Examining the Extraction Efficiency of Petroleum-Derived Dissolved Organic Matter in Contaminated Groundwater Plumes

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Abstract

The extraction efficiency of petroleum-derived dissolved organic matter (DOM) was examined for groundwater samples from an aquifer contaminated with crude oil. Five different types of extraction techniques were investigated to determine which method is best suited for the analysis of potentially toxic petroleum-derived DOM. The five types were a liquid-liquid extraction (LLE) with dichloromethane (DCM) and total petroleum hydrocarbons-diesel range (TPH_d) with DCM (EPA method 3510C), and three solid-phase extraction (SPE) stationary phases that are routinely used for extraction of polar analytes from water. For the LLE and TPH_d that is selective for nonpolar compounds, the extraction efficiency of petroleum-derived DOM decreased downgradient as the petroleum-derived DOM becomes increasingly polar due to biodegradation. In contrast, the average extraction efficiency by the SPE methods was greater than 65% across the gradient. The results showed that SPE is more efficient for extracting petroleum-derived DOM at hydrocarbon-contaminated sites. The use of a method with greater extraction efficiency for partially oxidized hydrocarbons may prove useful in determining relationships between their composition and structure and potential for risks to human health or the environment.

Introduction

Over 25% of the United States relies on groundwater as a source of drinking water (Dieter et al. 2018). According to a 2012 report from the National Research Council (NRC), there are 126,000 groundwater sites in the United States that are contaminated (NRC 2012). Contamination by petroleum hydrocarbons results from human activities such as petroleum production and refining, hydraulic fracturing, underground storage tanks (USTs) and pipeline ruptures (Cozzarelli et al. 2014). Biotransformation begins once the oil/fuel infiltrates the subsurface (soil and groundwater) creating a series of degradation intermediate compounds that are more polar than the parent hydrocarbons from which they are derived (Thorn and Aiken 1998; Zemo et al. 2013; Islam et al. 2016; Podgorski et al. 2018). One of the most widely accepted remediation strategies for groundwater contamination is the process of monitored natural attenuation (MNA) (EPA 1999; NRC 2000). Natural attenuation is defined by the U.S. Environmental Protection Agency (USEPA) as “a variety of physical, chemical or biological processes that, under favorable conditions, act without human intervention to reduce the mass, toxicity, mobility, volume or concentration of contaminants in soil or groundwater” (EPA 1999). These in situ processes include “biodegradation; dispersion; dilution; sorption; volatilization; radioactive decay; transformation or destruction of contaminants” (EPA 1999). Methods of monitoring natural attenuation are dictated by the EPA, state appointed water management boards as well as internal procedures from oil companies (NRC 2000). Regulators often request total petroleum hydrocarbon (TPH) data for delineation of petroleum hydrocarbon plumes in environmental media (soil or water) or for a more comprehensive evaluation of risks from a petroleum release (CDC 2011; Zemo et al. 2013).

There is a lot of debate regarding the definition of TPH and the limitations of the current analytical techniques used to measure it. According to the Center of Disease Control (CDC), TPH is a term used to describe a large class of hydrocarbons that may be present in crude oil or petroleum products; these hydrocarbons include hexane, benzene, and toluene (CDC 2011). The Interstate Technology Regulatory Council (ITRC) defines TPH as “the analytical method used to measure TPH and the methods can be variable from state to state and even from lab to lab” (ITRC 2018). Some agencies such as the San Francisco Bay Regional Water Quality Control Board believe, regardless of the definition, everything that comes from the source zone should be evaluated at petroleum-contaminated...
sites (Fry and Steenson 2019). This also includes the analytical technique selected to measure the more polar compounds that are not amenable by gas chromatography (Zito et al. 2019). Zemo and Foote (2003) showed that most of the organic matter in groundwater at 22 fuel spill sites quantified using the USEPA method for TPH in the diesel range, C_{22}-C_{35} (TPH_{S}) was comprised of polar compounds (Zemo and Foote 2003). They concluded that these were likely biodegradation intermediates of petroleum hydrocarbons (Zemo and Foote 2003). Zemo et al. (2013, 2017) argued on the basis of 760 tentatively identified degradation intermediates identified by Mohler et al. (2013) that the intermediates pose little to no risk in groundwater, but the issue remains controversial (Mohler et al. 2013; Brewer and Hellmann-Blumberg 2014; ITRC 2018). The dissolved organic plume is of interest because it acts as a conduit to mobilize compounds derived at the petroleum source to downgradient receptors such as drinking water supplies and surface waters (Bekins et al. 2016; Mackay et al. 2018). This dissolved organic plume is comprised of native (background) dissolved organic matter (DOM; Luzius et al. 2018; Baker and Lamont-Biack 2001), dissolved petroleum hydrocarbons (Zemo and Foote 2003; Mohler et al. 2013; Zemo et al. 2013, 2017), water-soluble metabolites (Gieg and Sulfita 2002; Jobelius et al. 2011) and what the San Francisco Bay Regional Water Quality Control Board calls hydrocarbon oxidation products (HOPs) (Fry and Steenson 2019).

