QuantArray®

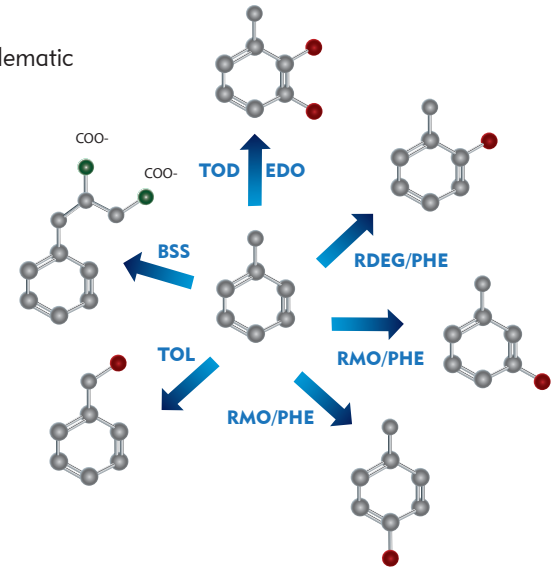
MOLECULAR BIOLOGICAL TOOL

Simultaneously quantify functional genes responsible for aerobic and anaerobic biodegradation of petroleum hydrocarbons in a single analysis

Comprehensive evaluation of biodegradation potential at petroleum impacted sites is inherently problematic due to two factors: (1) Petroleum products are complex mixtures of hundreds of aliphatic, aromatic, cyclic and heterocyclic compounds (2) Even for common classes of contaminants like benzene, toluene, ethylbenzene, and xylenes (BTEX), biodegradation can proceed by a multitude of pathways. For example, biodegradation of toluene can proceed via five known aerobic pathways and one known anaerobic pathway as shown.

The Petroleum QuantArray has been designed to address both of these issues by providing the simultaneous quantification of the specific functional genes responsible for both aerobic and anaerobic biodegradation of BTEX, PAHs, and a variety of short and long chain alkanes.

Thus, when combined with chemical and geochemical groundwater monitoring programs, the QuantArray allows site managers to simultaneously yet economically evaluate the potential for biodegradation of a spectrum of petroleum hydrocarbons through a multitude of aerobic and anaerobic pathways to give a much more clear and comprehensive view of contaminant biodegradation.



BTEX and MTBE

- Benzene/toluene dioxygenases (TOD) monooxygenases (RMO, RDEG, PHE) and other functional genes responsible for aerobic biodegradtion of BTEX
- Includes MTBE utilizing strain *Methylibium petroleiphilum* PM1 and TBA monooxygenase
- Benzylsuccinate synthase (BSS) for anaerobic biodegradation of toluene, ethylbenzene, and xylenes
- Benzene carboxylase (ABC) initiates the only known pathway for anaerobic benzene biodegradation

Naphthalene and PAHs

- Includes three groups of naphthalene dioxygenase genes (NAH, NAG, PHN) for aerobic biodegradation
- Naphthylmethylsuccinate synthase (NMS) for anaerobic biodegradation of methyl-naphthalenes
- Naphthalene carboxylase (ANC) initiates the only known pathway for anaerobic naphthalene biodegradation

Alkanes/TPH

- The *n*-alkanes are a substantial portion of petroleum products
- The Petroleum QuantArray includes quantification of alkane monooxygenase genes (alkB)
- Also includes quantification of alkylsuccinate synthase (assA) genes to evaluate anaerobic biodegradation of alkanes

Quantification of a multitude of key functional genes responsible for aerobic and anaerobic biodegradation of petroleum hydrocarbons.

Aerobic Biodegradation

- Benzene/toluene dioxygenase (TOD)
- Toluene/benzene monoxygenases (RMO, RDEG)
- Phenol hydroxylase (PHE)
- Ethylbenzene and isopropylbenzene dioxygenases (EDO, BPH4)
- Naphthalene dioxygenases (NAH, NAG, PHN)
- MTBE-utilizing strain PM1
- TBA monooxygenase
- Alkane monooxygenases

Anaerobic Biodegradation

- Benzylsuccinate synthase (BSS)
- Benzene carboxylase (ABC)
- Naphthalene carboxylase (ANC)
- Naphthylmethylsuccinate synthase (NMS)
- Alkylsuccinate synthase
- Benzoyl Coenzyme A reductase (BCR)

Other Groups

- Total Bacteria (EBAC)
- Sulfate reducing bacteria (APS)

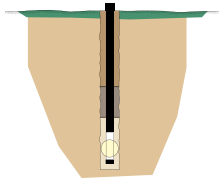
How does it work?

