

Original article:

Study of oral lactobacillus towards developing a comprehensive structured for integrated exponential regression model

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Abstract:

Introduction: Probiotics are well-defined as live microorganisms that usefully affect the host and probiotic bacteria have been used intensely. For years to target gastrointestinal disease by rebalancing the compound microflora. Besides the gastrointestinal tract also the oral cavity is highly colonized by bacteria and many different bacterial species are part of the microbiota in the mouth, as it offers ideal conditions for bacteria with a stable temperature, moist surface with a relatively stable pH and regular supply of nutrients. Probiotic bacteria like Lactobacillus are a promising treatment strategy for oral disease with a microbiological etiology. To gain better results, many researchers that study and emphasize specific methods been tried to build a new or improved methodology. **Objectives:** The aimed of this study is to improve the performance of exponential growth by adding bootstrap and fuzzy techniques (Integrated exponential regression method). The aim of the research work is to develop a comprehensive framework for an integrated exponential regression model. **Material and Methods:** The data were taken from the present data available from the recently done by a researcher for nurturing selected microorganisms. The gathered data will be used for the exponential modeling and the efficiency of the model will be compared accordingly due to the predicted interval from the exponential regression method and an integrated exponential regression method. This paper also provides the algorithm for the prediction of cell growth and inferences. **Results:** The result shows that the average width for the exponential regression model was 19.2228 while an integrated exponential regression method was 0.0075. The average width of integrated exponential regression was smaller than the exponential regression. This clearly shows that the integrated exponential regression method is more efficient than exponential regression technique. **Conclusion:** This proposed method can be applied to small sample size data, especially when limited data is obtained.

Keywords: Exponential Growth; Probiotics Oral Cavity; Lactobacillus; Bootstrap; Exponential Regression Method; Integrated exponential regression Method; and Fuzzy Regression methodology.

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Introduction

Bacteria in Dental Caries

The prevailing ecological view of the etiology of dental caries is more attuned to the polymicrobial nature of the dental plaque biofilm. However, only a limited number of bacteria are consistently recovered

from caries lesions and have thus been recognized to be specifically associated with dental caries¹. The association between Lactobacilli was the leading candidate in the causation of dental caries prior to the 1950s, when the mutans Streptococci(MS) started to dominate the literature². Despite numerous

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studies linking Lactobacilli to caries in both adults and children, our understanding of their role in this disease remains incomplete. Some of the major gaps in knowledge span the transmission and colonization of the oral cavity, as well as the genetic basis for the adaptation to this niche. Probiotics are well-defined as live microorganisms that usefully affect the host. Probiotic bacteria have been used intensely. For years to target gastrointestinal disease by rebalancing the compound microflora. Besides the gastrointestinal tract also, the oral cavity is highly colonized by bacteria, and many different bacterial species are part of the microbiota in the mouth, as it offers ideal conditions for bacteria with a stable temperature, moist surface with a relatively stable pH and regular supply of nutrients³.

Probiotic bacteria like Lactobacillus are a promising treatment strategy for oral disease with a microbiological etiology. Those include plaque-associated diseases like dental caries, which is an infectious disease with microbial processes eroding and destroying the hard dental tissue or inflammation of periodontal tissue, namely gingivitis and the more severe periodontitis⁴. In the oral cavity, *Lactobacilli* comprise about 1% of the cultivable microbiota. Commonly isolated strains include *L. Casei*, *L. Paracasei*, *L. Lantarum*, *L. Rhamnosus*, *L. Fermentum*, *L. Acidophilus*, and *L. Salivarius*. In contrast to MS, lactobacilli have poor adherence properties and have traditionally been considered as having an opportunistic role in the caries disease⁵. High levels of lactobacilli have been linked with high caries activity and high carbohydrate consumption. Simark-Mattsson and colleagues isolated *lactobacilli* from subjects with different levels of dental caries⁶. They demonstrated that naturally occurring lactobacilli inhibited the growth of MS; the effect of lactobacilli was most pronounced in subjects without dental caries, who were lacking S. Mutans.