The extraction method used by risk assessors to quantify TPH_{S} is the USEPA method 3510C titled “Separatory funnel liquid-liquid extraction (LLE)” (EPA 1996). This method outlines a procedure for “isolating organic compounds” from water samples and is applicable to “the isolation and concentration of water-insoluble and slightly water-soluble organics” (EPA 1996). The data from TPH and 3510C determine the decision making for cleanup and risk assessment of an impacted site involving petroleum hydrocarbons and their transformation products (Zemo et al. 2013, 2017). However, the USEPA method 3510C for characterizing compounds in groundwater petroleum plumes is analytically limited because the compounds in the plumes become increasingly more polar during biotransformation (Thorn and Aiken 1998; Islam et al. 2016; Podgorski et al. 2018), and partitioning into the extraction solvent, dichloromethane (DCM) becomes less likely. Islam et al. (2016) analyzed polymeric Amberlite (XAD) resin extracted water samples from four Bemidji wells using Fourier transform ion cyclotron resolution mass spectrometry and reported compounds containing up to 10 oxygens (Islam et al. 2016). Harriman et al. (2017) also reported high oxygen content using Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) on solid phase extracted (SPE) DOM from biodegraded oil sand patties (Harriman et al. 2017). Therefore, analytically, the existing LLE-based methods are potentially missing most of the polar compounds hampering the effort to study the safety of MNA for petroleum-contaminated groundwater. There is a need for improved extraction techniques to target petroleum-derived DOM produced from the biotic and abiotic degradation of petroleum and refined fuels.

In the past few decades, there have been many extraction techniques to isolate DOM. A few examples are SPE techniques (XAD, PPL, C18) (Aiken et al. 1979; Lara and Thomas 1994; Dittmar et al. 2008), reversed osmosis and electrodialysis (Vetter et al. 2007) and silica discs (Kim et al. 2003). DOM is a complex mixture of aromatic, phenolic and aliphatic hydrocarbon structures with multiple heteroatom functionalities such as ketones, carboxyl, amides and hydroxy groups (Leenheer and Croué 2003). The operational definition of DOM is that which can pass through a 0.45μm membrane, although a range from 0.2 to 0.7 μm is commonly used based on the application (Aiken and Leenheer 1993; Leenheer and Croué 2003). Therefore, techniques used to extract DOM are very compatible for the extraction of water-soluble polar petroleum compounds, metabolites, and HOPs. Early on, extraction techniques used for DOM profiling were resin sorbents such as XAD and LLEs (Fontanals et al. 2004). However, SPE is becoming the more progressive extraction technique replacing LLE for isolation of more polar compounds from samples in all areas of chemistry spanning from environmental to food analyses (Thurman and Mills 1998; Poole 2003; Fontanals et al. 2004). More recent studies use solid-phase extraction (SPE) for the isolation of DOM (Kim et al. 2003; Dittmar et al. 2008). The use of SPE cartridges is widely popular due to the ease of the extraction methods, feasibility, reduced cost, reusability, and portability (Madikizela et al. 2018; David et al. 2019).

