NONLINEAR PARAMETER ESTIMATION IN CONTINUOUS TOWER FERMENTATION SYSTEMS

by

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B. Sc., Banaras Hindu University, 1969

A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Chemical Engineering

KANSAS STATE UNIVERSITY

Manhattan, Kansas

1971

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Chapter 1

INTRODUCTION*

Recently, several researchers investigated a tower fermentor for conducting growth processes [1, 2, 3, and 4].** Prokop et al. [1] investigated an eight stage tower fermentor with co-current flow of air and medium from the bottom to the top of the column. Their work consisted of an experimental study of the residence time distribution of liquid media and cells as well as a steady state investigation of the substrate and cell concentrations throughout the fermentor. They found that the residence time characteristics of the continuous phase and the dispersed (microorganisms) phase are distinct. Falch and Gaden [2] studied continuous culture in a 4 stage tower fermentor, in which air and medium were introduced into the column counter-currently. They also developed a method for analyzing the reactor behavior of a continuous multistage tower fermentor and set up a model consisting of a system of interconnected ideal subreactors on the basis of the fermentor's configuration and flow pattern [5]. A least-squares fitting procedure was used to identify the proper model. The effects of the plate and agitator design on backflow, oxygen transfer rate and gas holdup have been reported by Kitai et al. [3].

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*A portion of this chapter has been taken from reference (9).

**Equations, figures, tables, appendices, and references cited will all be found within the chapter in which they are cited except where specific references is made to another chapter.

High productivity, reduced operating cost and flexibility of operation and control are the major advantages of a tower type fermentor. The important factors influencing the operating performance in a tower type fermentor are feed geometry, system dilution rate, backflow, and sedimentation. The degree of mixing between stages is influenced by the backflow rate while the sedimentation greatly influences the cell concentration in each stage. The feed geometry allows for a wide range of growth rates and cell physiological states.

The tower system considered here is assumed to be constructed such that compartments are separated by perforated plates. Complete mixing is assumed in each stage. The oxygen for the biological growth is supplied from air bubbles which are introduced at the bottom. Oxygen transfer and mass transfer rates which depend upon the distribution of bubbles, bubble size and air velocity have been studied by other researchers [3, 6, 7 and 8].

Continuous processes can be classified into three different flow models with respect to their macroscopic degree of mixing; (1) plug flow, (2) complete mixing and (3) partial mixing. The flow behavior corresponding to each of these three different flow models can be approached by properly designing and operating the tower system. Plug flow is achieved if the number of stages is large and fluid backflow is prevented. Complete mixing is achieved when fluid backflow is increased. Partial mixing is the most general case found in tower systems, as it is the intermediate case between the two extreme cases.

Lee [9] has discussed the modelling of microbial growth, analysis of steady state performance and the stability of growth processes in tower

systems, and the optimization of biological waste treatment processes using the tower system.

A tower type biological system has certain advantages over the conventional stirred tank system. Redispersion and circulation of air bubbles makes the individual stage almost equivalent to a completely mixed tank. Appearance of backflow and sedimentation can be realized in the tower system. The principle disadvantages of the tower fermentor are its relative instability and foaming. Perhaps the greatest potential value to the tower fermentor is its range of operation and the possibility of varying the substrate, cell and fluid age distributions relatively independent of each other. Besides providing a potentially useful piece of equipment for optimizing certain fermentation systems, a tower type system may require less space and land.

This work is concerned with the handling of batch and continuous tower system data. Data analysis is often useful in understanding process mechanisms and predicting process behavior for the design of continuous tower fermentation processes. An analysis was conducted of nonlinear parameter estimation for both algebraic and differential models based on experimental and simulated data of known error distributions. A brief description of the various nonlinear parameter estimation techniques can be found in reference [10].

Sequential simplex pattern search [11] which minimizes or maximizes a selected criterion function was used for estimating parameters in a four stage tower system using unsteady state simulated data and a differential model. Various types of experimental errors including normally

distributed error, instrument drift, zero error in the instrument and various combinations of these were taken into account for simulating the data. Flow nonidealities like fluid backflow and dispersed (microorganism) phase sedimentation were included in the mathematical model.

One of the problems often encountered in experimental procedures using a tower type of system is the measurement of back flow rate from each stage. Hence once again parameter identification techniques were employed to estimate the backflow parameters and to observe the relationship between the flows from individual stages. A linear flow model was used in this study.

The problem of curve fitting, where one tries to fit the available experimental data to a certain curve is often encountered in practice. If the experimental data can be approximated by a straight line, the problem is simple, but on the other hand if the data shows the trend of a curve, which is the case in most practical situations, the problem is slightly complicated. The obvious thing to do next, is to try a certain degree polynomial to fit such data. If one fails to find a suitable polynomial to approximate the data, an attempt is made to include the cross-terms, reciprocal terms and logarithmic terms etc. There is no unique statistical procedure which will tell us the degree of the polynomial to use or when the addition of cross-terms, reciprocal terms or logarithmic terms will improve the model for predictive purposes. Personal judgement will always be a necessary part in the analysis. A number of methods have been proposed to discriminate between models. Examination and analysis of residuals is often useful in selecting the

best regression equation. A brief description of this appears in reference [12]. The other procedures for selecting the best regression equation are also discussed in reference [12], and can be listed as follows

(1) all possible regressions

(2) backward elimination

(3) forward selection

(4) stepwise regression

(5) stagewise regression.

A similiar problem was encountered in the analysis of tower fermentor performance using a flow model. The object was to estimate the backflow parameters from the available tracer data in a multistage tower system. The problem was formulated in such a way that it was necessary to approximate the tracer data in one of the individual stages with a curve. Fortunately it was possible to fit the available data by a polynomial. Personal experience combined with statistical techniques formed the basis of judgement to find the exact degree of polynomial. In particular, graphical techniques were used to examine and analyze the residuals obtained from various regression models, and an attempt was made to suggest the most adequate and satisfactory regression model for the available experimental data.

REFERENCES

1. Prokop, A., L. E. Erickson, J. Fernendez, and A. E. Humphrey, Biotechnol. Bioeng., 11, 945 (1969).

2. Falch, E. A., and E. L. Gaden, Biotechnol. Bioeng., 11, 927 (1969).

3. Kitai, A., S. Goto and A. J. Ozaki, Fermentation Technol., 47, 340, 348, 356 (1969).

4. Kitai, A., H. Tone, and A. Ozaki, Biotechnol. Bioeng., 11, 911 (1969).

5. Falch, E. A., and E. L. Gaden, Biotechnol. Bioeng., 12, 465-482 (1970).

6. Tsao, T. N., Biotechnol. Bioeng., 11, 1071 (1969).

7. Zieminski, S. A. and R. L. Hill, J. of Chem. and Eng. Data, 7, 51 (1962).

8. Flynn, D. S., and M. D. Lilly, Biotechnol. Bioeng., 9, 515 (1967).

9. Lee, S. S., M. S. Thesis, Kansas State University (1970).

10. Stamets, L. Ph.D. Thesis, Kansas State University (1969).

11. Nelder, J. A., and R. Mead, Computer J., 7, 308 (1965).

12. Draper, N. R., and H. Smith, "Applied Regression Analysis," John Wiley and Sons, Inc. (1968).

Chapter 2

ANALYSIS OF UNSTEADY STATE, CONTINUOUS MULTISTAGE TOWER FERMENTOR PERFORMANCE

2.1 INTRODUCTION

It is generally agreed that continuous processing offers a number of advantages over batch operation. These include higher productivity through reduced "downtime," elimination of some labor requirements, and the opportunity for improved process control [1]. Continuous processing has found few applications in the microbiological process industries. The most important factor in limiting the use of continuous processing is the fact that fermentation plants have small capacities. Furthermore, flexibility has been a major concern in the design of fermentation plants in the past and this consideration certainly favors batch operation [2].

Falch and Gaden [2] have discussed the design of a multistage tower fermentor. They have also discussed a reactor system for continuous microbial processes, in which they say that the behavior of a continuous reactor system ranges between the "ideal" extremes of plug flow and complete backmixing. The backmixed, continuous stirred tank reactor (CSTR) has found considerable application in the development of fermentation technology. For many of the industrially important fermentations, however, the plug flow or a combined reactor system has been shown to be optimal [3]. Most of the experimental continuous cultivation studies have been carried out in highly backmixed (CSTR) systems. The plug flow reactor has not been extensively studied for continuous fermentation. The practical

design of continuous fermentors with plug flow characteristics involves considerable problems because of the difficulty of providing adequate aeration while maintaining plug flow conditions [2]. Falch and Gaden [2] have also suggested that plug flow behavior can be approximated by a reaction system comprised of a large number of stirred tanks connected in series. By varying the number of stages it is also possible to obtain reaction systems with characteristics intermediate between the two idealized continuous models. The number of stages may then be optimized [4].

In this chapter a four stage tower fermentor has been investigated. The mathematical model considered is relatively simple and takes into account the sedimentation of cells besides the other factors. Medium is fed to the second stage. The parameters of the model have been estimated using simulated data and a simplex-pattern search technique [5]. The same program can be used for estimating parameters for any unsteady state experimental data available on a continuous tower fermentor. In this work no attempt has been made to discuss the continuous tower fermentor system in general, the major objective being to demonstrate the applicability of simplex pattern search techniques [5] in estimating parameters in a multistage tower system.

2.2 PARAMETER IDENTIFICATION IN TOWER SYSTEMS

Mathematical models are the basic symbolic forms which describe the relationships between physically measured variables. Mathematical models are often necessary to quantitatively describe the mechanism or predict the behavior of a chemical or biochemical system or process. These are mathematical equations, algebraic or differential in nature which attempt

to describe as far as possible the physical system. These mathematical equations contain dependent variables, independent variables and constants. These constants are known as parameters, and are generally determined experimentally by employing parameter estimation techniques. Since experimental results include some experimental error, the values of the parameters selected to be used in the mathematical model are called the estimates of the parameters. It is the objective of all parameter estimation techniques to find the most accurate estimate of parameters. Consider the following mathematical model

$$Y' = f(\xi_1, \xi_2, \ldots, \xi_M; P_1, P_2, \ldots, P_p) + E \tag{1}$$

where

Y' = measured dependent variable

ξ_i = measured independent variable, $i = 1, \ldots M$

P_j = parameter of the model, $j = 1, \ldots, p$

E = experimental error

Equation (1) is said to be linear (in parameters) if it is of the form [6].

$$Y' = P_o + P_1 Z_1 + \ldots + P_p Z_p + E \tag{2}$$

where

Z_L = function of ξ_i, $L = 1, 2, \ldots, p$

Any parameter estimation problem which cannot be converted into this form is considered to be nonlinear [6]. Parameter estimation in a linear

system is simple and can be found in many standard treatments of regression analysis. In this chapter a non-linear differential model has been considered. The effect of experimental error and number of parameters is considered for the non-linear model, using simulated data.

A simplex pattern search technique [5] which minimizes or maximizes a selected criterion function has been used for estimating parameters in the tower system. A brief description of this technique is included in Appendix II.

2.3 KINETIC MODEL

In 1942 Monod [7] presented a model for the kinetics of biological growth. The equation proposed by Monod [7] to describe the relationship between growth rate and concentration of limiting nutrient has the same form as the Michaelis-Menten equation, which describes the kinetics of enzymatic reactions. Although this is a gross oversimplification of the complex phenomena that occur, considerable effort will be required before a model which accounts for the various complex phenomena occuring in growth processes can be developed. Hence Monod's [7] model is used in this study.

The growth of microorganisms will be expressed in terms of a single growth rate equation which is at all times a function of the concentrations of the growth limiting substrate (nutrients) and organisms. If oxygen and trace nutrients are available in sufficient quantities, the growth rate of the microorganisms can be expressed as follows:

$$\frac{dX}{dt} = \mu X \qquad (3)$$

where μ is the specific growth rate and X is the concentration of the bacterial cells. In 1942 Monod [7] showed that the value of μ is not constant, but depends on the concentration of growth limiting substrate, S, according to the equation

$$\mu = \frac{\mu_{max} S}{(K_S+S)} \tag{4}$$

where

μ_{max} = maximum specific growth rate, hr^{-1}

K_S = saturation constant, concentration of organics at which the specific growth rate is one half the maximum value, gms/lit.

Substituting Equation (4) in Equation (3) gives

$$\frac{dX}{dt} = \frac{\mu_{max} SX}{(K_S+S)} \tag{5}$$

Monod [7] was the first to establish that, for a given organism and limiting substrate, the weight of the bacterial cells produced per weight of nutrient utilized is a constant under the same environmental conditions. The relationship is expressed in Equation (6) where Y is the yield constant.

$$Y = -\frac{dX}{dS} = \frac{\text{weight of organism formed}}{\text{weight of limiting substrate utilized}} \tag{6}$$

From Equations (5) and (6) one can obtain the following equation for the rate of substrate utilization:

$$-\frac{dS}{dt} = \frac{\mu_{max} SX}{Y(K_S+S)} \tag{7}$$

It is important to note, however, that the work of Hetling et.al. [8], Marr et. al. [9], and Rao and Gaudy [10] have shown that the yield coefficient is not necessarily a true constant either for pure cultures or mixed cultures.

2.4 MATHEMATICAL MODEL

Figure 1 shows the flow diagram for the four stage tower fermentor system, with feed to the second stage. Writing the material balances for substrate and cells around stages one through four, respectively, one can get the following equations by taking into account the sedimentation of cells:

$$V_1 \frac{dS_1}{dt} = fS_2 - fS_1 - \frac{\mu_{max} S_1 X_1 V_1}{Y(K_S+S_1)} \tag{8}$$

$$V_1 \frac{dX_1}{dt} = fX_2 - f\beta X_1 + \frac{\mu_{max} S_1 X_1 V_1}{(K_S+S_1)} \tag{9}$$

$$V_2 \frac{dS_2}{dt} = fS_1 + qS_0 + fS_3 - (q+2f)S_2 - \frac{\mu_{max} S_2 X_2 V_2}{Y(K_S+S_2)} \tag{10}$$

$$V_2 \frac{dX_2}{dt} = f\beta X_1 + fX_3 - (q+f)\beta X_2 - fX_2 + \frac{\mu_{max} S_2 X_2 V_2}{(K_S+S)} \tag{11}$$

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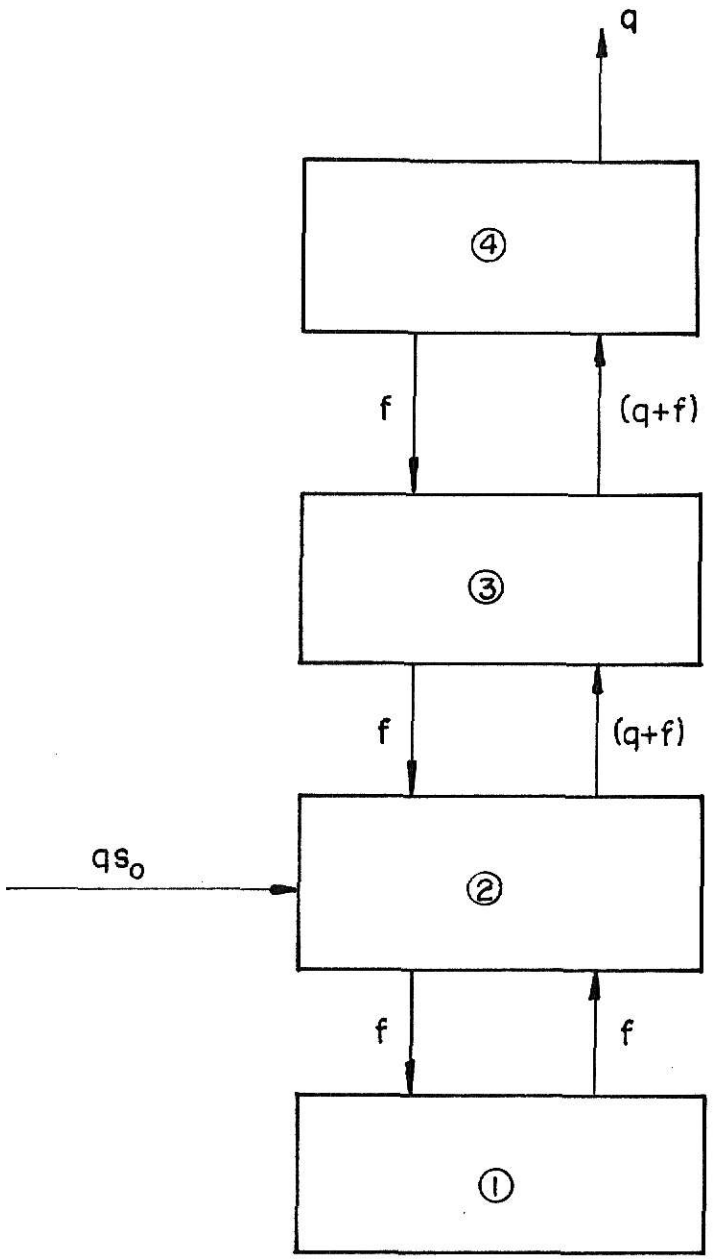

Fig. 1. Flow diagram for four stage tower fermentor.

$$V_3 \frac{dS_3}{dt} = (q+f)S_2 + fS_4 - (q+2f)S_3 - \frac{\mu_{max} S_3 X_3 V_3}{Y(K_S+S_3)} \tag{12}$$

$$V_3 \frac{dX_3}{dt} = (q+f)\beta X_2 + fX_4 - (q+f)\beta X_3 - fX_3 + \frac{\mu_{max} S_3 X_3 V_3}{(K_S+S)} \tag{13}$$

$$V_4 \frac{dS_4}{dt} = (q+f)S_3 - (q+f)S_4 - \frac{\mu_{max} S_4 X_4 V_4}{Y(K_S+S_4)} \tag{14}$$

$$V_4 \frac{dX_4}{dt} = (q+f)\beta X_3 - q\beta X_4 - fX_4 + \frac{\mu_{max} S_4 X_4 V_4}{(K_S+S)} \tag{15}$$

where

S_i = substrate concentration in the i^{th} stage (gms/lit.), i=1, 2, ...4

X_i = cell mass concentration in the i^{th} stage (gms/lit.), i=1, 2, ...4

t = time which is the independent variable (hr.)

q = rate of medium flow (lit./hr)

f = rate of backflow (lit./hr)

β = sedimentation coefficient (dimensionless)

Y = yield constant (dimensionless)

V_i = effective liquid volume in the i^{th} stage, i=1, 2, ... 4

S_0 = substrate concentration in the feed, (gms/lit.)

The following initial conditions are known:

$S_i(0) = 1.0$, i=1, 2, ... 4

$X_1(0) = 0.1$

$X_2(0) = X_3(0) = X_4(0) = 0$

Note that some microorganisms are assumed to be present in the first stage.

2.5 SIMULATION OF EXPERIMENTAL DATA

Generated experimental data can be useful in demonstrating the usefulness and applicability of a certain parameter estimation technique. Experimental data are often not available for some chemical or biological systems. It is worthwhile, in such a situation to use simulated data to investigate the feasibility of a particular parameter estimation technique. Experimental results often include some experimental error or uncertainity. Simulated data may be employed to consider the effects of various types of experimental errors, which might possibly be present. Data was generated taking into account several types of experimental errors and parameters were estimated taking them into account.

2.5a Normally distributed error

It has often been found that an error distribution is a special case of the normal distribution with mean zero and standard deviation σ. Several sets of data were generated taking different values for the standard deviation in order to study the effect of standard deviation of the error on the parameter estimates. The system of non-linear differential equations was first numerically integrated assuming the following numerical values of the parameters using fourth-order-Runge-Kutta integration with fixed step size. Subroutine RKGS [11] was employed for numerical integration.

q = 4.0 lit./hr.

f = 0.6 lit./hr.

S_0 = 1.0 gms/lit.

Y = 1.0

μ_{max} = 1.0 hr^{-1}

V_1 = 1 lit.

V_2 = 1 lit.

V_3 = 1 lit.

V_4 = 1 lit.

K_S = 0.05 gms/lit.

β = 0.8

The system of simultaneous equations was integrated from initial time of 0-hours to a final time of 20-hours taking a step size of 0.1 and with known initial conditions of all the dependent variables. Error was then introduced in these integrated results. Subroutine GAUSS [12] and RANDU [13] were used for this purpose. Random numbers were generated using the RANDU subroutine and these random numbers were then distributed normally with some standard deviation and mean zero with the help of GAUSS subroutine. The data was essentially generated for substrate and cell mass concentrations, where

$$S'_{iE} = (Sic \pm E) \qquad i = 1, 2, \ldots, 4 \qquad (16)$$

$$X'_{iE} = (Xic \pm E) \qquad i = 1, 2, \ldots, 4 \qquad (17)$$

where

S'_{iE} = generated experimental substrate concentration of the i^{th} stage.

X'_{iE} = generated experimental cell mass concentration of the i^{th} stage.

Sic = calculated substrate concentration of the i^{th} stage with assumed parameters.

Xic = calculated cell mass concentration of the i^{th} stage with assumed parameters.

E = experimental error introduced with mean zero and standard deviation σ.

Data was generated for the following values of standard deviations.

$\sigma = .01$ (Data Set 1)

$\sigma = .001$ (Data Set 2)

$\sigma = .0001$ (Data Set 3)

$\sigma = .00001$ (Data Set 4)

2.5b Zero error in the instrument

The presence of a zero error in the measuring instrument might result in undesirable experimental measurements. It was assumed that a zero error is present and this was superimposed over the normally distributed error. Now

$$S''_{iE} = (S'_{iE} + 0.001) \qquad i = 1, 2, \ldots 4 \tag{18}$$

$$X''_{iE} = (X'_{iE} + 0.001) \qquad i = 1, 2, \ldots 4 \tag{19}$$

where

S''_{iE} = experimental substrate concentration of the i^{th} stage.

X''_{iE} = experimental cell mass concentration of the i^{th} stage.

Here 0.001 is a constant number (the assumed zero error) which was combined with Data Set 2 to obtain Data Set 5.

2.5c Instrument drift

Here account was taken of the fact, that the experimental error changes with time. As experimentation proceeds the error due to instrument drift increases. Once again this type of error was superimposed over the normally distributed error. One now has

$$S'''_{iE} = S'_{iE} + 0.05\ t/t' \tag{20}$$

$$X'''_{iE} = X'_{iE} + 0.05\ t/t' \tag{21}$$

where

S'''_{iE} = experimental substrate concentration of the i^{th} stage.

X'''_{iE} = experimental cell mass concentration of the i^{th} stage.

t = any instant of time (between 0-20 hours)

t' = final time (20 hours)

Here 0.05 is an assumed constant. By adding instrument drift to Data Set 3, Data Set 6 was obtained. It is known that 99.73% of the random numbers should lie between $(\mu' + 3\sigma)$ to $(\mu' - 3\sigma)$. In other words, these

are the upper and lower bounds of the error introduced. Figures 2 through 4 show the variation of introduced error with time for Data Sets 1, 2, 5, and 6. Inspection of these plots shows that in all the cases except Data Set 6, the introduced error lies between the upper and lower bounds. Tabular values of the six sets of data may be found in Appendix I.

2.6 FORMULATION OF THE CRITERION FUNCTION

Three different models were considered for the identification of parameters:

1. 2-Parameter Model: the parameters were maximum growth rate (μ_{max}) and sedimentation coefficient (β).
2. 4-Parameter Model: the parameters were maximum growth rate (μ_{max}), sedimentation coefficient (β), yield constant (Y), and saturation constant (K_S).
3. 6-Parameter Model: the parameters were maximum growth rate (μ_{max}), sedimentation coefficient (β), yield constant (Y), saturation constant (K_S), rate of medium flow (q), and rate of backflow (f).

The simulated experimental data can essentially be represented by the following matrix, which has 21 rows corresponding to the 21 data points in time and 8 columns corresponding to the eight dependent variables.

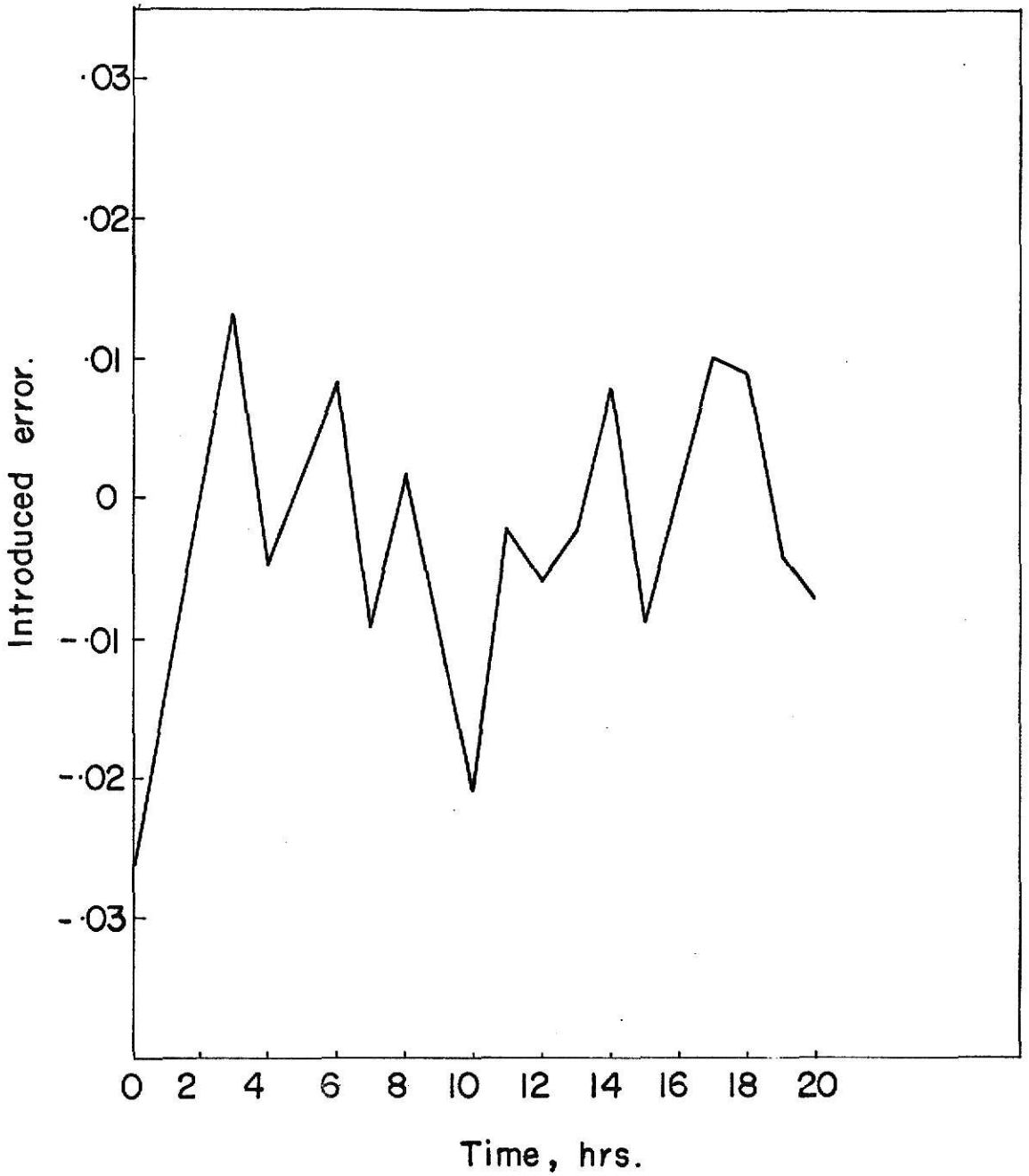

Fig. 2. Introduced error vs. time for data set 1; standard deviation (σ) = 0.01.

