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## Microbial Degradation of Gasoline in Soil: Comparison by Soil Type

D A Turner <sup>1</sup>, J. Pichtel <sup>2</sup>, Y Rodenas, <sup>3,2</sup> J McKillip <sup>3</sup> and J V Goodpaster <sup>1\*</sup>

<sup>1</sup>Department of Chemistry and Chemical Biology, Forensic and Investigative Sciences Program, Indiana University Purdue University Indianapolis (IUPUI), Indianapolis, USA

<sup>2</sup>Department of Natural Resources and Environmental Management, Ball State University, Muncie, USA <sup>3</sup>Department of Biology, Ball State University, Muncie, USA

#### Abstract

'XULQJ WKH LQYHVWLJDWLRQ RI D VXVSLFLRXV ¿UH GHEULV LV RIWHQ FROOHI of ignitable liquids (e.g., gasoline). In cases where the debris is contaminated with soil, it is known that heterotrophic soil microorganisms can alter the chemical composition of the ignitable liquid residue over time. The effects of soil type and season upon this phenomenon are not known, however. Hence, soil collected from locations under WKUHH GLIIHUHQW XVHV UHVLGHQWLDO DJULFXOWXUDO EURZQ;HOG ZHUH VSI GD\V 7KH VRLOV ZHUH DOVR FKHPLFDOO\ DQG ELRORJLFDOO\ PRQLWRUHG IRU VKRZHG WKDW UHVLGHQWLDO VRLO ZDV PRVW DFWLYH DQG EURZQ; HOG VRLO OHE 7KH EURZQ;HOG VRLO SRVVHVVHG UHODWLYHO\ KLJK PJ NJ FRQFHQWUDWLR activity. Predominant viable bacterial populations enumerated using real-time reverse transcriptase polymerase chain reaction (RT-PCR) included members of the Alcaligenes, Acinetobacter, Arthrobacter, Bacillus, Flavobacterium, and Pseudomonas genera. Principal Components Analysis (PCA) was found effective in elucidating trends of microbial degradation among the different soil types and seasons. The results of this study demonstrate the necessity of SURPSW DQDO/VLV RI ÍRUHQVLF HYLGHQFH IRU SURSHU LGHQWL¿FDWLRQ RI SRVV

Several factors a ect bacterial numbers and activities in soil Keywords:Soil chemistry; Bacteria; Microbial degradation; Ignitable including soil type and season. Chemical and physical characteristics of liquids

#### Introduction

soils including pH, nitrogen level and phosphorus content will vary, as do soil physical properties (e.g., texture). In turn, varying populations

e number of incendiary res in the U.S. averages approximately of bacteria may impact the degree of microbial degradation observed in 210,300 every year, which comprises about 13% of the total of an debris samples containing soil.

reported res, according to FEMA's Topical Fire Report Series[1]. On an Previous work has demonstrated that bacteria readily degrade annual basis, incendiary res claim 375 lives, injure over one thousand mormal alkanes (e.g. decane) and lesser substituted alkyl benzenes (e.g. people, and cause approximately \$1 billion in direct property damage livene, ethyl benzene, propyl benzene) while more highly substituted [1]. In many cases, the arsonist uses an ignitable liquid to accelerate arkyl benzenes (e.g., 1,2,4-trimethylbenzene) and highly branched the re. Gasoline is the most commonly used ignitable liquid as it is likanes are more resistant to microbial attack [4-6]. While treatment readily accessible, inexpensive and ignites easily [2]. Gasoline and other of hydrocarbon-contaminated soils by bacteria is a well-known ignitable liquids are classi ed according to the American Society of hydrocarbon-contaminated soils by bacteria is a well-known phenomenon in the environmental engineering community [7-17], Testing and Materials (ASTM) guidelines by their boiling point range microbial processes are not wellunderstood in forensic science, is and chemical composition [3]. In practice, a forensic chemist will use various extraction methods coupled with gas chromatography/mass spectrometry (GC/MS) to determine if an Ignitable Liquid Residue (ILR) is present in a re debris sample. e ILR will then be classi ed heterotrophic bacteria. according to ASTM guidelines [2,4].

e overall objectives of this study were to assess the degradation Media rich in organic matter such as soil provides a rich source a common ignitable liquid (i.e., 87 octane gasoline) in soil as a ected of carbon and typically contains substantial quantities of actively soil type. e focus of this paper will be upon the e ect of soil bacterial biomass. Since ignitable liquids are composed of a range to include: (1) analysis of GC/MSdata from gasoline added to hydrocarbons, they may be suitable as a carbon substrate by bactehige di erent soils; (2) identi cation and quanti cation of bacterial Such transformations are problematic for re debris analysis as samples pulations present in the study soils; and (3) semi-quanti cation are o en stored for many weeks at room temperature before they are

analyzed due to case backlog and lack of cold storage. As a result,

selective loss of hydrocarbon species due to bacterial metabolism canresponding author: JV Goodpaster, Department of Chemistry and Chemical occur, making the identi cation and classi cation of ignitable liquid Biology, Forensic and Investigative Sciences Program, Indiana University Purdue residues di cult or even impossible. For example, ve speci<sub>3</sub> c C

alkylbenzenes (3-ethyltoluene, 4-ethyltoluene, 1,3,5-trimethylbenzeneeceived December 20, 2013; Accepted February 04, 2014; Published February 2-ethyltoluene and 1,2,4-trimethylbenzene)must be identied in10, 2014

a sample in order to determine if residues of gasoline are presedutation: Turner DA, Pichtel J, Rodenas Y, McKillip J, Goodpaster JV (2014) Furthermore, because these compounds also occur in other material crobial Degradation of Gasoline in Soil: Comparison by Soil Type. J Bioremed they must be present in relative amounts that are similar to that of a

gasoline standard [3]. Among the serious consequences of microbial pyright: © 2014 Turner DA, et al. This is an open-a ccess article distributed degradation are the selective losses of some of these compounds an Under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the changes in the ratios of these compounds in a gasoline sample. original author and source are credited.

Page 2 of 7

of organic and inorganic compounds present in the study soils by Principal Component Analysis (PCA).

### Materials and Methods

#### Soil chemical analyses

Soil material was obtained from an agricultural eld (Pella clay), a residential property (Miamian sandy clay), and a brown eld site

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Page 3 of 7

contained high levels of phosphorous (P) and potassium (K) due to treatment with commercial fertilizers.

Levels of extractable Cd, Cr, Fe and Zn were all within range for non-contaminated soils (Table 1). However, extractable Pb levels in the brown eld soil measured 497 mg/kg. An upper limit for Pb content of a normal soil is approximately 70 mg/kg [27]. e levels of Pb in soils that are toxic to soil microorganisms and plants are a function of species, Pb concentration and soil factors (e.g., pH, fertility status, presence

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Conclusions

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Page 7 of 7

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