The three SPE techniques covered in the current study were selected based on their wide use in the environmental field for extraction of DOM (Dittmar et al. 2008). The C18 SPE sorbent comprises of silica beads modified with octadecyl groups bonded to their surface. The octadecyl groups are capable of isolating nonpolar to modified polar compounds from pharmaceuticals to pesticides in water (Dittmar et al. 2008) and have reported extraction efficiencies around 50% (Kaiser et al. 2003; Dittmar et al. 2008). Stereore divinyl benzene polymer or PPL SPE sorbent targets highly polar to nonpolar compounds in water and is widely used for DOM extraction in marine and riverine matrices with extraction efficiencies greater than 65% (Dittmar et al. 2008). Oasis hydrophilic-lipophilic balance (HLB) SPE sorbent is comprised of a copolymer stationary phase containing N-vinylpyrrolidone (hydrophilic) and divinylbenzene (lipophilic) monomers and is capable of isolating acidic, basic and neutral analytes with extraction efficiencies higher than 80% (Poole 2003; Fontanals et al. 2004, 2005; Delhomme et al. 2006). Given that petroleum-derived DOM has similar functional groups and aromatic cores to DOM, we believe that SPE is a more suitable isolation technique than LLE.

We hypothesize the use of methods developed for analyses of DOM would be more applicable and capture more carbon for the isolation of petroleum-derived DOM when compared to LLE and TPH. We believe that a significant fraction of polar compounds are missed when LLE (EPA 3510C) and TPH_{S} (EPA 8015S) are used to assess toxicity in petroleum contaminated sites. The use of an SPE extraction technique to isolate petroleum-derived DOM present in contaminated sites will give us better insight on the persistence and fate of DOM. The objective of this paper is to provide evidence that the SPE extraction technique should be considered to isolate petroleum-derived DOM from petroleum-contaminated plumes undergoing biodegradation processes in order to accurately monitor natural attenuation and potential threats to drinking water and aquatic ecosystems.
Methods

Site Description and Sample Collection

An oil pipeline rupture in 1979 located outside the city of Bemidji, Minnesota sprayed approximately 1.7 million liters of light (33° API) crude oil across an area of 6500 m² (Essaid et al. 2011). The spilled oil contained 0.56% sulfur and 0.28% nitrogen with a composition of 58 to 61% saturated hydrocarbons, 33 to 35% aromatics, 4 to 6% resins, and 1 to 2% asphaltenes (Eganhouse et al. 1993). The oil subsequently collected into depressions and approximately 25% of the unrecovered oil percolated through an unconsolidated silt, sand, gravel, and glacial outwash formation to form three residual oil bodies at the water table of the underlying aquifer (Essaid et al. 2011; Bekins et al. 2016). Samples for this study were collected from the north oil pool groundwater plume where the water table is 6–8 m below the land surface and the groundwater flows east-northeast at an average velocity of 22 m/year towards the Unnamed Lake (Essaid et al. 2011). Detailed maps and descriptions of the sample site can be found elsewhere (Baedecker et al. 1993; Eganhouse et al. 1993; Essaid et al. 2011; Fahrenfeld et al. 2014; Bekins et al. 2016). A Keck pump was used to purge each well with at least threewell volumes and 3 L of sample was collected after field measurements of pH, dissolved oxygen, temperature, and specific conductance stabilized. The samples collected for this study were from a background well (310E) located 200 m upgradient which contains native DOM, and from wells along a transect of the petroleum-derived DOM plume centerline located 39 m (533E), 102 m (9315B), and 254 m (925D) downgradient of the center of the oil body (Baedecker et al. 1993; Eganhouse et al. 1993; Essaid et al. 2011; Fahrenfeld et al. 2014; Bekins et al. 2016).

Solid Phase Extraction Procedure

PPL, C18, and HLB stationary phases were investigated to understand which produced the highest extraction efficiency for petroleum-derived DOM. For the PPL (100mg) and C18 (600mg), the method is described in Zemo et al. (2013). Briefly: The unfiltered samples were extracted using methylene chloride (DCM) in accordance with USEPA Method 3510C. Extracts were analyzed for TPH\textsubscript{d} using USEPA Method 8015B. In order to compare the TPH\textsubscript{d} data with the study, the data were corrected for the amount of carbon (1 mg TPH\textsubscript{d} = 0.87 mg C) to get TPH\textsubscript{d} per unit carbon (TPH\textsubscript{d}/C).

