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## Microbial contamination and its control in fuels and fuel systems since  $1980 - a$  review

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#### 1. Introduction

#### 1.1. The problem

First documented by [Miyoshi \(1895\)](#page--1-0), fuel biodeterioration has been well documented for more than a century ([Gaylarde et al.,](#page--1-0) [1999](#page--1-0)). Bacteria and fungi proliferate and are most metabolically active at interfaces within fuel systems [\(Passman, 2003](#page--1-0)). Selectively depleting primary aliphatic compounds, contaminant populations adversely affect a variety of fuel performance properties [\(Passman,](#page--1-0) [1999](#page--1-0)). Moreover, metabolically active microbial communities produce metabolites that can accelerate fuel deterioration ([Rosenberg et al., 1979; Morton and Surman, 1994\)](#page--1-0). Fuel deterioration is more likely to be problematic in bulk storage systems in which turnover rates are slow  $(>30 \text{ d})$ ; [Chesneau, 1983](#page--1-0)). This reflects the longer contact time between the stored fuel and biodeteriogenic microbial populations. In fuel systems with faster

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#### **ABSTRACT**

Although the documentation of fuel biodeterioration dates back to the late 19th century, general recognition of the value of microbial contamination control evolved slowly until the 1980s. Since the early 1980s a number of factors have converged to stimulate greater interest in fuel and fuel system biodeterioration. This, in turn, has stimulated applied research in the ecology of biodeteriogenic processes and biodeterioration control. This presentation reviews progress in both of these areas since 1980. The aforementioned factors that have provided the impetus for improved microbial control, the evolution of our understanding of the nature of the biodeteriogenic processes will be discussed. Activities of consensus organizations to develop guidelines and practices will also be reviewed.

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turnover rates, the risk of infrastructure damage is substantially greater than the risk of product deterioration.

The two primary types of infrastructure problems caused by microbes are microbiologically influenced corrosion (MIC) and fouling. [Little and Lee \(2007\)](#page--1-0) have recently reviewed MIC in considerable detail. Fouling includes the development of biofilms on system surfaces, consequent flow-restriction through small diameter piping, and premature filter plugging. MIC is linked inextricably with biofilm development ([Little and Lee, 2007\)](#page--1-0). Biofilms on tank gauges cause inaccurate readings [\(Williams and Lugg,](#page--1-0) [1980](#page--1-0)). The concept of premature filter plugging will be explored in greater detail below.

This review will discuss current knowledge of the factors involved in fuel and fuel system biodeterioration.

#### 12. The remedies

Water is an essential factor for microbial activity [\(Allsopp et al.,](#page--1-0) [2004](#page--1-0)). Consequently, the most commonly recommended measure for mitigating against microbial activity in fuel systems is water control ([Swift, 1987; Arnold, 1991\)](#page--1-0). As will be discussed below, preventing water accumulation in fuel systems is not a trivial







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process. Once significant microbial contamination is present, the two primary processes for removing accumulated biomass and for eradicating contaminant microbes are tank cleaning and treatment with microbicides ([Chesneau, 2003\)](#page--1-0). Process selection depends on fuel system configuration, fuel application and fuel grade. Regulatory considerations also impact microbial control strategy selection. All of these factors will be addressed in this paper.

#### 2. Fuel biodeterioration

#### 2.1. Fuels as nutrient sources

The differentiation between bioremediation (typically reported as biodegradation) and biodeterioration is primarily commercial. Both are consequences of microbiological activity. When fuel degradation is desired (for example, after spills or tank leaks) the operative term is bioremediation. When fuel loses commercial value then we identify the phenomenon as biodeterioration. From a microbial ecology perspective, there is little difference between biodeterioration and bioremediation. [Passman et al. \(1979\)](#page--1-0) reported that approximately 90% of the heterotrophic population recovered from surface waters of the North Atlantic Ocean could use  $C^{14}$ -dodecane as a sole carbon source. As explained by [Gaylarde](#page--1-0) [et al. \(1999\),](#page--1-0) all petroleum fuels are comprised of hydrocarbons, organonitrogen and organosulfur molecules and a variety of trace molecules, including organometals, metal salts and phosphorous compounds. These molecules provide nitrogen, sulfur, phosphorus  $e$  essential macronutrients and well as a range of mineral micronutrients. Petroleum distillate fuels are derived from distillation fractions (cuts) of crude. Table 1 summarizes a number of primary properties of petroleum distillate fuels. The molecular size distributions shown in the table belie the complexity of petroleum fuels. Gasolines are blends of n-, iso- and cyclo-alkanes  $(31-55%)$ ; alkenes  $(2-5%)$  and aromatics  $(20-50%)$  [\(IARC, 1989\)](#page--1-0). Chemical complexity increases dramatically as the carbon number and carbon number range increase. Middle distillate fuels typically have thousands of individual compounds including alkanes (64%; including n-, iso- and cyclo-alkane species), alkenes  $(1-2\%)$ , aromatics ( $\sim$ 39%) and heteroatomic compounds ([Bacha et al.,](#page--1-0) [1998\)](#page--1-0). As noted previously, the heteroatomic compounds include organonitrogen and organosulfur molecules. [Robbins and Levy](#page--1-0) [\(2004\)](#page--1-0) have also reviewed the fuel biodeterioration literature, concluding that all grades of conventional, bio and synthetic fuel are subject to biodeterioration. The following subsections will review recent studies demonstrating biodeterioration of various grades of commercial fuels.

