

## CASE STUDY: MTBE AND BTEX TREATABILITY REPORT

### INTRODUCTION

Nordevco Associated Ltd. (Nordevco) conducted treatability testing during the period of May to September 2004 under controlled laboratory conditions to establish if Nordevco's BactiDomus<sup>®</sup> Technology would thoroughly and efficiently biodegrade methyl tertiary butyl ether (MTBE). The treatability testing conducted at an independent accredited laboratory, has demonstrated that Nordevco's Bio4Water<sup>™</sup>: MTBE microbiological module: (1) biodegrades MTBE and BTEX, (2) biodegrades MTBE thoroughly (93.00% to 99.99% biodegradation); (3) biodegrades BTEX thoroughly (99.95% to 99.99%), (4) biodegrades MTBE efficiently (3 to 8 hours for more than 93.00% and up to 99.99% biodegradation); (5) biodegrades BTEX efficiently (4 to 8 hours for more than 99.95% up to 99.99% biodegradation), and (6) creates water and carbon dioxide as the final metabolites in the biodegradation process (7). Nordevco has established the likely pathway by which this MTBE biodegradation process occurs.

It was also established that some form of aeration is a pre-requisite for successful biodegradation, and that under oxygen-lean conditions; biodegradation occurs only to the extent that oxygen is available.

### PROBLEM DEFINITION

The use of MTBE started in 1979 when it was initially used to increase the octane level in gasoline. More recently, MTBE was used as a fuel additive to meet fuel oxygen requirement levels mandated by the United States Environmental Protection Agency (EPA) and similar regulatory bodies in other countries.

It is now established that MTBE is a chronic persistent pollutant in soil and groundwater. The use of MTBE has, therefore, been discontinued.

MTBE is blamed for water contamination in more than two dozen U.S. states. Often the problem arises from leaking underground storage tanks that contained MTBE-blended gasoline. In many cases, MTBE will be found in the presence of conventional gasoline with BTEX (benzene, ethylbenzene, toluene and mixed xylenes) soil and groundwater contaminants. MTBE and BTEX are regarded as serious soil and groundwater contaminant requiring thorough and efficient remediation to preserve and protect public drinking water supplies and sensitive receptors.

### THE TECHNOLOGY

Nordevco's BactiDomus<sup>®</sup> Technology was developed by a diversified group of research scientists working together at Universities in Belgium and France. Their goal was to create a mechanism with the flexibility to deliver biological solutions to a range of environmental issues more effectively and efficiently. The foundation for the success of the BactiDomus<sup>®</sup> Technology was the development team's clear understanding that for any carrier material to be successful it had to meet specific underlying needs of the organisms:

- Regardless of the organisms used, they would be cultured in a sterile laboratory and would require time to acclimate to the environment they were activated in.



- Microorganisms, like humans, do not exist or thrive in isolation of each other but rather rely on others for stimulation and competition;
- Organisms prefer to grow and live in colonies or flocs and prefer to attach to something to anchor these colonies;
- Individual species of microorganisms do not work in isolation to break down organic compounds. To successfully break down any organic completely to CO<sub>2</sub> and H<sub>2</sub>O, a variety of different organisms are required;

The result of that work is the BactiDomus® Technology which is based on the use of an inorganic limestone-like porous carrier material. The porosity of the material allows it to be bathed in a nutrient broth, absorbing key micro-nutrients that act as an initial food source when the product is activated. It is then impregnated with a range of different naturally occurring and non-pathogenic organisms, selected for their ability to breakdown specific organic contaminants.

The organisms selected for inclusion are selected based on the understanding that each contaminated environment can be aerobic, anaerobic or facultative anaerobic. Therefore, aerobic, anoxic and anaerobic organisms are selected and used in each product to ensure that they can function successfully in a broad range of environments.

The carrier material's large surface area to size ratio provides the organisms with both internal and external floc points where they grow and create large effective colonies of biodegraders working together to break down the organic contaminant into carbon dioxide and water. The carrier material's hydrophilic nature allows it to absorb both the water and contamination. This provides a steady strong contact between the imbedded organisms and organic contaminant. This ensures that the organisms have a continuous food source as they grow and create flocs within the protective confines of the capillary network of the carrier material.