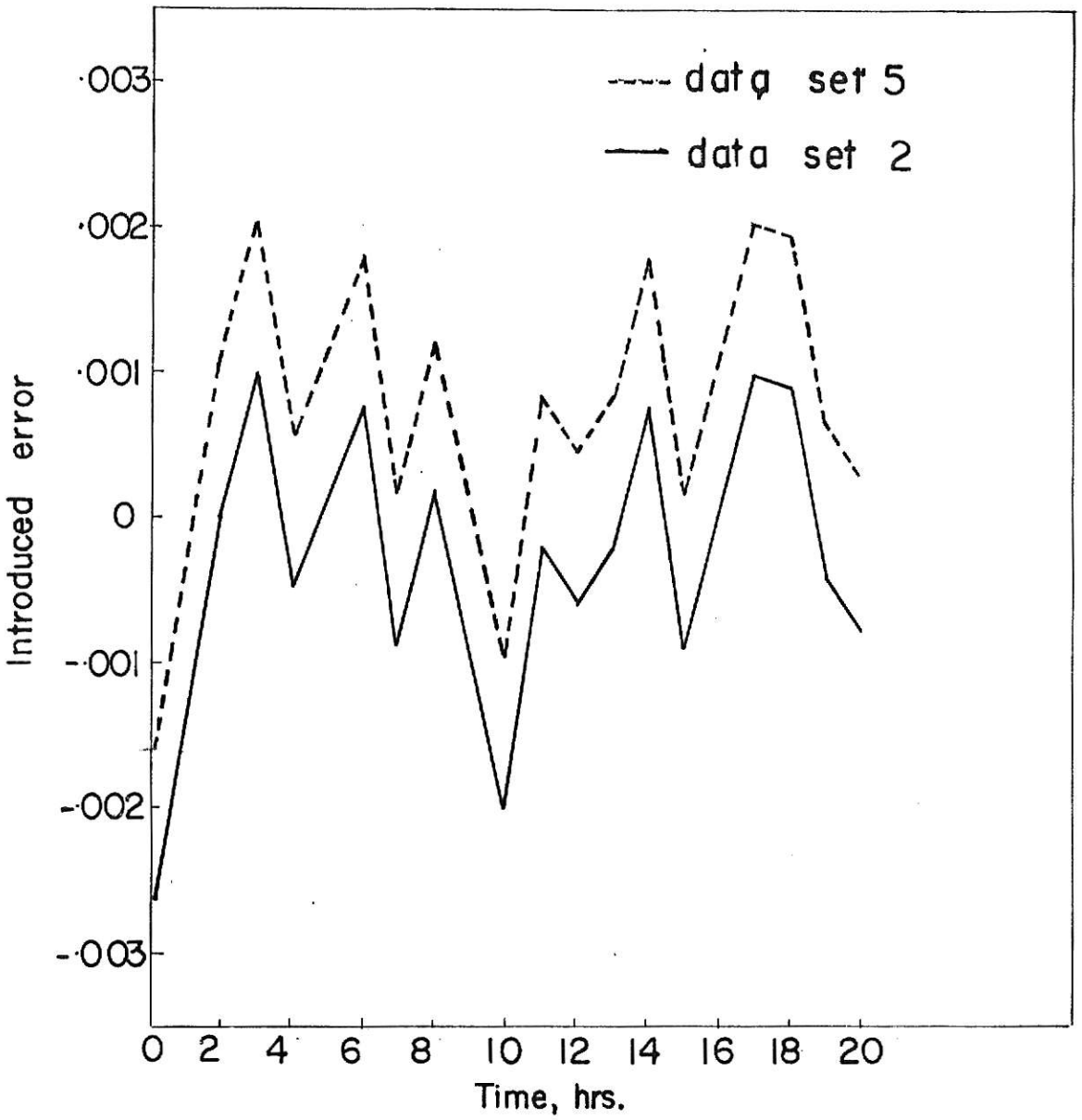

Fig. 3. Introduced error vs. time for data set 2 and 5; standard deviation (σ) = 0.001 and zero error = 0.001.

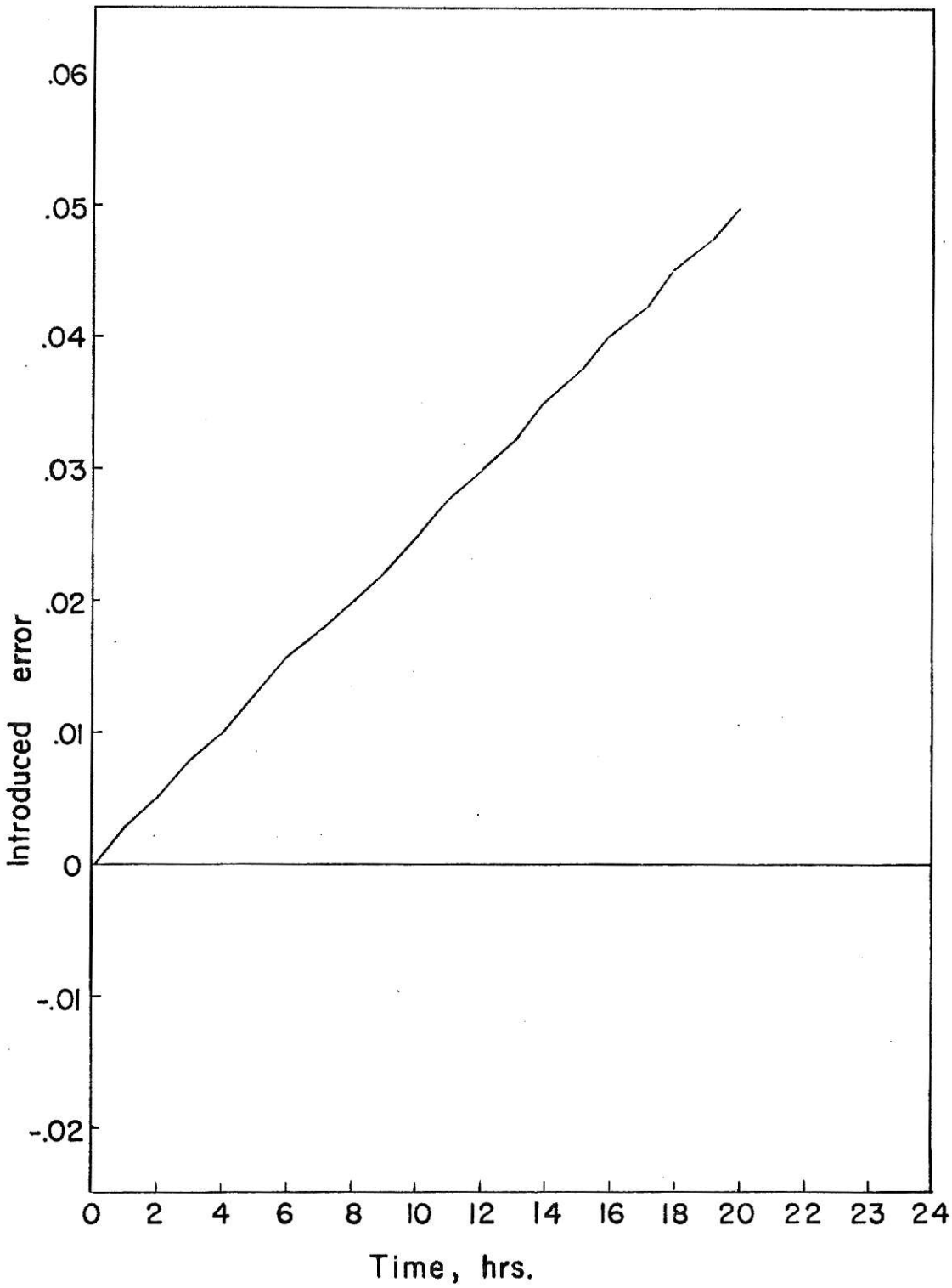

Fig. 4 Introduced error vs. time for data of set 6; standard deviation (σ) = 0.00001 and instrument drift.

$$Dy(J,I) = \begin{bmatrix} Dy(1,1) & Dy(1,2) & \cdots & Dy(1,8) \\ Dy(2,1) & Dy(2,2) & \cdots & Dy(2,8) \\ \vdots & \vdots & & \vdots \\ Dy(21,1) & Dy(21,2) & \cdots & Dy(21,8) \end{bmatrix}, \quad \begin{matrix} J=1, 2, \ldots 21 \\ I=1, 2, \ldots 8 \end{matrix} \qquad (22)$$

The matrix which was generated from the mathematical model at each iteration using assumed values of parameters can be represented as

$$Ey(J,I) = \begin{bmatrix} Ey(1,1) & Ey(1,2) & \cdots & Ey(1,8) \\ Ey(2,1) & Ey(2,2) & \cdots & Ey(2,8) \\ \vdots & \vdots & & \vdots \\ Ey(21,1) & Ey(21,2) & \cdots & Ey(21,8) \end{bmatrix}, \quad \begin{matrix} J=1, 2, \ldots 21 \\ I=1, 2, \ldots 8 \end{matrix} \qquad (23)$$

The objective function which was minimized can be represented as follows;

$$y_i = \sum_{J=1}^{N} \sum_{I=1}^{M} [Dy(J,I) - Ey(J,I)]^2 \qquad (24)$$

M = number of variables (8 in our case)

N = number of data points (21 in our case)

2.7 PARAMETER ESTIMATION RESULTS FOR NON-LINEAR DIFFERENTIAL MODELS

The optimal results for two, four and six parameter models are presented in Table 1 through Table 3. Results at intermediate iterations

Table 1. Parameter Estimates for Cases with Two Parameters

Data Set	Iteration No.	μ_{max}	β	y
1	0	1.000	0.800	0.0381
	20	0.976	0.796	0.0234
	34	0.978	0.797	0.0233
2	0	2.000	2.000	15.1117
	20	2.089	1.208	7.7558
	60	0.974	0.794	0.0044
	81	0.977	0.796	0.0041
3	0	3.000	3.000	21.2583
	20	3.720	2.096	17.8026
	60	0.986	0.803	0.0077
	91	6.977	0.796	0.0039
4	0	1.000	0.800	0.0190
	20	1.001	0.802	0.0190
	36	0.979	0.798	0.0039
5	0	1.000	0.800	0.01906
	20	0.978	0.796	0.00399
	46	0.976	0.795	0.00391
6	0	1.000	0.800	0.1595
	20	0.952	0.774	0.1021
	33	0.957	0.775	0.1015

Table 2. Parameter estimates for Cases with Four Parameters

Date Set	Iteration No.	μ_{max}	K_S	Y	β	y
1	0	1.000	0.05	1.000	0.800	0.0381
	20	0.987	0.052	1.013	0.808	0.0265
	60	0.984	0.061	0.984	0.785	0.0227
	80	0.983	0.062	0.978	0.782	0.0225
	120	0.955	0.045	0.963	0.777	0.0204
	139	0.954	0.045	0.961	0.775	0.0204
2	0	1.000	0.050	1.000	0.800	0.0192
	20	0.985	0.052	1.013	0.805	0.0067
	60	0.973	0.057	0.974	0.779	0.0023
	80	0.972	0.057	0.971	0.778	0.0023
	120	0.969	0.558	0.969	0.777	0.0021
	166	0.960	0.050	0.965	0.776	0.0019
3	0	1.000	0.050	1.000	0.800	0.0190
	20	0.985	0.052	1.013	0.805	0.0064
	60	0.973	0.057	0.974	0.779	0.0021
	80	0.972	0.057	0.971	0.778	0.002
	120	0.970	0.055	0.972	0.778	0.0019
	160	0.961	0.050	0.966	0.776	0.0017
4	0	1.000	0.050	1.000	0.800	0.0190
	20	0.985	0.052	1.013	0.805	0.0065
	60	0.981	0.063	0.968	0.773	0.0036
	80	0.980	0.063	0.97	0.776	0.0028
	120	0.976	0.056	0.978	0.782	0.0027
	159	0.977	0.050	0.975	0.779	0.0017
5	0	1.000	0.050	1.000	0.800	0.0190
	20	0.985	0.052	1.013	0.805	0.0065
	60	0.973	0.056	0.974	0.778	0.0022
	80	0.972	0.057	0.972	0.777	0.0021
	146	0.962	0.051	0.967	0.776	0.0020
6	0	1.000	0.050	1.000	0.800	0.1595
	20	0.965	0.055	1.026	0.788	0.0988
	60	0.984	0.064	1.041	0.793	0.0901
	80	1.058	0.122	1.052	0.783	0.0663
	120	1.088	0.139	1.055	0.784	0.0639
	168	1.092	0.140	1.050	0.784	0.0637

Table 3. Parameter Estimates for Cases with Six Parameters

Data Set	Iteration No.	μ_{max}	K_S	Y	β	q	f	y
1	0	1.000	0.050	1.000	0.800	4.000	0.600	0.0381
	20	0.997	0.050	1.100	0.803	4.010	0.627	0.0293
	60	0.990	0.052	1.100	0.804	4.000	0.615	0.0206
	100	0.986	0.052	1.100	0.802	4.000	0.615	0.0202
	200	0.960	0.055	1.100	0.802	3.780	0.567	0.0190
	227	0.958	0.055	1.001	0.800	3.786	0.566	0.0190
2	0	1.000	0.050	1.000	0.800	4.000	0.600	0.0192
	20	0.997	0.050	1.004	0.803	4.013	0.627	0.0099
	60	0.990	0.052	1.013	0.808	3.990	0.616	0.0018
	100	0.989	0.052	1.010	0.806	3.994	0.613	0.0015
	200	0.971	0.052	0.982	0.788	3.975	0.608	0.0007
	290	0.955	0.052	1.003	0.801	3.796	0.568	0.0002
3	0	1.000	0.050	1.000	0.800	4.000	0.600	0.0190
	20	0.998	0.050	1.004	0.803	4.013	0.627	0.0096
	60	0.991	0.052	1.011	0.807	3.997	0.616	0.0015
	100	0.988	0.052	1.011	0.806	3.995	0.614	0.0012
	200	0.970	0.048	0.984	0.791	4.001	0.611	0.0005
	300	0.952	0.049	0.999	0.800	3.811	0.571	0.0002
4	0	1.000	0.050	1.000	0.800	4.000	0.600	0.01904
	20	0.997	0.050	1.004	0.803	4.013	0.627	0.00095
	60	0.990	0.052	1.011	0.807	3.997	0.616	0.00015
	100	0.989	0.052	1.011	0.806	4.002	0.615	0.00014
	160	0.965	0.047	0.986	0.792	3.956	0.602	0.00003
	193	0.964	0.048	0.988	0.793	3.950	0.599	0.00002
5	0	1.000	0.050	1.000	0.800	4.000	0.600	0.0190
	20	0.997	0.050	1.004	0.803	4.013	0.627	0.0094
	60	0.990	0.052	1.014	0.808	3.991	0.616	0.0016
	100	0.989	0.052	1.012	0.806	4.000	0.614	0.0014
	200	0.970	0.050	0.992	0.794	3.954	0.600	0.0005
	239	0.963	0.055	0.999	0.797	3.843	0.578	0.0003
6	0	1.000	0.050	1.000	0.800	4.000	0.600	0.1595
	20	0.994	0.050	1.054	0.799	4.012	0.606	0.1172
	60	0.978	0.053	1.040	0.800	3.997	0.607	0.0959
	100	0.974	0.055	1.039	0.797	3.943	0.596	0.0935
	200	0.940	0.070	1.052	0.802	3.546	0.511	0.0885
	300	0.978	0.084	1.101	0.829	3.569	0.519	0.0799
	550	1.067	0.154	1.111	0.817	3.573	0.522	0.0622

have also been shown to facilitate a close observation of the variation of the parameters as the iterative computations proceed. Results are presented for each of the six data sets in each table. In Tables 1 through 3 the parameter values at the zeroth iteration are the initial estimates.

2.8 DISCUSSION AND CONCLUDING REMARKS

The main advantage of using the simplex pattern search for estimating parameters is the fact that it is neither based on gradients (first-order derivatives) nor on quadratic forms (second-order derivatives). Box [14] has suggested that in using this technique to find the optimum point, the feasible region must be convex, which essentially means that the function should be unimodal (either concave or convex) in the feasible region. However with the multidimensional nature of the problem, it is practically impossible to test for convexity of the function in the feasible region. In this situation experience combined with ingenuity are helpful in finding a satisfactory solution of the problem.

It has been noted by Nelder and Mead [5] and Spendler and Himsworth [15] that the effect of step size on the number of evaluations is not very large.

The values of the constants σ, η and γ appearing in the search technique equations are taken to be 1, 1/2 and 2 respectively for all computational work, as suggested by Nelder and Mead [5].

The stopping criterion used for convergence is

$$\left\{ \sum_{i=1}^{n+1} (y_i - \bar{y})^2 \; / \; n \right\}^{1/2} < 10^{-6} \qquad (25)$$

Figure 5 shows how the optimum function value changes as the standard deviation of introduced error is varied, for two, four and six parameters.

Comparison of optimum function value obtained from two, four and six parameters models using data sets 1 through 6 indicates that it decreases appreciably as the number of parameters in the model are increased from two to six. The results reveal that a better fit for the experimental data can be obtained if one uses the six parameter model.

The effect of standard deviation of introduced error on the final estimates of parameters can be studied with the help of Tables 1 through 3. The parameter estimates should diverge, in general, from the parameter values used for generating the data ($\sigma = 0.0$), as the standard deviation of introduced error increases. This was, however not found very predominant from the results obtained, but still is noticeable in the case of four and six parameter models. These curves suggest that for all parameter models the optimum function value decreases as the standard deviation of the error is decreased.

Figure 6 shows the variation in the computing time with the number of parameters in the model. It can be seen that in going from the 2-parameter model to the 4-parameter model the computing time is almost tripled. Further increasing the number of parameters does not increase the computing time in the same manner, as can be seen from the plot. This technique took 5.5 mins. of computing time on the IBM 360/50 computer for the 2-parameter model, 16.4 mins. for the 4-parameter model and 21 mins. for the 6-parameter model. The considerable amount of computing time taken for estimating parameters in the tower system occurs because the system of

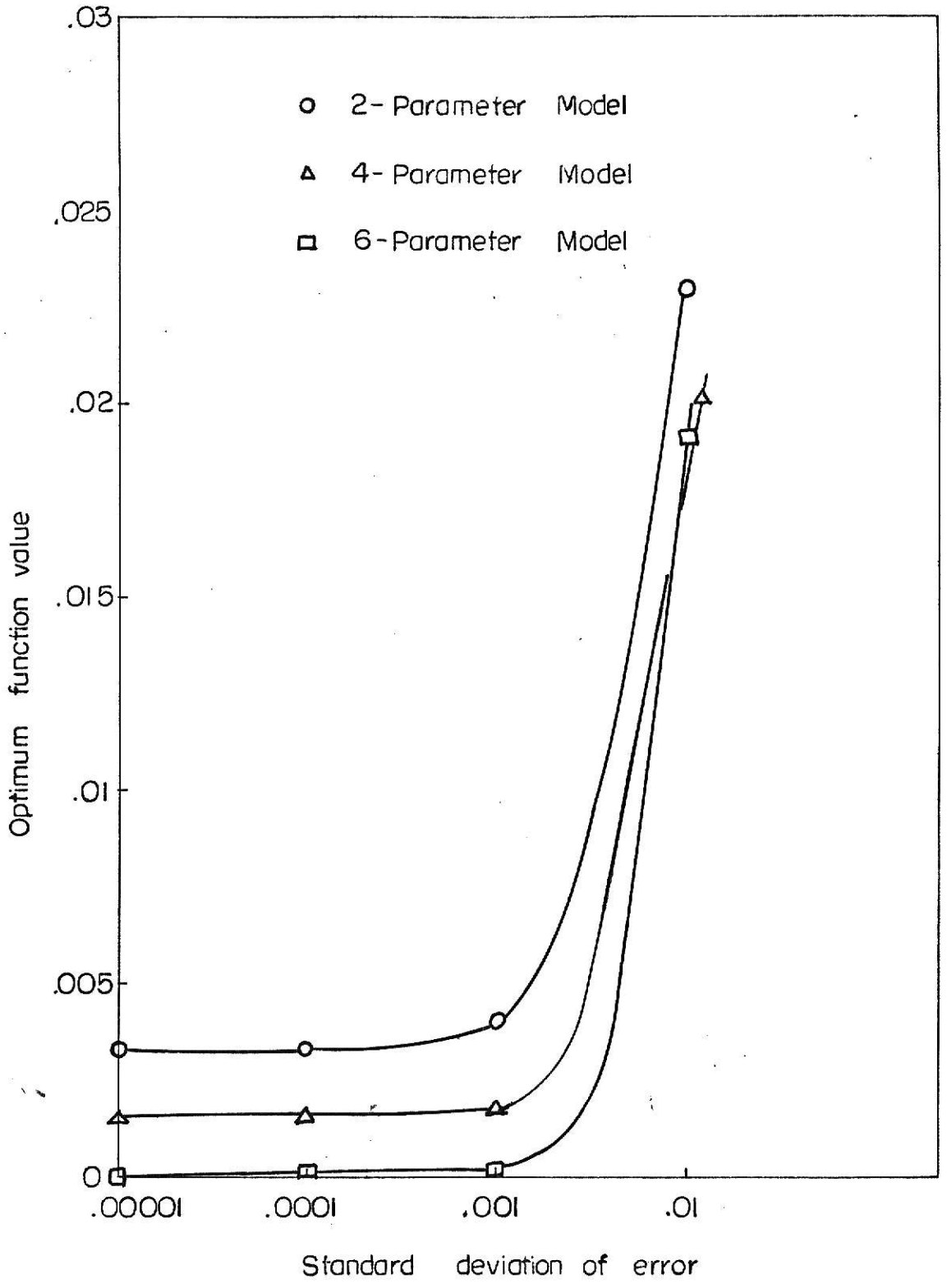

Fig. 5. Standard deviation of normally distributed error vs. optimum function value.

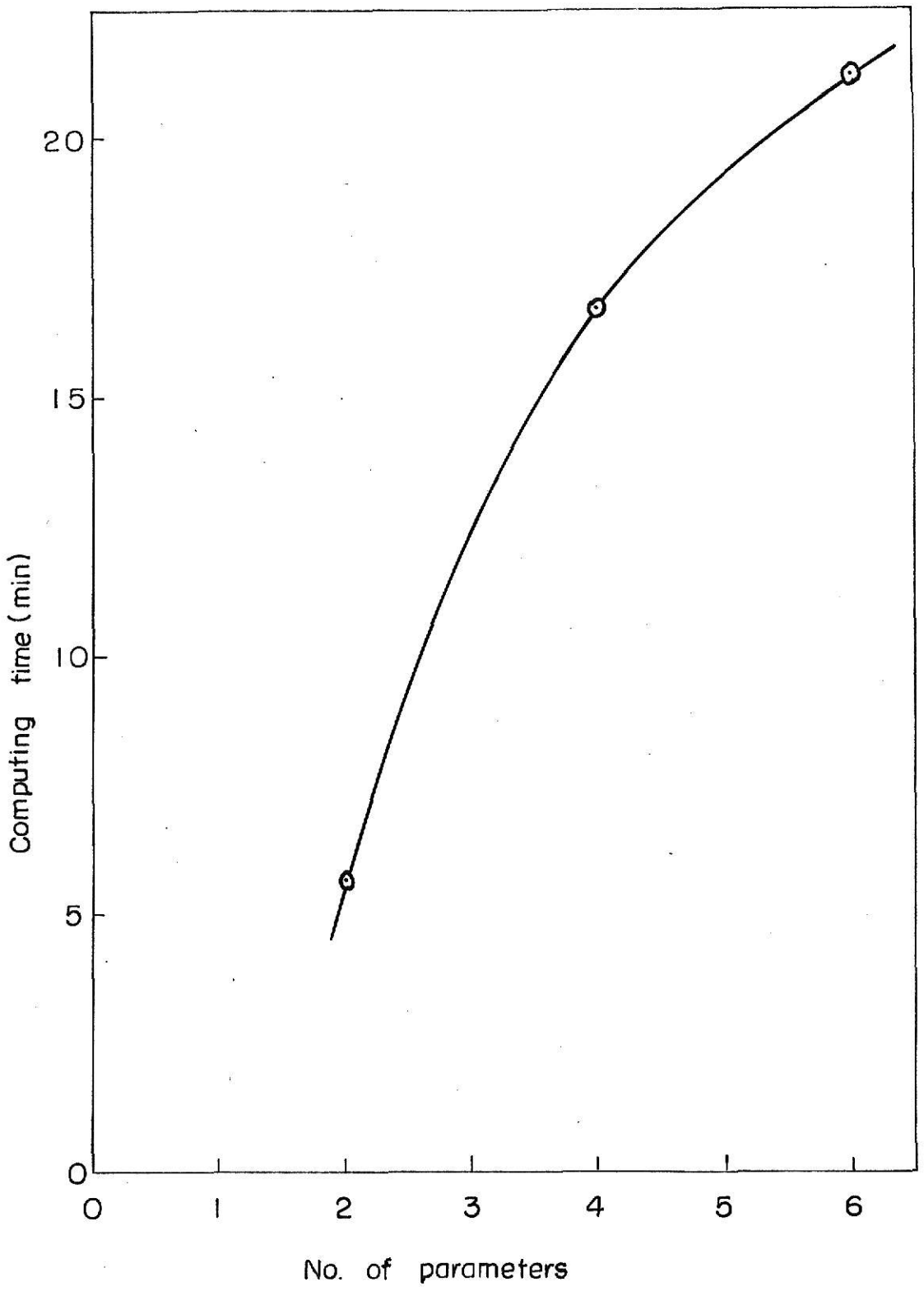

Fig. 6. No. of parameters in the model vs. computing time.

non-linear simultaneous differential equations must be numerically integrated at each iteration. It would be worthwhile to estimate parameters for the tower system using other non-linear parameter estimation techniques like Bard's modification [16] and Davidon's method [17] etc. and to compare the results, as the employment of a very efficient search technique is needed in this problem.

Figure 7 shows the variation of the number of function evaluations with the number of parameters in the model for different values of standard deviation of introduced error. The general trend of the curves shows that as the number of parameters in the model increase, the number of evaluations are increased for all the cases considered. The curves, however, indicate how this change occurs. For example for a case with standard deviation of error (σ) = 0.01, the number of evaluations are almost directly proportional to the number of parameters in the model.

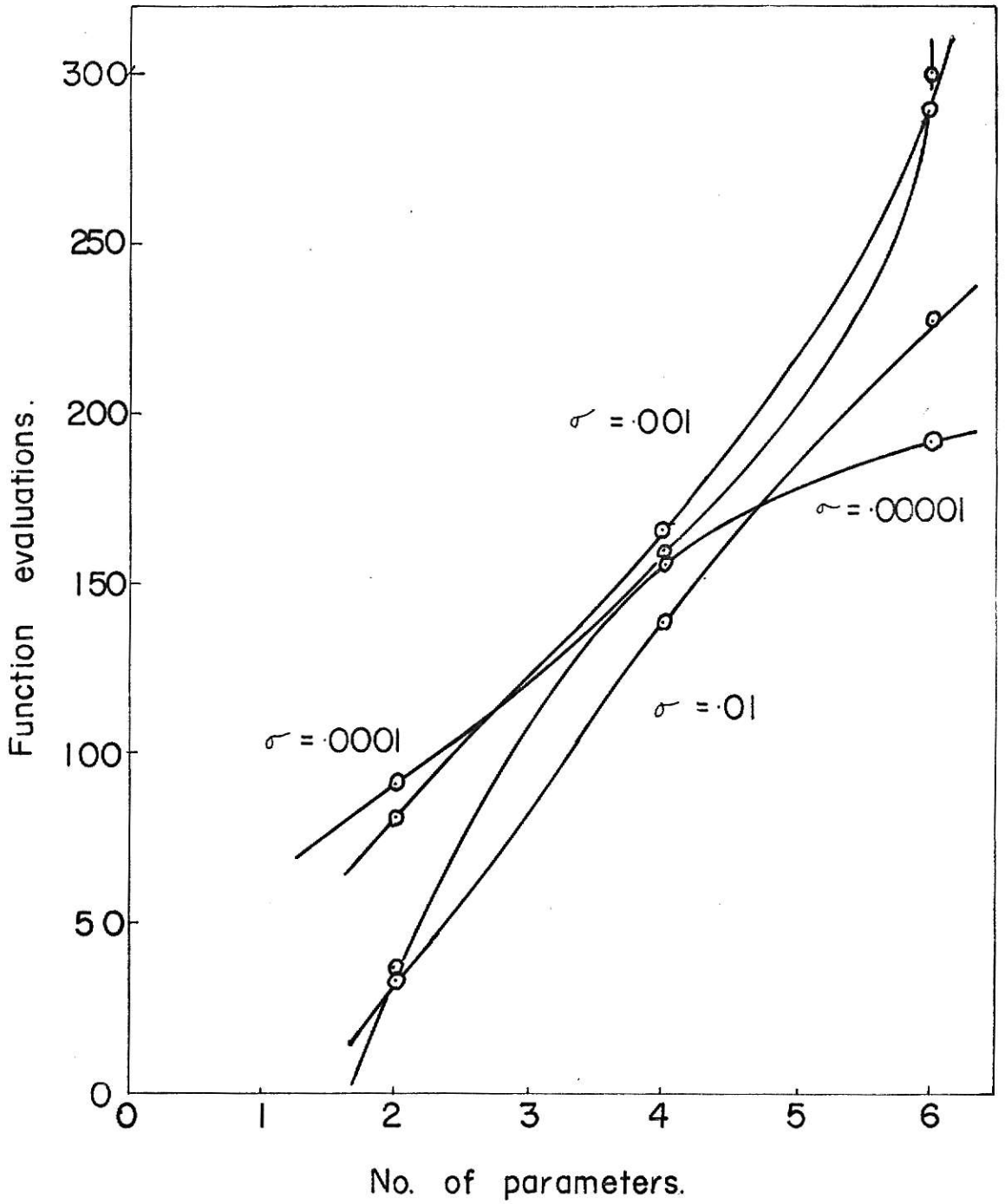

Fig. 7. Number of parameters in the model vs. function evaluations for different values of deviations of normally distributed error.

