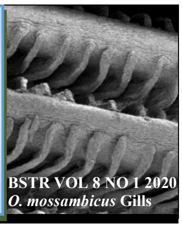
HIBISCUS PUBLISHER

BIOREMEDIATION SCIENCE AND TECHNOLOGY RESEARCH

Website: https://journal.hibiscuspublisher.com/index.php/BSTR



Kinetics Modelling of Tributyltin Toxicity on The Growth of *Bacillus stearothermophilus*

Abdussamad Abubakar[1], Salihu Ibrahim[2], Ilelah Kabiru Garba[3], Abubakar Sadiq Tanko[4], Mansur Abdulrasheed[5], Abdullahi Adamu[6], Bose Mahmud Mohammed[7], Garba Uba[8]*

[1]Department of Microbiology, Faculty of Sciences, Bauchi State University Gadau, PMB 65 Itas Gadau, Bauchi, Nigeria.
[2]Center for Biotechnology Research, Bayero University, PMB 3011 Kano, Nigeria.
[3]Department of Environmental Management Technology, Faculty of Environmental Studies, Abubakar Tafawa Balewa University, PMB 0248 Bauchi, Nigeria.
[4]Department of Biochemistry, Faculty of Sciences, Bauchi State University Gadau, P. M. B. 65 Itas Gadau, Bauchi, Nigeria.
[5]Department of Microbiology, Facuty of Science, Gombe State University, Gombe Nigeria.
[6]Department of Environmental and Resource Management, Faculty of Engineering and Environmental Design, Usman Danfodiyo University, PMB 2346 Sokoto, Nigeria.
[7]Department of Geography and Environmental Sciences, Federal University Kashere PMB 0182 Gombe, Nigeria.
[8]Department of Science Laboratory Technology, College of Science and Technology, Jigawa State Polytechnic, PMB 7040 Dutse, Nigeria.

*Corresponding author:
Garba Uba
Department of Science Laboratory Technology,
College of Science and Technology,
Jigawa State Polytechnic, PMB 7040
Dutse,
Nigeria..
Email: garbauba@jigpoly.edu.ng

HISTORY

Received: 20th March 2020
Received in revised form: 30th April 2020
Accepted: 1st July 2020

KEYWORDS

Tributyltin
Toxicity
Bacillus stearothermophilus
Kinetics

ABSTRACT

The growth of the bacterium, *Bacillus stearothermophilus* was intensely inhibited by tributyltin (TBT). As the TBT concentration increases, the overall specific growth rate was inhibited. The growth rates obtained were then modelled according to the modified Han-Levenspiel, Wang, Liu, Shukor and modified Andrews. Among the five models, Wang shows the best fitting while the Andrew model shows poor fitting. Results of the statistical analysis showed that the Wang model was the best model based on the lowest values for RMSE and AICc, highest adjusted correlation coefficient (AdR^2) and values of AF and BF closest to unity. The parameters obtained from the Wang model, which are *Ccrit,* μ_{max} and *m* which represent critical TBT concentration (nM), maximum growth rate (nM h) and empirical constant values were 177.99 (95%, confidence interval 144.26 to 211.71), 2.41 (95%, confidence interval of 2.17 to 2.64) and 2.76 (95%, confidence interval of 1.44 to 4.07) respectively.

INTRODUCTION

Tributyltin (TBT) has been extensively used as an antifouling biocide in maritime paints to prevent the attachment of fouling organisms from the surface of ships and boats [1]. Tributyltin is found to be highly persistent especially in sediment and very toxic to even non targeted organisms [2]. Use of TBT as marine biocide has been barn long ago globally in 2008 by the International Maritime Organization (IMO), however due to it persistent nature its residue has been reported in many places across the world including South Africa, Strait of Johore Malaysia, Portugal, Australia [3–6]. High concentrations still persistent in fresh water and sediments and can stays up to 30 years [3,7]. Organotin used in industries have also been detected in terrestrial environment [8]. Bacteria have been well-known to degrade organic pollutants [9]. Martin et al. [10] studied the interactions between *Bacillus stearothermophilus* with different TBT concentrations (0, 100, 200, 300, 400 and 500 nM). As the TBT concentration increases, the growth of *Bacillus stearothermophilus* was severely inhibited. The bacterium, *Bacillus stearothermophilus* have been used as suitable model for drug and pesticide toxicity assessment [11].