#### **TREATABILITY TESTS**

The treatability studies were performed at an independent, accredited laboratory (Enviro-Test Laboratories (Winnipeg, Manitoba, Canada) was completely in charge of the testing from beginning to end in order to insure the complete integrity of the tests) Nordevco conducted four series of bench treatability tests to establish that its Bio4Water™: MTBE microbiological module: (1) biodegrades MTBE and BTEX, (2) biodegrades MTBE and BTEX thoroughly; (3) biodegrades MTBE and BTEX efficiently; and (4) and to determine the final metabolites in the MTBE biodegradation process. Furthermore, the test objective was to establish the likely MTBE biodegradation pathway.

The testing was conducted in 1 liter Erlenmeyer flasks. The stopper of the flasks contained two openings: one for an air supply line, the other was used to locate a charcoal filter to quantify MTBE losses attributable to volatilization.

Each test series included a control without the addition of BactiDomus® Technology product. The test flasks were run for varying periods of time.

**Test Series 1:** **MTBE** Initial concentration: 10,000 ppm.  
Treatments were run for 2 and 6 hours.

**Test Series 2:** **MTBE** Initial concentration: 3,400 ppm.  
Treatments were run for 1, 3 and 8 hours.

**Test Series 3:** **MTBE** Initial concentration: 1,600 ppm.  
Treatment was run for 8 hours.

For this series no aeration was provided. Hence the only oxygen supply available was the dissolved oxygen in the water and the air above the water in the closed flask. For this series, the tests were run in duplicate plus a control.

**Test Series 4:** **MTBE** Initial concentration: 90 ppm.  
**BTEX** Initial concentration: 36 ppm.  
Treatment was run for 4 and 8 hours

This test did not include controls and carbon filters. During the test, the flasks were continuously stirred and aerated.

#### **RESULTS OF THE TREATABILITY TESTS**

Testing established that Nordevco's BactiDomus<sup>®</sup> Technology product: (1) biodegrades MTBE and BTEX, (2) biodegrades MTBE thoroughly (93.00% to 99.71% biodegradation); (3) biodegrades BTEX thoroughly (99.95% to 99.99%), (4) biodegrades MTBE efficiently (3 to 8 hours for more than 93.00% and up to 99.71% biodegradation); (5) biodegrades BTEX efficiently (4 to 8 hours for more than 99.95% up to 99.99% biodegradation), and (6) creates water and carbon dioxide as the final metabolites in this biodegradation process.

The Series 1 tests started with a MTBE concentration of 10,000 ppm. After 2 hours of treatment, 2,000 ppm of MTBE remained in the test flask, indicating that 80.00% of the MTBE was biodegraded. After 6 hours of treatment, 700 ppm of MTBE remained in the test flask, further indicating that MTBE was 93.00% biodegraded.

The Series 2 tests started with a MTBE concentration of 3,500 ppm. After 1 hour of treatment, 1,300 ppm of MTBE remained in the tested flask, indicating that 61.76% of the MTBE was biodegraded. After 3 hours of treatment, 210 ppm of MTBE remained in the test flask, indicating that MTBE was 93.82% biodegraded. After 8 hours of treatment, 10 ppm of MTBE remained in the test flask, further indicating that MBTE was 99.71% biodegraded.

The results of the tests conducted are summarized in Table 1 and the attached charts.

The MTBE control concentration did not change over the course of the treatment periods. In other words, there was no MTBE detectable natural degradation during the test periods.

**TABLE 1: DEGRADATION OF MTBE**

Treatment Time	Control	Series 1 Tests		Series 2 Tests	
		MTBE (ppm)	Degradation (%)	MTBE (ppm)	Degradation (%)
Initial Values	10,000	10,000	0.00	3,400	0.00
1 hour				1,300	61.76
2 hours		2,000	80.00		
3 hours				210	93.82
6 hours	10,000	700	93.00		
8 hours				10	99.71

Charcoal filters were put on the Erlenmeyer flasks for the Series 2 Test to quantify possible MTBE losses attributable to the aeration process.<sup>2</sup> MTBE was not detected at the back of the filters for the 1, 3, and 6 hours test periods.. At the front of the filters (flask side), insignificant

Note: Series one did not have a charcoal filter

MTBE values were detected (see Table 2). This demonstrates that the MTBE biodegradation occurred from treating MTBE with Nordevcos microbiological Bio4Water™: MTBE module., Furthermore, there was negligible impact from aerating the flasks.