Liquid-Liquid Extraction Procedure

Samples and pure water (Table S1) were extracted following the EPA method 3510C. Briefly, water samples were filtered through a precombusted 0.27µm glass fiber filter and the pH was adjusted to 10 using 10 M sodium hydroxide. The bases and neutrals were extracted using 1:4 (v/v) DCM:sample. Each DCM fraction was collected and combined into a round bottom flask. The water sample was then adjusted to pH 2 with 10 M sulfuric acid and the acidic species were extracted using 1:4 DCM:sample. Each fraction was combined and the DCM was evaporated to a small volume and transferred to a 40-mL vial. The round bottom flask was rinsed with three 5 mL aliquots of DCM to ensure all residues was transferred and added to the 40mL vial. The DCM was evaporated to dryness to prepare for nonvolatile DOC analysis. The extractions were performed in triplicate for each well sample.

Dissolved Organic Carbon Analysis

Dried extracts of the four treatments (LLE, PPL, C18, and HLB) were redissolved into nanopure water and sodium hydroxide base was added to help dissolve any residue. Each sample was vortexed, adjusted with base a second time and sonicated for 60 min to ensure all residue was dissolved. Each sample was filtered and acidified to pH 2 for DOC analysis. DOC concentrations were measured with a Shimadzu TOC Analyzer using a high temperature combustion method and a platinitized alumina catalyst. The acidified samples (pH 2) were sparged for 5 min at 75 mL/min with ultra-pure air to remove inorganic carbon from samples prior to the measurement. The mean of three to five injections of 25 µL is reported for every sample and the coefficient of variance was <2% for replicate injections. Bracking six point potassium hydrogen phthalate standard curves were run every 10 samples and a single check standard was run every 5 samples to ensure system suitability during analysis. The concentration of DOC in the extract was multiplied by the volume of the redissolved extract (0.030 L) to calculate the amount of DOC (mg) in the extract. The initial and final amounts of DOC (mg) are reported in Tables 1 and S2 as the “DOC in water that was extracted” and “DOC extracted from the water,” respectively.

TPH\textsubscript{d} Analysis

Samples and pure water (Table S1) were extracted following the EPA method 3510C. Extracts were analyzed for TPH\textsubscript{d} using USEPA Method 8015B. In order to compare the TPH\textsubscript{d} data with the study, the data were corrected for the amount of carbon (1 mg TPH\textsubscript{d} = 0.87 mg C) to get TPH\textsubscript{d} per unit carbon (TPH\textsubscript{d}/C).

Percent Extraction Efficiency Formula

\[
\% \text{ Extraction efficiency} = \frac{\text{DOC extracted from the water (mg)}}{\text{DOC in the water that was extracted (mg)}} \times 100
\]

Statistical Analyses

Microsoft Excel 2016, Redmond, Washington data analysis add-in was used to run a one-way ANOVA analysis (Table S3) and a t-test (Table S4) to compare the different extraction techniques (LLE, TPH\textsubscript{d}, PPL, C18, and HLB).

Results

Figure 1 compares DOC and TPH\textsubscript{d}/C as a function of distance from the study site oil body where biodegradation...
of the oil has been occurring for 40 years (Thorn and Aiken 1998; Essaid et al. 2011; Islam et al. 2016; Podgorski et al. 2018).

These data show that the DOC (mg C) is higher than TPH\textsubscript{d}/C (mg-C) each well. This result indicates that the extraction and/or the analytical methods used for the TPH\textsubscript{d} measurement are not capturing all of the organic carbon in the polar plume. To understand if the large gap of missing carbon between the TPH\textsubscript{d} and DOC concentrations is a result of extraction selectivity, we selected four representative wells across the plume of the site and assessed the extraction efficiency of the LLE method, TPH\textsubscript{d} and three SPE sorbents. The four wells selected were 310E, 533E, 9315B, and 925D, and cover the site from 200 m upgradient to 39, 102, and 254 m downgradient from the oil body, respectively.