In this study, numerous models such as the modified Han-Levenspiel [12], Wang , Liu [13], modified Andrews [14], Amor [15] have been utilised [16] to evaluate the result of TBT toxicity on *B. stearothermophilus* bacterium [17]. From these models inhibition related constants, which include *C, Ccrit,* μ*,* μ_{max}*, Kc, Ks, Ki and m* which represent TBT concentration (nM), critical TBT concentration (nM), initial growth rate (nM h), maximum growth rate (nM h), inhibition constant (nM), Monod constant (nM), TBT inhibition constant (nM) and empirical constant values, respectively, can be found.

To date aside from these reports, there are almost no other reports on the kinetic model effect of TBT toxicity on the growth rate of microorganisms as most reports were on the effect of heavy metals on the primary models of the growth of microorganisms and not on secondary models [18,19]. The aim of this work is to model the effect of tributyltin on the growth rate of *Bacillus stearothermophilus* through the use of several inhibition models. `

MATERIALS AND METHODS

Data source

Data from **Table 1 of Martins et al** was processed using the software Webplotdigitizer 2.5 [22] which digitizes the scanned figure and has been utilized by many researchers and acknowledged for its reliability [17,23–25].

Effect of TBT on growth rate of *B. steearothermophilus*

The models utilized in this study is as follows;

Models	Equation	Ref
Modified Han-levenspiel	$r = u_{max}\left(1 - \frac{C}{C_{crit}}\right)^m$	[12]
Wang	$r = \frac{u_{max}}{1 + \left(\frac{C}{K_C}\right)^m}$	[13]
Liu	$r = \frac{u_{max}K_C}{K_C + C}$	[26]
Modified Andrews	$r = \frac{u_{max}C}{K_s + C + \left(\frac{C^2}{K_i}\right)}$	[14]
Shukor	$r = v_{max}\left(1 - \left(\frac{C}{S_m}\right)^n\right)$	[21]

Fitting of the data

The nonlinear equations were fitted with a Marquardt algorithm using CurveExpert Professional software (Version 1.6). The algorithm searches the best method that minimizes the sum of the squares between predicted and measured values. The software calculates the starting values automatically via the steepest ascent method.

Statistical analysis

To choose the best model, numerous statistical methods including the corrected AICc (Akaike Information Criterion), Root-Mean-Square Error (RMSE), Bias Factor (BF), Accuracy Factor (AF), and Adjusted Coefficient of Determination (R^2) was utilized as before [17,23,25].

RESULTS AND DISCUSSION

The growth rates at various concentrations of Tributyltin was then modelled using the available inhibition models. Out of the five models, only Wang, modified Han-Levenspiel and the Liu models were able to fit the curve, whilst the modified Andrews and Amor models were unable to fit the curves **(Figs. 3 to 5).** Both the Wang and modified Han-Levenspiel models show acceptable fitting while the Liu model shows poor fitting. Results of the statistical analysis showed that the Wang model was the best model based on the lowest values for RMSE and AICc, highest Adjusted Correlation Coefficient (AdjR^2) and values of AF and BF closest to unity **(Table 1).**

Table 1. Error function analysis of the effect of increasing concentrations of tributyltin to the maximum specific growth rate of *Bacillus stearothermophilus* as fitted to various secondary models.

Model	*P*	RMSE	R^2	$AdjR^2$	AF	BF	AICc
Wang	3	0.13	0.99	0.97	1.06	0.98	1.97
Levenspiel	3	0.20	0.97	0.93	1.08	1.01	7.79
Liu	2	0.42	0.73	0.59	1.12	1.00	3.38
Andrews	4	1.40	-0.07	-2.22	3.18	0.33	76.81
Shukor	3	0.25	0.95	0.90	1.11	1.03	10.91

Note:
p no of parameter
AdjR^2 Adjusted Correlation Coefficient
RMSE Root Mean Square Error
AF Accuracy Factor
BF Bias Factor
AICc corrected Akaike Information Criteria

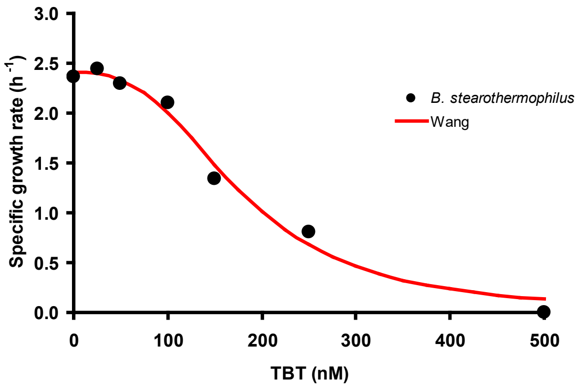


Fig. 1. The effect of increasing concentrations of tributyltin to the specific growth rate of *Bacillus stearothermophilus* as fitted to the Wang model.