Mathematical Theory

Fuzzy Linear Regression

Fuzzy linear regression analysis was first introduced by⁷ in which two factors namely the degree of fitness and the fuzziness of data sets, are considered⁸. In the Fuzzy method, regression plays a crucial role when dealing with imprecise data. A simple regression equation involving a single independent fuzzy variable can be used for such situation. The result from studies⁹, showed that fuzzy regression can be a better replacement for statistical

regression when the data are vague with poor model specification. According¹⁰, the regression problem in form of gradient descent optimization was used to show the effectiveness of an iterative algorithm for multiple regressions with fuzzy data. A fuzzy linear regression model is commonly presented $\tilde{y} = \tilde{f}_t(x) = \tilde{A}_0 + \tilde{A}_1 x_1 = \tilde{A}_x$ where $x = [1, x_1]^T$ is a crisp vector of independent variables and \tilde{y} is the estimated fuzzy output. $\tilde{A} = [\tilde{A}_0, \tilde{A}_1]$ is a vector of fuzzy parameters of the fuzzy linear regression model. \tilde{A}_j is presented in the form of symmetric triangular fuzzy numbers denoted by $\tilde{A}_j = (\hat{a}_j, c_j)$, $j = 0, 1, 2, \dots, N$ where its membership function is shown as $\mu_{\tilde{A}_j}(a) = \begin{cases} 1 - \frac{a - \hat{a}_j}{c_j} & \hat{a}_j - c_j \leq a \leq \hat{a}_j + c_j \\ 0 & \text{otherwise} \end{cases}$, where \hat{a}_j is the central value of the fuzzy number and c_j is the spread. Therefore the fuzzy linear regression model can be rewritten as shown $\tilde{y} = (a_0, c_0) + (a_1, c_1)x_1$. However, the interaction between variables and higher-order terms are not included in the fuzzy linear regression defined in¹. In fact, the interaction between variables and higher order terms often exist in physical systems. A simple procedure is commonly used to solve the linear programming problem¹¹. Linear regression is used to study the linear relationship between a dependent variable Y and one or more independent variables X. The dependent variable Y must be continuous, while the independent variables may be either continuous, binary, or categorical. The initial judgment of a possible relationship between two continuous variables should always be made on the basis of a scatter plot¹².

Bootstrap Proposed by Efron (1979), the bootstrap is a simulation technique to evaluate the standard error of the estimation of a parameter¹³. The idea is to perform various resampling with the replacement of the dataset. Let $\hat{\theta} = t(x)$ the estimator θ calculated from a sample (x_1, \dots, x_n) . A sample bootstrap $x^*(i) = (x_1^*, \dots, x_n^*)$ is a resample with the replacement of size from (x_1, \dots, x_n) , the index $i = 1, \dots, B$ refer to the number of wanted replicas B . This way the bootstrap estimator of the variance of $\hat{\theta}$ is given by $\text{var}(\hat{\theta}) = \frac{1}{B-1} \sum_{i=1}^B (t(x^*(i)) - \hat{\theta})^2$ where $t(x^*(i))$ is the estimator θ based on the i -th replic bootstrap and $\hat{\theta}^* = B^{-1} \sum_{i=1}^B t(x^*(i))$. For a sufficiently large number of replicas, we can calculate a confidence interval (with significance level 2α) for $\hat{\theta}$ through normal approximation, which is given by $C_B[\hat{\theta}, (1-2\alpha)] = [\hat{\theta}^* - z_\alpha [\text{var}(\hat{\theta})]^{1/2}; \hat{\theta}^* + z_\alpha [\text{var}(\hat{\theta})]^{1/2}]$ where z_α corresponds to the α -the quantile of standard normal distribution. We can estimate the empiric density \hat{F} as a normal approximation of real F.