Percent extraction efficiency was calculated for the TPH\textsubscript{d}/C (black), LLE (red), PPL (blue), C18 (green), and HLB (purple) extractions for the selected wells (Figure 2). The wells are plotted in order of increasing Polarity of DOM or farthest distance from oil body with 310E located upgradient from the oil body (−200 m). Table 1 lists each well with their corresponding distance from the oil body (m), extraction efficiency (%) for each technique.

| Well | Distance From Source (m) | Extraction type | Initial DOC (mg/L) | Volume Extracted (L) | DOC in water that was extracted (mg) | DOC extracted from the water (mg) | Extraction Efficiency (%) |
|------|--------------------------|----------------|-------------------|---------------------|-------------------------------------|----------------------------------|--------------------------|
| 533E | 38.8                     | TPH\textsubscript{d}/C | 22.2              | 1.0                 | 22.2                               | 7.05                             | 31.7                     |
|      |                          | LLE             | 0.05              | 1.11                | 0.27                               | 24.6 ±10.1                       |
|      |                          | PPL             | 0.005             | 0.11                | 0.09                               | 83.0 ±4.1                        |
|      |                          | C18             | 0.005             | 0.11                | 0.10                               | 87.8 ±5.2                        |
|      |                          | HLB             | 0.005             | 0.11                | 0.13                               | 119 ±22                          |
| 9315B| 102                      | TPH\textsubscript{d}/C | 9.78              | 1.0                 | 9.78                               | 2.0                              | 20.5                     |
|      |                          | LLE             | 0.125             | 1.22                | 0.23                               | 18.4 ±3.3                        |
|      |                          | PPL             | 0.009             | 0.09                | 0.08                               | 93.2 ±12                        |
|      |                          | C18             | 0.009             | 0.09                | 0.08                               | 93.5 ±2.7                        |
|      |                          | HLB             | 0.009             | 0.09                | 0.09                               | 98.1 ±9.0                        |
| 925D | 254                      | TPH\textsubscript{d}/C | 2.31              | 1.0                 | 2.31                               | 0.13                             | 5.65                     |
|      |                          | LLE             | 0.125             | 1.16                | 0.20                               | 16.9 ±2.4                       |
|      |                          | PPL             | 0.053             | 0.12                | 0.10                               | 83.6 ±4.9                       |
|      |                          | C18             | 0.053             | 0.12                | 0.09                               | 74.6 ±1.9                       |
|      |                          | HLB             | 0.053             | 0.12                | 0.09                               | 69.6 ±18                        |
| 310E | –200\textsuperscript{a} | TPH\textsubscript{d}/C | 1.26              | 1.0                 | 1.26                               | 0.08                             | 6.56                     |
|      |                          | LLE             | 0.75              | 0.95                | 0.04                               | 4.5 ±0.4                        |
|      |                          | PPL             | 0.078             | 0.10                | 0.08                               | 78.4 ±5.4                       |
|      |                          | C18             | 0.078             | 0.10                | 0.07                               | 76.1 ±5.7                       |
|      |                          | HLB             | 0.078             | 0.10                | 0.08                               | 83.4 ±16                        |

Figure 1. Amount of carbon (mg) versus distance from oil body (m) for DOC (black) and TPH\textsubscript{d}/C (blue) for Bemidji wells 533E, 9315B, 925D, and 310E.

Table 1
Comparison of the Efficiency of Liquid-Liquid Extraction Techniques (TPH\textsubscript{d}/C and LLE) and Solid Phase Extraction Techniques (PPL, C18, and HLB) to Extract Polar Organic Compounds from Groundwater

Percent extraction efficiency was calculated for the TPH\textsubscript{d}/C (black), LLE (red), PPL (blue), C18 (green), and HLB (purple) extractions for the selected wells (Figure 2). The wells are plotted in order of increasing Polarity of DOM or farthest distance from oil body with 310E located upgradient from the oil body (~200 m). Table 1 lists each well with their corresponding distance from the oil body (m), extraction efficiency (%) for each technique.

Notably, as the distance from the oil body increased, the extraction efficiency of the LLE and TPH\textsubscript{d}/C decreased moving downgradient from the well with the most nonpolar petroleum-derived DOM, 533E, to 925D. The background well with the most polar DOM native to the groundwater, 310E, had the lowest value. Overall, the SPE methods achieved significantly higher ($p<0.05$, Table S3) extraction efficiency than LLE and TPH\textsubscript{d} for every well and did not show the same trend in decreasing efficiency with distance downgradient. The average extraction efficiency for all of the wells ($n = 12$ for four wells run in triplicate) was $16.1 ± 8.4\%$ for the LLE, $84.6 ± 6.2\%$ and $83.0 ± 9.1\%$ for
the PPL and C18, respectively and 92.4 ± 20.9% for the HLB. The C18 extraction was higher for well 533E compared to both the LLE and PPL and comparable for wells 9315B, 925D, and 310E. The HLB had the highest extraction efficiency for wells 533E and 9315B and comparable values to PPL and C18 for wells 925D and 310E.