2.9 NOTATION

DY(J,I)	= J^{th} element of I^{th} column of data matrix
E	= experimental error
EY(J,I)	= J^{th} element of I^{th} column of generated matrix
f	= rate of backflow, (lit./hr)
K_S	= saturation constant, (gms/lit.)
n	= dimension of the problem
P_J	= J^{th} parameter in the model
P'_1, P'_2, P'_3	= trial points in the starting simplex
P'_4	= centroid of points P'_1 and P'_2
P'_5	= reflection of the point P'_3 with respect to P'_4
P'_6	= expansion of P'_5
P'_7	= contraction of the highest valued point P'_3 with respect to P'_4
P'_c	= centroid of a set of n points in a simplex
q	= rate of medium flow, (lit./hr)
S_0	= initial substrate concentration, (gms/lit.)
S_i	= substrate concentration in i^{th} stage, (gms/lit.)
S'_{iE}	= generated experimental substrate concentration for i^{th} stage with normally distributed error, (gms/lit.)
S_{ic}	= calculated substrate concentration for i^{th} stage with normally distributed error, (gms/lit.)
S''_{iE}	= generated experimental substrate concentration for i^{th} stage with zero error superimposed over normally distributed error, (gms/lit.)

S'''_{iE} = generated experimental substrate concentration for i^{th} stage with instrument drift superimposed over normally distributed error, (gms/lit.)

t = any time, (hrs)

t' = final time, (hrs)

V_i = effective volume in the i^{th} stage

X'_{iE} = generated experimental cell mass concentration for the i^{th} stage with normally distributed error, (gms/lit.)

X''_{iE} = generated experimental cell mass concentration for the i^{th} stage with zero error superimposed over normally distributed error, (gms/lit.)

X'''_{iE} = generated experimental cell mass concentration for the i^{th} stage with instrument drift superimposed over normally distributed error, (gms/lit.)

y = objective function value

y_1, y_2, y_3 = objective function value at P'_1, P'_2, and P'_3

$\bar{y}$ = average of y_1, y_2, and y_3

Y = yield constant

Z_L = functions of ξ_i

Greek Letters

α = reflection coefficient

β = sedimentation coefficient

γ = contration coefficient

η = expansion coefficient

μ = specific growth rate, (hr^{-1})

μ_{max} = maximum growth rate when the organic concentration is not limiting the rate of growth, (hr^{-1})

μ' = mean

σ = standard deviation

2.10 REFERENCES

1. Maxon, W. D., "Continuous Fermentation," Advances in Applied Microbiology, 2, 335 (1960).

2. Falch, E. A., and E. L. Gaden, Jr., "A Continuous Multistage Tower Fermentor," Biotech. and Bioeng., 2, 927-943 (1969).

3. Bischoff, K. B., Can. J. Chem. Engr., 44, 281 (1966).

4. Deindoerfer, F. H., and A. E. Humphrey, Ind. Eng. Chem., 51, 810 (1959).

5. Nelder, J. A., and R. Mead, Computer J., 7, 308 (1965).

6. Draper, N. R. and H. Smith, "Applied Regression Analysis," John Wiley and Sons, Inc. (1968).

7. Monod, J., "Recherces sur la Croissances des Cultures Bacteriennes," Hermann and Cie, Paris, (1942).

8. Hetling, L. J., D. R. Washington, and S. S. Rao, Proc. 19th Ind. Waste Conf., Purdue Univ., Purdue Eng. Extension Series No. 117, 49, 687 (1965).

9. Marr, A. G., E. H. Nilson, and D. V. Clark, Ann, N. Y. Acad. Sci., 102, 536 (1936).

10. Rao, B. S., and A. F. Gaudy, Jr., J. Water Pollution Control Federation 38, 794 (1966).

11. RKGS, IBM Scientific Subroutine Package, The IBM Company, White Plains, New York, Version II.

12. GAUSS, IBM Scientific Subroutine Package, The IBM Company, White Plains, New York, Version II.

13. RANDU, IBM Scinetific Subroutine Package, The IBM Company, White Plains, New York, Version II.

14. Box, M. J., "A New Method of Constrained Optimization and A Comparison with other Methods," Comp. J., 8, 42 (1965).

15. Spendley, W., G. R. Hext, and F. R. Himsworth, "Sequential Applications of Simplex Design in Optimization and Evolutionary Operation," Technometrics, 4 (1962).

16. Bard, Y., IBM New York Scientific Centre Report No. 320-2902 (1967).

17. Fletcher, R., and M. J. D. Powell, "A Rapidly Convergent Descent Method for Minimization," The Computer J. 6 (2) 163-168 (1963).

18. Gauss, C. F., "Theory of Least Squares," English Translation by Hale F. Trotter, Princeton University, Statistical Techniques Research Group, Technical Report No. 5 (1957)(original (1821).

19. Box, G. E. P., and G. A. Coutie, Proc. Inst. Elect. Engers., 103, Part B, Supplement No. 1, 100 (1956).

20. Marquardt, D. W., J. Soc. Ind. Appl. Math. 11 (2), 431 (1963).

21. Rosenbrock, H. H., Computer J., 3, 175 (1960).

22. Hooke, R., and T. A. Jeeves, J. Assn. Comp. Math., 8, (2), 212 (1961).

23. Fan, L. T., C. L. Hwang, and F. A. Tillman, AIIE Transactions, Industrial Engineering Research and Development, 1, (3), 267-273 (1969).

24. Chen, G. K., M. S. Thesis, Kansas State University (1968).

APPENDIX I

Table 4. Simulated Experimental Data For Four Stage Tower Systems with Standard Deviation = 0.01 (Data Set 1)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.973667	0.073667	0.973667	-0.026333	0.973667	-0.026333	0.973667	-0.026333
1.10	0.876784	0.166524	0.971423	0.012091	0.971393	0.011222	0.972662	0.007916
2.10	0.754243	0.311888	0.964147	0.047062	0.959684	0.051981	0.956312	0.054044
3.10	0.559257	0.546683	0.945298	0.094104	0.935366	0.105031	0.926546	0.111953
4.10	0.234640	0.869407	0.878180	0.129636	0.859136	0.150428	0.841969	0.164346
5.10	0.041508	1.128447	0.836674	0.194986	0.795163	0.240817	0.758604	0.272488
6.10	0.042709	1.186998	0.831378	0.226345	0.767792	0.301465	0.704413	0.363529
7.10	0.022906	1.203250	0.808941	0.219852	0.736182	0.308901	0.659658	0.388379
8.10	0.033211	1.238585	0.817976	0.238143	0.740758	0.334061	0.658069	0.421889
9.10	0.018229	1.240939	0.802257	0.228311	0.722519	0.328075	0.636505	0.420347
10.10	0.007768	1.242466	0.791326	0.221287	0.710022	0.323421	0.622005	0.418334
11.10	0.027172	1.270161	0.810416	0.243027	0.728086	0.346707	0.638779	0.443306
12.10	0.023350	1.272070	0.806370	0.240805	0.723357	0.345523	0.633190	0.443244
13.10	0.026431	1.279116	0.809312	0.244988	0.725819	0.350415	0.635066	0.448897
14.10	0.037072	1.292500	0.819850	0.256389	0.736032	0.362302	0.644878	0.461306
15.10	0.019372	1.276699	0.802079	0.239215	0.718038	0.345464	0.626607	0.444827
16.10	0.029434	1.288074	0.812092	0.249640	0.727896	0.356121	0.636273	0.455732
17.10	0.039536	1.299082	0.822160	0.259993	0.737856	0.366635	0.646102	0.466417
18.10	0.037938	1.298110	0.820538	0.258568	0.736161	0.365321	0.644315	0.465222
19.10	0.024310	1.284915	0.806894	0.245060	0.722465	0.351889	0.630557	0.451872
20.10	0.020787	1.281687	0.803359	0.241618	0.718895	0.348501	0.626943	0.448540

Table 5. Simulated Experimental Data For Four Stage Tower System with Standard Deviation = 0.001 (Data Set 2)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.997367	0.097367	0.997367	-0.002633	0.997367	-0.002633	0.997367	-0.002633
1.10	0.888399	0.178140	0.983038	0.023706	0.983008	0.022837	0.984277	0.019531
2.10	0.753404	0.311050	0.963309	0.046223	0.958845	0.051142	0.955473	0.053205
3.10	0.546767	0.534193	0.932808	0.081615	0.922877	0.092542	0.914056	0.099463
4.10	0.238946	0.873713	0.882486	0.133941	0.863442	0.154733	0.846275	0.168652
5.10	0.040158	1.127097	0.835324	0.193636	0.793813	0.239467	0.757254	0.271138
6.10	0.034738	1.179028	0.823407	0.218374	0.759821	0.293494	0.696442	0.355559
7.10	0.031084	1.211428	0.817119	0.228030	0.744360	0.317079	0.667836	0.396557
8.10	0.031091	1.236465	0.815855	0.236022	0.738637	0.331940	0.655949	0.419768
9.10	0.028933	1.251643	0.812961	0.239015	0.733223	0.338779	0.647208	0.431051
10.10	0.027450	1.262149	0.811009	0.240970	0.729704	0.343104	0.641687	0.438017
11.10	0.029098	1.272086	0.812342	0.244952	0.730012	0.348632	0.640705	0.445232
12.10	0.028517	1.277238	0.811546	0.245972	0.728525	0.350690	0.638357	0.448412
13.10	0.028689	1.281374	0.811570	0.247246	0.728077	0.352673	0.637324	0.451155
14.10	0.029660	1.285088	0.812438	0.248977	0.728620	0.354891	0.637466	0.453894
15.10	0.027826	1.285152	0.810533	0.247668	0.726491	0.353918	0.635060	0.453281
19.30	0.028788	1.287428	0.811446	0.248993	0.727249	0.355475	0.635627	0.455086
19.50	0.029768	1.289313	0.812391	0.250224	0.728088	0.356866	0.636333	0.456649
19.70	0.029587	1.289759	0.812187	0.250216	0.727809	0.356969	0.635964	0.456871
19.90	0.028210	1.288814	0.810793	0.248959	0.726364	0.355789	0.634456	0.455771
20.10	0.027847	1.288748	0.810419	0.248679	0.725956	0.355561	0.634004	0.455601

Table 6. Simulated Experimental Data For Four Stage Tower System with Standard Deviation = 0.0001 (Data Set 3)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.999737	0.099737	0.999737	-0.000263	0.999737	-0.000263	0.999737	-0.000263
1.10	0.889561	0.179301	0.984200	0.024867	0.984169	0.023999	0.985439	0.020693
2.10	0.753320	0.310966	0.963225	0.046140	0.958761	0.051058	0.955390	0.053121
3.10	0.545519	0.532944	0.931559	0.080366	0.921628	0.091293	0.912807	0.098214
4.10	0.239277	0.874144	0.882916	0.134372	0.863873	0.155164	0.846705	0.169082
5.10	0.040023	1.126962	0.835189	0.193501	0.793678	0.239332	0.757119	0.271003
6.10	0.033941	1.178230	0.822610	0.217577	0.759024	0.292697	0.695645	0.354761
7.10	0.031902	1.212245	0.817937	0.228848	0.745177	0.317897	0.668654	0.397375
8.10	0.030879	1.236252	0.815643	0.235810	0.738425	0.331728	0.655737	0.419556
9.10	0.030003	1.252713	0.814031	0.240085	0.734293	0.339849	0.648278	0.432121
10.10	0.029419	1.264117	0.812977	0.242938	0.731673	0.345072	0.643656	0.439985
11.10	0.029290	1.272279	0.812534	0.245145	0.730204	0.348825	0.640898	0.445425
12.10	0.029033	1.277754	0.812063	0.246488	0.729041	0.351207	0.638874	0.448928
13.10	0.028915	1.281599	0.811796	0.247471	0.728302	0.352898	0.637549	0.451381
14.10	0.028919	1.284347	0.811697	0.248236	0.727879	0.354149	0.636725	0.453153
15.10	0.028671	1.285997	0.811378	0.248514	0.727336	0.354763	0.635905	0.454126
19.30	0.028723	1.287363	0.811381	0.248929	0.727184	0.355410	0.635562	0.455021
19.50	0.028791	1.288336	0.811414	0.249247	0.727111	0.355889	0.635356	0.455672
19.70	0.028752	1.288923	0.811351	0.249381	0.726974	0.356134	0.635129	0.456035
19.90	0.028600	1.289204	0.811183	0.249349	0.726754	0.356178	0.634846	0.456161
20.10	0.028553	1.289454	0.811125	0.249385	0.726662	0.356267	0.634710	0.456307

Table 7. Simulated Experimental Data For Four Stage Tower System with Standard Deviation = 0.00001 (Data Set 4)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.999974	0.099974	0.999974	-0.000026	0.999974	-0.000026	0.999974	-0.000026
1.10	0.889677	0.179417	0.984316	0.024984	0.984286	0.024115	0.985555	0.020809
2.10	0.753312	0.310958	0.963216	0.046131	0.958753	0.051050	0.955381	0.053113
3.10	0.545394	0.532819	0.931434	0.080241	0.921503	0.091168	0.912682	0.098089
4.10	0.239420	0.874187	0.882959	0.134415	0.863916	0.155207	0.846748	0.169125
5.10	0.040010	1.126948	0.835176	0.193488	0.793664	0.239319	0.757106	0.270989
6.10	0.033861	1.178151	0.822531	0.217497	0.758944	0.292618	0.695565	0.354682
7.10	0.031984	1.212327	0.818019	0.228930	0.745259	0.317979	0.668736	0.397457
8.10	0.030858	1.236231	0.815622	0.235789	0.738404	0.331707	0.655715	0.419535
9.10	0.030110	1.252820	0.814138	0.240192	0.734400	0.339956	0.648386	0.432228
10.10	0.029616	1.264315	0.813174	0.243135	0.731870	0.345269	0.643853	0.440182
11.10	0.029310	1.272298	0.812554	0.245164	0.730224	0.348844	0.640917	0.445444
12.10	0.029085	1.277806	0.812114	0.246540	0.729093	0.351258	0.638925	0.448980
13.10	0.028937	1.281622	0.811818	0.247494	0.728325	0.352921	0.637572	0.451403
14.10	0.028845	1.284272	0.811623	0.248162	0.727805	0.254075	0.636651	0.453079
15.10	0.028756	1.286082	0.811463	0.248598	0.727421	0.354848	0.635990	0.454211
16.10	0.028717	1.287356	0.811374	0.248922	0.727178	0.355404	0.635556	0.455015
17.10	0.028693	1.288239	0.811316	0.249150	0.727013	0.355792	0.635259	0.455574
18.10	0.028668	1.288839	0.811268	0.249298	0.726891	0.356051	0.635045	0.455952
19.10	0.028639	1.289243	0.811222	0.249388	0.726793	0.356217	0.634885	0.456200
20.10	0.028624	1.289524	0.811196	0.249456	0.726732	0.356338	0.634780	0.456378

Table 8. Simulated Experimental Data For Four Stage Tower System with Standard Deviation = .001 and Zero Error of 0.001 (Data Set 5)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.998367	0.098367	0.998367	-0.001633	0.998367	-0.001633	0.998367	-0.001633
1.10	0.889399	0.179140	0.984038	0.024706	0.984008	0.023837	0.985277	0.020531
2.10	0.754404	0.312050	0.964309	0.047223	0.959845	0.052142	0.956473	0.054205
3.10	0.547767	0.535193	0.933808	0.082615	0.923877	0.093542	0.915056	0.100463
4.10	0.239946	0.874713	0.883486	0.134941	0.864442	0.155733	0.847275	0.169652
5.10	0.041158	1.128097	0.836324	0.194636	0.794813	0.240467	0.758254	0.272138
6.10	0.035738	1.180027	0.824407	0.219374	0.760821	0.294494	0.697442	0.356559
7.10	0.032084	1.212427	0.818119	0.229030	0.745360	0.318079	0.668836	0.397557
8.10	0.032091	1.237464	0.816855	0.237022	0.739637	0.332940	0.656949	0.420768
9.10	0.029933	1.252643	0.813961	0.240015	0.734223	0.339779	0.648208	0.432051
10.10	0.028450	1.263148	0.812009	0.241970	0.730704	0.344104	0.642687	0.439017
11.10	0.030098	1.273086	0.813342	0.245952	0.731012	0.349632	0.641705	0.446232
12.10	0.029517	1.278237	0.812546	0.246972	0.729524	0.351690	0.639357	0.449412
13.10	0.029689	1.282373	0.812570	0.248246	0.729077	0.353673	0.638324	0.452155
14.10	0.030660	1.286087	0.813438	0.249977	0.729620	0.355891	0.638466	0.454894
15.10	0.028826	1.286152	0.811533	0.248668	0.727491	0.354918	0.636060	0.454281
16.10	0.029788	1.288427	0.812446	0.249993	0.728249	0.356475	0.636627	0.456086
17.10	0.030768	1.290313	0.813391	0.251224	0.729088	0.357866	0.637333	0.457649
18.10	0.030587	1.290758	0.813187	0.251216	0.728809	0.357969	0.636964	0.457871
19.10	0.029210	1.289813	0.811793	0.249959	0.727364	0.356789	0.635456	0.456771
20.10	0.028847	1.289747	0.811419	0.249679	0.726956	0.356561	0.635004	0.456601

Table 9. Simulated Experimental Data For Four Stage Tower System with Standard Deviation = .00001 and Instrument Drift (Data Set 6)

Time, hrs.	S_1	X_1	S_2	X_2	S_3	X_3	S_4	X_4
0.00	0.999737	0.099737	0.999737	-0.000263	0.999737	-0.000263	0.999737	-0.000263
1.10	0.892311	0.182051	0.986950	0.027617	0.986919	0.026749	0.988189	0.023443
2.10	0.758570	0.316216	0.968475	0.051390	0.964011	0.056308	0.960640	0.058371
3.10	0.553268	0.540694	0.939309	0.088116	0.929378	0.099043	0.920557	0.105964
4.10	0.249626	0.884394	0.893166	0.144622	0.874122	0.165414	0.856955	0.179332
5.10	0.052773	1.139711	0.847939	0.206251	0.806428	0.252082	0.769869	0.283752
6.10	0.049191	1.193480	0.837860	0.232827	0.774274	0.307947	0.710895	0.370011
7.10	0.049652	1.229994	0.835687	0.246598	0.762927	0.335646	0.686404	0.415125
8.10	0.051129	1.256501	0.835893	0.256060	0.758675	0.351978	0.675986	0.439806
9.10	0.052753	1.275462	0.836781	0.262835	0.757043	0.362599	0.671028	0.454871
10.10	0.054668	1.289367	0.838226	0.268188	0.756922	0.370322	0.668905	0.465235
11.10	0.057040	1.300028	0.840284	0.272895	0.757954	0.376575	0.668647	0.473174
12.10	0.059283	1.308002	0.842312	0.276738	0.759291	0.381456	0.669123	0.479178
13.10	0.061664	1.314348	0.844545	0.280221	0.761052	0.385648	0.670299	0.484130
14.10	0.064168	1.319595	0.846946	0.283485	0.763129	0.389399	0.671974	0.488403
15.10	0.066421	1.323747	0.849128	0.286263	0.765086	0.392513	0.673655	0.491876
16.10	0.068973	1.327612	0.851630	0.289178	0.767434	0.395660	0.675811	0.495271
17.10	0.071540	1.331084	0.854163	0.291996	0.769859	0.398638	0.678105	0.498421
18.10	0.074000	1.334171	0.856600	0.294629	0.772222	0.401382	0.680377	0.501283
19.10	0.076347	1.336950	0.858930	0.297096	0.774502	0.403926	0.682593	0.503909
20.10	0.078800	1.339700	0.861372	0.299632	0.776909	0.406514	0.684956	0.506554

Appendix II

SIMPLEX PATTERN SEARCH TECHNIQUE FOR ESTIMATION OF PARAMETERS IN DIFFERENTIAL MODELS

The purpose of various parameter estimation techniques and search techniques is to minimize the selected criterion function. This criterion function is a measure of the difference between the measured experimental value of the response at particular values of the independent variable and the predicted response (from the mathematical model) based on the values of independent variables and parameters. The linearization techniques are based on the particular form of the conventional sum of squares criterion function with the assumption that the linearity assumption around the minimum criterion function value allows efficient progress. Sometimes for some non-linear models and for poor initial estimates of parameters the usual parameter estimation techniques like Gauss method [18], Steepest descent method [19], Marquardt modification [20], generalized Newton-Raphson method [21], and Bard's modification [16] may be quite inefficient. Hence, here a search technique developed to minimize an objective function can be used.

The pattern search techniques are efficient and simple to use because they do not require derivatives. The Simplex method [5], Box method [14] and Hooke-Jeeves method [22] are commonly used methods of this type. The particular method proposed by Nelder and Mead [5] will be described here briefly. A simplified description of the same can be found in the paper by Fan, Hwang, and Tillman [23]. To use this method for the minimization of a function of n variables, it is necessary to set up a Simplex of

(n + 1) vertices, that is, to select (n + 1) trial points in n-dimensional space. The values of the objective function are then calculated at each of these points. By comparing the objective function value of these (n + 1) points, the point with the highest value is replaced by a point with a lower value of the objective function. A lower function value is selected by a reflection, expansion or contraction operation through the centroid of the current Simplex. If none of these operations are successful, the Simplex is reduced in size around the lowest functional value before starting the next iteration.

For a two-dimensional problem where the objective function $y = f(x_1,x_2)$ is to be minimized, a Simplex with $(n + 1) = 3$ points is required. Let P'_1, P'_2 and P'_3 be three trial points, such that $y_1 < y_2 < y_3$. where

y_1 = objective function value at point 1

y_2 = objective function value at point 2

y_3 = objective function value at point 3

The various operations to get a point with lower objective function value are defined as

$$\text{Reflection:} \quad P'_5 = P'_4 + \alpha(P'_4 - P'_3) \tag{II-1}$$

$$\text{Expansion:} \quad P'_6 = P'_4 + \eta(P'_5 - P'_4) \tag{II-2}$$

$$\text{Contraction:} \quad P'_7 = P'_4 + \gamma(P'_3 - P'_4) \tag{II-3}$$

where

P_4' = centroid of points P_1' and P_2', in general the centroid of a set of n points in a simplex is

$$P_c' = \sum_{i=1}^{n} (P_i/n) \qquad \text{(II-4)}$$

P_5' = reflection of the point P_3' with respect to P_4'

P_6' = expansion of P_5'

P_7' = contraction of the highest valued point P_3' with respect to P_4'

The values of the coefficients, α, η and γ, considered best by Nelder and Mead [5] are

$$\alpha = 1, \ \eta = \frac{1}{2}, \text{ and } \gamma = 2$$

However the best values of these coefficients may be different for different problems and should be found by experience. The details of the procedure and a flow diagram to facilitate the description has been given by Fan, Hwang, and Tillman [23].

One stopping criterion is the occurrence of five consecutive values of the objective function which are considered "equal". Another stopping criterion would be to compare the "standard error" of the y's in the form

$$\left(\sum_{i=1}^{n+1} (y_i - \bar{y})/n \right)^{1/2} \qquad \text{(II-5)}$$

with a preset value and stop the program when it falls below this value [5].

The success in employing the criterion for stopping computations depends upon the simplex not becoming too small in relation to the curvature of the surface as the final minimum is reached. The reasoning behind this criterion is given by Nelder and Mead [5]. They have suggested that in statistical problems where one is concerned with finding the minimum of a negative likelihood, the curvature near the minimum gives the information on unknown parameters. If the curvature is slight, the sampling variance of the estimates will be large and there is no sense in finding the coordinate of the minimum very accurately, while, if the curvature is very large, there is justification for pinning down the minimum more exactly.

Chen [24] describes a modified technique, called the "modified simplex method." This method is similar to the simplex method except that if the expansion step produces a new minimum, then the centroid of the simplex for the next iteration is moved closer to this improved value to allow more rapid movement in the favorable direction. The computer program that is used to estimate the parameters is written by Chen [24], the only difference being that subroutine RKGS [11] was coupled with this program to numerically integrate the system of non-linear differential equations which constitute the mathematical model for the tower system.

Figure II-1 shows the flow diagram which must be coupled with the one given by Fan, Hwang, and Tillman [23] to estimate parameters in a multistage tower fermenter model.

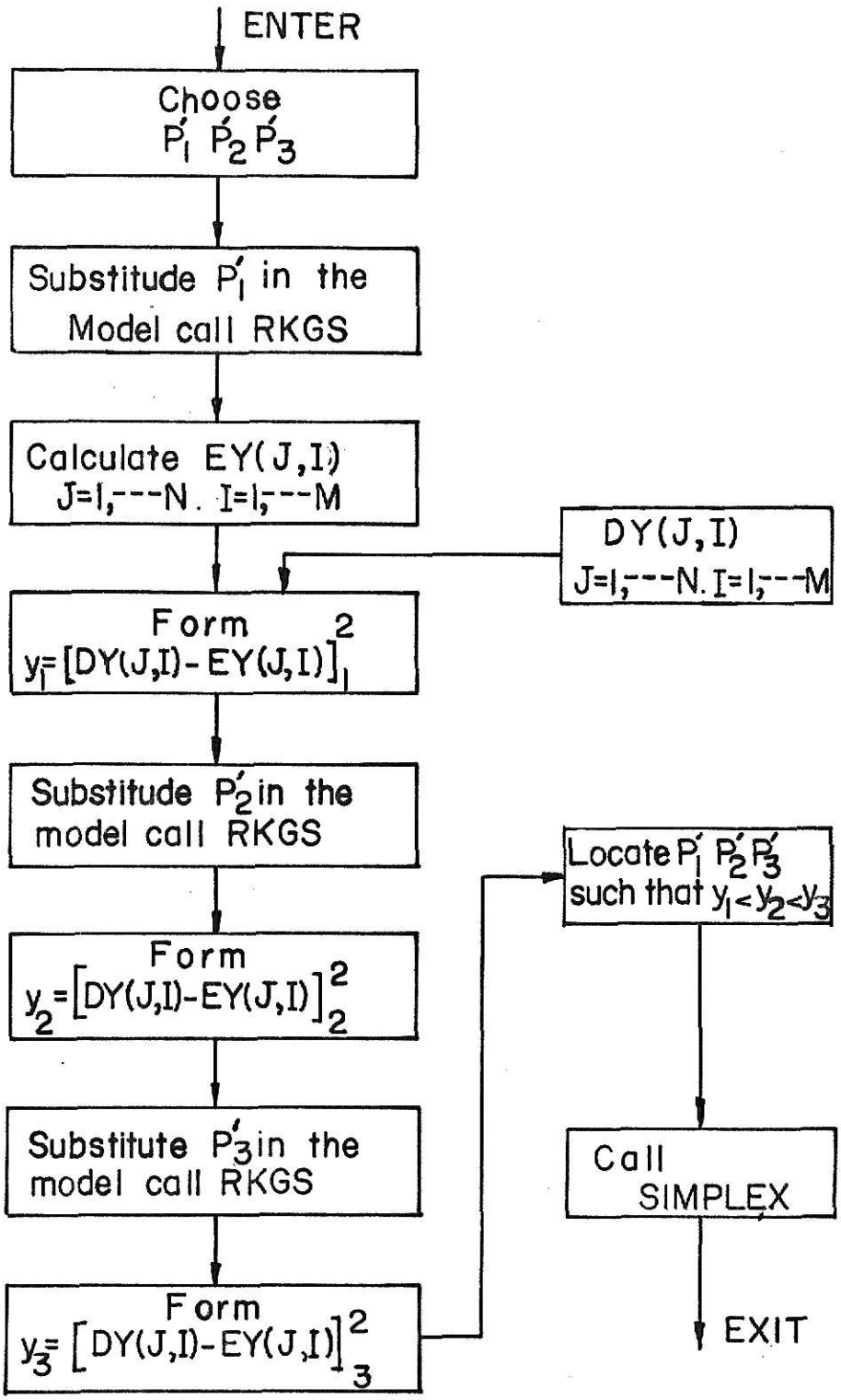

Fig. II-1. Flow diagram for estimation of parameters in tower system.

Chapter 3

ANALYSIS OF FLOW BEHAVIOR IN A CONTINUOUS MULTISTAGE TOWER FERMENTOR: ESTIMATION OF PARAMETERS

3.1 INTRODUCTION

Many types of continuous reactors, which could be used for continuous fermentation reactions, were proposed by Herbert [1]. The behavior of continuous multistage tower fermentors cannot, in general, be simulated by either of the ideal continuous reactor models, plug-flow or completely backmixed, and, therefore, a nonideal model such as a "mixed model" [2] must be employed. The tower system considered here is assumed to be constructed such that compartments are separated by perforated plates. Complete mixing of the liquid phase was assumed in each compartment. Interstage backflow was included in the model. Rotating disc contactors [3], and multistage fluid bed reactors [4] are examples of industrial flow systems which use a cascade of stirred vessels with back flow between stages.