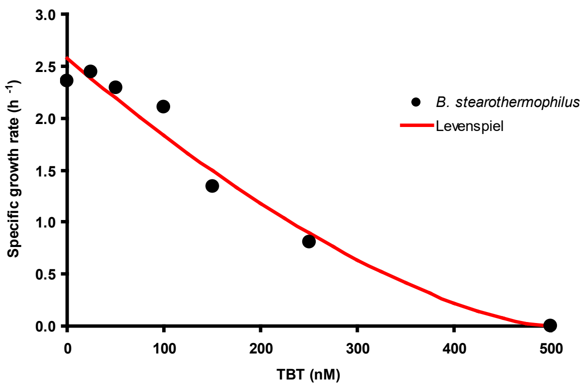


Fig. 2. The effect of increasing concentrations of tributyltin to the specific growth rate of *Bacillus stearothermophilus* as fitted to the Levenspiel model.

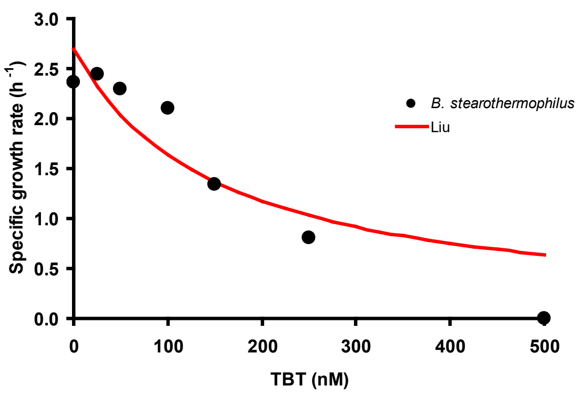


Fig. 3. The effect of increasing concentrations of tributyltin to the specific growth rate of *Bacillus stearothermophilus* as fitted to the Liu model.

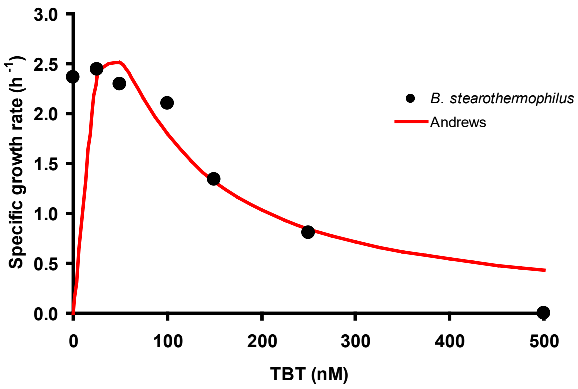


Fig. 4. The effect of increasing concentrations of tributyltin to the specific growth rate of *Bacillus stearothermophilus* as fitted to the Andrews model.

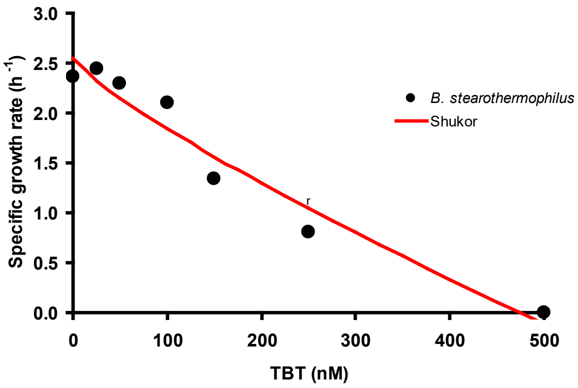


Fig. 4. The effect of increasing concentrations of tributyltin to the specific growth rate of *Bacillus stearothermophilus* as fitted to the Shukor model.

The parameters obtained from the Wang model, which are C_{crit}, μ_{max} and m which represent critical TBT concentration (nM), maximum growth rate (nM) and empirical constant values were 177.99 (95%, confidence interval 144.26 to 211.71), 2.41 (95%, confidence interval of 2.17 to 2.64) and 2.76 (95%, confidence interval of 1.44 to 4.07) respectively. The Wang model allows for the prediction of the critical TBT concentration which can completely inhibited *Bacillus stearothermophilus* growth.

