Personal Care Products Council (INCI Dictionary). Accessed April 2015 at http://www.ctfa-gov.org/jsp/gov/GovHomePage.jsp

Safe Work Australia (SWA). Hazardous Substances Information system (HSIS). Accessed June 2015 at http://hsis.safeworkaustralia.gov.au/HazardousSubstance

Scientific Committee on Consumer Products (SCCP) 2006. Opinion on HC Violet no 1 (Colipa No B66). Accessed April 2015 at http://ec.europa.eu/health/ph_risk/committees/04_sccp/docs/sccp_o_063.pdf

US Environmental Protection Agency's Aggregated Computational Toxicology Resource (ACToR). Accessed April 2015 at http://actor.epa.gov/actor/faces/ACToRHome.jsp

Last update 03 July 2015

Share this page