A tracer study is often useful in investigating the flow behavior of a column fermentor. An understanding of the flow behavior and the growth kinetics is necessary if one wishes to develop a simulation model for the fermentor. In the tower fermentor, flow models can be developed for each phase (gas, liquid, and solid). The model developed here is for the liquid phase.

A tracer study was undertaken at the Department of Chemical Engineering at the University of Pennsylvania by Humphrey and his associates, using the

eight stage tower fermentor described previously [5]. Experimental measurements of tracer concentration vs. time were recorded for the first three stages and the eighth stage to determine the extent of backflow in the tower system. In this chapter, the parameter estimation techniques which can be used to estimate the extent of backflow are presented. One set of their experimental data was used to illustrate uses of the techniques.

3.2 MATHEMATICAL MODEL

An eight stage tower fermentor with feed introduced at the second stage was used for the tracer study [5]. Experimental measurements were made using salt as a tracer for the first three stages and the eighth stage only. The flow model of the tower fermentor is shown in Figure 1. A dotted boundary is used to designate the subsystem consisting of the first three stages from which the data employed in estimating the parameters were obtained. Complete mixing was assumed in each compartment and backflow from each stage was included in the model. One of the important factors considered was the rate of backflow, and this will be represented by $F_i\gamma_i$, where γ_i is the fraction of the total flow F_i from the ith stage which appears in the backflow.

The unsteady state tracer material balances around each stage of the tower fermentor are as follows:

$$\text{First stage:} \quad V_1 \frac{dC_1}{dt} = F_2\gamma_2 C_2 - F_1 C_1 \tag{1}$$

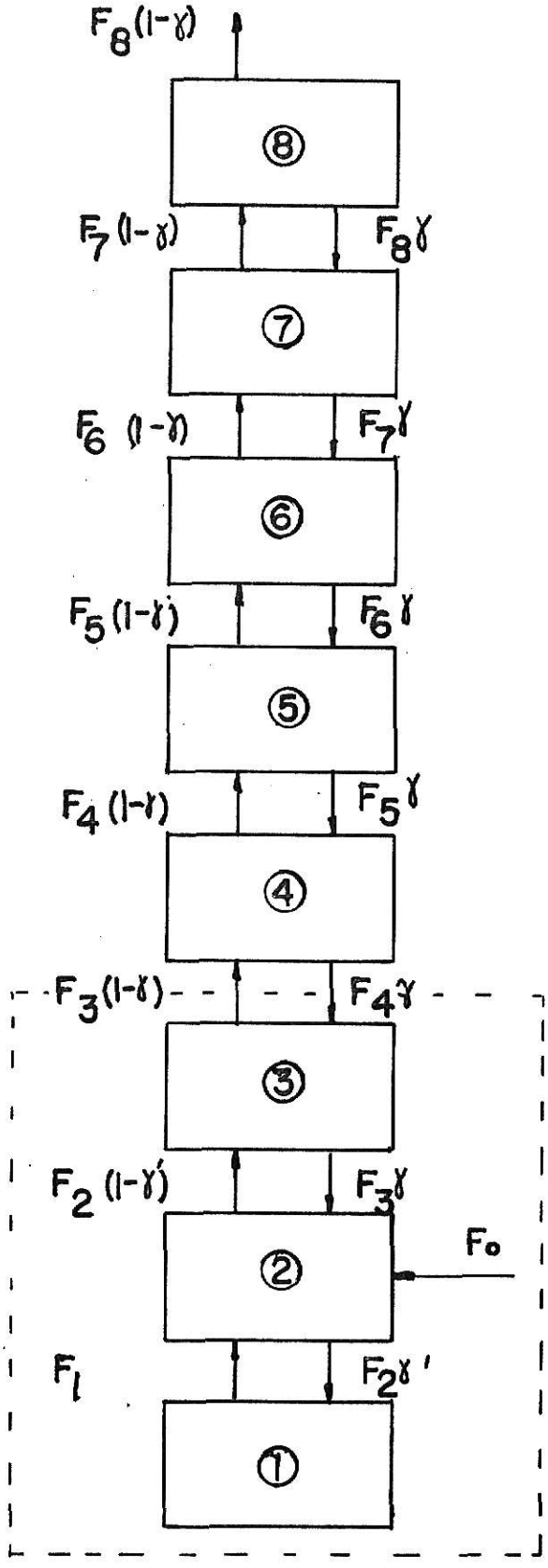

Fig. 1.. Flow model of the tower fermentor.

Second stage: $$V_2 \frac{dC_2}{dt} = F_1C_1 + F_3\gamma_3C_3 - F_2C_2 \quad (2)$$

Stages 3 to 7: $$V_i \frac{dC_i}{dt} = F_{i-1}(1 - \gamma_{i-1})C_{i-1} + F_{i+1}\gamma_{i+1}C_{i+1} - F_iC_i,$$

$$i = 3, 4, 5, 6, 7 \quad (3)$$

Eighth stage: $$V_8 \frac{dC_8}{dt} = F_7(1 - \gamma_7)C_7 - F_8C_8 \quad (4)$$

The initial conditions are

$c_i = 0$ at $t = 0$, $i = 1, 3, 4, 5, 6, 7, 8$

$c_2 = 0$ finite at $t = 0$

During the tracer experiment, the liquid flow rate was constant. The steady state material balances are as follows:

Overall balance: $$F_0 = F_8(1 - \gamma_8) \quad (5)$$

Stage 1: $$F_2\gamma_2 - F_1 = 0 \quad (6)$$

Stage 2: $$F_1 + F_3\gamma_3 - F_2 = - F_0 \quad (7)$$

Stages 3 to 7: $$F_{i-1}(1 - \gamma_{i-1}) + F_{i+1}\gamma_{i+1} - F_i = 0,$$

$$i = 3, 4, 5, 6, 7 \quad (8)$$

Stage 8: $$F_7(1 - \gamma_7) - F_8 = 0 \quad (9)$$

where

V_i = volume of the ith stage (lit.), i = 1, 2, ..., 8

C_i = tracer concentration in the ith stage, (gm./c.c.), i = 1, 2, ..., 8

F_i = flow out of stage i, (lit./hr.), i = 1, 2, ..., 8

F_0 = feed flow rate to the second stage, (lit./hr.)

t = time, (hrs)

γ_i = backflow coefficient for ith stage (i = 2, 3, ..., 8)

Two simplifications were made in the above mathematical model. First of all the individual stage volumes are assumed to be equal i.e. $V_1 = V_2 = V_3 = ... = V_8 = V$. The backflow coefficients for stages three through eight are assumed to be equal i.e. $\gamma_3 = \gamma_4 = \gamma_5 = ... = \gamma_8 = \gamma$. The backflow coefficient for stage two may differ from γ because the feed is introduced to the second stages; γ_2 will be represented by γ'.

3.3 ANALYSIS OF EXPERIMENTAL DATA

Experimental measurements were made for tracer concentrations in the first three stages and eighth stage at different times as shown in Table 1. One of the important aspects to note is the fact that the tracer concentrations C_1, C_2, C_3, and C_8 were measured at different instants of time. The time lag between measurements of C_1 and C_2 is practically constant.

One of the objectives of this tracer study was to compare the backflow from the third stage to the second stage with that from the second stage to the first stage. This can be done by estimating the backflow parameters

Table 1. Tracer Data for Eight Stage Tower Fermentor

Time	C_1	Time	C_2	Time	C_3	Time	C_8
0.0330	0.0000044	0.0666	0.0003300	0.1166	0.0001200	0.1333	0.0000002
0.2000	0.0002200	0.2333	0.0001650	0.2666	0.0001120	0.3000	0.0000013
0.3660	0.0002750	0.4000	0.0000980	0.4333	0.0001120	0.4666	0.0000074
0.5330	0.0003100	0.5666	0.0000600	0.6000	0.0000860	0.6333	0.0000164
0.7000	0.0003300	0.7333	0.0000400	0.7666	0.0000650	0.8000	0.0000300
0.8667	0.0003350	0.9000	0.0000430	0.9333	0.0000500	0.9666	0.0000420
1.0330	0.0003300	1.0667	0.0000210	1.1166	0.0000390	1.1333	0.0000480
1.2000	0.0003150	1.2333	0.0000175	1.2667	0.0000315	1.3000	0.0000540
1.3666	0.0003060	1.4000	0.0000145	1.4333	0.0000275	1.4666	0.0000540
1.5333	0.0002900	1.5666	0.0000115	1.6000	0.0000210	1.6333	0.0000545
1.7000	0.0002750	1.7333	0.0000100	1.7667	0.0000180	1.8000	0.0000525
1.8667	0.0002600	1.9000	0.0000086	1.9333	0.0000150	1.9666	0.0000480
2.0333	0.0002450	2.0667	0.0000074	2.1000	0.0000130	2.1333	0.0000400
2.2000	0.0002300	2.2333	0.0000064	2.2667	0.0000112	2.3000	0.0000390
2.3666	0.0002250	2.4000	0.0000058	2.4333	0.0000100	2.4666	0.0000360
2.5333	0.0002050	2.5667	0.0000051	2.6000	0.0000086	2.6333	0.0000320
2.7000	0.0001950	2.7333	0.0000047	2.7667	0.0000078	2.8166	0.0000287
2.8667	0.0001820	2.9000	0.0000041	2.9333	0.0000068	2.9666	0.0000255
3.0333	0.0000017	3.0667	0.0000037	3.1000	0.0000059	3.1333	0.0000225
3.2000	0.0001600	3.2333	0.0000035	3.2667	0.0000053	3.3000	0.0000200
3.3666	0.0001470	3.4000	0.0000031	3.4333	0.0000047	3.4666	0.0000172
3.5500	0.0001370	3.5833	0.0000028	3.6166	0.0000043	3.6500	0.0000142
3.7000	0.0001300	3.7333	0.0000026	3.7667	0.0000039	3.8000	0.0000135
3.8667	0.0001200	3.9167	0.0000026	3.9500	0.0000035	3.9833	0.0000118
4.1667	0.0001070	4.2000	0.0000021	4.2333	0.0000029	4.2666	0.0000096
4.4167	0.0000970	4.4500	0.0000018	4.4833	0.0000025	4.5166	0.0000080
4.6667	0.0000880	4.7000	0.0000016	4.7333	0.0000022	4.7666	0.0000068
5.0000	0.0000780	5.0333	0.0000014	5.0667	0.0000018	5.1000	0.0000053
5.3333	0.0000660	5.3667	0.0000012	5.4000	0.0000015	5.4333	0.0000043
5.6666	0.0000600	5.7000	0.0000009	5.7333	0.0000012	5.7666	0.0000034
6.0000	0.0000520	6.0333	0.0000008	6.0667	0.0000010	6.1000	0.0000028
6.3333	0.0000450	6.3667	0.0000007	6.4000	0.0000009	6.4333	0.0000022
6.6666	0.0000410	6.7000	0.0000006	6.7333	0.0000007	6.7666	0.0000018
7.0000	0.0000350	7.0333	0.0000005	7.0667	0.0000007	7.1000	0.0000015
7.5000	0.0000290	7.5333	0.0000004	7.5666	0.0000005	7.6000	0.0000012
8.0000	0.0000230	8.0333	0.0000004	8.0667	0.0000004	8.1000	0.0000009
9.0000	0.0000130	9.0333	0.0000003	9.0667	0.0000003	9.1000	0.0000006

appearing in the mathematical model. Since the initial tracer concentration in the second stage is a finite unknown quantity, it was considered as a third unknown parameter.

Method I:

Two distinct approaches were used to identify the unknown parameters. In the first one (Method I), the differential mathematical model consisting of Equations (1) through (4) was considered and the parameters were estimated using a modified simplex pattern search technique [6] together with the numerical integration subroutine RKGS [7]. The fact, that the independent variable time is different for each set of experimental data makes it rather difficult to estimate the parameters using a differential model. Simplifications were made at this point to get a satisfactory solution to this problem. The tracer concentrations in the first, second, third, and eighth stages were plotted on an enlarged scale and numerical values at equal intervals of time were tabulated from the smooth curves. Linear interpolation was used to obtain the tracer concentrations, at desired time values, if otherwise not available from the experimental measurements. The idea behind this was to transform the experimental data such that the usual parameters estimation techniques for differential models can be applied.

The steady state material balance equations may be solved to obtain flow rates from individual stages as a function of unknown parameters γ, γ', and $C_2(0)$ and known flow rate F_0. Equations (10) through (17) represent the flows F_1 through F_8 as obtained by solving Equations (6) through (9).

$$F_1 = F_0\gamma'\left[\frac{1}{(1-\gamma)^7} - \frac{5\gamma}{(1-\gamma)^6} + \frac{6\gamma^2}{(1-\gamma)^5} - \frac{\gamma^3}{(1-\gamma)^4}\right] \quad (10)$$

$$F_2 = F_0\left[\frac{1}{(1-\gamma)^7} - \frac{5\gamma}{(1-\gamma)^6} + \frac{6\gamma^2}{(1-\gamma)^5} - \frac{\gamma^3}{(1-\gamma)^4}\right] \quad (11)$$

$$F_3 = F_0\left[\frac{1}{(1-\gamma)^6} - \frac{4\gamma}{(1-\gamma)^4} + \frac{3\gamma^2}{(1-\gamma)^4}\right] \quad (12)$$

$$F_4 = F_0\left[\frac{1}{(1-\gamma)^5} - \frac{3\gamma}{(1-\gamma)^4} + \frac{\gamma^2}{(1-\gamma)^3}\right] \quad (13)$$

$$F_5 = F_0\left[\frac{1}{(1-\gamma)^4} - \frac{2\gamma}{(1-\gamma)^3}\right] \quad (14)$$

$$F_6 = F_0\left[\frac{1}{(1-\gamma)^3} - \frac{\gamma}{(1-\gamma)^2}\right] \quad (15)$$

$$F_7 = F_0\left[\frac{1}{(1-\gamma)^2}\right] \quad (16)$$

$$F_8 = F_0\left[\frac{1}{(1-\gamma)}\right] \quad (17)$$

These flows can be substituted back into the differential model to reduce the number of unknown parameters. Since the experimental measurements

are available for first three stages and the eighth stage only, it is assumed that the experimental tracer concentrations for fourth, fifth, sixth, and seventh stages are the same as predicted by the differential model. With the above simplifying assumptions, theoretically the backflow parameters and the unknown initial condition ($C_2(0)$) can be estimated using parameter estimation techniques for differential models. The mathematical objective function to be minimized is given by Equation (18)

$$S = \sum_{i=1}^{N} (EC_{1i} - C_{1i})^2 + \sum_{i=1}^{N} (EC_{2i} - C_{2i})^2 + \sum_{i=1}^{N} (EC_{3i} - C_{3i})^2$$

$$+ \sum_{i=1}^{N} (EC_{8i} - C_{8i})^2 \qquad (18)$$

where

S = objective function value

EC_1 = experimental tracer concentration in stage 1, (gm./c.c.)

EC_2 = experimental tracer concentration in stage 2, (gm./c.c.)

EC_3 = experimental tracer concentration in stage 3, (gm./c.c.)

EC_8 = experimental tracer concentration in stage 8, (gm./c.c.)

N = number of data points

Method II:

The alternative approach (Method II) which was employed to solve this problem is to use a partial model consisting of Equations (1) and (2) only.

Substituting Equation (6) in Equation (1) one gets

$$V \frac{dC_1}{dt} = F_1(C_2 - C_1) \tag{19}$$

Substituting Equation (7) in Equation (2), one gets

$$V \frac{dC_2}{dt} = F_1 C_1 - \frac{F_1 C_2}{\gamma_2} + \frac{F_1 C_3}{\gamma_2} - F_1 C_3 - F_0 C_3 \tag{20}$$

Rearranging Equations (19) and (20), one gets

$$\frac{dC_1}{dt} = \frac{F_1(C_2 - C_1)}{V} \tag{21}$$

$$\frac{dC_2}{dt} = \frac{F_1}{V}(C_1 - C_3) + \frac{F_2}{V}(C_3 - C_2) - \frac{F_0}{V} C_3 \tag{22}$$

Equations (21) and (22) have the following initial conditions.

$C_i = 0$ at $t = 0$ $\quad i = 1, 3$

$C_2 =$ finite at $t = 0$

The mathematical model consisting of Equations (21) and (22) can be used to estimate the above mentioned parameters if an expression for C_3 is available. In this method a polynomial equation and the experimental data were used to find a suitable expression for C_3. An unsteady state balance for C_3 cannot be written, as was done for tracer concentrations C_1 and C_2, because of the fact that C_4 (tracer concentration in

the fourth stage) starts appearing in the material balances and experimental data is not available for C_4. Thus, parameter estimation techniques were used to develop a polynomial equation for C_3 in Method II. Statistical techniques described in Chapter 4 were used to select the degree of the polynomial to be used for approximating the tracer concentrations in the third stage.

With the polynomial equation for C_3 it is possible through integration to analytically reduce the differential model represented by Equation (21) and (22) to an algebraic model. Equations (21) and (22) may be integrated with the given initial conditions and a fourth order polynomial for C_3,

$$C_3 = A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4 \tag{23}$$

where A_0, A_1, A_2, A_3, A_4 are constants to be estimated from the experimental data on C_3. The results of the integration are

$$C_1 = \frac{1}{(\lambda_1 - \lambda_2)} \left[\frac{C_2(0)F_1}{V}\right] + (\lambda_2 K_0 - K_1)\Big] e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left(\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right) + K_0\right] e^{\lambda_2 t}$$

$$+ (K_0 + K_1 t + K_2 t^2 + K_3 t^3 + K_4 t^4) \tag{24}$$

$$C_2 = \frac{1}{(\lambda_1 - \lambda_2)} \left[\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right] e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\} + K_0\right] e^{\lambda_2 t}$$

$$+ \frac{V}{F_1} \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\}\right] \lambda_1 e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\} + K_0\right] \lambda_2 e^{\lambda_2 t}$$

$$+ \frac{V}{F_1} \left[K_1 + 2K_2 t + 3K_3 t^2 + 4K_4 t^3\right] \qquad (25)$$

where

$$\lambda_1 = \frac{- \left(1 + \frac{F_2}{F_1}\right) + \sqrt{\left(1 + \frac{F_2}{F_1}\right)^2 - 4 \frac{V}{F_1} \left(\frac{F_2 - F_1}{V}\right)}}{2 \frac{V}{F_1}}$$

$$\lambda_2 = \frac{- \left(1 + \frac{F_2}{F_1}\right) - \sqrt{\left(1 + \frac{F_2}{F_1}\right)^2 - 4 \frac{V}{F_1} \left(\frac{F_2 - F_1}{V}\right)}}{2 \frac{V}{F_1}}$$

and K_0, K_1, K_2, K_3, K_4 are constants. The form of these constants is shown in Appendix II.

The computational procedure used with Method II is as follows:

(1) Find constants A_0, A_1, A_2, A_3, A_4 in equation (23) by "Least Squares Curve Fitting." For this part the objective function to be minimized is

$$S = \sum_{i=1}^{N} [EC_{3i} - (A_0 + A_1 t_i + A_2 t_i^2 + A_3 t_i^3 + A_4 t_i^4)]^2 \qquad (26)$$

where N is the number of data points for C_3.

(2) Find the parameters F_1, F_2 and $C_2(0)$ so as to minimize the following objective function,

$$S = \sum_{i=1}^{N} |EC_1^i - C_1^i| + \sum_{i=1}^{N} 0.5|EC_2^i - C_2^i| + \sum_{i=1}^{N} 0.5(EC_2^i - C_2^i)^2 \qquad (27)$$

(3) Simulate the system using the optimal estimates of the parameters to predict the tracer concentrations in individual stages. C.S.M.P. (continuous systems modeling program) [8] or RKGS [7] can be employed for this.

3.4 CRITERIA FOR ESTIMATION: The parameter estimates are determined by minimizing a selected criterion function, S, which is a measure of the difference between the measured experimental value of the response and the predicted response from the mathematical model. Various criterion functions differ in their appropriateness for engineering interpretation and ease of determining optimum parameter estimates.

The method of maximum likelihood is a general method for parameter estimation [9]. Box [10] has presented a discussion for selecting a

criterion function based on this approach. The conventional sum of squares criterion function is widely used and allows sufficient statistical analysis. This criteria seems to be more favorable when the errors can be assumed to be uncorrelated and normally distributed with mean zero and variance σ^2. The objective function in this case can be written as follows:

$$S(\bar{p}) = \sum_{i=1}^{N} [y_i - f(\bar{x}_i, \bar{p})]^2 \tag{28}$$

where y_i are the dependent variables. The subscript i denotes the ith experiment in a total of N experiments. $\bar{x}_i = (x_{1i}, x_{2i}, \ldots, x_{Mi})$ are the independent variables and $\bar{p} = (p_1, p_2, \ldots, p_p)$ are the parameters. Weighted sum of least squares where

$$S = \sum_{i=1}^{N} [y_i - f(\bar{x}_i, \bar{p})W_i]^2 \tag{29}$$

is frequently a more useful criterion for parameter estimation. This arises when the level of experimental error is dependent on the values of the independent variables, $\bar{x}_i$. Hence the same variance σ_i^2, for all i may be an unacceptable assumption.

Another approach is to use the sum of absolute differences as the criterion function [11].

$$S = \sum_{i=1}^{N} |y_i - f(\bar{x}_i, \bar{p})| \tag{30}$$

This criterion puts equal weights on all differences.

For Method I, the conventional sum of squares was the only criterion function used in this work; however, for Method II several criterion functions were considered. The conventional sum of squares was the first criterion function which was selected to get the optimum estimates of backflow parameters. It was found that the optimal estimates obtained from this approach overestimated the experimental response curves of tracer concentrations in various tanks. On the other hand, the use of sum of absolute differences as the criterion function underestimated them. However, a weighted combination of these two criterion functions gave satisfactory results. This criteria is represented by Equation (27).

3.5 RESULTS AND DISCUSSION: Modified sequential simplex pattern search [6] was used to estimate the unknown parameters in the differential model (Method I). Subroutine RKGS [7] was coupled with this search technique in order to numerically integrate the differential equations. The computer program for simplex pattern search was written by Chen [12]. Using the estimated values of the parameters the set of linear algebraic Equations (6) through (9) is solved to obtain the flows out from each stage. This information was fed back into the unsteady system of differential equations and the whole system was simulated using C.S.M.P. (continuous systems modeling program) [8] to predict the values of the response. This process was repeated until the optimal estimates of the parameters are obtained. The computer program for this simulation is included in Appendix I. Figures 3 through 6 indicate the fit for the experimental data for tracer concentrations in stages 1, 2, 3 and 8, respectively, using this approach.

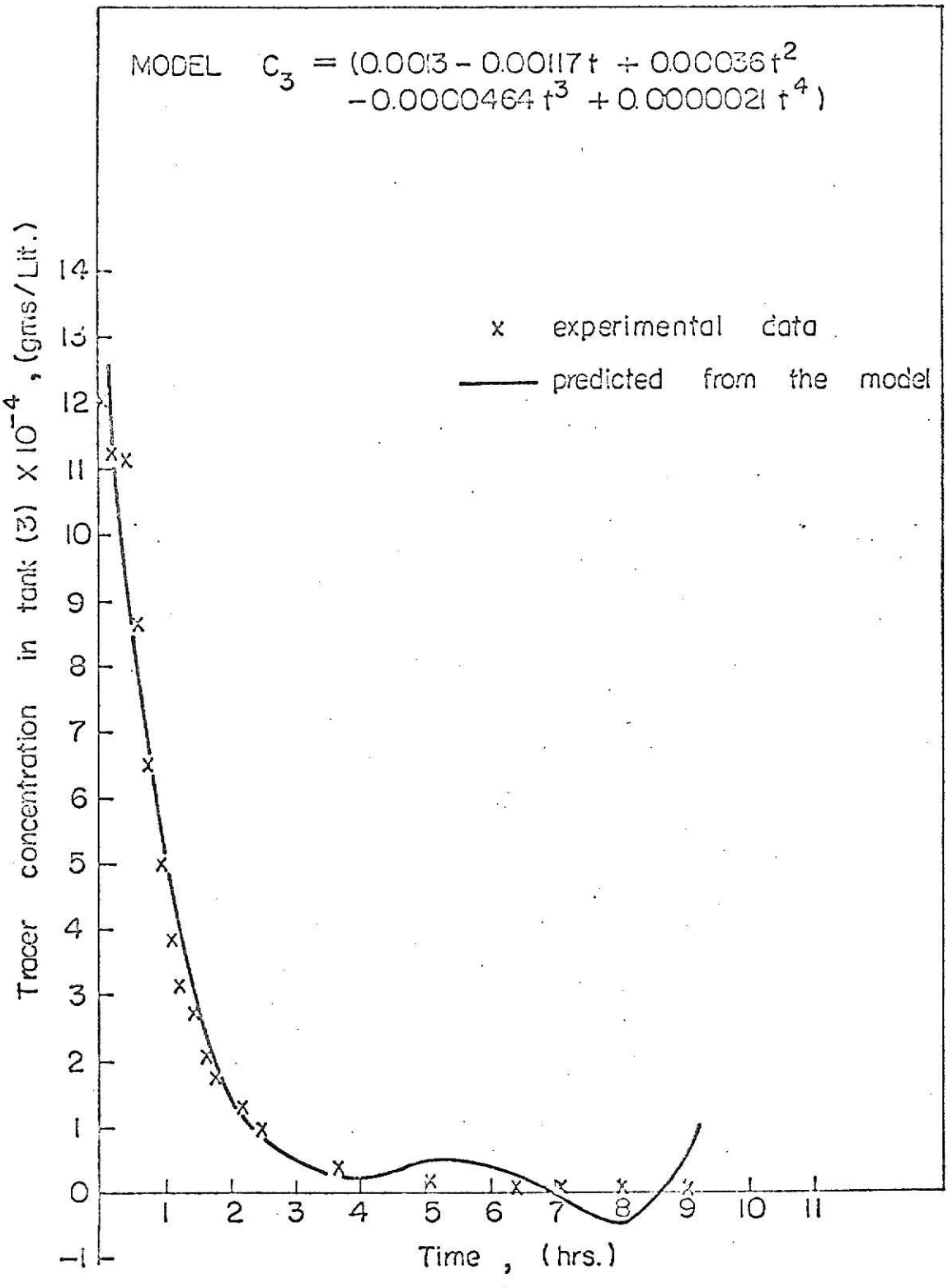

Fig. 2. Tracer concentration in tank (3) vs time.

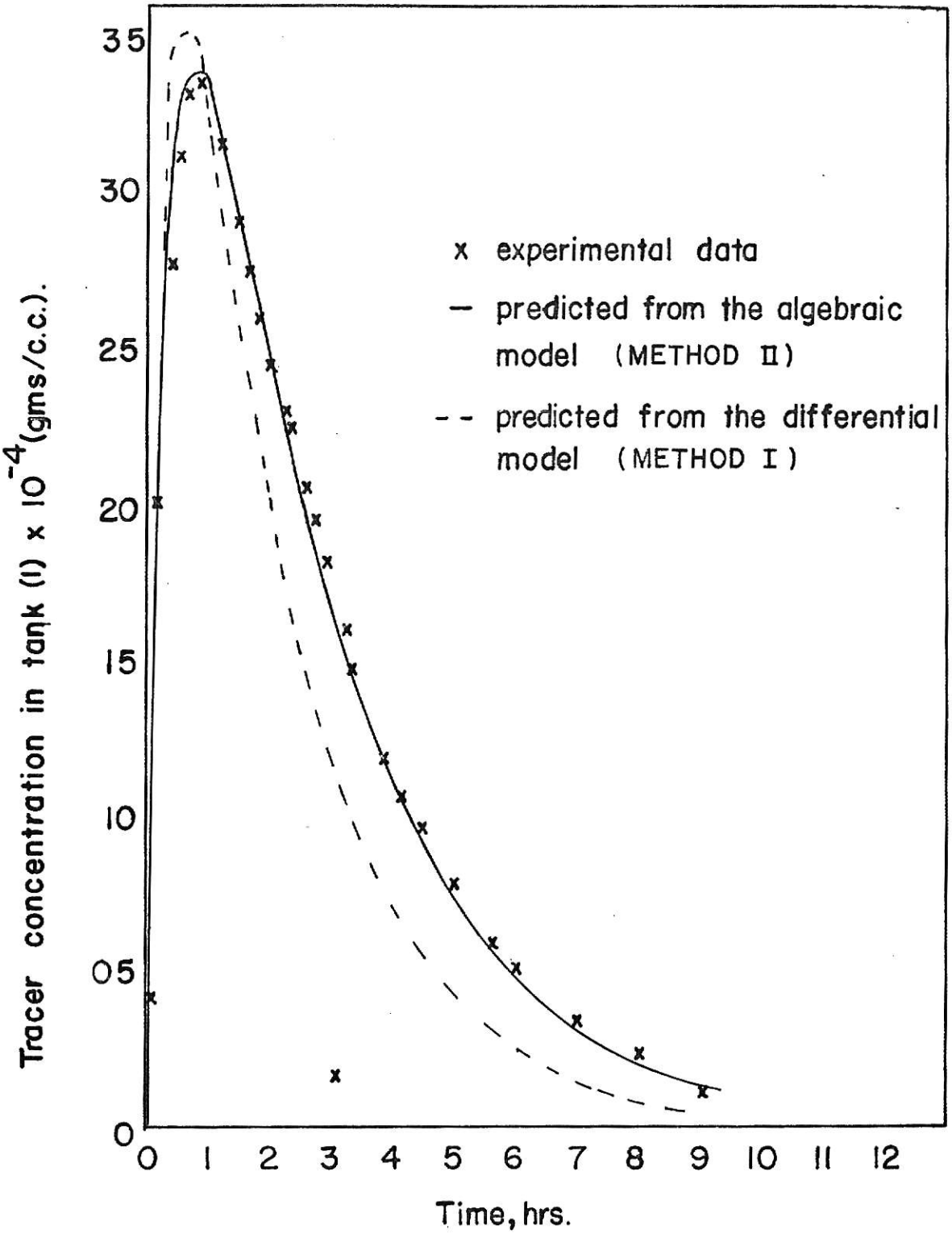

Fig. 3. Tracer concentration in tank (I) vs time.