Discussion

The data from this study illustrate a gap of missing carbon when the TPH_d and LLE methods are used together to assess the concentration of polar petroleum transformation products in contaminated groundwater (Figure 1). Figure 2 shows that only 16% of the carbon on average is being extracted using LLE and TPH_d isolation techniques, which rely on DCM as an extraction solvent. The efficiency of LLE for wells in the plume of petroleum-derived DOM is lowest at the well farthest downgradient. Not only does this mean the plume extent is undefined, but the risk to downgradient receptors for DOM containing HOPs is unknown. The concentrations of DOC determined using LLE are biased low or are non-detect. This is important information since the LLE and TPH_d methods are being used to monitor natural attenuation sites and contaminated groundwater. The EPA data on active UST leaks indicates that over 65,000 sites are designated to be cleaned up. Nationwide there have been over half a million UST releases reported to the USEPA since 1984 (EPA 2018). Although 88% have completed cleanups, Wisconsin and Minnesota state data show that residual fuel is still present in the subsurface at 27–44% of sites (Evanston et al. 2009; MPCA 2019). If these percentages apply in all states then upwards of 135,000 legacy UST release sources may exist nationwide. Our data show that the routinely used extraction solvent DCM is not capturing a large fraction of petroleum-derived DOM (Figure 2). Therefore, any studies that only use TPH_d to monitor these sites are missing a significantly large pool of polar compounds because they are using DCM extracts to evaluate the extent of contamination and potential adverse effects to receptors. It is also noteworthy to mention that the groundwater for the TPH_d method is not filtered prior to analysis; therefore, the results may include suspended oil droplets and/or particulate organic matter (Zsolnay 2003).

When comparing the different SPE sorbents used in this study, the HLB sorbent had the highest extraction efficiency for the petroleum-derived DOM for all wells with the highest for well 533E. This may be due to the hydrophilic and lipophilic monomers on the sorbent which target broad classes of compounds. The average extraction efficiency for all of the wells provides evidence that SPE technique, regardless of sorbent is better for isolation of petroleum-derived DOM collected from sites contaminated with petroleum.

The HLB extraction had the highest error compared to the LLE, C18, and PPL. However, it is unclear where the source of error came from other than the long elution times during the extraction procedure. Future work looking at the compositional selectivity of each technique will provide more insight.

Conclusion

Although limited in scope, these results show that conventional extraction techniques used to monitor natural attenuation and characterization of petroleum-contaminated groundwater are antiquated given the recent understanding that the majority of the petroleum-derived DOM are polar partial transformation products. This study improves our understanding of extraction techniques used for extraction of petroleum-derived DOM and which are best to use for monitoring natural attenuation of petroleum-contaminated sites. Future work will investigate the use of analytical methods optimized for nonpolar compounds used to measure petroleum-derived DOM and how they may be analytically blind especially when using DCM extraction techniques.

Figure 2. Percent extraction efficiency comparing TPH_d/C (black), LLE (red), PPL (blue), C18 (green), and HLB (purple) techniques. Error bars reflect 95% confidence interval of the mean (n = 3).
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Supporting Information

Additional Supporting Information may be found in the online version of this article. Supporting Information is generally not peer reviewed.

Table S1. Dissolved organic carbon concentrations (mgC/L) for nanopure water with 95% confidence interval (n = 3) for each technique.

Table S2. Comparison of the efficiency of liquid-liquid extraction techniques (TPH/C, LLE, PPL, C18, and HLB) to extract polar organic compounds from groundwater.

Table S3. ANOVA single factor analysis comparing extraction techniques (TPH/C, LLE, PPL, C18, and HLB) for each well. The data are highly statistically different therefore, the null hypothesis can be rejected. F value > F crit (39 > 3.1) and p-value was <alpha value = 0.05 (9.5 x 10^-8 < 0.05).

Table S4. t-Test results comparing each extraction technique (TPH/C, LLE, PPL, C18, and HLB). Data are statistically different when P(T≤t) two-tail < 0.05.

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