



# Massapequa Water District

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**RE: Northrop Grumman and Naval Weapons Industrial Reserve Plant (NWIRP)  
Sites**

Dear Ms. Fly and Mr. Hannon:

The Massapequa Water District (MWD) is seeking permission to sample select monitoring wells to perform a Compound Specific Isotope Analysis (CSIA) to test for a correlation between the TCE in the monitoring wells and the TCE emanating from the source plume. MWD will pay for all sampling and analysis costs and will share the data with the Navy, Grumman and all parties listed below. The monitoring well locations requested for sampling include Onsite Well 1, RE 103 D3, TT101 D2, BPOW 3-4, BPOW 5-6 and BPOW 5-4.

Attached please find references for the USEPA methodology for sample collection and analysis related to the CSIA scope. MWD plans on analyzing the samples using CSIA technology for both TCE and 1,4 Dioxane.

Thank you in advance for your consideration of this request. Please feel free to contact the undersigned if you should have any questions or require any additional information.

Very truly yours,  
**MASSAPEQUA WATER DISTRICT**

Stan Carey  
Superintendent

*Committed to deliver and preserve our water supply for the welfare, health, and safety  
of the inhabitants of the Massapequa Water District*

cc:

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# A Guide for Assessing Biodegradation and Source Identification of Organic Ground Water Contaminants using Compound Specific Isotope Analysis (CSIA)



## 9.0

# Recommendations for the Application of CSIA

Compound Specific Isotope Analysis (CSIA) provides another dimension of information on pollutants in the environment to supplement knowledge of their chemical identity and their concentration. CSIA has matured into a technique that can be used on a routine basis, in particular for carbon isotope analysis, for a wide range of organic contaminants relevant to hydrogeology and environmental geochemistry. Modern instrumentation can provide valid determinations of isotope ratios at low concentrations of contaminants that are near their regulatory standards or clean-up goals.

Application of CSIA at a contaminated site should start with a clear idea of the information that is sought from stable isotope analysis. Basically, there are three distinct goals: (i) source characterization or differentiation; (ii) qualitative proof of biodegradation or abiotic transformation; or, (iii) quantification of biodegradation or abiotic transformation processes. Advantages and limitations of the use of CSIA for these purposes have been discussed in detail throughout this guideline. If the specific interest is *quantification* of degradation, the first step is to consult the literature (summarized in Table 8.1) to determine whether an appropriate isotopic enrichment factor is available. Using the enrichment factor, the second step is to estimate whether the observed changes in concentration of the contaminant at the field site are sufficient to produce a measurable change in the isotope ratio. If these two prerequisites are met there is a good chance that it will be possible to put a conservative boundary on the extent of biodegradation or abiotic transformation at the field site.

On a per sample basis, the cost of an individual isotope analysis is substantially higher than the cost of a VOC analysis to identify the chemical, and determine its concentration. This is due to the price of the equipment required to perform CSIA, the costs of consumables, the level of training and experience required for the analytical chemist, and the number of standards and sample duplicates that are needed to ensure reliable data as discussed in section 2.4. However, a simple comparison of direct costs for analyses is not very meaningful. Instead, consider the total cost of a site investigation. If the

additional information from CSIA leads to a robust conceptual model for a site, it can lead to savings in conventional monitoring. If there is greater faith in the site conceptual model, adequate monitoring can be attained with fewer rounds of sampling and analysis, and fewer monitoring wells.

Furthermore, CSIA can guide decisions on selection and implementation of remediation strategies, and can be used to monitor the performance of remedial technology in an early stage of implementation. Thus, the huge waste in resources that are associated with the selection of an unsuitable remediation strategy might be avoided.

Compound specific isotope analysis can be applied on a routine basis when precautions are taken to ensure high quality data and the appropriate interpretation of the data. However, CSIA cannot replace a proper hydrological and geochemical characterization or measurements of contaminant concentrations. Multiple lines of evidence will continue to be necessary to come to a meaningful assessment of the risks associated with the contaminants and the selection of an appropriate remedy.

It is our hope that this Guide will be a useful introduction for beginners in environmental isotope analysis. We expect that CSIA will have a growing role in investigations at hazardous waste sites. The growth in the application of CSIA is driven by continued improvements in analytical methods, by more widespread availability of the instruments used in CSIA, by an increasing number of publications showing the broad applicability of CSIA to a variety of contaminants, and by an increasing appreciation for the unique information provided by CSIA. We have only “scratched the surface” of the potential of CSIA to provide a better understanding of the source, distribution, and behavior of organic compounds at contaminated field sites.