The QuantArray is a hybrid technology combining the highly parallel detection of DNA microarrays with the accurate and precise quantification of qPCR into a single platform. The key to the approach is nanoliter fluidics for low volume, solution phase qPCR allowing simultaneous quantification of different gene targets and therefore more comprehensive site assessment.

In many other respects, the QuantArray is the same as conventional qPCR with TaqMan® probes so you can expect the same level of accuracy and precision. qPCR is a process whereby many copies of a specific gene are generated. The gene copied during the process (target gene) is determined by short segments of DNA called "primers" and a TaqMan® "probe". As each gene copy is made, a fluorescent marker is released from the TaqMan® probe, measured, and used to quantify the number of target genes present in the sample.

Other methods like multiplex qPCR have been described that achieve some level of parallel quantification. There is a fundamental difference between the QuantArray and multiplex qPCR however. For multiplex qPCR, multiple primer sets are added to a reaction mixture to quantify multiple gene targets. Unlike multiplex qPCR, the QuantArray employs discrete through-holes for individual qPCR reactions ensuring that reaction kinetics are not compromised.

Sample Collection

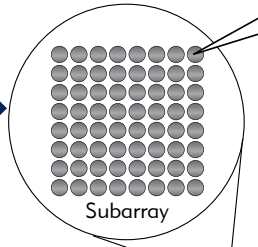


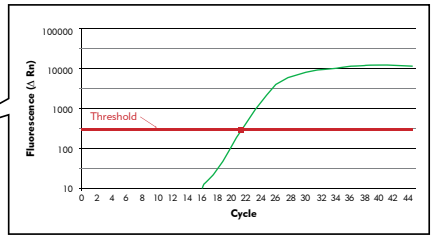
Groundwater, soil, or Bio-Trap® Sampler collected and shipped overnight on ice (4°C)

DNA Extraction

DNA is extracted from samples upon arrival

Amplification

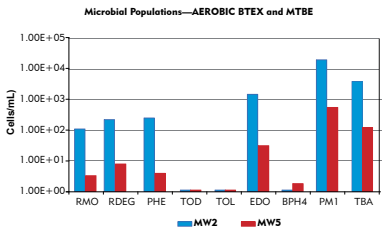




Quantification of a multitude of gene targets

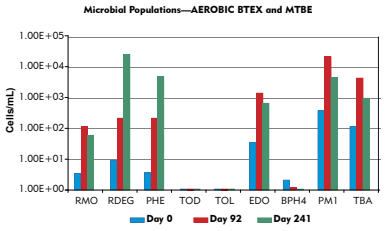
Results

Sample Information	MW2	MW5
Aerobic BTEX and MTBE		
Toluene Ring Hydroxylating Monooxygenases (RMO)	1.50E+02	4.17E+00
Toluene 2 Monooxygenase/Phenol Hydroxylase (RDEG)	2.80E+02	1.00E+01
Phenol Hydroxylase (PHE)	3.20E+02	4.57E+00
Toluene/Benzene Dioxygenase (TOD)	<1.00E+00	<1.00E+00
Xylene/Toluene Monooxygenase (TOL)	<1.00E+00	<1.00E+00
Ethylbenzene/Isopropylbenzene Dioxygenase (EDO)	2.10E+03	4.47E+01
Biphenyl/Isopropylbenzene Dioxygenase (BPH4)	1.20E+00	1.94E+00
Methylibium petroliphilum PM1 (PM1)	2.87E+04	5.22E+02
TBA Monooxygenase (TBA)	5.21E+03	1.63E+02



Quantification of a broad spectrum of different microorganisms and key functional genes responsible for various biodegradation pathways critical for site remediation.

Assessment



QuantArray results are integrated with other site parameters to optimize site management

microbialinsights
www.microbe.com

10515 Research Drive
Knoxville, TN 37932
Phone: 865.573.8188