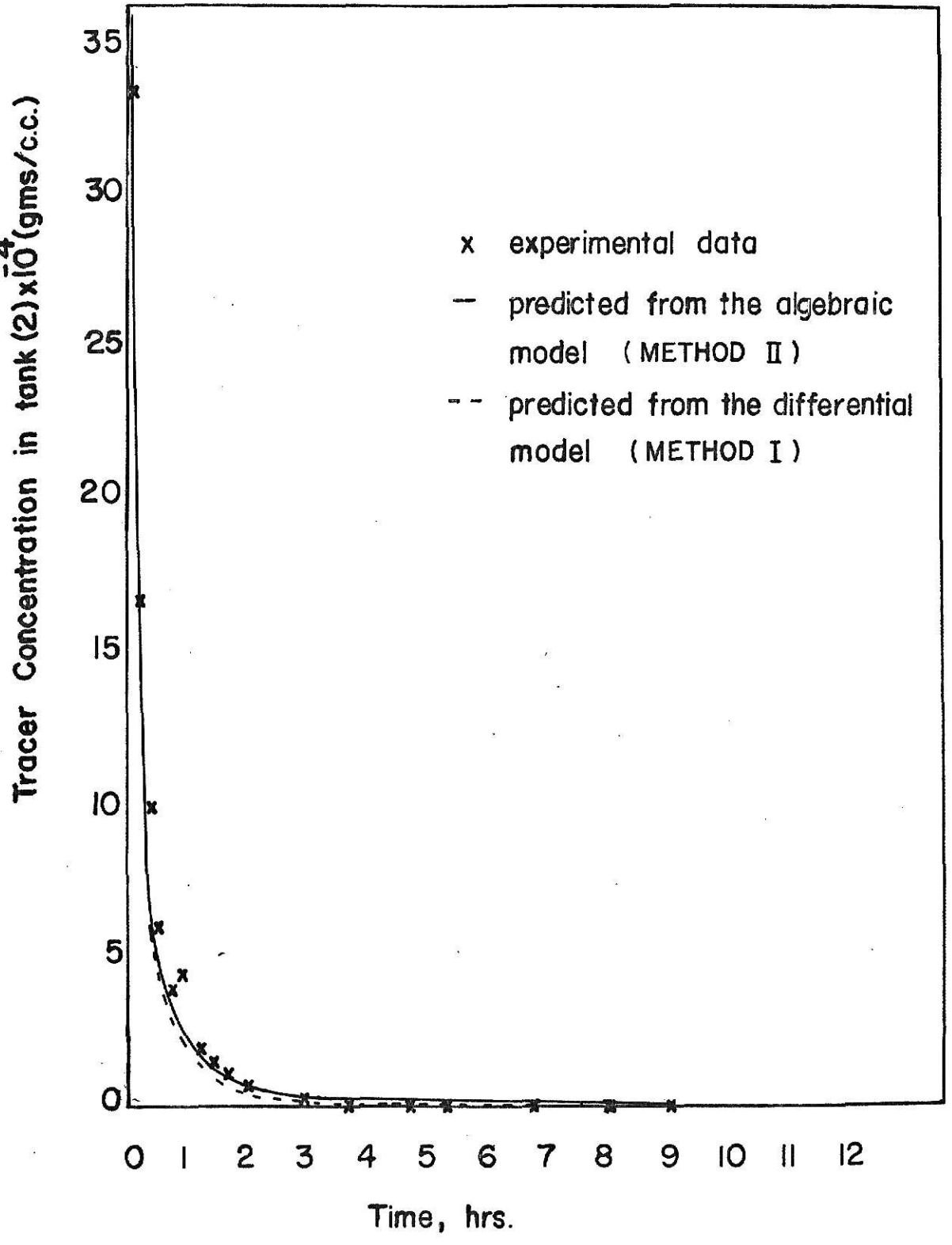

Fig. 4. Tracer concentration in tank (2) vs time.

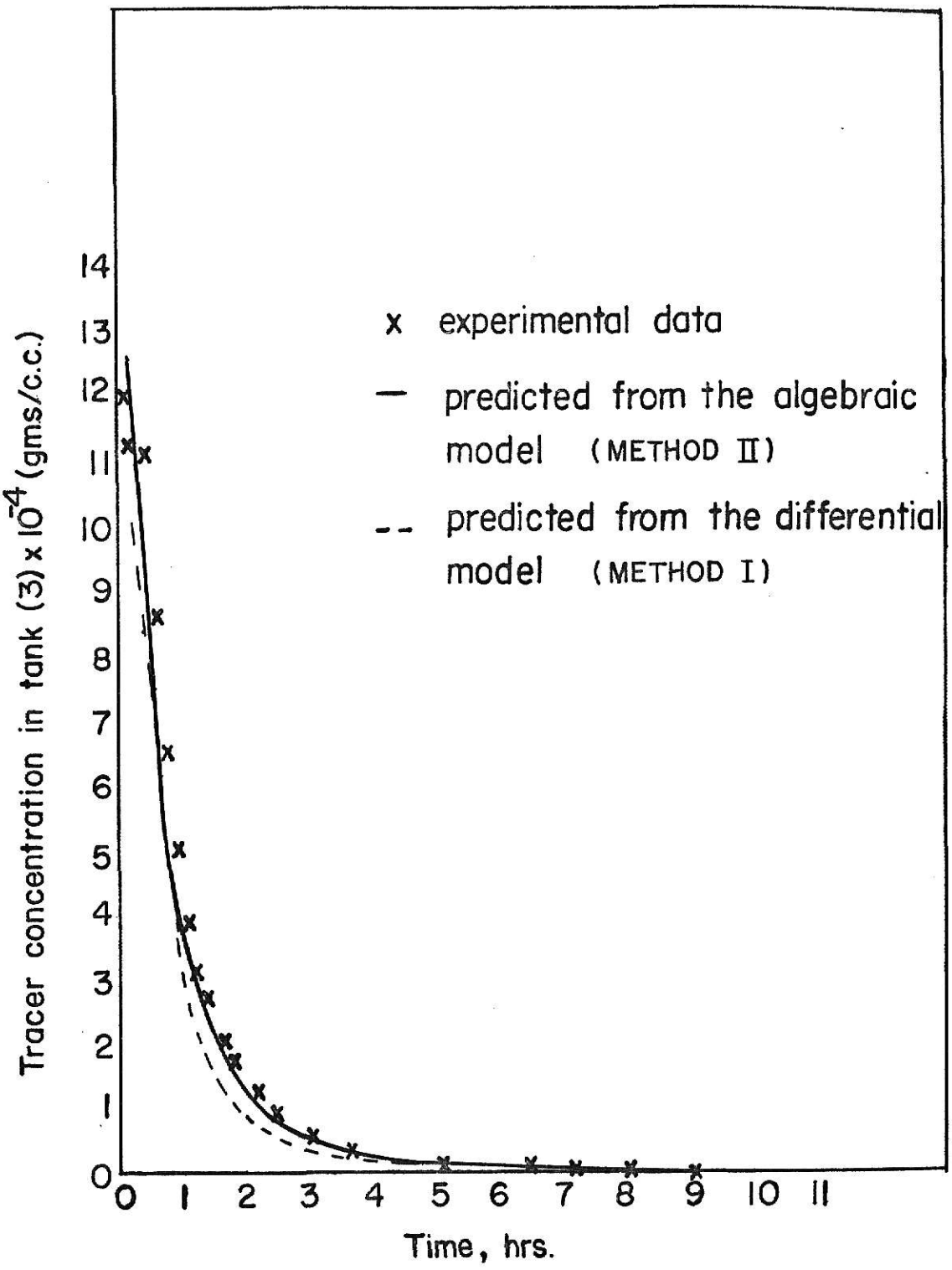

Fig. 5. Tracer concentration in tank (3) vs. time.

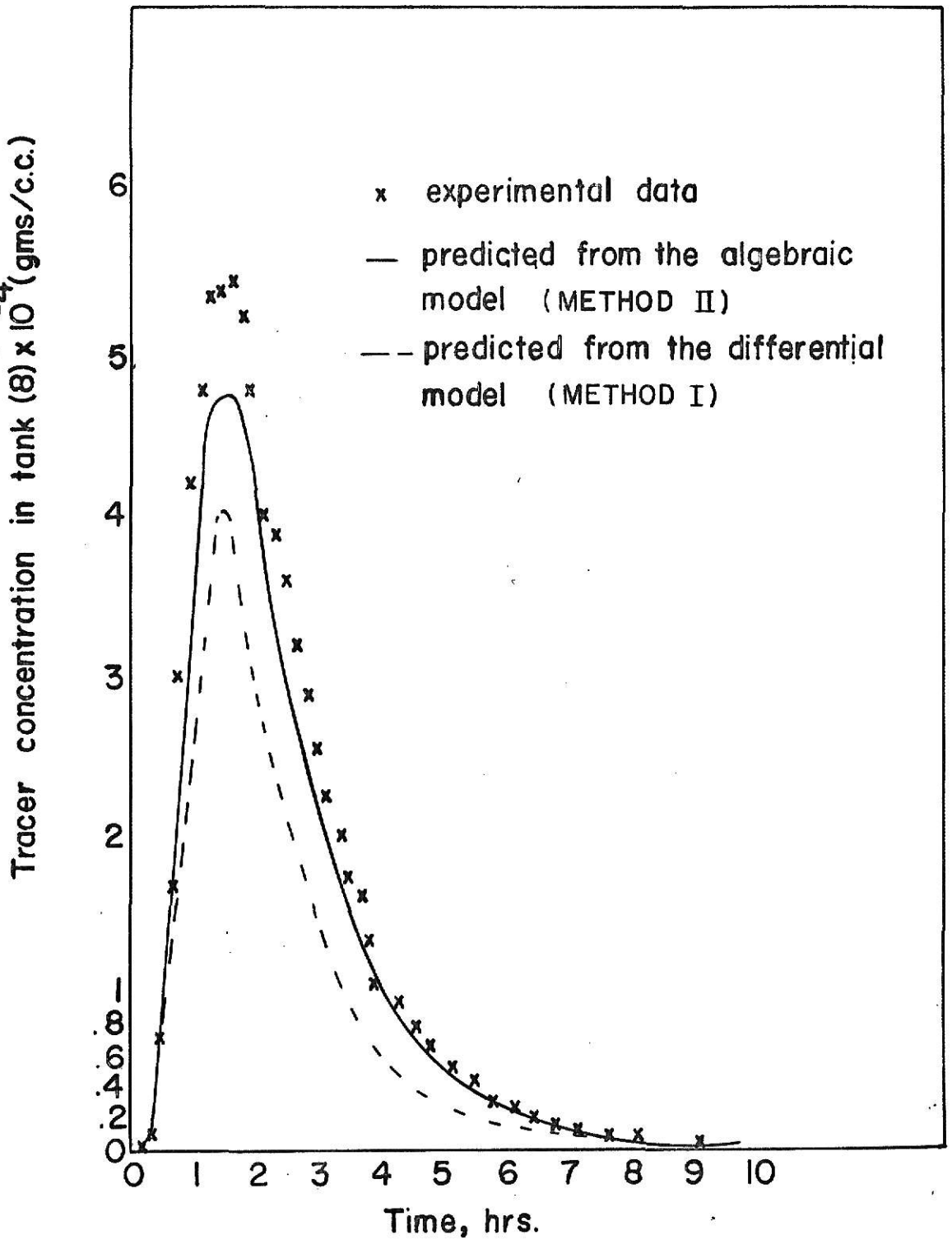

Fig.6. Tracer concentration in tanks (8) vs. time.

Table 2. Estimated Values of the Constants in the Polynomial

Parameter	Mean	Std. Deviation
A_0	0.13914 x 10-2	0.5691×10^{-4}
A_1	-0.11796×10^{-2}	0.2637×10^{-4}
A_2	0.36202×10^{-3}	0.4463×10^{-5}
A_3	-0.46428×10^{-4}	0.2470×10^{-6}
A_4	0.90998×10^{-5}	0.3589×10^{-4}

Alternatively the simplex pattern search [6] was employed to estimate the unknown parameters using the algebraic model (Method 2) consisting of Equations (24) and (25). In the first part of the estimation Bard's program [13] for "Single equation least square curve fitting" was used. The central function of this program is to maximize a selected criterion function. There are two alternative subroutines available for this purpose. Gauss-Newton method with modifications by Greenstadt-Eisenpress [14], Bard [13] and Carroll [15] was used for maximization. Since the problem considered is a minimization problem, the negative of the selected criterion function was maximized. The estimated values of the constants A_0, A_1, A_2, A_3, A_4 and their standard deviations are shown in Table 2.

These constants were substituted back into Equation (23) and the values of C_3 were calculated at the time values (t) at which the experimental data was taken. Figure 2 shows the curves for C_3 predicted from experimental data. It can be seen that experimental data and the predicted values match well. However, at this point one does not have any theoretical basis for assuming a fourth order polynomial for C_3. If a higher degree polynomial is assumed one might sometimes overestimate the parameters and if on the other hand the degree of the polynomial is less one might face the difficulty of underestimation. Hence as an extension of this work, polynomials of varying degrees will be assumed and statistical methods will be employed to investigate the degree of polynomial to be used in our estimations. It should be realized that this is an important step in the estimation procedure as the accuracy of the parameter estimates depends on the tracer concentration in the third tank (C_3).

In the next step the modified simplex pattern search [6] was used to estimate the parameters $C_2(0)$, F_1 and F_2, using Equations (24) and (25). Plots of tracer concentrations in stages 1, 2, 3, and 8 as obtained from the experimental data and mathematical model are shown in Figures 3 through 6. Once again the dynamic system of differential equations was simulated using C.S.M.P. [8] to predict the values of response. Comparison of results obtained from the differential model and the algebraic model indicate that a better fit for the experimental data can be obtained when the parameters are estimated using an algebraic model, and a partial system. The optimal results obtained from the algebraic and differential models are compared in Table 3. The ratio $(F_3\gamma/F_2\gamma')$ represents the relationship between the flows from the third stage to second stage and second stage to first stage.

The application of parameter identification techniques to estimate the extent of backflow appears to give useful information for fermentor design. If sufficient experimental information is available, one can study the effect of backflow rate on various design variables such as hole diameter and hole void area of the sieve plates, air and medium flow rate etc., which may be useful in improving the overall performance of the tower fermentor.

Table 3. Comparison of Parameter Estimates Obtained from Algebraic and Differential Models.

Parameter	Algebraic model	Differential model
$C_2(0)$	0.00375	.00375
γ	0.289	0.30
γ'	0.089	0.099
F_1	330	340
F_2	3700	3830
F_3	4730	4960
F_4	4700	4920
F_5	4620	4830
F_6	4430	4600
F_7	3960	4080
F_8	2810	2850
$[\frac{F_3\gamma}{F_2\gamma'}]$	4.15	3.93

3.6 NOTATION

A_0, A_1, A_2, A_3, A_4 = constants in Equation (23)

B_1, B_2 = constants in Equation (II-25)

C_i = tracer concentration in i^{th} tank, (gm./cc)

$C_i(0)$ = initial tracer concentration in i^{th} tank, (gm./cc)

EC_i = experimental tracer concentration in i^{th} tank, (gm./cc)

F_0 = feed flow rate, (lit./hr)

F_i = flow out from the i^{th} stage, (lit./hr)

K_0, K_1, K_2, K_3, K_4 = constants in Equations (24) and (25)

N = number of data points

p = parameters

S = objective function value

t = time (hrs.)

V_i = volume of the i^{th} tank (lit.)

x_i = i^{th} independent variable

y_i = i^{th} dependent variable

GREEK LETTERS

γ_i = backflow coefficient in i^{th} stage

λ_1, λ_2 = roots of Equation (II-22)

3.7 REFERENCES

1. Herbert, D., Continuous Culture of Microorganisms, S.C.I. Monograph No. 12, 21 (1961).

2. Levenspiel, O., Chemical Reaction Engineering, John Wiley & Sons, New York (1962).

3. Stainthorp, F. P., and N. Sudall, Trans. Instn. Chem. Engrs., 42, T 198 (1964).

4. Haddad, A. H. and D. Wolf, Can. J. Chem. Eng., 45, 100 (1967).

5. Prokop, A., L. E. Erickson, J. Fernandez, and A. E. Humphery, "Desing and Physical Characteristics of a Multistage, Continuous Tower Fermentor," Biotech. and Bioeng., 11, 945-966 (1969).

6. Nelder, J. A., and R. Mead, Computer J., 7, 308 (1965).

7. RKGS, IBM Scientific Subroutine Package, The IBM Company, White Plains, New York, Version II.

8. Continuous Systems Modeling Program, IBM Application Program, The IBM Company, White Plains, New York 10601.

9. Hogg, R. V., and A. T. Craig, "Introduction to Mathematical Statistics," MacMillan Company (1965).

10. Box, G. E. P., Annals N. Y. Acad. Sci., 86, Art. 3, 729 (1960).

11. Bellman, R., Proc. Natl. Acad. Sci., 41, 743 (1955).

12. Chen, G. K., M.S. Thesis, Kansas State University (1968).

13. Bard, Y., "A Function Maximization Method with Application to Parameter Estimation," New York Scientific Centre Report 322-0910, IBM (May 1967).

14. Eisenpress, H., and Greenstadt, J., "The Estimation of Nonlinear Econometric Systems," New York Scientific Centre Report 322-0910, IBM (July 1966).

15. Carroll, C. W., "The Created Response Surface Technique for Optimizing Nonlinear Restrained Systems," Operations Research, 9(2), 169-184 (1961).

APPENDIX I

Continuous systems modelling program (C.S.M.P.) used for simulating the tower fermentor flow model has been included in this Appendix. Computer output also shows the tracer concentration profiles in different tanks as predicted from the mathematical model for one of the experiments.

```
                ****CONTINUOUS SYSTEM MODELING PROGRAM****

          ***PROBLEM INPUT STATEMENTS***

     *DATA STATEMENTS
     CONST F0=2.0,F1=.33,F2=3.7,F3=4.73081,F4=4.69913,F5=4.62139,...
     F6=4.4307,F7=3.96289,G1=.0891892,G2=.28959,F8=2.81528
     PARAM V=.633
     *DATA STATEMENTS
     *INITIAL CONDITION
     INCON C10=0.,C20=.00375000,C30=0.,C40=0.,C50=0.,C60=0.,C70=0.,C80=0.
     *OUTPUT CONTROL STATEMENTS
     PRTPLT C1
     LABEL TRACER CONCENTRATION PROFILE IN TANK(1)
     PRTPLT C2
     LABEL TRACER CONCENTRATION PROFILE IN TANK(2)
     PRTPLT C3
     LABEL TRACER CONCENTRATION PROFILE IN TANK(3)
     PRTPLT C4
     LABEL TRACER CONCENTRATION PROFILE IN TANK(4)
     PRTPLT C5
     LABEL TRACER CONCENTRATION PROFILE IN TANK(5)
     PRTPLT C6
     LABEL TRACER CONCENTRATION PROFILE IN TANK(6)
     PRTPLT C7
     LABEL TRACER CONCENTRATION PROFILE IN TANK(7)
     PRTPLT C8
     LABEL TRACER CONCENTRATION PROFILE IN TANK(8)
     *STRUCTURE STATEMENTS
     C1=INTGRL(C1C,(F2*G1*C2-F1*C1)/V)
     C2=INTGRL(C2C,(F1*C1+F3*G2*C3-F2*C2)/V)
     C3=INTGRL(C3C,(F2*(1.-G1)*C2+F4*G2*C4-F3*C3)/V)
     C4=INTGRL(C4C,(F3*(1.-G2)*C3+F5*G2*C5-F4*C4)/V)
     C5=INTGRL(C5C,(F4*(1.-G2)*C4+F6*G2*C6-F5*C5)/V)
     C6=INTGRL(C6C,(F5*(1.-G2)*C5+F7*G2*C7-F6*C6)/V)
     C7=INTGRL(C7C,(F6*(1.-G2)*C6+F8*G2*C8-F7*C7)/V)
     C8=INTGRL(C8C,(F7*(1.-G2)*C7-F8*C8)/V)
     *EXECUTION CONTROL STATEMENTS
     TIMER OUTDEL=0.25,FINTIM=10.0,DELT=0.1
     END
     STOP

OUTPLT VARIABLE SEQUENCE
ZZOCC2 C1      ZZC004 C2      ZZC006 C3      ZZ0008 C4      ZZC010 C5
ZZCC12 C6      ZZC014 C7      ZZ0016 C8

 OUTPUTS     INPUTS    PARAMS  INTEGS + MEM BLKS  FORTRAN  DATA CDS
 20(5C0)   43(14C0)  23(400)   8+  0=   8(3CC)   17(6C0)     22
```

TRACER CONCENTRATION PROFILE IN TANK(1) PAGE 1

TIME	C1	MINIMUM 0.0 C1 VERSUS TIME MAXIMUM 3.3909E-04
0.C	0.C	+
2.5C0E-01	2.5681E-04	----------+
5.CCCE-01	3.2532E-04	----------+
7.5CCE-01	3.39C9E-04	----------+
1.CCCE 00	3.313CE-04	----------+
1.25C0E 00	3.141CE-04	----------+
1.5CCCE 00	2.9289E-04	----------+
1.75CCE 00	2.7037E-04	----------+
2.CC00E 00	2.4792E-04	----------+
2.25CCE 00	2.2629E-04	----------+
2.5CCCE 00	2.0588E-04	----------+
2.75CCE 00	1.8685E-04	----------+
3.CCCCE 00	1.6926E-04	----------+
3.25CCE 00	1.5311E-04	----------+
3.5CCCE CO	1.3835E-04	----------+
3.75CCE 00	1.2489E-04	----------+
4.CCCCE CO	1.1267E-04	----------+
4.25C0E 00	1.0157E-04	----------+
4.5CCCE 00	9.1528E-05	----------+
4.75CCE CO	8.2443E-05	----------+
5.CCCCE CC	7.4234E-05	----------+
5.25C0E 00	6.6823E-05	----------+
5.5CCCE 00	6.0139E-05	----------+
5.75CCE CO	5.411CE-05	----------+
6.CCCCE CC	4.8679E-05	----------+
6.25CCE 00	4.3786E-05	----------+
6.5CCCE 00	3.9381E-05	----------+
6.75CCE 00	3.5415E-05	----------+
7.CC00E 00	3.1845E-05	----------+
7.25C0E 00	2.8633E-05	----------+
7.5CCCE 00	2.5744E-05	----------+
7.75C0E 00	2.3145E-05	----------+
8.CCCCE 00	2.08C7E-05	----------+
8.25CCE 00	1.87C5E-05	----------+
8.5CCCE 00	1.6815E-05	----------+
8.75CCE 00	1.5115E-05	----------+
9.CCCCE 00	1.3587E-05	----------+
9.25CCE 00	1.2213E-05	----------+
9.5CCCE 00	1.0977E-05	----------+
9.75CCE 00	9.8669E-06	----------+
1.CCCCE 01	8.86E7E-06	----------+

TRACER CONCENTRATION PROFILE IN TANK(2) PAGE 1

TIME	C2	MINIMUM 1.6106E-06 C2 VERSUS TIME MAXIMUM 3.7500E-03
		I I
0.0	3.7500E-03	----------+
2.5000E-01	1.1919E-03	----------+
5.0000E-01	5.7414E-04	-------+
7.5000E-01	3.3934E-04	----+
1.0000E 00	2.2580E-04	--+
1.2500E 00	1.6214E-04	--+
1.5000E 00	1.2275E-04	-+
1.7500E 00	9.6567E-05	-+
2.0000E 00	7.8181E-05	-+
2.2500E 00	6.4684E-05	+
2.5000E 00	5.4411E-05	+
2.7500E 00	4.6358E-05	+
3.0000E 00	3.9892E-05	+
3.2500E 00	3.4597E-05	+
3.5000E 00	3.0192E-05	+
3.7500E 00	2.6478E-05	+
4.0000E 00	2.3315E-05	+
4.2500E 00	2.0597E-05	+
4.5000E 00	1.8245E-05	+
4.7500E 00	1.6198E-05	+
5.0000E 00	1.4408E-05	+
5.2500E 00	1.2836E-05	+
5.5000E 00	1.1450E-05	+
5.7500E 00	1.0226E-05	+
6.0000E 00	9.1411E-06	+
6.2500E 00	8.1780E-06	+
6.5000E 00	7.3214E-06	+
6.7500E 00	6.5584E-06	+
7.0000E 00	5.8778E-06	+
7.2500E 00	5.2700E-06	+
7.5000E 00	4.7268E-06	+
7.7500E 00	4.2409E-06	+
8.0000E 00	3.8060E-06	+
8.2500E 00	3.4164E-06	+
8.5000E 00	3.0672E-06	+
8.7500E 00	2.7542E-06	+
9.0000E 00	2.4735E-06	+
9.2500E 00	2.2216E-06	+
9.5000E 00	1.9956E-06	+
9.7500E 00	1.7927E-06	+
1.0000E 01	1.6106E-06	+

TRACER CONCENTRATION PROFILE IN TANK(3) PAGE 1

TIME	C3	MINIMUM 0.0 C3 VERSUS TIME MAXIMUM 1.1875E-03
		I I
0.C	0.0	+
2.5CCOE-01	1.1875E-03	----------+
5.CCCCE-01	8.2336E-04	----------+
7.5CCCE-01	5.4772E-04	----------+
1.CCCCE 00	3.7791E-04	----------+
1.25CCE 00	2.7176E-04	----------+
1.5CCCE 00	2.0261E-04	---------+
1.75CCE 00	1.5566E-04	------+
2.CCCCE 00	1.2262E-04	-----+
2.25CCE 00	9.8605E-05	----+
2.5CCCE 00	8.0664E-05	---+
2.75C0E 00	6.6933E-05	--+
3.CCCCE 00	5.6203E-05	--+
3.25CCE 00	4.7664E-05	--+
3.5CCCE 00	4.0762E-05	-+
3.75C0E 00	3.5108E-05	-+
4.CCCCE 00	3.0421E-05	-+
4.25CCE 00	2.6496E-05	-+
4.5CCCE 00	2.3179E-05	+
4.75CCE 00	2.0355E-05	+
5.CCCCE 00	1.7934E-05	+
5.25C0E 00	1.5845E-05	+
5.5C00E 00	1.4034E-05	+
5.75C0E 00	1.2456E-05	+
6.CCCCE 00	1.1075E-05	+
6.2500E 00	9.8623E-06	+
6.5CCCE 00	8.7944E-06	+
6.75CCE 00	7.8511E-06	+
7.CCCCE 00	7.0158E-06	+
7.25C0E 00	6.2747E-06	+
7.5CCCE 00	5.6160E-06	+
7.75C0E 00	5.0295E-06	+
8.CCCCE 00	4.5066E-06	+
8.25CCE 00	4.0399E-06	+
8.5CCCE 00	3.6229E-06	+
8.75C0E 00	3.2501E-06	+
9.CCCCE 00	2.9164E-06	+
9.25CCE 00	2.6176E-06	+
9.5CCCE 00	2.3499E-06	+
9.75C0E 00	2.1099E-06	+
1.CCCCE 01	1.8948E-06	+

TRACER CONCENTRATION PROFILE IN TANK(8) PAGE 1

TIME	C8	MINIMUM 0.0 C8 VERSUS TIME MAXIMUM 4.7603E-04
0.C	0.C	+
2.5CCCE-01	4.60C5E-06	+
5.CCCCE-01	6.9373E-05	-------+
7.5CCCE-01	2.1247E-04	----------+
1.CCCCE 00	3.5728E-04	----------+
1.25CCE 00	4.475CE-04	----------+
1.5CCCE 00	4.76C3E-04	----------+
1.75CCE 00	4.5939E-04	----------+
2.CCCCE 00	4.1722E-04	----------+
2.25CCE 00	3.6454E-04	----------+
2.5CCCE 00	3.107CE-04	----------+
2.75CCE 00	2.6071E-04	----------+
3.CCCCE 00	2.1675E-04	----------+
3.25CCE 00	1.7932E-04	----------+
3.5CCCE 00	1.4811E-04	----------+
3.75CCE 00	1.2241E-04	----------+
4.CCCCE 00	1.C141E-04	----------+
4.25CCE 00	8.4318E-05	--------+
4.5CCCE 00	7.0416E-05	--------+
4.75CCE 00	5.9102E-05	-------+
5.CCCCE 00	4.9869E-05	-----+
5.25CCE 00	4.2308E-05	-----+
5.5CCCE 00	3.6088E-05	---+
5.75CCE 00	3.0944E-05	---+
6.CCCCE 00	2.6667E-05	--+
6.25CCE 00	2.3C9CE-05	--+
6.5CCCE 00	2.CC8CE-05	--+
6.75CCE 00	1.7532E-05	-+
7.CCCCE 00	1.5363E-05	-+
7.25CCE 00	1.35C6E-05	-+
7.5CCCE 00	1.19C8E-05	-+
7.75CCE 00	1.0527E-05	-+
8.CCCCE 00	9.3262E-06	+
8.25CCE 00	8.2791E-06	+
8.5CCCE 00	7.3623E-06	+
8.75CCE 00	6.5569E-06	+
9.CCCCE 00	5.8472E-06	+
9.25CCE 00	5.22C2E-06	+
9.5CCCE 00	4.6649E-06	+
9.75CCE 00	4.1722E-06	+
1.CCCCE 01	3.7342E-06	+

APPENDIX II

The complete analytical solution of the performance equations used with the partial model are presented in this appendix.

