



SDI FINAL EVALUATION FORM 1.1

PART 1:

Journal Name:	International Research Journal of Pure and Applied Chemistry
Manuscript Number:	MS: 2012/IRJPAC/2489
Title of the Manuscript:	COMPARATIVE STUDY OF BENZYL AND PHENYL COMPOUNDS EFFECTS ON THE INHIBITION OF METHANE GAS BIOSYNTHESYS

PART 2:

FINAL EVALUATOR'S comments on revised paper (if any)	Authors' response to final evaluator's comments
The revised manuscript failed to adequately address major concerns raised by the reviewer. Namely, the relevancy of this work as related to previous research and the quality assurance/quality control (QA/QC) issue. 1. Lots of similar if not identical research have been done before by environmental engineers and scientists in the 80's with numerous publications. The authors need to acknowledge those and address the innovativeness of this work as related to the previous ones. 2. Methane gas production was the only parameter monitored and used in reaching conclusion. The QA/QC issue should be addressed.	1 The innovativeness of this work as related to previous one is: a) At our knowledge, most of these works were performed with the granular sludge as inoculum from an industrial anaerobic reactor called "Upflow Anaerobic Sludge Bed" (UASB-reactor). Generally, the digesters are heated to about 30° C and more, but in this work digested pig manure from a laboratory scale digester was used as inoculum and the experiment was conducted at room temperature of an African tropical country (27±1°C). Unlike granular sludge, digested pig manure is a natural inoculum and has never been in contact with xenobiotic aromatic compounds and thus, not acclimated to them. But, there is almost no literature data concerning the methanogenic inhibition of aromatic compounds determined with digested pig manure as inoculum. b) The present work aims to study the effect of aromatic structure on the inhibition of the acetoclastic methanogenesis by digested pig manure methanogenic archaea. The aromatic structure depends of the nature, the number and the position of substituents on aromatic ring. The effects of hydrobicity, hydrophilicity, electronic effects, formation of intramolecular and intermolecular hydrogen bonds and steric hindrances of substituents on aromatic ring have to be investigated. The diffusion of a toxic compound across a microbial membrane depends on the permeability of the membrane. However, the membrane permeability is a function of the partition coefficient logP_{oct} and the diffusion coefficient (substance mobility). The objective of this study was also to examine the structure-toxicity relationships of aromatic compounds to acetoclastic methanogens. The ultimate goal of this work and our previous [] works is the modeling the effect of the aromatic structure on the inhibition of methane biosynthesis and to predict the behavior of aromatic compounds. These results will also be applied in medical field as drug, antibiotic and pesticide molecules design and synthesis. c) The system is also used for presence detection of toxic compounds in rivers and lakes even in drink water and beverages.. 2. Assurance quality/Quality control (QA/QC). a) All the used chemicals were pure pro analysis and supplied by MERCK. b) The method used is a standard method of serum bottles used by many authors in anaerobic toxicity assay [5,6,7,17] and in our previous papers [13,14,15,18,19,20,21] The methane gas production was not the only parameter monitored; the presence of oxygen in the digesters was monitored with rezasurin during the anaerobic toxicity test. The concentration of acetate was dosed and compared to the quantity of methane produced during the test. It was found that the methane production was proportional to the concentration of acetate in the digester and the quantity acetate COD converted was equal to the quantity of methane COD measured. c) Precision in determination of IC₅₀: According to Field and Sierra [6] the 50% inhibiting concentration (CI₅₀) cannot be accurately estimated if the inhibition of the highest concentration treatment is less than 50%. It cannot also be accurately estimated if the



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	inhibition of the lowest concentration treatment is greater than 80%.But the best precision of the IC_{50} value is obtained when at least one concentration treatment inhibits less than 50% and at least one other that inhibits between 50 and 80%. This procedure has been strictly adhered to in this work. d) The measurement of CI_{50} was made in triplicate and the standard derivations are given in the manuscript.
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