As such, we can estimate the confidence interval using \hat{F} quantiles, so the interval is given by $C_B[\hat{\theta}, (1 - 2\alpha)] = [\hat{F}(\alpha), \hat{F}(1 - \alpha)]$.

Material and Methods

Data Collection

The data composed of two variables which are the days of the culture (x), bacterial growth (y), and the rate of the exponential growth of bacterial culture (lny). The data were taken from the present data available from the recently done by a researcher for nurturing selected microorganisms (Lactobacillus) in School of Dental Sciences, Universiti Sains Malaysia. Then, the data were recorded in a SAS Program to evaluate microorganism growth before analyzing by using the new proposed statistical methodology. The selected microorganisms were kept in storage accordingly (Table 1 and Figure 1). And so, all microorganisms were disposed of carefully in the clinical waste bin.

Table 1. Description of variables

x	Days of the culture
y	Lactobacillus Salivarius (5×10^5)
lny	The rate of exponential growth

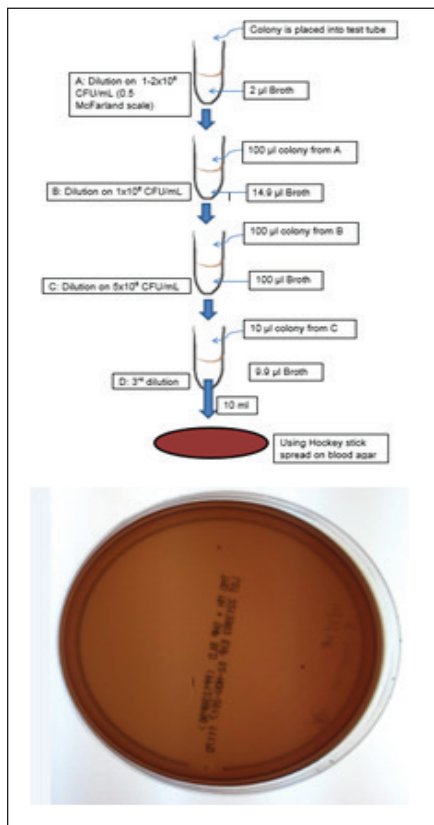


Figure 1. Step for Broth Microdilution Method and blood agar of Lactobacillus Salivarius(5×10^5)

Statistical Analysis

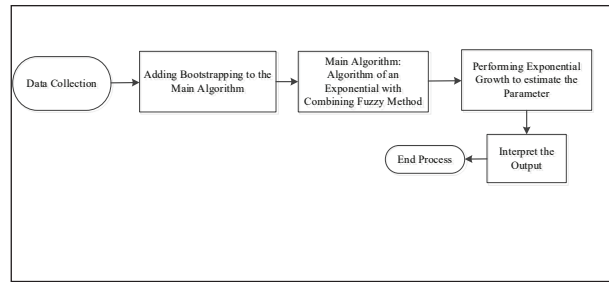


Figure 2: Flowchart of the Integrated Exponential Calculation

Calculations of an Exponential Bacteria growth using SAS Algorithm

`/*ADDING BOOTSTRAPPING ALGORITHM TO THE METHOD */`

`%MACRO bootstrap(data=_last_, booted=booted, boots=100, seed=1234);`

`DATA &booted;`

`pickobs = INT(RANUNI(&seed)*n)+1;`

`SET &data POINT = pickobs NOBS = n;`

`REPLICATE=int(i/n)+1;`

`i+1;`

`IF i> n*&boots THEN STOP;`

`RUN;`

`%MEND bootstrap;`

`/*DATA ENTRY FOR EXPONENTIAL REGRESSION*/`

`Data Lactobacillus;`

`input x y lny;`

`datalines;`

1 23 3.14

2 23 3.14

3 40 3.69

5 60 4.09

9 80 4.38

.

.

.