# Compound Specific Isotope Analysis

## EMD Team Fact Sheet—November 2011

This fact sheet, developed by the ITRC Environmental Molecular Diagnostics (EMD) Team, is one of 10 designed to provide introductory information about and promote awareness of EMDs. Please review the Introduction to EMDs Fact Sheet along with this one. A glossary is included at the end of this fact sheet.

### Why is compound specific isotope analysis relevant?

Compound specific isotope analysis (CSIA) is an analytical method that measures the ratios of naturally occurring stable isotopes in environmental samples. As described in this fact sheet, CSIA can be used to gain information (e.g., potential contaminant sources, extent of degradation, comingling of contaminant plumes) relevant to environmental remediation decision makers. Stable isotope probing (SIP), which is a separate and distinct EMD method and is discussed in its own fact sheet, uses isotopically enriched (labeled) contaminants and examines the incorporation of stable isotopes into biomolecules and by-products that are generated during biochemical processes associated with contaminant biodegradation.

### What does CSIA do?

CSIA is a laboratory method in which samples collected from the field are analyzed to give information that can be valuable for assessing environmental forensics or contaminant fate. Each element in a compound has a distinct isotopic ratio. For a given element the isotopic ratio is known to within a few percent; however, that ratio can change in systematic ways during the course of biodegradation or other processes. CSIA measures these small changes in isotopic ratios very precisely. Those changes can be exploited to gain important information about the source, transport, and fate of a compound. Table 1 includes information about environmentally relevant elements to which CSIA is often applied.

Since the isotopic ratio in the compound is a function of the starting material and the manufacturing process as well as the degradation of that compound after it was made, CSIA has applications in environmental forensics, biodegradation, and abiotic degradation. Examples include the following:

**Table 1. Environmentally relevant ratios for CSIA**

Atom	Ratio
Hydrogen	$^2\text{H}/^1\text{H}$ (D/H)
Carbon	$^{13}\text{C}/^{12}\text{C}$
Nitrogen	$^{15}\text{N}/^{14}\text{N}$
Oxygen	$^{18}\text{O}/^{16}\text{O}$ and $^{17}\text{O}/^{16}\text{O}$
Chlorine	$^{37}\text{Cl}/^{35}\text{Cl}$

- **Biodegradation**
  - Apparent *cis*-dichloroethene (*cis*-DCE) stall: Is the *cis*-DCE biodegrading, or are concentration changes the result of dilution?
  - Dense, nonaqueous-phase liquids: Is there biodegradation?
  - Biodegradation of methyl *tert*-butyl ether (MTBE): Is it occurring?
  - Aerobic *cis*-DCE and vinyl chloride (VC): How do we prove progress without daughter or end products?
  - Cometabolic degradation of chlorinated ethenes: Will the accumulation of VC really be skipped?
- **Environmental Forensics**—Especially when combined with other tests, CSIA can reveal detailed information not just about remedial progress or remedial potential but also about forensic issues such as the potential for multiple sources.
  - Methane: From shallow biodegradation or pipeline gas?
  - Perchlorate: Is it natural or synthetic?
  - Volatile organic compounds (VOCs): Origin from one source or multiple sources?
  - Nitrate: Is it runoff or naturally occurring?
- **Abiotic Degradation**
  - Is biogeochemical transformation occurring?
  - In situ chemical oxidation (ISCO): Was the ISCO successful in destroying contaminant mass and the “rebound” is really just newly desorbed product?
  - In situ chemical reduction, iron wall, nanoscale iron, or other reducing mixtures: Has the contaminant been destroyed or displaced?

## How are the data used?

CSIA can be used to make informed decisions for site characterization, monitoring, remedy selection, and closure. CSIA can differentiate between degradation of a compound (e.g., ISCO) and other processes that can also reduce contaminant concentration but do not reduce contaminant mass (e.g., displacement or dilution). CSIA has also been used to distinguish contaminant sources (e.g., trichloroethene [TCE] from different plumes).