The performance of the partial system can be represented by Equation (21) and (22)

$$\frac{dC_1}{dt} = \frac{F_1}{V} (C_2 - C_1) \qquad \text{(II-1)}$$

$$\frac{dC_2}{dt} = \frac{F_1}{V}(C_1 - C_3) + \frac{F_2}{V} (C_3 - C_2) - \frac{F_0}{V} C_3 \qquad \text{(II-2)}$$

From equation (II-1)

$$C_2 = C_1 + \frac{V}{F_1} \frac{dC_1}{dt} \qquad \text{(II-3)}$$

assuming

$$C_3 = (A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4) \qquad \text{(II-4)}$$

Substituting equations (II-3) and (II-4) in equation (II-2)

$$\frac{d}{dt} \left[C_1 + \frac{V}{F_1} \frac{dC_1}{dt}\right] = \frac{F_1}{V} \left[C_1 - (A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4)\right]$$

$$+ \frac{F_2}{V} \left[(A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4) - (C_1 + \frac{V}{F_1} \frac{dC_1}{dt})\right]$$

$$- \frac{F_0}{V} \left[(A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4)\right]$$

$$\frac{dC_1}{dt} + \frac{V}{F_1} \frac{d^2 C_1}{dt^2} = \frac{C_3}{V} (F_2 - F_1 - F_0) + \frac{C_1}{V} (F_1 - F_2) - \frac{F_2}{F_1} \frac{dC_1}{dt}$$

$$\frac{V}{F_1}\frac{d^2C_1}{dt^2} + \frac{dC_1}{dt}\left[1 + \frac{F_2}{F_1}\right] + \frac{C_1}{V}\left[F_2 - F_1\right] = \frac{C_3}{V}\left[F_2 - F_1 - F_0\right] \qquad \text{(II-5)}$$

making the following substitutions

Let

$$a = \frac{V}{F_1}$$

$$b = \left(1 + \frac{F_2}{F_1}\right)$$

$$g = \frac{(F_2 - F_1)}{V}$$

$$m = \frac{(F_2 - F_1 - F_0)}{V}$$

Equation (II-5) becomes

$$a\frac{d^2C_1}{dt^2} + b\frac{dC_1}{dt} + gC_1 = mC_3 \qquad \text{(II-6)}$$

Equation (II-6) is a linear, non-homogenous, second order differential equation which can be solved without much difficulty. The solution of equation (II-6) consists of the sum of a particular solution and a homogenous solution. The particular solution of equation (II-6) is assumed to be of the following form

$$C_1(P) = (K_0 + K_1 t + K_2 t^2 + K_3 t^3 + K_4 t^4) \tag{II-7}$$

$$\frac{dC_1(P)}{dt} = (K_1 + 2K_2 t + 3K_3 t^2 + 4K_4 t^3) \tag{II-8}$$

$$\frac{d^2 C_1(P)}{dt^2} = (2K_2 + 6K_3 t + 12K_4 t^2) \tag{II-9}$$

where K_0, K_1, K_2, K_3, K_4 are constants to be found out.

Substituting equations (II-4), (II-7), (II-8), and (II-9) into equation (II-6) one gets

$$a[2K_2 + 6K_3 t + 12K_4 t^2] + b[K_1 + 2K_2 t + 3K_3 t^2 + 4K_4 t^3]$$

$$+ g[K_0 + K_1 t + K_2 t^2 + K_3 t^3 + K_4 t^4] = m[A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4] \tag{II-10}$$

Comparing coefficients of equal powers of t in equation (II-10) one gets

Coefficients of t^0

$$2K_2 a + bK_1 + gK_0 = mA_0 \tag{II-11}$$

Coefficients of t

$$6K_3 a + 2K_2 b + K_1 g = mA_1 \tag{II-12}$$

Coefficient of t^2

$$12K_4 a + 3K_3 b + K_2 g = mA_2 \tag{II-13}$$

Coefficient of t^3

$$4K_4b + K_3g = mA_3 \tag{II-14}$$

Coefficient of t^4

$$K_4g = mA_4 \tag{II-15}$$

Constants K_0, K_1, K_2, K_3, K_4 can be found out from equations (II-11) through (II-15) since there are five equations and five unknowns.

$$K_0 = [\frac{mA_0}{g} - \frac{2amA_2}{g^2} + \frac{24a^2mA_4}{g^3} + \frac{6abmA_3}{g^3} - \frac{24ab^2mA_4}{g^4} - \frac{bmA_1}{g^2}$$

$$+ \frac{6abmA_3}{g^3} - \frac{24ab^2mA_4}{g^4} + \frac{2b^2mA_4}{g^3} - \frac{24ab^2mA_4}{g^4} - \frac{6b^3mA_3}{g^4} + \frac{24b^4mA_4}{g^5} \tag{II-16}$$

$$K_1 = [\frac{mA_1}{g} - \frac{6amA_3}{g^2} + \frac{24abmA_4}{g^3} - \frac{2bmA_2}{g^2} + \frac{24abmA_4}{g^3} + \frac{6b^2mA_3}{g^3} - \frac{24b^3mA_4}{g^4}] \tag{II-17}$$

$$K_2 = [\frac{mA_2}{g} - \frac{12amA_4}{g^2} - \frac{3bmA_3}{g^3} + \frac{12b^2mA_4}{g^3}] \tag{II-18}$$

$$K_3 = [\frac{mA_3}{g} - \frac{4bmA_4}{g^2}] \tag{II-19}$$

$$K_4 = [\frac{mA_4}{g}] \tag{II-20}$$

Knowing the constants K_0, K_1, K_2, K_3 and K_4 the particular solution for C_1 can be found out.

Homogenous solution

$$a\frac{d^2C_1}{dt^2} + b\frac{dC_1}{dt} + gC_1 = 0 \qquad \text{(II-21)}$$

The auxiliary equation can be written as:

$$(a\lambda^2 + b\lambda + g) = 0 \qquad \text{(II-22)}$$

$$\lambda = \frac{-b \pm \sqrt{b^2 - 4ag}}{2a}$$

Hence

$$\lambda_1 = \frac{-b + \sqrt{b^2 - 4ag}}{2a} \qquad \text{(II-23)}$$

$$\lambda_2 = \frac{-b - \sqrt{b^2 - 4ag}}{2a} \qquad \text{(II-24)}$$

whence

$$C_1(h) = (B_1e^{\lambda_1 t} + B_2e^{\lambda_2 t}) \qquad \text{(II-25)}$$

$$\therefore \quad C_1 = C_1(P) + C_1(h)$$

$$C_1 = (B_1e^{\lambda_1 t} + B_2e^{\lambda_2 t}) + (K_0 + K_1t + K_2t^2 + K_3t^3 + K_4t^4) \qquad \text{(II-26)}$$

$$\because C_2 = C_1 + \frac{V}{F_1}\frac{dC_1}{dt}$$

$$C_2 = \frac{V}{F_1}\left[(B_1\lambda_1 e^{\lambda_1 t} + B_2\lambda_2 e^{\lambda_2 t}) + (K_1 + 2K_2 t + 3K_3 t^2 + 4K_4 t^3)\right]$$

$$+ \left[(B_1 e^{\lambda_1 t} + B_2 e^{\lambda_2 t}) + (K_0 + K_1 t + K_2 t^2 + K_3 t^3 + K_4 t^4)\right] \qquad \text{(II-27)}$$

Applying the initial conditions in equations (II-26) and (II-27)

at $t = 0$ $C_1 = 0$

@ $t = 0$ $C_2 = C_2(0)$

$$0 = B_1 + B_2 + K_0$$

$$\therefore B_2 = -(B_1 + K_0) \qquad \text{(II-28)}$$

From equation (II-27)

$$C_2(0) = B_1 - (B_1 + K_0) + K_0 + \frac{V}{F_1}\left[B_1\lambda_1 - \lambda_2 (B_1 + K_0) + K_1\right] \qquad \text{(II-29)}$$

Rearrangement of terms in equation (II-29) gives

$$B_1 = \frac{1}{(\lambda_1 - \lambda_2)}\left[\frac{C_2(0)\ F_1}{V} + (\lambda_2 K_0 - K_1)\right] \qquad \text{(II-30)}$$

Substituting equations (II-28) and (II-30) in equations (II-26) and (II-27) gives the final solutions for C_1 and C_2 in terms of unknown parameters and time, which are given by Equations (24) and (25) in the main text.

Chapter 4

ANALYSIS OF FLOW BEHAVIOR IN A CONTINUOUS, MULTISTAGE TOWER FERMENTOR: EXAMINATION AND ANALYSIS OF RESIDUALS

4.1 INTRODUCTION

In recent years, a number of techniques have been proposed for the examination and analysis of residuals. These give information on various questions of interest. Often, one makes use of these techniques to assess the validity of appropriateness of the conventional analysis techniques such as analysis of variance and least-squares curve fitting [1].

Although the work of this chapter is restricted to linear regression models only, the same methods can be applied to nonlinear regression models and analysis of variance models. In general, the techniques for the examination and analysis of residuals can be classified into three broad categories: graphical, numerical, and mixed. However, emphasis is placed on the graphical examination of residuals in this chapter. The residuals are defined as the differences $e_i = (Y_i - y_i)$, $i = 1, 2, \ldots, n$, where Y_i is an observation, y_i is the corresponding fitted value obtained by the fitted regression model and n is the number of observations. According to Draper and Smith [2] the residuals e_i are the differences between what is actually observed, and what is predicted by the regression equation. In other words, this is the amount which the regression equation has not been able to explain. This can also be thought of as the observed errors, if the model is correct. The usual assumptions made about the errors while performing the regression analysis are that the errors are independent,

have zero mean and constant variance, σ^2, and follow a normal distribution. The assumption that errors follow a normal distribution is often required in making F-tests. If after the examination of residuals one observed that the residuals show tendencies that tend to confirm the basic assumptions, then one has sufficient reason to believe that the model is adequate and correct.

4.2 GENERAL CONSIDERATIONS

Once the residuals have been calculated, a number of alternative approaches are possible. These have been discussed in considerable detail by Anscombe and Tukey [1]. A rather short description of them is presented here.

1. The relationship of residuals to external variables, such as time of observation and geographical position, can be examined best graphically. Sometimes it is useful to look for a variety of trends including both straight and curved trends.

2. Two important things which can be examined by eye-judgment from the residuals are (i) outliers (ii) shape of the distribution of the deviations which the residuals reflect.

3. The individual residuals can be plotted against individual fitted values to find out (i) removable nonadditivity (ii) dependence of variability of response on level of response.*

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*Discussion of these terms (removable nonadditivity and dependence of variability on level) and techniques for their measurement appear elsewhere [1].

4. Groups of residuals from different parts of data can be used to indicate difference in variability of response in these parts.

Anscombe and Tukey [1] have also suggested that, as a result of the above mentioned approaches, one may try to

1. Improve the precision of results by rejecting or modifying observations corresponding to residuals identified as extremes.
2. By measuring the extent of removable nonadditivity and the extent of dependence of variability on level, one can obtain information as to how we should modify our model to fit the experimental data better.

Anscombe and Tukey [1] are of the opinion that the graphical techniques for the examination of residuals offer valuable assistance in differential diagnosis. According to them it is best to begin the analysis with the graphical method. Once the most important sort of misbehavior has been found, it is usually important to deal with it. The usual thing to do at this stage is to reject or modify observations associated with the outliers or transform the observations. After this has been done attempts should be made to find out if there are any other types of misbehavior.

Anscombe and Tukey [1] have described the various ways by which the improvement in the analysis is possible (i) by modifying the form of the "model" and calculating the fitted values from this model which is supposed to be a better representation of the experimental observations (ii) by changing the mode of expression of the observation and then applying the original analysis in the new mode (for example when log Y_i's are analysed in place of Y_i's) (iii) by discriminating among the individual observations,

this may include the assignment of different weights to different observations or a set of observations, rejection of certain observations or the replacement of some observed values by modified values before the analysis.

4.3 CORRELATION AMONG THE RESIDUALS

Draper and Smith [2] have presented a mathematical approach to find the correlation among the residuals. According to them, when θ parameters are estimated from n observations, the n residuals are associated with only $(n-\theta)$ degrees of freedom. This suggests that the residuals cannot be independent and correlations exist among them. The correlation between e_i and e_j is given by

$$\rho_{ij} = \frac{\text{covariance } (e_i, e_j)}{[v(e_i) \cdot v(e_j)]^{1/2}} \qquad (1)$$

where

ρ_{ij} = correlation coefficient between the i^{th} residual the j^{th} residual

i = i^{th} residual

j = j^{th} residual

$v(e_i)$ = variance of the i^{th} residual

$v(e_j)$ = variance of the j^{th} residual

In this chapter, however, no attempt has been made to consider the correlations among the residuals. The reason is that the investigation was restricted to the application of graphical techniques only. The justification for this is given by Anscombe [3] as follows: although correlations and constraints affect distribution of functions of the residuals, the corresponding effects on the graphical procedures can usually be neglected. Draper and Smith [2] have stated that, in general regression situations, the effect of correlations between residuals need not be considered when plots are made, except when the ratio $(n-\theta)/n$ is quite small.

4.4 OUTLIERS

One of the most important purposes of calculating the residuals is to detect the outliers. Outliers are the observations having large residuals, in comparison to others, which suggests to us that they need very careful examination. The outlier is a peculiarity and represents a data point which is not at all typical of the rest of the data. This is the reason why one submits the outlier to careful examination so as to find the cause of this peculiarity. There are many sources of outliers, for example, mistake in reading a scale or an error in conducting an experiment, such as unknowingly measuring the wrong thing, or failing to notice whether some intended condition is satisfied [1].

Various rules have been proposed for the rejection of outliers, and will be described here briefly. A rather exhaustive treatment is given to this subject by Anscombe [3]. According to Draper and Smith [2] automatic rejection of outliers is not always a very wise procedure.

Sometimes the outlier is providing information, which other data points cannot, due to the fact that it arises from an unusual combination of circumstances, which may be of vital interest and require further investigation rather than rejection.

4.4a Rejection of Outliers

Variability or dispersion in a set of observations can arise from several different sources. These may include inherent variability, measurement error and execution error etc. Inherent variability is sometimes observed in the population even if all measurements are perfectly correct. This variability cannot be reduced without changing the population itself or the object of study. Measurement errors are caused by the measuring instruments. Execution error might include any discrepancy between what one intends to do and what is actually done [3].

If a rather pronounced degree of abnormality is expected in our experimental observations, to work with medians instead of means is often useful according to Edgeworth (1887). A modified least-squares method, with weights depending upon the residuals, has been suggested by Jeffreys [4].

The rejection rules discussed here are the ones proposed by Anscombe [3]. According to him rejection rules are not significance tests. Significance tests are appropriate, where the object of study is how often spurious observations occur in a certain field.

Before the rejection rules are investigated, it will be assumed that the following conditions are met.

(i) All observations are independent of each other. It was also assumed that spuriousness is uncorrelated with the reading that would have

been obtained, had the observation been made without abnormal error.

(ii) No prior knowledge of the means or regression coefficients that are to be estimated from the data is incorporated in the rejection rule, which is, therefore, "impartial."

According to Anscombe [3], it is useful to consider rejection criteria based on the magnitude of residuals. In the least squares analysis, the computation of residuals is a standard procedure when Bard's method [5] is used, and hence a rejection criterion based on residuals is particularly convenient.

4.4b Rules for Rejection

Consider observations $Y_1, Y_2, \ldots, Y_n$ $(n \geq 3)$, where n is the total number of observations. It is hoped that they are a random sample from a normal population $N(\mu, \sigma^2)$, where μ is known and σ is to be estimated. But one of the Y_i's is spurious, and ought to be rejected. Consider the effect of applying a rejection rule routinely to samples of fixed size n. Let

$$Z_i = (Y_i - \bar{Y}), \quad n\bar{Y} = \sum_{i=1}^{n} Y_i \tag{2}$$

where

Z_i (i=1, 2, ... n) are the residuals and hereafter ν will denote the number of residual degrees of freedom.

If Y_i is omitted, the average of the remaining observations is

$$\sum_{j \neq i} Y_j/\nu = (\bar{Y} - Z_i/\nu), \qquad \nu = (n-1) \tag{3}$$

More generally when several observations, say $Y_1, Y_2, \ldots, Y_r$, are

omitted the average of the rest is

$$\bar{Y} - (Z_1 + Z_2 + \ldots + Z_r)/(n - r) \tag{4}$$

where r is the number of observations omitted. Let m be the serial number of the observation having the greatest residual, so that

$$|Z_m| > |Z_i| \text{ for all } i \neq m \tag{5}$$

assuming no two residuals are equal in magnitude if they are recorded to a sufficient number of decimal places.

Rule (i) For given C, reject Y_m if $|Z_m| > C\sigma$; otherwise no rejections. Estimate μ by the means of the retained observations; thus

$$\hat{\mu} = \bar{Y} \text{ if } |Z_m| < C\sigma \tag{6}$$

$$= (\bar{Y} - Z_m/\nu) \text{ if } |Z_m| > C\sigma \tag{7}$$

Under this rule not more than one observation can be rejected. If more than one observation out of a very small sample (having 3 or 4 observations) appears too spurious, the observer would most likely wish to scrap all of them. For large samples, however, the possibility of multiple selective rejections needs to be considered, and Anscombe [3] has suggested the following rule:

Rule (ii): Apply Rule (i). If an observation is rejected, consider the remaining observations as a sample of size (n-1) and apply Rule (i) again; and so on. Estimate μ by the mean of the retained observations. It would be possible for the values of C to differ in successive applications of

Rule (i), but there is no obvious advantage in this, and, in so far as Rule (ii) is considered, C will be assumed constant. In fact, it is difficult to study Rule (ii) exactly, apart from Monte Carlo computation. Rule (i) is easier, and sometimes has almost the same effect as Rule (ii) - namely, when there is not more than one spurious observation present in the sample, C is not very small and n is not very large.

4.5 MATHEMATICAL MODEL

The problem of polynomial approximation is often encountered wherein one tries to fit the experimental data to a polynomial. A similar problem was encountered in Chapter 3 where a fourth degree polynomial was assumed for predicting the tracer concentration in tank (3). This choice was random. No theoretical basis for assuming a fourth degree polynomial was present. It appears to be plausible that the higher the degree of the polynomial, the better will be the predicted values of the response. However, this is not true in some situations. If one assumes a higher degree polynomial one might sometimes overestimate the parameters in the model and if on the other hand the degree of the polynomial is less, one might face the difficulty of underestimation. The compromise between these two extremes is what is usually called "selecting the best regression equation." There is no unique statistical procedure for doing this, and personal judgment is always a necessary part of any method.

Equations (8) through (13) represent the various polynomials assumed for the tracer concentration in stage (3). Graphical techniques were used to investigate which model gives the best fit for the data.

$$C_3 = a'_o + a'_1 t \tag{8}$$

$$C_3 = b_o + b_1 t + b_2 t^2 \tag{9}$$

$$C_3 = m_o + m_1 t + m_2 t^2 + m_3 t^3 \tag{10}$$

$$C_3 = d_o + d_1 t + d_2 t^2 + d_3 t^3 + d_4 t^4 \tag{11}$$

$$C_3 = e'_o + e'_1 t + e'_2 t^2 + e'_3 t^3 + e'_4 t^4 + e'_5 t^5 \tag{12}$$

$$C_3 = h_o + h_1 t + h_2 t^2 + h_3 t^3 + h_4 t^4 + h_5 t^5 + h_6 t^6 \tag{13}$$

where

C_3 = tracer concentration in tank (3), (gms./c.c.)

t = time in hrs.

a'_o, a'_1 = parameters for first order model.

b_o, b_1, b_2 = parameters for second order model.

m_o, m_1, m_2, m_3 = parameters for third order model.

d_o, d_1, d_2, d_3, d_4 = parameters for fourth order model.

e'_o, e'_1, e'_2, e'_3, e'_4, e'_5 = parameters for fifth order model.

h_o, h_1, h_2, h_3, h_4, h_5, h_6 = parameters for sixth order model.

4.6 ESTIMATION OF PARAMETERS IN VARIOUS MODELS

The parameters appearing in various models represented by equations

(8) thru (13) must be found out before one can proceed to analyze the residuals. The Gauss-Newton method with modifications by Bard [5], Carroll [6] and Eisenpress and Greenstadt [7] was used to estimate these parameters. A brief description of this method is presented here. Detailed description of the method and programming details can be found in reference [5]. This method will be referred to as Bard's method hereafter.

Bard's program is designed to estimate unknown parameters in a variety of mathematical models using a variety of best-fit criteria. Whenever Bard's method [5] is used, the measure of fit depends upon the residuals. Sum of squares of residuals is one of the most common criterion usually employed, although more sophisticated measures exist.

4.6a Best Fit Conditions

If the mathematical model is represented by $y_\mu = f(a_\mu,\theta)$ then the residuals can be stated mathematically as follows;

$$U_\mu = f(a_\mu,\theta) - Y_\mu \qquad (\mu = 1,2,\ldots,n) \tag{14}$$

$$y_\mu = f(a_\mu,\theta) \qquad (\mu = 1,2,\ldots,n) \tag{15}$$

where

a_μ = independent variables (μ = 1, 2, ..., n).

$\theta = (\theta_1, \theta_2, \ldots, \theta_\ell)$ = parameters.

Y_μ = observed variables (μ = 1, 2, ..., n).

n = number of observations.

U_μ = residuals (μ = 1, 2, ..., n).

The main object of parameter estimation techniques is to find a set of parameters, θ, which minimize or maximize a certain selected criterion function of the residuals, U_μ. This function will be denoted by $F(U_\mu) = F(f(a_\mu,\theta) - Y_\mu)$ when a_μ and Y_μ are given, $F(U_\mu)$ becomes a function of the parameters alone. This function will be denoted by $G(\theta)$. Some of the commonly employed functions are given below.

(i) Least Squares (L. S.)

Consider the case where there is only one observed variable per experiment. Defining

$$F_{L.S.}(U_\mu) = \sum_{\mu=1}^{n} U_\mu^2 \tag{16}$$

i.e.,

$$G_{L.S.}(\theta) = \sum_{\mu=1}^{n} [f(a_\mu,\theta) - Y_\mu]^2 \tag{17}$$

one can determine θ so as to minimize $G_{L.S.}(\theta)$. In some situations it is advantageous to use a weighted least squares criterion, in which case the G-function to be minimized can be written as

$$G_{W.L.S.}(\theta) = \sum_{\mu=1}^{n} \sum_{i=1}^{k} W_i [f_i(a_\mu,\theta) - Y_{\mu i}]^2 \tag{18}$$

where k is the number of variables.

One can also assign weights to the cross product of variables. The

problem often encountered here is that one frequently does not know what weights to assign. This is one of the shortcomings of least squares estimation. However this problem is overcome by maximum likelihood methods.

(ii) Maximum Likelihood (M. L.)

One may assume that the residuals U are random variables possessing a joint probability density function $P(U,\phi)$ of known mathematical form, possibly containing some unknown parameters ϕ. Here U denotes a matrix of the residuals. According to the maximum likelihood principle, one seeks those values of θ and ϕ which maximize the likelihood of having made the actual observations, i.e., which maximize P or more conveniently its logarithm. Thus

$$G_{M.L.}\ (\theta,\phi) = \log P\ (f(a_\mu,\theta) - Y_\mu,\phi) \tag{19}$$

Bard [5] has shown that if there is one observed variable per experiment then maximum likelihood is equivalent to least squares and the residuals, U, are normally distributed with zero mean and variance σ^2. If the number of observed variables per experiment is more ($k>1$) then maximum likelihood reduces to weighted least squares.

(iii) Bayesian Estimation

An expression such as $P(U,\phi)$ states the probability distribution of Y_μ given the parameters θ. In fact, however, observations Y_μ are given and the parameters θ are sought. One may conceive of many possible universes, each with its own values of θ. Given the observations, one may assign relative probabilities to these universes, and ask which of these

universes is most probable. The above explanation comes from statistical theory. Cornfield [8] has summarized the current thought on this subject.

We maximize $P(\theta|Y_\mu)$ i.e., the probability density of θ given the observations Y_μ. According to the Bayes formulae

$$P(\theta|Y_\mu) = \frac{1}{C'} P(Y_\mu|\theta) P_o(\theta) \tag{20}$$

where

$C' = \int P(Y_\mu|\theta) P_o(\theta) d\theta$ = a normalizing constant.

$P(Y_\mu|\theta)$ = probability density of Y_μ given θ

$P_o(\theta)$ = prior distribution of θ i.e., it is the probability distribution one would have assigned to θ prior to having made the observations Y_μ.

Ignoring the irrelevant constant c, we seek then to maximize

$$G_B(\theta,\phi) = \log P[f(a_\mu,\theta) - Y_\mu,\phi)] + \log P_o(\theta) \tag{21}$$

If absolutely nothing is known a priori about θ, one assumes a uniform prior distribution. Then $P_o(\theta)$ = constant, and maximizing $P(\theta|Y_\mu)$ is equivalent to maximizing $P(U,\phi)$, i.e., the Bayesian and maximum likelihood estimates will be identical.

4.6b Method of Solution

The central function of Bard's [5] program is to maximize a selected criterion function. Since the problem considered in this chapter is essentially a minimization problem, the negative of the mathematical objective

function was maximized. The objective functions which were minimized for the models represented by Equations (8) through (13) are given by T_1 through T_6 given below.

$$T_1 = \sum_{i=1}^{n} [C_{3i} - (a'_0 + a'_1 t_i)]^2 \tag{22}$$

$$T_2 = \sum_{i=1}^{n} [C_{3i} - (b_0 + b_1 t_i + b_2 t_i^2)]^2 \tag{23}$$

$$T_2 = \sum_{i=1}^{n} [C_{3i} - (m_0 + m_1 t_i + m_2 t_i^2 + m_3 t_i^3)]^2 \tag{24}$$

$$T_4 = \sum_{i=1}^{n} [C_{3i} = (d_0 + d_1 t_i + d_2 t_i^2 + d_3 t_i^3 + d_4 t_i^4)]^2 \tag{25}$$

$$T_5 = \sum_{i=1}^{n} [C_{3i} - (e'_0 + e'_1 t_i + e'_2 t_i^2 + e'_3 t_i^3 + e'_4 t_i^4 + e'_5 t_i^5)]^2 \tag{26}$$

$$T_6 = \sum_{i=1}^{n} [C_{3i} - (h_0 + h_1 t_i + h_2 t_i^2 + h_3 t_i^3 + h_4 t_i^4 + h_5 t_i^5 + h_6 t_i^6)]^2 \tag{27}$$

Two alternative subroutines are provided for effecting the maximization. These are

(i) The Gauss-Newton method, with modifications by Greenstadt-Eisenpress [7], Bard [5], and Carroll [6].

(ii) The Davidon-Fletcher-Powell method [9]

The modified Gauss-Newton method was used for maximization. Davidon's method is fully described in reference [9]. A brief description of the Gauss-Newton method is given in Appendix V.

4.7 GRAPHICAL EXAMINATION OF RESIDUALS

The usual assumptions made about the errors while performing the regression analysis are that the errors are independent, and follow a normal distribution with mean zero and variance, σ^2. Hence the residuals should exhibit tendencies that tend to confirm the assumptions, provided the fitted model is correct. While examining the residuals one should try to investigate whether the assumptions made, appear to be violated or not; of course the latter does not mean that the assumptions are correct, but it merely means that on the basis of the experimental data, it appears that one does not have any valid reason to say that the assumptions are incorrect.