47 30867 10.34

50 45420 10.72

53 59092 11.00

55 65095 11.10

57 89998 11.41

59 127285 11.80

60 159880 11.98;

`run;`

`/*GENERATE OUTPUT IN MICROSOFT WORD*/`

`ods rtf file='abc.rtf' style=journal;`

`/**GENERATE BOOTSTRAP SAMPLE**/`

`%bootstrap(data= Lactobacillus,boots=100);`

```
run;
/**BOOTSTRAP SAMPLE**/
procprint data=booted;
run;
/*FITTING EXPONENTIAL WITH BOOTSTRAP*/
title "Corrected sum of squares";
procsql;
select css(y) into :CSSy
from booted;
quit;
title "Exponential fit with boot";
ods graphics / imagename="ExponentialFit";
procnlin data=booted plots=fit;
parameters A=1 b=0;
model y = A*exp(b*x);
ods output EstSummary=summExp;
run;
/*FITTING REGRESSION BOOTSTRAPPING DATA*/
Procreg data=booted;
model lny=x;
run;
/*Fuzzy Regression Methodology Building */
```

```
procoptmodel;
set j= 1..2500;
number lny{j}, x{j};
read data booted into [_n_] lny x;
/*Print x */
print lny x;
number n init2500; /* Total number of Observations*/
/*Decision Variables*/
/*Theses two variables are bounded*/
/*These two variables are not bounded*/
var aw{1..2}>=0;
var ac{1..2};
/* Objective function*/
min z1= aw[1] * n + sum{i in j} x[i] * aw[2];
/*Linear Constraints*/
con c{i in 1..n}:
ac[1] +x[i] *ac[2] -aw[1] -x[i] *aw[2] <= lny[i];
con c1{i in 1..n}: ac[1] +x[i] *ac[2] +aw[1]+x[i]
*aw[2] >= lny[i];
/*This provides all equations */
expand;
solve;
print ac aw;
quit;
ods rtf close;
run;
```

Exponential Bacteria Growth Transforming to Linear Form

Exponential growth formula and exponential decay formula are given by $Y=Ae^{bx}$ and $Y=Ae^{-box}$ and exponential bacteria growth after transform to linear is $h Y = \ln(Ae^{bx}) = \ln(A) + \ln(e^{bx}) = \ln(A) + bx$.

Ethical Approval

This study was approved for ethical permission by the Human Research Ethics Committee of Universiti Sains Malaysia (USM/JEPeM/18080391).

Result

The result shows that the graph of *Lactobacillus Salivarius*.

(A) Exponential Regression Model

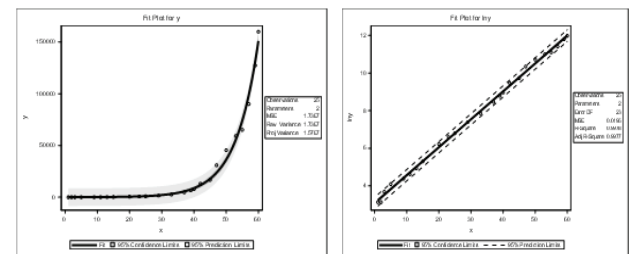


Figure 3: Graph *Lactobacillus Salivarius* for Exponential regression and linear regression

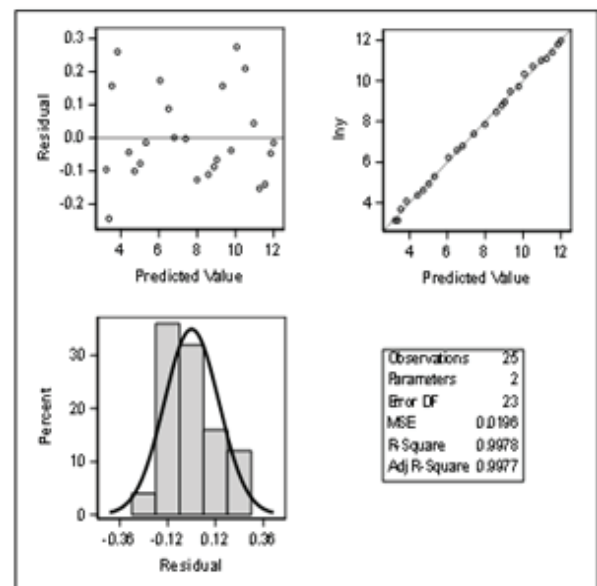


Figure 4: Diagnostic graph for *Lactobacillus Salivarius*

(B) Integrated Exponential Regression Model (Combining Bootstrap and Fuzzy Method)