### Example Environmental Remediation Questions CSIA Can Help Answer

- **Site Characterization**
  - Has biological or abiotic degradation occurred? If so, how much and where?
  - Is methane from near-surface biodegradation or natural gas production?
  - Is the TCE a parent from one source or a daughter product of perchloroethene (PCE) from another source?
  - Is there evidence of multiple sources?
  - Is the contaminant in the dissolved or nonaqueous phase?
  - Is there evidence of a rate-limiting step (i.e., accumulation of contaminant intermediates)?
- **Remediation**
  - Is monitored natural attenuation (MNA) feasible?
- **Monitoring**
  - Is remediation progressing as planned?

CSIA has been in use for more than two decades since the merger of two 50-year-old techniques that were each well established: gas chromatography and isotopic analysis. Isotopic analysis has been used extensively in geology and petroleum exploration. Due to the extremely dilute concentrations typically encountered in environmental samples (as compared to those from traditional geology and petroleum exploration applications), the early methods were not suitable for environmental work. However, within the last 10–12 years advances in analytical instrumentation have resulted in the ability to apply CSIA to very dilute samples, and the technique has been broadly applied to evaluate environmental questions.

## How does it work?

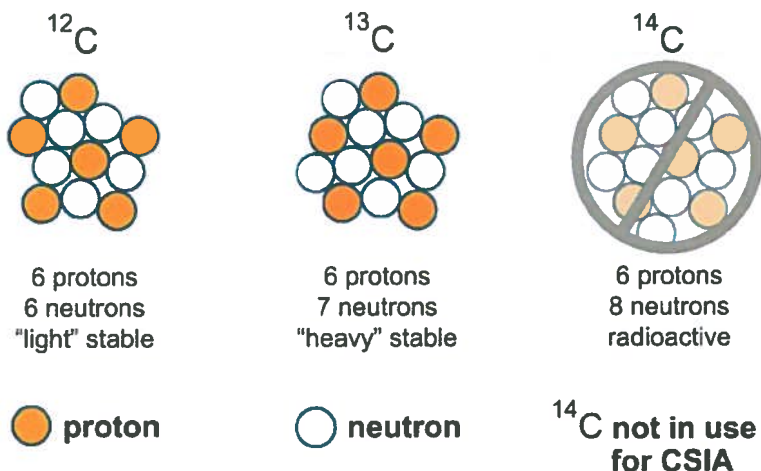
Changes in isotopic ratios are caused by the breaking of bonds between atoms. Physical processes such as dilution, diffusion, and volatilization do not change the isotopic ratios in compounds to the same extent as chemical or biochemical processes such as degradation. For VOCs in groundwater, this means that degradation of a compound is, by far, the major cause of significant changes in isotopic ratios. This change in isotopic ratio happens in both biological and abiotic reactions, and CSIA is used to measure those changes. The CSIA laboratory method is implemented using a number of instruments, including a gas chromatograph (GC) and an isotope ratio mass spectrometer (IRMS).

Because compounds are made up of multiple elements, CSIA can be performed on multiple isotopes to gain further insight into origin or mechanisms of degradation. For example, both carbon ( $^{13}\text{C}/^{12}\text{C}$ ) and hydrogen ( $^2\text{H}/^1\text{H}$ ) are often used for MTBE, whereas chlorine ( $^{37}\text{Cl}/^{35}\text{Cl}$ ) as well as two isotopic ratios of oxygen ( $^{18}\text{O}/^{16}\text{O}$  and  $^{17}\text{O}/^{16}\text{O}$ ) are often used for perchlorate. Table 1 lists environmentally relevant ratios on which CSIA can be used.

Every contaminant is made of atoms of various elements. The isotopes of a given element (e.g., carbon, hydrogen, chlorine) have the same number of protons and electrons but a different number of neutrons and thus different atomic mass. Each element has a most-abundant isotope (for example,  $^{12}\text{C}$ , or "carbon-twelve," for carbon) and one or more less-abundant isotopes ( $^{13}\text{C}$ , or "carbon-thirteen," for carbon) (Figure 1). The less-abundant isotopes are sometimes heavier (i.e., contain one or more extra neutrons). Some of these heavier, less-abundant isotopes are nonradioactive (i.e., stable), and the only significant difference between these isotopes and their more-abundant counterpart is the increased mass. The