There are several graphical methods of examining the residuals in order to check the models and see which model gives the best fit for the experimental data. These procedures indicate to a great extent whether the assumptions are violated or not. A rather detailed description of these techniques can be found in reference [2]. A brief description of them will be given here. The residuals can be plotted in the following ways;

A. Overall plots

B. Time sequence plots

C. Against the fitted values y_i, for i=1, 2, ..., n

D. Against the independent variable $a_{j\mu}$, for j = 1, 2, ..., K

4.7a Overall plots of residuals

If the model is correct and appropriate for predictive purposes the residuals should resemble thirty-seven observations (equal to the number of data points) from a normal distribution with mean zero. The mean should necessarily be zero for any regression model with the constant term in it. The constant terms in our models are a_o, b_o, m_o, d_o, e_o and h_o.

One can make use of a table of random deviates for judging the overall plots of residuals. An extensive table is published by the Rand Corporation [10]. Thirty-seven normal deviates were selected randomly from this table and plotted. Figure 1 shows the plot of random normal deviates. Now the overall residual plots obtained from different models were compared with this plot. Visual observation reveals that the plot obtained by using a fifth order model (Figure 2) is best as compared to the others.

An alternative approach will be to construct the "normal plot" or a "half normal" plot of the residuals on standard probability paper. The points should fall approximately on a straight line. Daniel [11] has described the use of these plots.

Overall residual plots obtained from other models are included in the Appendix and can be used for comparison.

4.7b Time sequence plot of residuals

If the "step back" view of the time sequence plots gives an impression of a horizontal "band" of residuals as shown in Figure 3, one can conclude that a long-term time effect is not influencing data. However, if we obtain "step back" views from the plots of residuals resembling those of Figure 4, it is indicative of the fact that a time-effect was not accounted for. The

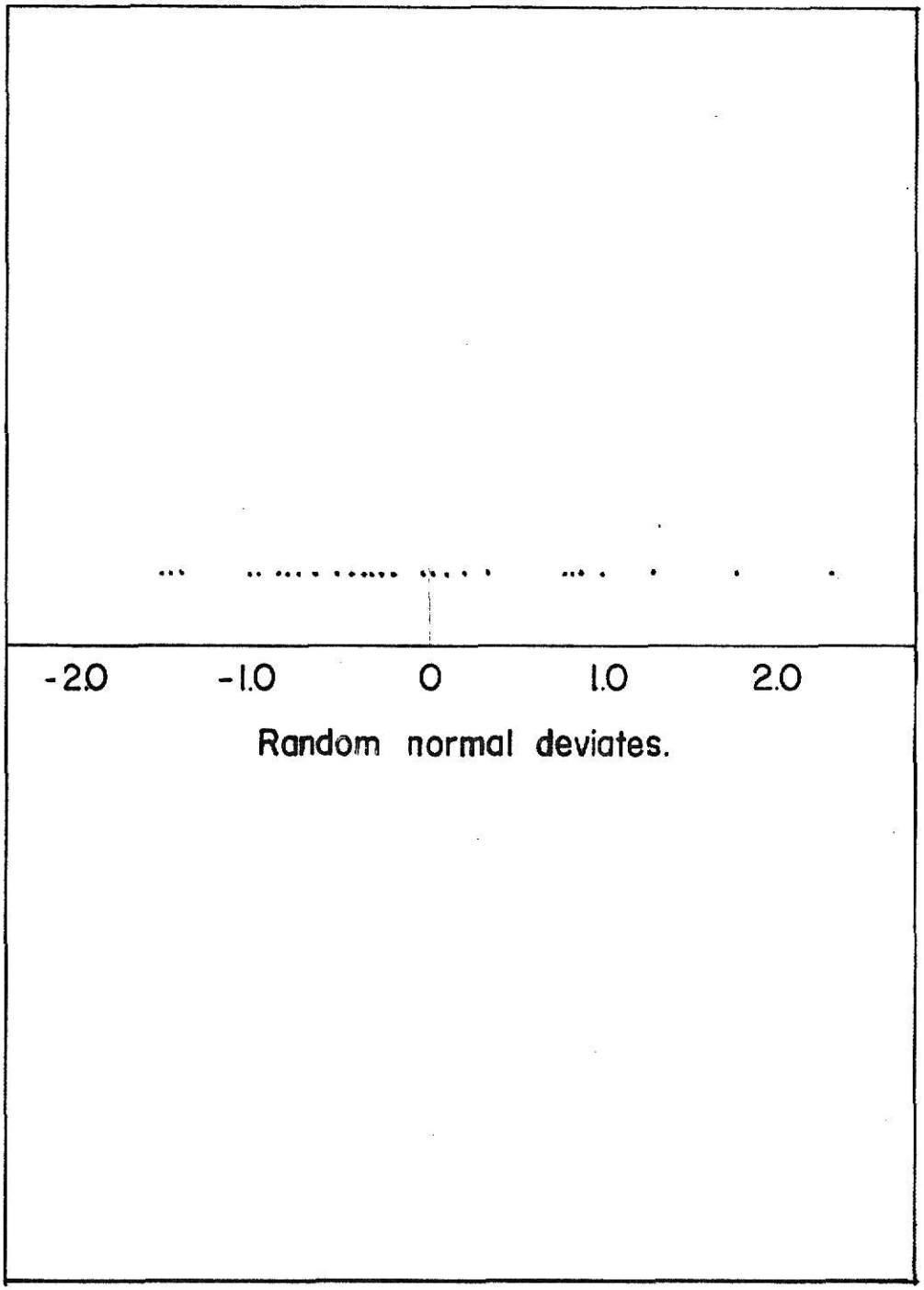

Fig. 1. Plot of random normal deviates.

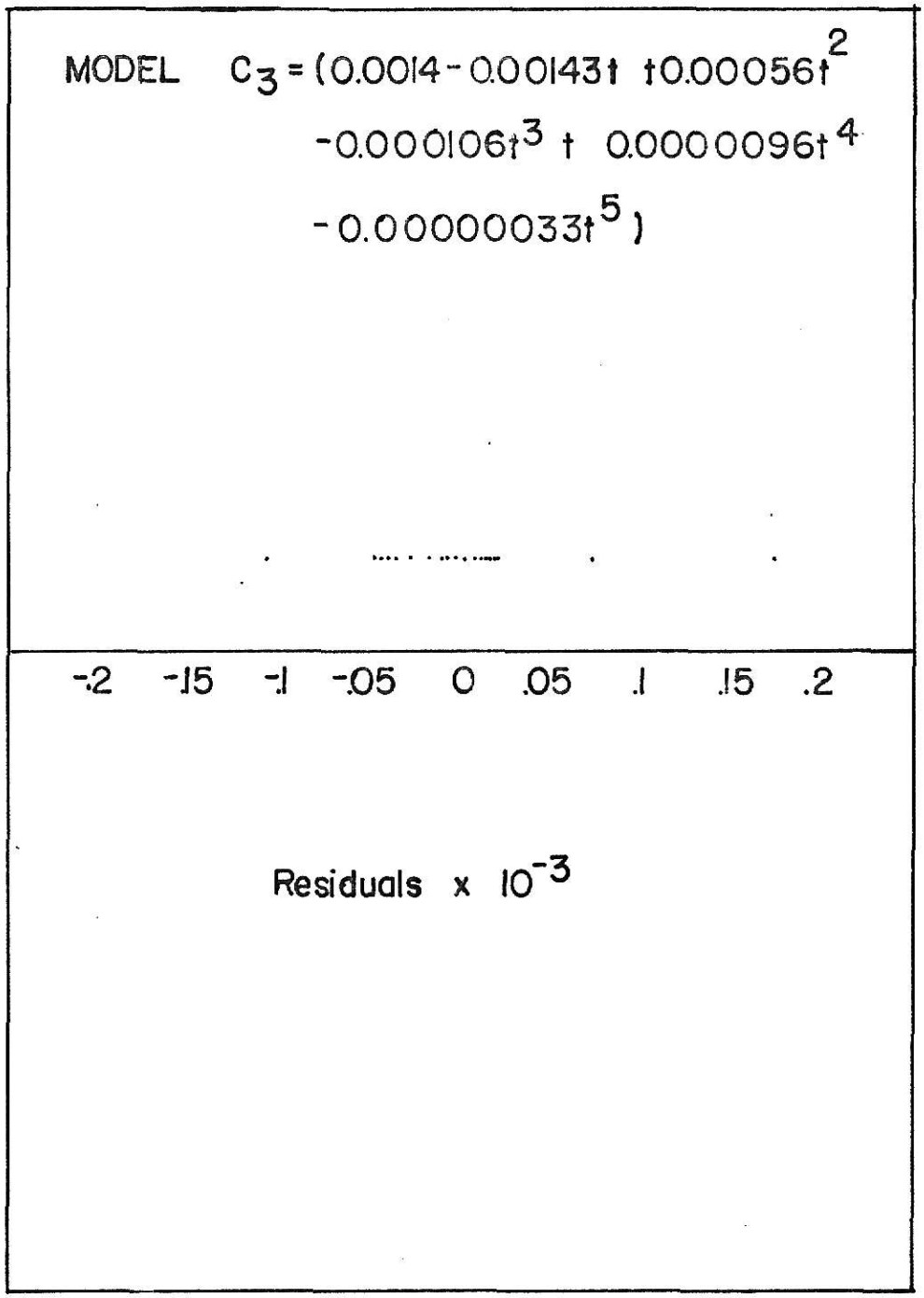

Fig. 2. Overall plot of residuals.

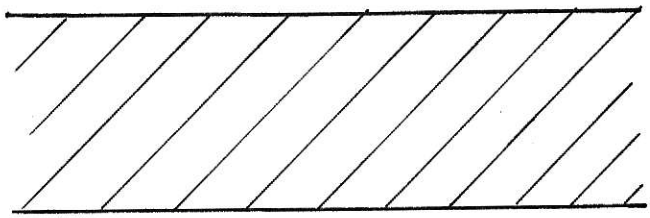

Fig.3. Horizontal band of residuals.

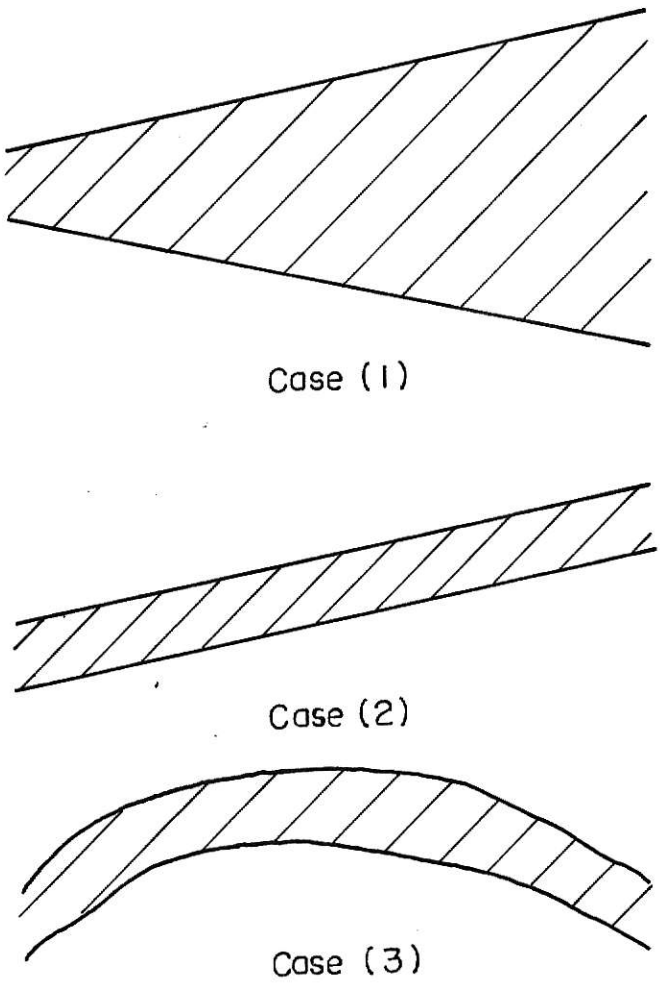

Fig.4. Discrepencies indicated by "step" back view.

various cases in Figure 4 indicate the following facts.

Case 1. The variance is not constant but increases with time. This suggests that weighted least squares analysis should have been used.

Case 2. This indicates that a linear term in time should have been included in the model.

Case 3. This suggests that linear and quardratic terms in time should have been included in the model.

The observation of time sequence plots for the various models indicates that the "step-back" view of a plot of the fifth-order model (Figure 5) can be closely approximated by Figure 3. Most of the residuals lie within a horizontal strip, which is indicative of the fact, as explained earlier that a long-term time effect is not influencing the data. This conclusion is restricted to the fifth order model only. Comparison between the fifth order model and the sixth order model indicates that increasing the order of the model does not necessarily make it better for predictive purposes (Figure 5).

4.7c Plots of residuals vs. predicted values of response

Tables 9 through 14 show the predicted values of tracer concentrations in tank (3) and residuals for various models. Once again the "horizontal band" indicates no abnormality. Defects or abnormalities will be indicated by plots of the form shown in cases (1), (2) and (3) in Figure 4. The following facts are indicated by the various cases in Figure 4.

Case 1. The variance is not constant, as assumed. This suggests the use of weighted least squares or a transformation on the observations

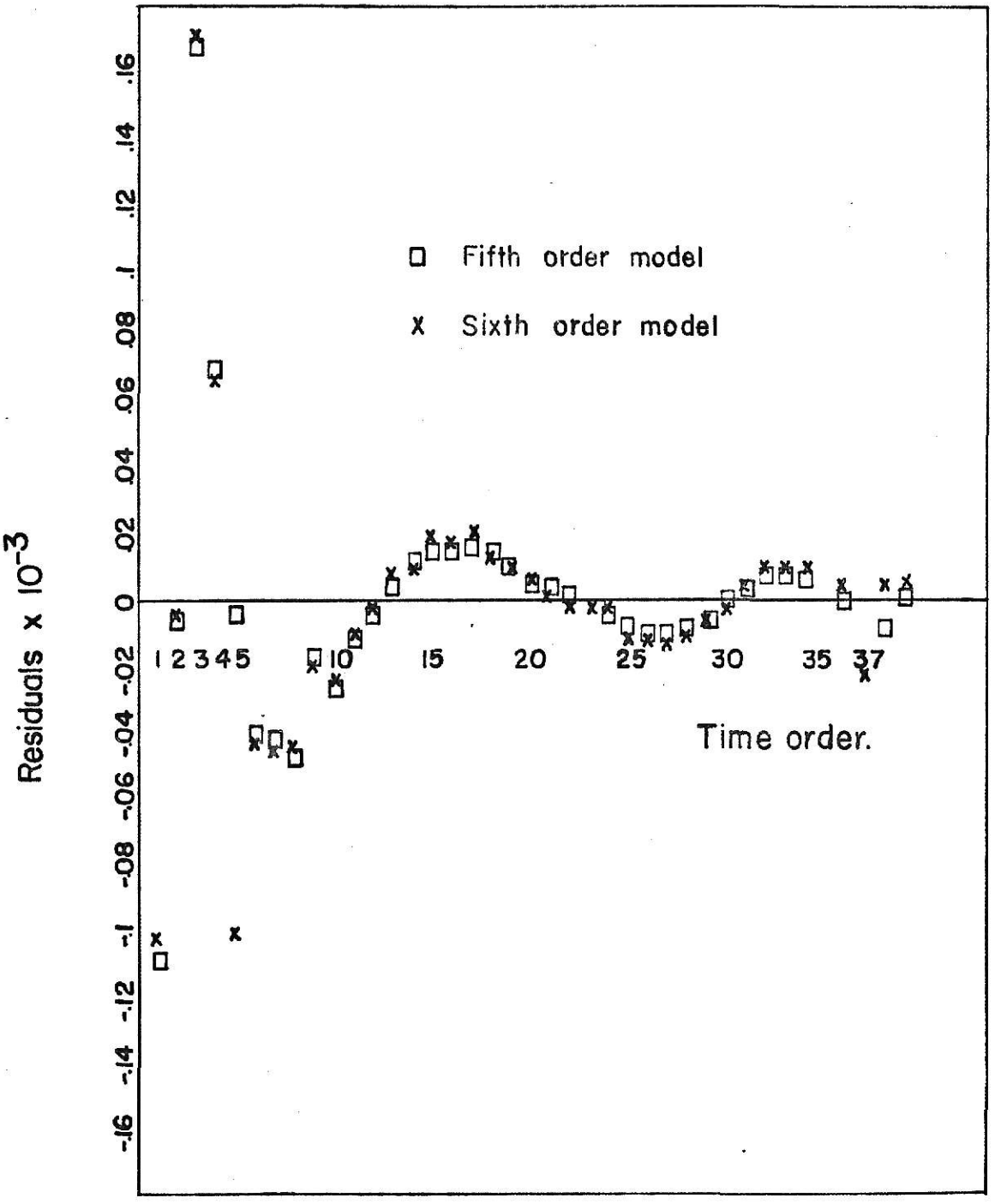

Fig.5. Time sequence plot of residuals.

Table 1. Mean and Standard Deviation of Parameters.

MODEL $C_3 = (0.00055 - 0.000097t)$

Parameter	Mean (Expected Value)	Standard Deviation
a'_0	0.5562×10^{-3}	0.7338×10^{-4}
a'_1	-0.9756×10^{-4}	0.1743×10^{-4}

Table 2. Mean and Standard Deviation of Parameters

MODEL $C_3 = (0.0009 - 0.00037t + 0.000033t^2)$

Parameter	Mean (Expected Value)	Standard Deviation
b_0	0.9258×10^{-3}	0.6768×10^{-4}
b_1	-0.3714×10^{-3}	0.3841×10^{-4}
b_2	0.3306×10^{-4}	0.4446×10^{-5}

Table 3. Mean and Standard Deviation of Parameters.

MODEL $C_3 = (0.0012 - 0.00076t + 0.000147t^2 - 0.0000088t^3)$

Parameter	Mean (Expected Value)	Standard Deviation
m_0	-0.7601×10^{-3}	0.5034×10^{-4}
m_1	0.1473×10^{-3}	0.1358×10^{-4}
m_2	-0.8794×10^{-5}	0.1027×10^{-5}
m_3	0.1204×10^{-2}	0.5025×10^{-4}

Table 4. Mean and Standard Deviation of Parameters.

MODEL $C_3 = (0.0013 - 0.00117t + 0.00036t^2 - 0.0000464t^3 + 0.0000021t^4)$

Parameter	Mean (Expected Value)	Standard Deviation
d_0	0.1391×10^{-2}	0.3589×10^{-4}
d_1	-0.1179×10^{-2}	0.5691×10^{-4}
d_2	0.3620×10^{-3}	0.2637×10^{-4}
d_3	-0.4642×10^{-4}	0.4463×10^{-5}
d_4	0.2099×10^{-5}	0.2470×10^{-6}

Table 5. Mean and Standard Deviation of Parameters.

MODEL $C_3 = (0.0014 - 0.00143t + 0.00056t^2 - 0.000106t^3 + 0.0000096t^4 - 0.00000033t^5$

Parameter	Mean (Expected Value)	Standard Deviation
e'_0	0.1468×10^{-2}	0.7470×10^{-4}
e'_1	-0.1432×10^{-2}	0.5070×10^{-4}
e'_2	0.5616×10^{-3}	0.1436×10^{-4}
e'_3	-0.1065×10^{-3}	0.1779×10^{-5}
e'_4	0.9694×10^{-5}	0.3509×10^{-7}
e'_5	-0.3386×10^{-6}	0.7940×10^{-7}

Table 6. Mean and Standard Deviation of Parameters.

MODEL $C_3 = (0.0014 - 0.00137t + 0.000496t^2 - 0.0000772t^3 + 0.00000352t^2 + 0.000000269t^5 - 0.0000000225t^6)$

Parameter	Mean (Expected Value)	Standard Deviation
h_0	0.1455×10^{-2}	0.3562×10^{-4}
h_1	-0.1374×10^{-2}	0.7547×10^{-4}
h_2	0.4965×10^{-3}	0.5308×10^{-4}
h_3	-0.7726×10^{-4}	0.1801×10^{-4}
h_4	0.3527×10^{-5}	0.3473×10^{-5}
h_5	0.2691×10^{-6}	0.3590×10^{-6}
h_6	-0.2258×10^{-7}	0.1472×10^{-7}

Table 7. Mean and Sum of Squares of Residuals for Different Models.

Model	No. of Parameters	Sum of Squares of Residuals	Mean of Residuals
$C_3 = a_0' + a_1't$	2	0.2183×10^{-5}	2.73×10^{-6}
$C_3 = b_0 + b_1t + b_2t^2$	3	0.8311×10^{-6}	3.39×10^{-10}
$C_3 = m_0 + m_1t + m_2t^2 + m_3t^3$	4	0.2581×10^{-6}	9.67×10^{-11}
$C_3 = d_0 + d_1t + d_2t^2 + d_3t^3 + d_4t^4$	5	0.7993×10^{-7}	9.53×10^{-10}
$C_3 = e_0' + e_1't + e_2't^2 + e_3't^3 + e_4't^4 + e_5't^5$	6	0.5691×10^{-7}	2.58×10^{-9}
$C_3 = h_0 + h_1t + h_2t^2 + h_3t^3 + h_4t^4 + h_5t^5 + h_6t^6$	7	0.5651×10^{-7}	4.78×10^{-7}

Table 8. Computing Time, No. of Iterations and Optimum Function Value for Different Models.

Model	No. of Parameters	Computing Time Sec.	Optimum Function Value	Function Evaluations	Devivative Evaluations
$C_3 = a_0' + a_1't$	2	25.20	0.975×10^{-4}	6	4
$C_3 = b_0 + b_1t + b_2t^2$	3	28.80	0.831×10^{-6}	6	4
$C_3 = m_0 + m_1t + m_2t^2 + m_3t^3$	4	31.2	0.258×10^{-6}	8	5
$C_3 = d_0 + d_1t + d_2t^2 + d_3t^3 + d_4t^4$	5	39.50	0.799×10^{-7}	15	7
$C_3 = e_0' + e_1't + e_2't^2 + e_3't^3 + e_4't^4 + e_5't^5$	6	57.50	0.569×10^{-7}	19	9
$C_3 = h_0 + h_1t + h_2t^2 + h_3t^3 + h_4t^4 + h_5t^5 + h_6t^6$	7	407	0.565×10^{-7}	237	81

Table 9. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.00055 - 0.000097t)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	0.5449×10^{-3}	0.1200×10^{-2}	0.6511×10^{-3}
0.2666	0.5302×10^{-3}	0.1120×10^{-2}	0.5897×10^{-3}
0.4333	0.5140×10^{-3}	0.1120×10^{-2}	0.6060×10^{-3}
0.6000	0.4977×10^{-3}	0.8600×10^{-3}	0.3622×10^{-3}
0.7666	0.4815×10^{-3}	0.6500×10^{-3}	0.1685×10^{-3}
0.9333	0.4652×10^{-3}	0.5000×10^{-3}	0.3481×10^{-4}
1.1166	0.4473×10^{-3}	0.3899×10^{-3}	-0.5730×10^{-4}
1.2667	0.4327×10^{-3}	0.3150×10^{-3}	-0.1176×10^{-3}
1.4333	0.4164×10^{-3}	0.2750×10^{-3}	-0.1414×10^{-3}
1.6000	0.4001×10^{-3}	0.2100×10^{-3}	-0.1901×10^{-3}
1.7667	0.3839×10^{-3}	0.1800×10^{-3}	-0.2038×10^{-3}
1.9333	0.3676×10^{-3}	0.1500×10^{-3}	-0.2176×10^{-3}
2.1000	0.3514×10^{-3}	0.1300×10^{-3}	-0.2213×10^{-3}
2.2667	0.3351×10^{-3}	0.1125×10^{-3}	-0.2226×10^{-3}
2.4333	0.3188×10^{-3}	0.1000×10^{-3}	-0.2188×10^{-3}
2.6000	0.3026×10^{-3}	0.8600×10^{-4}	-0.2165×10^{-3}
2.7667	0.2863×10^{-3}	0.7800×10^{-4}	-0.2083×10^{-3}
2.9333	0.2701×10^{-3}	0.6799×10^{-4}	-0.2020×10^{-3}
3.1000	0.2438×10^{-3}	0.5900×10^{-4}	-0.1947×10^{-3}
3.2667	0.2375×10^{-3}	0.5300×10^{-4}	-0.1845×10^{-3}
3.4333	0.2213×10^{-3}	0.4699×10^{-4}	-0.1742×10^{-3}
3.6166	0.2034×10^{-3}	0.4299×10^{-4}	-0.1603×10^{-3}
3.7667	0.1888×10^{-3}	0.3900×10^{-4}	-0.1497×10^{-3}
3.9500	0.1709×10^{-3}	0.3500×10^{-4}	-0.1358×10^{-3}
4.2333	0.1432×10^{-3}	0.2900×10^{-4}	-0.1142×10^{-3}
4.4833	0.1188×10^{-3}	0.2500×10^{-4}	-0.9383×10^{-4}
4.7333	0.9444×10^{-4}	0.2150×10^{-4}	-0.7294×10^{-4}
5.0667	0.6192×10^{-4}	0.1800×10^{-4}	-0.4392×10^{-4}
5.4000	0.2940×10^{-4}	0.1500×10^{-4}	-0.1440×10^{-4}
5.7333	-0.3120×10^{-5}	0.1200×10^{-4}	0.1511×10^{-4}
6.0667	-0.3564×10^{-4}	0.1050×10^{-4}	0.4614×10^{-4}
6.4000	-0.6816×10^{-4}	0.8900×10^{-5}	0.7706×10^{-4}
6.7333	-0.1007×10^{-3}	0.7400×10^{-5}	0.1080×10^{-4}
7.0667	-0.6500×10^{-5}	0.6500×10^{-5}	0.1397×10^{-3}
7.5666	-0.1820×10^{-3}	0.5200×10^{-5}	0.1871×10^{-3}
8.0667	-0.2308×10^{-3}	0.4450×10^{-5}	0.2352×10^{-3}
9.0667	-0.3283×10^{-3}	0.3300×10^{-5}	0.3316×10^{-3}

Table 10. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.0009 - 0.00037t + 0.000033t^2)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	0.8820×10^{-3}	0.1200×10^{-2}	0.3169×10^{-3}
0.2666	0.8292×10^{-3}	0.1120×10^{-2}	0.2907×10^{-3}
0.4333	0.7712×10^{-3}	0.1120×10^{-2}	0.3488×10^{-3}
0.6000	0.7149×10^{-3}	0.8600×10^{-3}	0.1450×10^{-3}
0.7666	0.6606×10^{-3}	0.6500×10^{-3}	-0.1057×10^{-4}
0.9333	0.6080×10^{-3}	0.5000×10^{-3}	-0.1080×10^{-3}
1.1166	0.5524×10^{-3}	0.3899×10^{-3}	-0.1623×10^{-3}
1.2667	0.5084×10^{-3}	0.3150×10^{-3}	-0.1934×10^{-3}
1.4333	0.4614×10^{-3}	0.2750×10^{-3}	-0.1864×10^{-3}
1.6000	0.4162×10^{-3}	0.2100×10^{-3}	-0.2062×10^{-3}
1.7667	0.3729×10^{-3}	0.1800×10^{-3}	-0.1928×10^{-3}
1.9333	0.3314×10^{-3}	0.1500×10^{-3}	-0.1813×10^{-3}
2.1000	0.2917×10^{-3}	0.1300×10^{-3}	-0.1616×10^{-3}
2.2667	0.2538×10^{-3}	0.1125×10^{-3}	-0.1413×10^{-3}
2.4333	0.2178×10^{-3}	0.1000×10^{-3}	-0.1178×10^{-3}
2.6000	0.1837×10^{-3}	0.8600×10^{-4}	-0.9766×10^{-4}
2.7667	0.1513×10^{-3}	0.7800×10^{-4}	-0.7333×10^{-4}
2.9333	0.1208×10^{-3}	0.6799×10^{-4}	-0.5283×10^{-4}
3.1000	0.9218×10^{-4}	0.5900×10^{-4}	-0.3318×10^{-4}
3.2667	0.6536×10^{-4}	0.5300×10^{-4}	-0.1236×10^{-4}
3.4333	0.4038×10^{-4}	0.4699×10^{-4}	0.6621×10^{-5}
3.6166	0.1503×10^{-4}	0.4299×10^{-4}	0.2797×10^{-4}
3.7667	-0.4077×10^{-5}	0.3900×10^{-4}	0.4307×10^{-4}
3.9500	-0.2540×10^{-4}	0.3500×10^{-4}	0.6039×10^{-4}
4.2333	-0.5397×10^{-4}	0.2900×10^{-4}	0.8296×10^{-4}
4.4833	-0.7477×10^{-4}	0.2500×10^{-4}	0.9977×10^{-4}
4.7333	-0.9144×10^{-4}	0.2150×10^{-4}	0.1129×10^{-3}
5.0667	-0.1072×10^{-3}	0.1800×10^{-4}	0.1252×10^{-3}
5.4000	-0.1157×10^{-3}	0.1500×10^{-4}	0.1306×10^{-3}
5.7333	-0.1168×10^{-3}	0.1200×10^{-4}	0.1287×10^{-3}
6.0667	-0.1105×10^{-3}	0.1050×10^{-4}	0.1210×10^{-3}
6.4000	-0.9695×10^{-4}	0.8900×10^{-5}	0.1058×10^{-3}
6.7333	-0.7600×10^{-4}	0.7400×10^{-5}	0.8340×10^{-4}
7.0667	-0.4771×10^{-4}	0.6500×10^{-5}	0.5421×10^{-4}
7.5666	0.8497×10^{-5}	0.5200×10^{-5}	-0.3297×10^{-5}
8.0667	0.8125×10^{-4}	0.4450×10^{-5}	-0.7680×10^{-4}
9.0667	0.2764×10^{-3}	0.3300×10^{-5}	-0.2730×10^{-3}