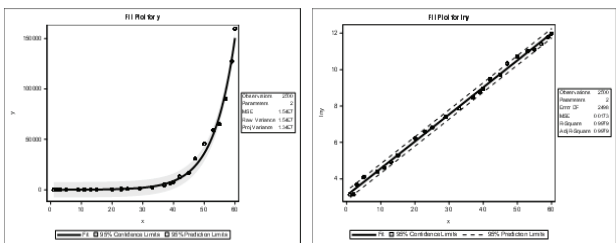


Figure 5: Graph *Lactobacillus Salivarius* for Exponential regression and linear regression

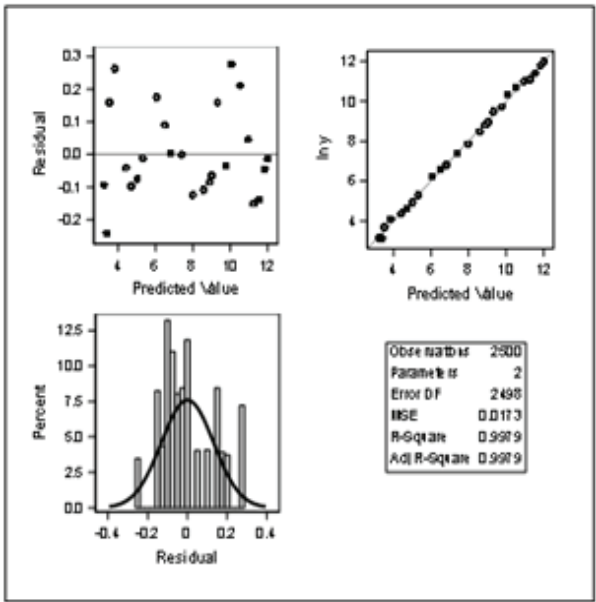


Figure 6: Diagnostic graph for *Lactobacillus Salivarius*

Table 2: Results of Exponential Regression Model and Integrated Exponential Regression

Model

Model/Explanation	Exponent Regression Model	Model/Explanation	Structure of Model KREB
Model n=25	Exponent $Y=29.2490e^{0.1425x}$ Transform to Linear $\ln y=3.3758+0.1425x$	Model with bootstrap Replicate = 100 n= 2500	Exponent = $Y=29.0454e^{0.1426x}$ Transform to linear= $\ln y=3.3689+0.1426x$
R ²	0.9978	R ²	0.9979
Standard Error	A= 29.2490 ± 9.4391 b = 0.1425 ± 0.00557	Standard Error	A= 29.0454 ± 0.8967 b = 0.1426 ± 0.0005
Upper limit interval	$\ln y=12.8149+0.14807x$	Upper limit interval	$\ln y=4.2656+0.1431x$

Model/Explanation	Exponent Regression Model	Model/Explanation	Structure of Model KREB
Lower Limit Interval	$\ln y=-6.0633+0.13693x$	Lower Limit Interval	$\ln y=2.4722+0.1421x$
Average Width	19.2228	Average Width	1.8241
-	-	Fuzzy Regression model	$Y= (3.3763, 0.00375)+ (0.1425, 0.0000)x$
-	-	Standard error	$\pm (3.3763,0.00375) \pm (0.1425,0.0000)$
-	-	Upper limit interval	$Y= 3.3801+ 0.1425x$
-	-	Lower limit interval	$Y= 3.3726+ 0.1425x$
-	-	Average Width	0.0075

Table 2 above shows that the results of exponential regression model and integrated exponential regression model. Therefore, the results show that the average width for the exponential regression model was 19.2228 while an integrated exponential regression method was 0.0075. The average width of integrated exponential regression was smaller than the exponential regression. According to the R-Square results obtained, the model is good and appropriate. This is because the value close to 1 is the best fit. For the nonlinear function (exponential) used here, the growth data was fitted to determine mathematical parameters with curve estimation. The model fitted by nonlinear regression gave reliable estimates of the correlation coefficient (R² values at p<0.01) for the functions used. They also assess the degree of fit of the model to each of the experimental data sets, which fell close to 1.

