

## **New substances: risk assessment summary, new substances notification 21661**

**Official title:** New substances: risk assessment summary, New Substances Notification 21661 – Schedule 1 of the *New Substances Notification Regulations (Organisms)*

**Notified organism:** Anaerobic 1,1,1-Trichloroethane Degrading Consortium KB-1 Plus 1,1,1-TCA (hereinafter referred to as the notified microbial consortium)

**Schedule of the NSNR(O):** Schedule 1 - Information Required in Respect of Micro-organisms

**Organism type:** Microbial consortium

**Use:** Manufacture of the notified microbial consortium for use in bioremediation through the biodegradation of chlorinated ethanes including 1,1,1-trichloroethane (TCA) and 1,1-dichloroethane in groundwater to non-toxic end products

**Anticipated quantity:** An estimated manufacture quantity up to 100 L during the first year and between 200 to 300 L in any 12-month period during the first three years.

### **Assessment level of concern:**

- Human health hazard: Low
- Human exposure: Low
- Environmental hazard: Low
- Environmental exposure: Low

**Assessment conclusion under section 64 of the *Canadian Environmental Protection Act, 1999*:** Low risk, not suspected to be toxic

**Category:** Added to the Domestic Substances List on May 22, 2024

**Recommended action:** None

### **Waivers:**

- A waiver to submit the data from a test conducted to determine the pathogenicity and toxicity of the notified microbial consortium under subparagraphs 5(a)(i) and 5(a)(ii) of Schedule 1 of the *New Substances Notification Regulations (Organisms)* [NSNR(O)], for aquatic and terrestrial plants, invertebrate and vertebrate species, respectively, was granted under paragraph 106(8)(c) of the *Canadian Environmental Protection Act, 1999* (CEPA) because the notified microbial consortium can't survive in the presence of oxygen and requires specific hydrocarbon substrates for growth.
- A waiver to submit the data from tests of antibiotic susceptibility under paragraphs 6(b) and 6(c) of Schedule 1 of the NSNR(O) was granted under paragraph 106(8)(a) of CEPA because the

notified microbial consortium is closely related to environmental isolates that are not known to be toxic or pathogenic to humans. Information was also provided to show that the notified microbial consortium is susceptible to several antimicrobials.

## Synopsis

The notified organism is a strictly anaerobic microbial consortium, consisting of a group of micro-organisms that cannot survive in the presence of oxygen. It was notified to the New Substances program for use in the biodegradation of certain pollutants (i.e., chlorinated ethanes, including 1,1,1-TCA) in groundwater to non-toxic end products. Other potential uses could include as a commercial product for application in anaerobic digesters, composting, treating groundwater or wastewater, alone or in combination with other consortia in Canada.

There is no evidence to suggest a potential risk of adverse environmental and indirect human health effects at the exposure levels predicted to the environment and general population in Canada from the manufacture and use of the notified microbial consortium.

It is determined that the notified strain is not toxic or capable of becoming toxic according to the criteria under section 64 of the Act as there is no evidence to suggest that the notified organism may enter the environment in a quantity or concentration or under conditions that:

- have or may have an immediate or long-term harmful effect on the environment or its biological diversity,
- constitute or may constitute a danger to the environment on which life depends, or
- constitute or may constitute a danger in Canada to human life or health.

No risk management is recommended.

## Background information

The notified consortium was isolated from soil and groundwater samples in 2001 from a site in the United States, that was contaminated with chlorinated solvents including 1,1,1-TCA. The consortium was selectively enriched by culturing them in supplementation with 1,1,1-TCA and maintaining under anaerobic (oxygen-free) conditions. The notified microbial consortium is intended for use in bioremediation of chlorinated ethanes, such as 1,1,1-TCA, for the treatment of contaminated groundwater sites. Other potential uses could include its use as a commercial product for anaerobic digesters, composting, to treat groundwater or wastewater, or for soil bioremediation, alone or in combination with other consortia in Canada.