#### 2.2. Gasoline biodeterioration

Historically, conventional wisdom held that the  $C_5-C_{12}$  molecules comprising gasoline somehow rendered gasoline inhibitory







to microbial growth ([Bartha and Atlas, 1987](#page--1-0)). This conventional wisdom apparently ignored the antimicrobial effect of tetraethyl lead – present at  $\sim 800$  mg kg<sup>-1</sup> in most gasoline products until the late 1970s when the U.S. EPA and governmental agencies other countries phased out its use ([Lewis, 1985\)](#page--1-0). A recent case study in China identified tetraethyl lead removal as a primary factor in highoctane gasoline deterioration in depot and retail site tanks ([Zhiping](#page--1-0) [and Ji, 2007](#page--1-0)). Passman and coworkers reported that uncharacterized microbial populations, obtained from microbially contaminated underground storage tanks (UST), selectively depleted  $C_5$  to  $C_8$  alkanes in gasoline ([Passman et al., 2001](#page--1-0)). However, in their survey of 96 regular, mid-grade and premium gasoline, and diesel fuel tanks, [Rodríguez-Rodríguez et al. \(2010\)](#page--1-0) did not detect any evidence of physicochemical changes in any of the sampled. It is likely that the dilution effect masks any such changes that might be occurring in storage tanks with  $>$ 35 m<sup>3</sup> capacity.

Ethanol and butanol use as oxygenates is growing ([Kanes et al.,](#page--1-0) [2010\)](#page--1-0). These alcohols are used as disinfectants at concentrations  $>$ 20% (v/v) [\(HSE, 2009\)](#page--1-0). At these concentrations some might feel reassured that given the disinfectant properties of these alcohols, it is unlikely that alcohol-blended gasolines will be susceptible to biodeterioration. [Mariano et al. \(2009\)](#page--1-0) have demonstrated that both butanol ( $\omega$  <10% by vol) and ethanol ( $\omega$  <20% by vol) stimulated gasoline mineralization in microcosms. In contrast, [Österreicher-Cunha et al. \(2009\)](#page--1-0) observed that selective metabolism of ethanol retarded BTEX (benzene, toluene, ethylbenzene and xylene) metabolism in soils contaminated from leaking UST that held E-blended (E-20 to E-26) gasoline. They found overall enhanced microbial activity but depressed BTEX degradation relative to soils in which ethanol was not present. [Solana and](#page--1-0) [Gaylarde \(1995\)](#page--1-0) had previously observed E-15 gasoline biodeterioration in laboratory microcosms. [Passman \(2009\)](#page--1-0) reported having observed metabolically active microbial populations in phaseseparated water under E-10 gasoline in underground storage tanks (UST) at gasoline retail sites (gas stations) in the U.S. In an unpublished poster presentation at the 11th International Conference on the Stability and Handling of Liquid Fuels held in Prague in 2009, English and Lindhardt presented data showing significant microbial contamination in the phase-separated aqueous layer under E-10 gasoline samples from retail UST in Europe. These field observations suggest that biodeterioration is a potential problem in fuel systems handling ethanol-blended gasoline, although reports of operational problems conclusively attributed to microbial activity are still relatively rare.

However, in two successive microcosm studies Passman observed opposite results. In one study [\(Passman, 2009](#page--1-0)), bottomwater biomass covaried with the fuel-phase ethanol concentration (E-0, E-10, E-15 and E-20;  $r^2 = 0.95$ ). In a second study, meant to corroborate the first series of triplicate experiments, [Passman](#page--1-0) [et al. \(2009\)](#page--1-0) observed an inverse relationship between fuel-phase ethanol concentration and bottom-water biomass  $(r^2 = 0.99)$ . Both studies used ethanol blends over 0, 0.5 and 5% bottom-water. For E-5, E-10 and E-20 fuels over 5% bottom-water, the ethanol concentration in the aqueous phase was  $50 \pm 2.5\%$  by vol, regardless of the ethanol concentration in the fuel phase. Clearly, additional work is needed to assess the impact of alcohol-fuel blends on fuel biodeterioration susceptibility.

#### 2.3. Diesel and biodiesel fuel biodeterioration

In contrast to the relatively limited literature describing gasoline biodegradation, there is a substantial body of work describing the biodegradation of middle distillate fuels ([Leahy and Colwell, 1990;](#page--1-0) [Hill and Hill, 1993; Bento and Gaylarde, 2001; Ghazali et al., 2004;](#page--1-0) [Robbins and Levy, 2004](#page--1-0)).

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