Summary And Discussion

From the result obtained, the integrated exponential regression method is more efficient than the exponential regression technique. This proposed method can be applied to small sample size data especially when limited data is obtained. We showed that the model can be used to find the potential biological parameters which may be able to predict the treatment outcome. We have demonstrated that the use of an exponential regression model along with the use of SAS software that can be a very

precise and useful tool to study the exponential growth of *Lactobacillus Salivarius*. Paperwork gives the explanation of exponential growth by adding improvement method which adding bootstrap and the fuzzy technique SAS software.

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Competing, financial interests:

The author declares no competing financial interests.

Conflict of Interest: The author declares that he has

no conflict of interest.

Author's contribution:

Data gathering and idea owner of this study: RAAR, WMAWA, NHI, MAYaqoob and FMMG.

Study design: RAAR, WMAWA, NHI, MAYaqoob

Data gathering: RAAR, WMAWA, NHI, MAYaqoob

Writing and submitting manuscript: RAAR, WMAWA, NHI, MAYaqoob, MKA and FMMG.

Editing and approval of final draft: RAAR, WMAWA, NHI, MAYaqoob, MKA and FMMG.

References:

1. Marsh PD, Moter A, Devine DA. Dental plaque biofilms: communities, conflict and control. *Periodontal* 2011; **55**(1):16-35
2. Badet C, Thebaud NB. Ecology of *Lactobacilli* in the oral cavity: a review of Literature. *Open Microbiol J* 2008; **2**:38-48.
3. Moukonen J, Matto J, Suihko ML, Saarela M. Intra-individual diversity and similarity of salivary and faecal microbiota. *J Med Microbiol* 2008; **57**:1560-1568.
4. Hoffmann A, Daniels R. *Lactobacilli* for the treatment of oral disease. *J Prob Health* 2017; **5**:181.
5. Bickel, P.J., and Freedman, D.A. Asymptotic normality and the bootstrap in stratified sampling. *Ann Stat.* 2002; **12**:470-482.
6. Simark-mattson C, Emilson CG, Hakansson EG, Jacobsson C, ROOS Kand HOLM S. Lactobacillus mediated interference of mutans streptococci in caries free vs caries active subjects. *Eur J Oral Sci.* 2007; **115**: 308-14.
7. Taheri S., and Mahmoud. Trends in Fuzzy Statistics. *Austrian Journal of Statistics*, 2003; **32**(3), 237-257.
8. Mcalinden, C., Khadka, J., and Pesudovs, K. Statistical methods for conducting agreement (comparison of clinical tests) and precision (repeatability or reproducibility) studies in optometry and ophthalmology. *Ophthalmic Physiol Opt.* 2011; **31**: 330-338.
9. Casella, G. Introduction to the Silver Anniversary of the Bootstrap. *Statistical Science*, 2003; **18**:133-134
10. Bargiela A, Pedrycz W. *Granular Computing*, Kluwer Academic Publisher, Dordrecht, 2002.
11. Kreft JU, Booth G, Wimpenny JWT. (1998). Racism a simulator for individual-based modeling of bacterial colony growth. *Microbiology*. Vol. **144**, pp:3575-3287.
12. Koo TK, Li MY. (2016). A Guideline of Selecting and Reporting Intraclass Correlation Coefficients for Reliability Research. *Journal of Chiropractic Medicine*. Vol. **15** (2), pp: 155-63.