The use of TBT inhibition models is poorly represented in the literature despite the importance of such study in light of the fact that TBT are ubiquitously present in polluted waters alongside other organic pollutants. A few studies have explored on the effect of TBT toxicity on the growth rate of bacteria [6,18]. For example, the biodegradation rate of monoaromatic hydrocarbons by *Bacillus sp.* and *Pseudomonas sp.* was inhibited strongly by zinc and nickel and the effect of these heavy metals on the degradation rate was successfully modelled using the Andrews model [15]. Heavy metals bind to important functional groups of enzymes such as the sulfhydryl group that are often found at the active sites of enzymes and this is probably the mechanism of inhibition [27].

In conclusion, the use of inhibition models to model the effect of toxicity on the growth rate of bacteria is very rare and largely ignored despite the importance of such study. In this study the effect of TBT toxicity on the growth of *B. stearothermophilus* bacterium was modelled according to several inhibition models, with the Wang model discovered as the best model. The Wang model allows for the prediction of the critical TBT concentration which can completely inhibited the bacterial growth. It is expected that in the presence of TBT, the growth rate on toxic substance will be even strongly affected as the bacteria have to resist the toxicity of both kind of toxicants at the same time. The results from this study can be very important in field trial works where TBT bioremediation is required.

REFERENCES

1. Abubakar A, Ferdaus B MY, Mustafa MB, Johari LWW, Zulkifli SZ, Ismail A, et al. Klebsiella sp. FIRD 2, a TBT-resistant bacterium isolated from contaminated surface sediment along Strait of Johor Malaysia. Mar Pollut Bull J [Internet]. 2015 Oct 1 [cited 2015 Dec 12];101(1):280–3. Available from: http://www.science-direct.com/science/article/pii/S0025326X15300576
2. Sudaryanto A, Takahashi S, Monirith I, Ismail A, Muchtar M, Zheng J, et al. Asia-Pacific mussel watch: Monitoring of butyltin contamination in coastal waters of Asian developing countries. Environ Toxicol Chem [Internet]. 2002 Oct [cited 2015 Jun 8];21(10):2119–30. Available from: http://doi.wiley-.com/10.1002/etc.5620211015
3. Cruz A, Caetano T, Suzuki S, Mendo S. Aeromonas veronii, a tributyltin (TBT)-degrading bacterium isolated from an estuarine environment, Ria de Aveiro in Portugal. Mar Environ Res [Internet]. 2007 Dec [cited 2013 Oct 2];64(5):639–50. Available from: http://www.ncbi.nlm.nih.gov/pubmed/17719084
4. Harino H, Ohji M, Wattayakorn G, Adulyanukosol K, Arai T, Miyazaki N. Concentrations of organotin compounds in tissues and organs of dugongs from Thai coastal waters. Arch Environ Contam Toxicol. 2007;53(3):495–502.
5. Radke B, Leczyński L, Wasik A, Namieśnik J, Bolalek J. The content of butyl- and phenyltin derivatives in the sediment from the Port of Gdansk. Chemosphere [Internet]. 2008 Sep [cited 2013 Nov 27];73(3):407–14. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18606436
6. Abubakar A, Muhammad A, Shehu D, Murtala Y, Babandi A, Salisu AI, Hadiza I, Sunusi U, Mohamat-yusuff F IS. Growth Kinetics Modelling of Klebsiella spp. FIRD 2 on TBT-Resistant Containing Zinc. Bayero J Pure Appl Sci. 2017;2(1):268–76.
7. Stewart C, de Mora SJ. A review of the degradation of tri(n -butyl)tin in the marine environment. Environ Technol [Internet]. 1990 Jun 17 [cited 2015 May 8];11(6):565–70. Available from: http://www.tandfonline.com/doi/abs/10.1080/09593333900938489 7#.VYZ4bROqqko
8. Fent K. Organotin compounds in municipal wastewater and sewage sludge: contamination, fate in treatment process and ecotoxicological consequences. Sci Total Environ [Internet]. 1996 Jun [cited 2015 Jun 8];185(1–3):151–9. Available from: http://www.sciencedirect.com/science/article/pii/00489697950504 85
9. Abubakar A, Mustafa MB, Johari WLW, Zulkifli SZ, Yusuff FBM. Tributyltin (TBT) Tolerance of Indigenous and Non-indigenous Bacterial Species. Water Air Soil Pollut. 2016;227(8).
10. Martins J, Jurado A, Moreno A, Madeira V. Comparative study of tributyltin toxicity on two bacteria of the genus Bacillus. Toxicol Vitr [Internet]. 2005 Oct [cited 2013 Oct 2];19(7):943–9. Available from: http://d.wanfangdata.com.cn/NSTLQK_NSTL_QK10481800.aspx
11. Martins JD, Jurado AS, Moreno AJM, Madeira VMC. Comparative study of tributyltin toxicity on two bacteria of the genus Bacillus. Toxicol Vitr. 2005;19(7):943–9.
12. Wang J, Wan W. Kinetic models for fermentative hydrogen production: A review. Vol. 34, Int J Hydrogen Energy. 2009. p. 3313–23.
13. Wang Y, Zhao QB, Mu Y, Yu HQ, Harada H, Li YY. Biohydrogen production with mixed anaerobic cultures in the presence of high-concentration acetate. Int J Hydrogen Energy. 2008 Feb;33(4):1164–71.
14. Andrews JF. A mathematical model for the continuous culture of microorganisms utilizing inhibitory substrates. Biotechnol Bioeng. 1968;10:707–23.
15. Amor L, Kennes C, Veiga MC. Kinetics of inhibition in the biodegradation of monoaromatic hydrocarbons in presence of heavy metals. Bioresour Technol. 2001;78:181–5.