## Hazard

The environmental and human health hazard potential of the notified microbial consortium is determined to be low because:

- 1) The notified consortium was isolated from the environment and has not been genetically modified.
- 2) The microbial constituents in the consortium were identified using relevant methods such as Next Generation Sequencing and targeted Quantitative PCR. Based on the Amplicon Sequence Variant (ASVs) DNA analysis, major, minor, rare, or trace constituents of the notified microbial consortium were identified at various taxonomic levels. These methods also effectively distinguished the ASVs of major and minor constituents of the consortium from organisms known to cause adverse effects to non-human species, the environment and human health.
- 3) Phylogenetic analyses conducted by the notifier showed that none of the major, minor, rare, or trace constituents of the notified microbial consortium are closely related to known animal or plant pathogens.
- 4) The notified consortium is composed of micro-organisms that are closely related to those found in a variety of environments, including rice paddies, contaminated aquifers, freshwater and marine sediments, wastewater treatment facilities, soils and animal gastrointestinal tracts. Results from the review of published literature of the major constituents identified in the consortium showed that the micro-organisms belonging to these groups are likely widespread throughout Canada in specific ecozones.
- 5) The New Substances program's in-house review of scientific literature did not reveal any studies suggesting that the major constituents identified in the consortium were pathogenic or toxic to aquatic or terrestrial plants, invertebrates, and vertebrates or humans. There are no reported cases of disease outbreaks to non-human species associated with these major constituents.
- 6) The notified consortium requires both anaerobic conditions and specific substrates for growth. The combinations of these properties render it unlikely to be able to cause infections. The microbial community composition and stability of the notified microbial consortium have been maintained and monitored for more than twenty years without any reports of adverse human health effects. Additionally, the notifier conducts routine testing for the presence of human or animal pathogens in the notified microbial consortium cultures to ensure that organisms of concern were not present.
- 7) Whereas there may be potential for some minor constituents of the consortium to be similar to known human pathogens, the notified microbial consortium is susceptible to clinically relevant antimicrobial drugs. Therefore, in an unlikely event of an infection, effective treatments are available.
- 8) In-house literature search shows that methanogenic archaea, which are micro-organisms known for their capability to produce methane gas, can trigger an immune response in both animals and humans. However, there have been no cases of allergic reaction to the major constituents of the notified consortium reported in the scientific literature.

## Exposure

The environmental and indirect human exposure potential of the notified microbial consortium is determined to be low because:

- 1) An estimated annual maximum of 200 to 300 L of the notified microbial consortium will be manufactured at a Canadian facility during the course of the first three years.
- 2) The manufacturing facility meets recommendations for a Containment Level (CL) 1 facility as defined by the Public Health Agency of Canada's [\*Canadian Biosafety Standard, Third Edition\*](#). All manufacturing personnel and field technicians will have adequate training and standard operating procedures are in place to deal with any accidental spills or loss of culture. Therefore, the notified microbial consortium is not expected to be released from the manufacturing facility to the environment and exposure of the general population is not expected.
- 3) The notified microbial consortium will be transported to contaminated sites to be bioremediated by direct injection into the soil subsurface where no receptor species are anticipated to be present other than micro-organisms themselves. Standard operational procedures are in place to limit environmental spills into surface waters and untreated groundwater used for irrigation.
- 4) Most of the constituents of the notified consortium cannot survive in the presence of oxygen. They are dependent on the growth and activity of each other and require a special source of carbon for survival. Although some constituents may persist in low populations, they are not expected to survive and proliferate upon depletion of 1,1,1-TCA or when exposed to higher concentrations of oxygen. These properties combined with their limited mobility in groundwater and the slow rate of subsurface flow, the consortium is not expected to reach major water sources or oxygen-rich, environmentally sensitive areas. In addition, the potential for exposure of the general human population is expected to be low.

## Risk characterization

Owing to the low potential hazard and the low potential exposure, the environmental and human health risks associated with the manufacture and use of the notified microbial consortium for biodegradation of chlorinated ethanes, including 1,1,1-TCA, in groundwater to non-toxic end products and for other potential uses such as a commercial product to treat groundwater or wastewater or soil bioremediation in Canada, are both low.

## Risk assessment conclusion

There is no evidence to suggest a potential risk of adverse environmental and human health effects at the exposure levels predicted to the environment and general population from the notified and potential uses of the notified microbial consortium. The risk to the environment and human health associated

with the notified microbial consortium is not suspected to meet the criteria in paragraphs 64(a), (b), or (c) of CEPA. No further action is recommended.