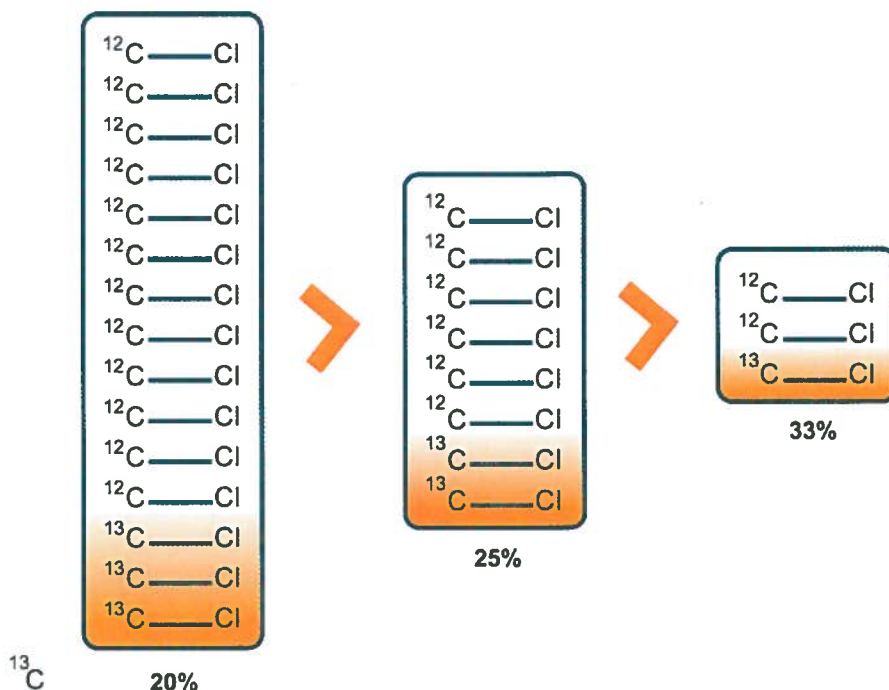
increased mass leads to the process called “isotopic fractionation.” Isotopic fractionation is monitored by measuring the isotopic ratio, i.e., by CSIA.

### Isotopes of Carbon



**Figure 1. Schematic of the atomic nuclei of the common carbon isotopes.**  
Carbon-fourteen is not used in CSIA. *Source: Microseeps, Inc., 2010, used with permission.*

Isotopic fractionation occurs because it takes slightly less energy to break a bond between a light isotope and another atom than it takes to break a bond between a heavy isotope and that same atom. As a result, the rates of reaction involving the heavier isotopes are slightly slower, so the percentage of heavy isotopes increases in the residual contaminant pool as the contaminant is degraded. Figure 2 shows this “isotopic fractionation” or enrichment for a carbon-chlorine (C-Cl) bond.



**Figure 2. Illustration of  $^{13}\text{C}$  enrichment during degradation of a contaminant with a C-Cl bond.**  
*Source: Microseeps, Inc., 2010, used with permission.*

## How are the data reported?

CSIA is a very sensitive technique, and because the differences in isotopic ratios are so small, it is more convenient to express the ratios in “per mil” (parts per thousand, or ‰, similar to the usual metric for comparison, parts per hundred, or percent) values, relative to a reference standard. (A variety of standards exist, and it is important that a common standard be used by the laboratory and reported with the analytical results to enable data comparison). This is accomplished by using “delta” notation. The standard is a constant and is the isotopic ratio of an internationally agreed-upon standard (for example, for  $^{13}\text{C}/^{12}\text{C}$  the standard is  $R_{\text{Std}} = 0.01118$ ). The definition of delta is as follows:

$$\delta_x = 1000 \times \frac{R_x - R_{\text{Std}}}{R_{\text{Std}}}$$

where the  $R_x$  is the isotopic ratio of sample “x” and  $\delta_x$  (called “delta of sample x”) is linearly related to the isotopic ratio. Thus, if the  $\delta(^{13}\text{C})$  for a TCE sample is “–31 per mil” (a typical value for undegraded TCE), this means that the  $^{13}\text{C}/^{12}\text{C}$  in the sample is 31 per mil, or 3.1 percent, lower than in the internationally agreed-upon standard ( $R_{\text{Std}}$ ).

## Advantages

- CSIA does not rely on concentration trends or the observation of daughter products.
- Because it isolates the contaminant, CSIA is relevant only to that compound.
- CSIA can detect very small changes in the isotopic ratio. This high level of precision allows for careful assessment across a site to reveal subtle but important differences in the contaminants.
- CSIA is extremely versatile. It can be used for many contaminants in a wide range of applications, as listed on p. 1.
- The advantages of CSIA can be increased by applying it to multiple isotopes in a given molecule (e.g., both  $^{13}\text{C}/^{12}\text{C}$  and  $^{37}\text{Cl}/^{35}\text{Cl}$  in TCE). See Table 1.