16. Gopishetty SR, Louie TM, Yu CL, Subramanian M V. Microbial degradation of caffeine, methylxanthines, and its biotechnological applications. In Thatoi HN, Mishra BB (ed), Microbial biotechnology methods and applications. 2011. 44–67 p.
17. Ibrahim S, Mansur A, Ahmad SA. Mathematical Modelling of the Growth of Caulobacter crescentus on Caffeine. J Environ Microbiol Toxicol. 2018;6(2):13–7.
18. Abdussamad A, Abdullahi MD, Shehu; Murtala, Ya'u; Abba, Babandi; Abubakar ST, Hadiza, Ibrahim; Usman, Sunusi; Ferdaus M-YSI. Modelling Growth Kinetics of Klebsiella sp . FIRD 2 on TBT-Resistant Containing Lead. J Appl Sci Environ Manag [Internet]. 2017;Vol. 21 (5. Available from: www.bioline.org.br/ja%0AModelling
19. Halmi MIE, Abdullah SRS, Johari WLW, Ali MSM, Shaharuddin NA, Khalid A, et al. Modelling the kinetics of hexavalent molybdenum (Mo 6+) reduction by the Serratia sp. strain MIE2 in batch culture. Rend Lincei. 2016;27(4):653–63.
20. Zahaba M, Izuan M, Halmi E, Othman AR, Shukor MY. Mathematical Modelling of the Growth Curve of Vibrio sp . Isolate MZ Grown in Seawater Medium. Bioremediation Sci Technol Res. 2018;6(1):31–4.
21. Manogaran M, Othman AR, Shukor MY, Halmi M. Modelling the Effect of Heavy Metal on the Growth Rate of an SDS-degrading Pseudomonas sp. strain DRY15 from Antarctic soil. Bioremediation Sci Technol Res. 2019;7:41–5.
22. Rohatgi A. WebPlotDigitizer.' (http://arohatgi.info/WebPlotDigitizer/app/. 2015.
23. Halmi M, Shukor MS, Johari WLW, Shukor MY. Modeling the growth curves of Acinetobacter sp. strain DRY12 grown on diesel. J Environ Bioremediation Toxicol. 2014;2:33–7.
24. Ibrahim S, Abdulrasheed M. Kinetics of caffeine inhibition to the growth of Caulobacter crescentus on caffeine. J Biochem Microbiol Biotechnol. 2019;7(1):43–7.
25. Abubakar A, Usman NI, Ibrahim H, Sunusi U, Mohamat-yusuff F, Ibrahim S. Growth Kinetics Modelling of Tributytin-Resistant Klebsiella SP . FIRD 2 In Cadmium Media. UMYU J Microbiol Res. 2017;2(1):157–65.
26. Liu Y. A simple thermodynamic approach for derivation of a general Monod equation for microbial growth. Biochem Eng J. 2006;31:102–5.
27. Gopinath KP, Kathiravan MN, Srinivasan R, Sankaranarayanan S. Evaluation and elimination of inhibitory effects of salts and heavy metal ions on biodegradation of congo red by Pseudomonas sp. mutant. Bioresour Technol. 2011;102:3687–3693.