**TABLE 2: LOSS OF MTBE DUE TO AERATION (SERIES 2 TEST)**

Treatment Time	MTBE (ppb)	MTBE (ppm)
1 hour	89	0.089
3 hours	210	0.210
6 hours	83	0.083

The results of the series 3 test show limited biodegradation: the 18.8 percent biodegradation is far below the 99 percent plus removal achieved when aeration is provided. The laboratory analysis reported no intermediate metabolites and the structure of the MBTE remaining intact. This raises the possibility that the limited degradation observed may be attributable to the laboratory analysis.

**TABLE 3: NO AERATION (SERIES 3 TEST)**

	<b>MTBE ppm after 8 hours</b>	<b>MTBE % degradation</b>
<b>TEST 1</b>	1400	12.5
<b>TEST 2</b>	1300	18.8
<b>CONTROL</b>	1600	0

**MTBE** Initial concentration: 90 ppm. **BTEX** Initial concentration: 36 ppm. Treatments were run for 4 and 8 hours.

The series 4 test started with a MTBE and BTEX concentration of 90 ppm and 36 ppm respectively. After treating MTBE and BTEX 4 hours, 1.7 ppm and 0.019 ppm respectively remained in the flask, indicating that 98.11% and 99.85% of the MTBE and BTEX was biodegraded. After treating MTBE and BTEX 8 hours, 0.0005 ppm and 0.0036 ppm respectively remained in the flask, indicating that 99.99% and 99.99% of the MTBE and BTEX was biodegraded.

The results of testing conducted are summarized in the following Table

**TABLE 4: DEGRADATION OF MTBE AND BTEX**

<b>Treatment Time</b>	<b>MTBE</b>		<b>BTEX</b>	
	<b>MTBE (ppm)</b>	<b>Degradation (%)</b>	<b>BTEX (ppm)</b>	<b>Degradation (%)</b>
Initial Conc.	90.00	0.00	36.00	0.00
4 hours	1.70	98.11	0.019	99.95
8 hours	0.0005	99.99	0.0036	99.99

#### **FINAL METABOLITES**

Water and carbon dioxide are the final metabolites of the MTBE biodegradation process arising from the application of Nordevcos microbiological Bio4Water™: MTBE module. No other final metabolites were identified. This is consistent with expectations that water and carbon dioxide are typical metabolites of any aerobic biodegradation.

#### **LIKELY PATHWAY**

The MTBE biodegradation process arising from the application of Nordevco s microbiological Bio4Water™: MTBE module likely occurs with the pathway described below. This pathway was determined with the aid of the laboratory GC-MS scanning results.

MTBE (C<sub>5</sub>H<sub>12</sub>O)

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2-Methyl-2-Propanol (C<sub>4</sub>H<sub>10</sub>O) {O contributed by aeration; loss of a CO<sub>2</sub>}

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Isobutylene / 2-methyl-1-Propene (C<sub>4</sub>H<sub>8</sub>): loss of 2[H<sub>2</sub>O]

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4[H<sub>2</sub>O] + 4[CO<sub>2</sub>] {O contributed by aeration}

### CONCLUSIONS

Nordevco has established that its Bio4Water™: MTBE microbiological module: (1) biodegrades MTBE and BTEX, (2) biodegrades MTBE thoroughly (93.00% to 99.99% biodegradation); (3) biodegrades BTEX thoroughly (99.95% to 99.99%), (4) biodegrades MTBE efficiently (3 to 8 hours for more than 93.00% and up to 99.99% biodegradation); (5) biodegrades BTEX efficiently (4 to 8 hours for more than 99.95% up to 99.99% biodegradation), and (6) creates water and carbon dioxide as the final metabolites in this biodegradation process (7). Nordevco has established the likely pathway by which this MTBE biodegradation process occurs.

The treatability results have demonstrated that treatment with Nordevco's Bio4Water™: MTBE microbiological module will be effective in the remediation of MTBE and BTEX contaminated groundwater and soil.

PHONE: 1-204-261-1801

FAX: 1-204-269-9097

EMAIL; [INFO@NORDEVCO.NET](mailto:INFO@NORDEVCO.NET)

WWW.NORDEVCO.NET