## Limitations

- At this time, only a limited number of laboratories provide CSIA services. Though CSIA may theoretically be the way to answer a question at a site, the particular isotopic analyses may not be commercially available. If this is the case, collaboration with academic laboratories and/or application development with commercial laboratory should be investigated.
- Because of the large number of compounds in petroleum products, there is the potential for interference at petroleum-release sites (as well as other sites with many compounds in the groundwater), especially near the contaminant source. SIP is one alternative to assess biodegradation at these sites (see the SIP Fact Sheet for more information). In addition, modifications to CSIA methods can be used to overcome interferences. For example, dual GC column separations have been used effectively to evaluate biodegradation of the lead scavenger 1,2-dibromoethane in groundwater at gasoline release sites (EPA 2008b). The same technique is used for MTBE (Kuder et al. 2005).
- While the method is quite sensitive, there are limitations to that sensitivity. Isotopic fractionation may be so minimal that little or no isotopic enrichment is detected. This effect can occur in molecules with many of the same atom (e.g., several C atoms in one molecule). For example, CSIA of either carbon or hydrogen is unlikely to provide valuable information for many high-molecular-weight polycyclic aromatic hydrocarbons during degradation.

## Developing an Appropriate CSIA Study

The U.S. Environmental Protection Agency's *A Guide for Assessing Biodegradation and Source Identification of Organic Ground Water Contaminants Using Compound Specific Isotope Analysis* (EPA 2008a) discusses many of the technical aspects of this technology, including how to best design a CSIA study to address a particular question.



## Sampling Protocols

The sampling protocols for CSIA depend on what questions are being asked and which compounds are of interest. For example, if CSIA is used to study the isotopic ratios of carbon, hydrogen, or chlorine of VOCs in groundwater, then sampling involves the collection of water samples into standard volatile organic analysis vials. Extra vials are often required due to the need for both concentration/identification and isotopic ratio analyses, as well as potential multiple dilutions and additional quality control (QC) samples. However, if CSIA is being used to study the origin of perchlorate in groundwater, sampling is more complex, often requiring several hundred liters of groundwater to be pumped through special ion exchange columns to trap perchlorate (Bohlke et al. 2009). For all CSIA analyses, the laboratory that will be analyzing the samples should be contacted for technical assistance and to ensure that proper sampling protocols for collecting, handling, and transporting the samples are followed.

## Quality Assurance/Quality Control

The quality assurance (QA)/QC program used for CSIA sampling depends on the application. EPA (2008a) offers QA/QC guidance for the application of CSIA to VOCs in groundwater. Further, the NELAC Institute has general standards for sample handling, data manipulation, training, documentation, and reporting, all of which are important issues in acquiring CSIA services but which are not often covered in technical methods or in method-specific SOPs (NELAC 2011). If NELAC Institute certification is not available from a laboratory, then the user should discuss each of these issues with the laboratory. Currently, users can best ensure data quality by detailing the laboratory requirements in a site-specific QA project plan (QAPP).

It should also be noted that the EPA guide (2008a) may not provide sufficient for application of CSIA to perchlorate or other non-VOC contaminants. QA/QC procedures and issues should be developed and discussed with the laboratory performing the analysis, the project manager, the regulators, and stakeholders prior to the collection of samples.

## Additional Information

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## Glossary

**cis-DCE stall**—In biodegradation through reductive dechlorination, the parent chlorinated ethene is sequentially dechlorinated via the following process: (PCE →) TCE → *cis*-DCE → vinyl chloride → ethene. For a variety of reasons, the slowest step in the process is often the dechlorination of the *cis*-DCE. This phenomenon is known as "*cis*-DCE stall."

**environmental forensics**—The process of distinguishing contaminants from different sources.

**isotopic fractionation**—Some processes (for example, those which involve breaking chemical bonds) have slightly different rates for different isotopes, leading to a more rapid consumption of one isotope over the other. This characteristic is manifested in a change in the isotopic ratio of the residual compound.

**isotopic ratio**—The concentration of the heavy isotope normalized by the concentration of the light isotope.

**stable isotope**—A form of an element that does not undergo radioactive decay at a measureable rate.

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Environmental Council of the States

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