Table 11. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.0012 - 0.00076t + 0.000147t^2 - 0.0000088t^3)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	$0.1118X10^{-2}$	$0.1200X10^{-2}$	$0.8215X10^{-3}$
0.2666	$0.1012X10^{-2}$	$0.1120X10^{-2}$	$0.1078X10^{-3}$
0.4333	$0.9021X10^{-3}$	$0.1120X10^{-2}$	$0.2179X10^{-3}$
0.6000	$0.7995X10^{-3}$	$0.8600X10^{-3}$	$0.6045X10^{-4}$
0.7666	$0.7044X10^{-3}$	$0.6500X10^{-3}$	$-0.5439X10^{-4}$
0.9333	$0.6162X10^{-3}$	$0.5000X10^{-3}$	$-0.1162X10^{-3}$
1.1166	$0.5272X10^{-3}$	$0.3899X10^{-3}$	$-0.1371X10^{-3}$
1.2667	$0.4602X10^{-3}$	$0.3150X10^{-3}$	$-0.1451X10^{-3}$
1.4333	$0.3918X10^{-3}$	$0.2750X10^{-3}$	$-0.1167X10^{-3}$
1.6000	$0.3295X10^{-3}$	$0.2100X10^{-3}$	$-0.1194X10^{-3}$
1.7667	$0.2730X10^{-3}$	$0.1800X10^{-3}$	$-0.9300X10^{-4}$
1.9333	$0.2221X10^{-3}$	$0.1500X10^{-3}$	$-0.7212X10^{-4}$
2.1000	$0.1766X10^{-3}$	$0.1300X10^{-3}$	$-0.4659X10^{-4}$
2.2667	$0.1362X10^{-3}$	$0.1125X10^{-3}$	$-0.2368X10^{-4}$
2.4333	$0.1006X10^{-3}$	$0.1000X10^{-3}$	$-0.6351X10^{-6}$
2.6000	$0.6971X10^{-4}$	$0.8600X10^{-4}$	$0.1629X10^{-4}$
2.7667	$0.4315X10^{-4}$	$0.7800X10^{-4}$	$0.3484X10^{-4}$
2.9333	$0.2073X10^{-4}$	$0.6799X10^{-4}$	$0.4726X10^{-4}$
3.1000	$0.2198X10^{-5}$	$0.5900X10^{-4}$	$0.5680X10^{-4}$
3.2667	$-0.1269X10^{-4}$	$0.5300X10^{-4}$	$0.6569X10^{-4}$
3.4333	$-0.2418X10^{-4}$	$0.4699X10^{-4}$	$0.7118X10^{-4}$
3.6166	$-0.3319X10^{-4}$	$0.4299X10^{-4}$	$0.7618X10^{-4}$
3.7667	$-0.3795X10^{-4}$	$0.3900X10^{-4}$	$0.7694X10^{-4}$
3.9500	$-0.4085X10^{-4}$	$0.3500X10^{-4}$	$0.7584X10^{-4}$
4.2333	$-0.3974X10^{-4}$	$0.2900X10^{-4}$	$0.6873X10^{-4}$
4.4833	$-0.3396X10^{-4}$	$0.2500X10^{-4}$	$0.5896X10^{-4}$
4.7333	$-0.2455X10^{-4}$	$0.2150X10^{-4}$	$0.4605X10^{-4}$
5.0667	$-0.7775X10^{-5}$	$0.1800X10^{-4}$	$0.2577X10^{-4}$
5.4000	$0.1205X10^{-4}$	$0.1500X10^{-4}$	$0.2955X10^{-5}$
5.7333	$0.3295X10^{-4}$	$0.1200X10^{-4}$	$-0.2095X10^{-4}$
6.0667	$0.5299X10^{-4}$	$0.1050X10^{-4}$	$-0.4249X10^{-4}$
6.4000	$0.7022X10^{-4}$	$0.8900X10^{-5}$	$-0.6131X10^{-4}$
6.7333	$0.8267X10^{-4}$	$0.7400X10^{-5}$	$-0.7526X10^{-4}$
7.0667	$0.8838X10^{-4}$	$0.6500X10^{-5}$	$-0.8188X10^{-4}$
7.5666	$0.8008X10^{-4}$	$0.5200X10^{-5}$	$-0.7487X10^{-4}$
3.0667	$0.4563X10^{-4}$	$0.4450X10^{-5}$	$-0.4117X10^{-4}$
9.0667	$-0.1280X10^{-3}$	$0.3300X10^{-5}$	$0.1313X1o^{-3}$

Table 12. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.0013 - 0.00117t + 0.00036t^2 - 0.0000464t^3 + 0.0000021t^4)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	0.1259×10^{-2}	0.1200×10^{-2}	-0.5878×10^{-4}
0.2666	0.1102×10^{-2}	0.1120×10^{-2}	0.1814×10^{-4}
0.4333	0.9446×10^{-3}	0.1120×10^{-2}	0.1753×10^{-3}
0.6000	0.8043×10^{-3}	0.8600×10^{-3}	0.5571×10^{-4}
0.7666	0.6797×10^{-3}	0.6500×10^{-3}	-0.2974×10^{-4}
0.9333	0.5697×10^{-3}	0.5000×10^{-3}	-0.6971×10^{-4}
1.1166	0.4643×10^{-3}	0.3899×10^{-3}	-0.7431×10^{-4}
1.2667	0.3892×10^{-3}	0.3150×10^{-3}	-0.7419×10^{-4}
1.4333	0.3166×10^{-3}	0.2750×10^{-3}	-0.4160×10^{-4}
1.6000	0.2545×10^{-3}	0.2100×10^{-3}	-0.4446×10^{-4}
1.7667	0.2019×10^{-3}	0.1800×10^{-3}	-0.2186×10^{-4}
1.9333	0.1579×10^{-3}	0.1500×10^{-3}	-0.7879×10^{-5}
2.1000	0.1217×10^{-3}	0.1300×10^{-3}	0.8327×10^{-5}
2.2667	0.9242×10^{-4}	0.1125×10^{-3}	0.2008×10^{-4}
2.4333	0.6934×10^{-4}	0.1000×10^{-3}	0.3066×10^{-4}
2.6000	0.5168×10^{-4}	0.8600×10^{-4}	0.3431×10^{-4}
2.7667	0.3876×10^{-4}	0.7800×10^{-4}	0.3924×10^{-4}
2.9333	0.2990×10^{-4}	0.6799×10^{-4}	0.3810×10^{-4}
3.1000	0.2448×10^{-4}	0.5900×10^{-4}	0.3452×10^{-4}
3.2667	0.2191×10^{-4}	0.5300×10^{-4}	0.3108×10^{-4}
3.4333	0.2165×10^{-4}	0.4699×10^{-4}	0.2534×10^{-4}
3.6166	0.2343×10^{-4}	0.4299×10^{-4}	0.1957×10^{-4}
3.7667	0.2606×10^{-4}	0.3900×10^{-4}	0.1293×10^{-4}
3.9500	0.3024×10^{-4}	0.3500×10^{-4}	0.4756×10^{-5}
4.2333	0.3765×10^{-4}	0.2900×10^{-4}	-0.8652×10^{-5}
4.4833	0.4402×10^{-4}	0.2500×10^{-4}	-0.1902×10^{-4}
4.7333	0.4925×10^{-4}	0.2150×10^{-4}	-0.2775×10^{-4}
5.0667	0.5324×10^{-4}	0.1800×10^{-4}	-0.3524×10^{-4}
5.4000	0.5278×10^{-4}	0.1500×10^{-4}	-0.3778×10^{-4}
5.7333	0.4732×10^{-4}	0.1200×10^{-4}	-0.3532×10^{-4}
6.0667	0.3693×10^{-4}	0.1050×10^{-4}	-0.2643×10^{-4}
6.4000	0.2232×10^{-4}	0.8900×10^{-5}	-0.1342×10^{-4}
6.7333	0.4785×10^{-5}	0.7400×10^{-5}	0.2611×10^{-5}
7.0667	-0.1373×10^{-4}	0.6500×10^{-5}	0.2022×10^{-4}
7.5666	-0.3754×10^{-4}	0.5200×10^{-5}	0.4273×10^{-4}
8.0667	-0.4638×10^{-4}	0.4450×10^{-5}	0.5083×10^{-4}
9.0667	0.4172×10^{-4}	0.3300×10^{-5}	-0.3841×10^{-4}

Table 13. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.0014 - 0.00143t + 0.00056t^2 - 0.000106t^3 + 0.0000096t^4 - 0.00000033t^5)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	0.1309×10^{-2}	0.1200×10^{-2}	-0.1092×10^{-3}
0.2666	0.1125×10^{-2}	0.1120×10^{-2}	-0.4830×10^{-5}
0.4333	0.9451×10^{-3}	0.1120×10^{-2}	0.1748×10^{-3}
0.6000	0.7895×10^{-3}	0.8600×10^{-3}	0.7045×10^{-4}
0.7666	0.6557×10^{-3}	0.6500×10^{-3}	-0.5722×10^{-5}
0.9333	0.5412×10^{-3}	0.5000×10^{-3}	-0.4122×10^{-4}
1.1166	0.4353×10^{-3}	0.3899×10^{-3}	-0.4326×10^{-4}
1.2667	0.3623×10^{-3}	0.3150×10^{-3}	-0.4726×10^{-4}
1.4333	0.2940×10^{-3}	0.2750×10^{-3}	-0.1897×10^{-4}
1.6000	0.2375×10^{-3}	0.2100×10^{-3}	-0.2752×10^{-4}
1.7667	0.1914×10^{-3}	0.1800×10^{-3}	-0.1139×10^{-4}
1.9333	0.1541×10^{-3}	0.1500×10^{-3}	-0.4143×10^{-5}
2.1000	0.1245×10^{-3}	0.1300×10^{-3}	0.5505×10^{-5}
2.2667	0.1013×10^{-3}	0.1125×10^{-3}	0.1122×10^{-4}
2.4333	0.8344×10^{-4}	0.1000×10^{-3}	0.1656×10^{-4}
2.6000	0.7002×10^{-4}	0.8600×10^{-4}	0.1598×10^{-4}
2.7667	0.6019×10^{-4}	0.7800×10^{-4}	0.1781×10^{-4}
2.9333	0.5318×10^{-4}	0.6799×10^{-4}	0.1481×10^{-4}
3.1000	0.4836×10^{-4}	0.5900×10^{-4}	0.1063×10^{-4}
3.2667	0.4514×10^{-4}	0.5300×10^{-4}	0.7850×10^{-5}
3.4333	0.4307×10^{-4}	0.4699×10^{-4}	0.3926×10^{-5}
3.6166	0.4161×10^{-4}	0.4299×10^{-4}	0.1393×10^{-5}
3.7667	0.4074×10^{-4}	0.3900×10^{-4}	-0.1745×10^{-5}
3.9500	0.3980×10^{-4}	0.3500×10^{-4}	-0.4804×10^{-5}
4.2333	0.3802×10^{-4}	0.2900×10^{-4}	-0.9020×10^{-5}
4.4833	0.3570×10^{-4}	0.2500×10^{-4}	-0.1069×10^{-4}
4.7333	0.3248×10^{-4}	0.2150×10^{-4}	-0.1097×10^{-4}
5.0667	0.2685×10^{-4}	0.1800×10^{-4}	-0.8843×10^{-5}
5.4000	0.2012×10^{-4}	0.1500×10^{-4}	-0.5116×10^{-5}
5.7333	0.1311×10^{-4}	0.1200×10^{-4}	-0.1095×10^{-5}
6.0667	0.6738×10^{-5}	0.1050×10^{-4}	0.3768×10^{-5}
6.4000	0.1994×10^{-5}	0.8900×10^{-5}	0.6925×10^{-5}
6.7333	-0.3990×10^{-6}	0.7400×10^{-5}	0.7811×10^{-5}
7.0667	0.2339×10^{-7}	0.6500×10^{-5}	0.6489×10^{-5}
7.5666	0.5652×10^{-5}	0.5200×10^{-5}	-0.4358×10^{-6}
8.0667	0.1402×10^{-4}	0.4450×10^{-5}	-0.9551×10^{-5}
9.0667	0.1455×10^{-5}	0.3300×10^{-5}	0.1933×10^{-5}

Table 14. Residuals and Predicted Values of Tracer Concentration in Tank (3).

MODEL $C_3 = (0.0014 - 0.00137t + 0.000496t^2 - 0.0000772t^3 + 0.00000352t^4 + 0.000000269t^5 - 0.0000000225t^6)$

Time, hrs.	C_3 (Predicted)	C_3 (Experimental)	Residuals
0.1166	0.1302×10^{-2}	0.1200×10^{-2}	-0.1021×10^{-3}
0.2666	0.1123×10^{-2}	0.1120×10^{-2}	-0.3187×10^{-5}
0.4333	0.9473×10^{-3}	0.1120×10^{-2}	0.1727×10^{-3}
0.6000	0.7936×10^{-3}	0.8600×10^{-3}	0.6637×10^{-4}
0.7666	0.6604×10^{-3}	0.6500×10^{-3}	-0.1036×10^{-4}
0.9333	0.5455×10^{-3}	0.5000×10^{-3}	-0.4548×10^{-4}
1.1166	0.4384×10^{-3}	0.3899×10^{-3}	-0.4842×10^{-4}
1.2667	0.3642×10^{-3}	0.3150×10^{-3}	-0.4923×10^{-4}
1.4333	0.2945×10^{-3}	0.2750×10^{-3}	-0.1954×10^{-4}
1.6000	0.2368×10^{-3}	0.2100×10^{-3}	-0.2676×10^{-4}
1.7667	0.1895×10^{-3}	0.1800×10^{-3}	-0.9474×10^{-5}
1.9333	0.1513×10^{-3}	0.1500×10^{-3}	-0.1332×10^{-5}
2.1000	0.1211×10^{-3}	0.1300×10^{-3}	-0.8903×10^{-5}
2.2667	0.9762×10^{-4}	0.1125×10^{-3}	0.1488×10^{-4}
2.4333	0.7982×10^{-4}	0.1000×10^{-3}	0.2017×10^{-4}
2.6000	0.6674×10^{-4}	0.8600×10^{-4}	0.1926×10^{-4}
2.7667	0.5748×10^{-4}	0.7800×10^{-4}	0.2051×10^{-4}
2.9333	0.5125×10^{-4}	0.6799×10^{-4}	0.1674×10^{-4}
3.1000	0.4734×10^{-4}	0.5900×10^{-4}	0.1166×10^{-4}
3.2667	0.4510×10^{-4}	0.5300×10^{-4}	0.7897×10^{-5}
3.4333	0.4401×10^{-4}	0.4699×10^{-4}	0.2989×10^{-5}
3.6166	0.4357×10^{-4}	0.4299×10^{-4}	-0.5737×10^{-6}
3.7667	0.4345×10^{-4}	0.3900×10^{-4}	-0.4457×10^{-5}
3.9500	0.4325×10^{-4}	0.3500×10^{-4}	-0.8253×10^{-5}
4.2333	0.4214×10^{-4}	0.2900×10^{-4}	-0.1315×10^{-4}
4.4833	0.3986×10^{-4}	0.2500×10^{-4}	-0.1486×10^{-4}
4.7333	0.3615×10^{-4}	0.2150×10^{-4}	-0.1464×10^{-4}
5.0667	0.2908×10^{-4}	0.1800×10^{-4}	-0.1108×10^{-4}
5.4000	0.2031×10^{-4}	0.1500×10^{-4}	-0.5310×10^{-5}
5.7333	0.1102×10^{-4}	0.1200×10^{-4}	0.9788×10^{-6}
6.0667	0.2693×10^{-5}	0.1050×10^{-4}	0.7805×10^{-5}
6.4000	-0.3170×10^{-5}	0.8900×10^{-5}	0.1207×10^{-4}
6.7333	-0.5344×10^{-5}	0.7400×10^{-5}	0.1274×10^{-4}
7.0667	-0.3070×10^{-5}	0.6500×10^{-5}	0.9569×10^{-5}
7.5666	0.8222×10^{-5}	0.5200×10^{-5}	-0.3024×10^{-4}
8.0667	0.2350×10^{-4}	0.4450×10^{-5}	-0.1905×10^{-4}
9.0667	-0.1866×10^{-5}	0.3300×10^{-5}	0.5163×10^{-5}

Y_i (experimental values of tracer concentration in tank (3)) before making a regression analysis.

Case 2. This indicates probable error in the analysis, and also tells that the departure from the fitted equation is systematic, which means that negative residuals correspond to low predicted values of Y_i. This effect can also be due to omitting a constant term in the model.

Case 3. This type of step-back view indicates that the assumed mathematical model is inadequate and extra terms need to be included in the model. These extra terms may be higher order terms or cross-product terms. This also suggests a need for the transformation of the experimental observations Y_i (experimental values of C_3 in our case) before analysis. It was observed that the fifth order model (Figure 5) shows least abnormalities.

4.7d Plots of residuals vs. independent variable

In these plots also, an overall impression of a horizontal band is considered to be satisfactory. Observation of plots for various models in this case also indicate that the fifth order model (Figure 6) approximates closely the ideal case. Most of the residuals here lie inside the horizontal band. The abnormalities indicated in Figure 4 here reveal that

Case 1. The variance is not constant. It is suggested to use weighted least squares or some transformation on Y_i (experimental values of C_3 in our case).

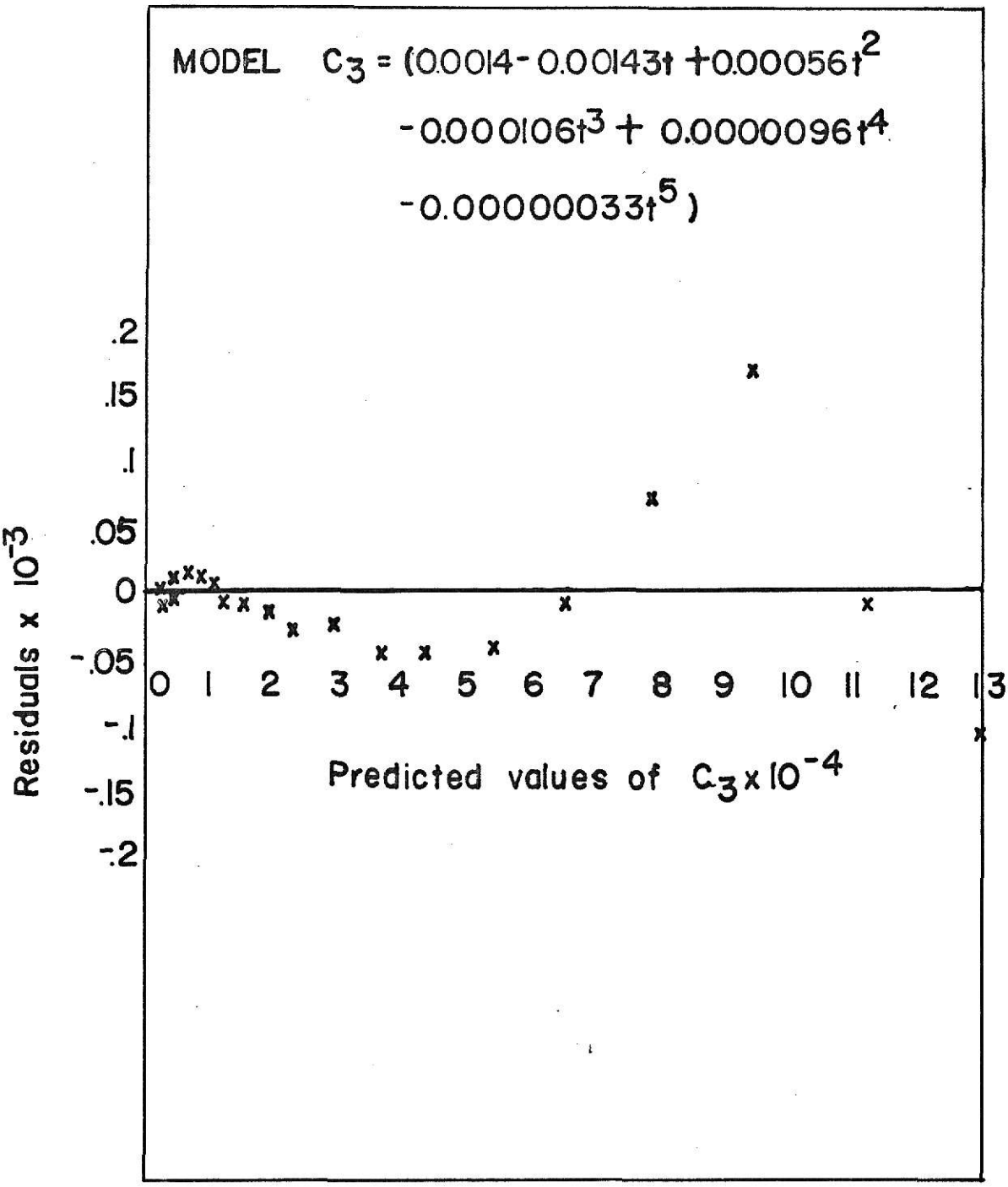

Fig. 6. Plot of residuals against predicted values of C_3.

Case 2. There is some error in the calculations and the linear effect of a_j (independent variable) is not removed.

Case 3. This suggests that we should have some extra terms in the model or should transform Y_i (experimental values of C_3 in our case) before analysis.

Time sequence plots of residuals, plots against predicted values of C_3, and independent variable, for models other than the fifth order model, are included in the Appendix and can be referred to for comparison.

A plot of residuals vs. time is shown in Figure 7.

4.8 NUMERICAL STATISTICAL TECHNIQUES FOR THE EXAMINATION OF RESIDUALS

The various graphical procedures which have been used in this chapter are essentially visual techniques for checking some of the basic regression assumptions. Draper and Smith [2] have described briefly the numerical measures for some of the discrepancies shown in Figure 4. They are of the opinion that in practical situations a detailed examination of the various residuals plots is sufficiently satisfactory. Consider the plots of e_i vs. y_i. Three major types of discrepancies were described and related to the diagrams of Figure 4. Draper and Smith [2] have suggested the following statistics for measuring these defects. Define

$$T_{pq} = \sum_{i=1}^{n} e_i^p \, y_i^q$$

Then

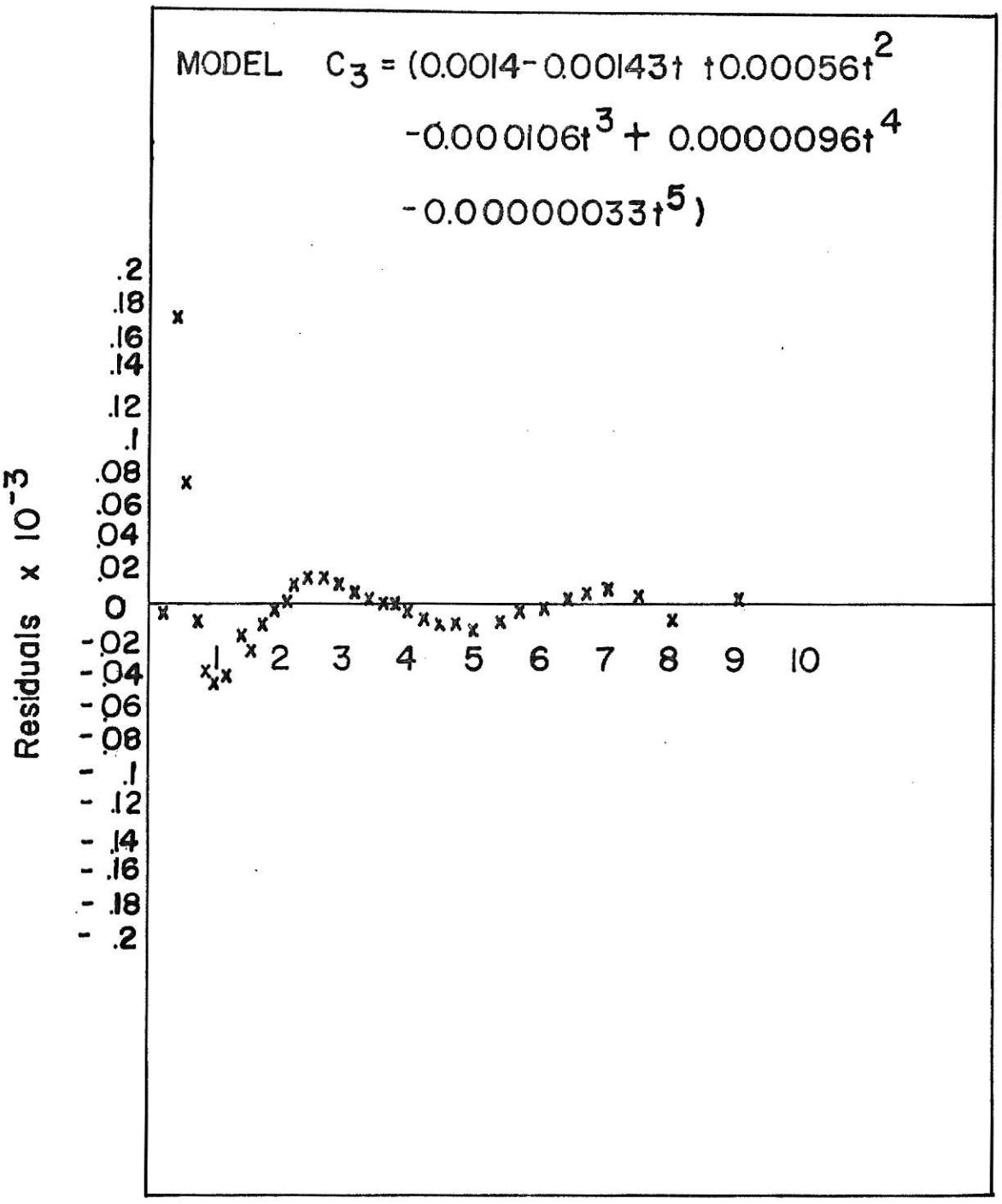

Fig. 7. Time vs. residuals.

(i) $T_{21} = \sum_{i=1}^{n} e_i^2 y_i$ provides a measure for the type of defect shown in Figure 4 (Case 1). A more general treatment of this defect can be found in the paper by Anscombe [13].

(ii) $T_{11} = \sum_{i=1}^{n} e_i y_i$ provides a measure for the type of defect shown in Figure 4 (Case 2), which should always be zero.

(iii) $T_{12} = \sum_{i=1}^{n} e_i y_i^2$ provides a measure for the type of defect shown in Figure 4 (Case 3).

Papers by Anscombe [12] and Anscombe and Tukey [2] have provided other numerical statistical procedures to check the basic regression assumptions. Anscombe [12] has discussed, besides other things, the empirical distribution of residuals, relation of residuals with the fitted values, etc. Their approach is mathematical in nature and very useful in certain situations. Anscombe and Tukey [1] have described numerical techniques for measuring removable nonadditivity, dependence of variability upon level of response and for assessing the distribution shape.

4.9 RESULTS AND DISCUSSION

There are two opposite criteria involved when one tries to select the best regression equation. The first being, as in the present case, to make the equation useful for predictive purposes, we should like the model to included as many higher order terms in t as possible so that reliable fitted values can be obtained, but on the other hand because of the cost involved, besides the other factors, one would like to include as few terms

in t as possible in the model. The purpose of this investigation was to find a compromise between these two situations and suggest the model which is most useful.

In the first part of this work the various parameters appearing in the models represented by Equations (8) through (13) were estimated using Bard's method of "Single Equation Least Squares" [5]. Tables 1 through 6 show the estimates of these parameters. The models were used to predict the tracer concentrations in tank (3). Figures 8 through 11 show the agreement between experimental values of C_3 and those predicted from the models.* It can be seen that the fourth, fifth and sixth order models give a reasonably good fit. Figures 12 and 13 compare some of the computational aspects for the various models. Figure 12 shows the variation of computing time as we increase the number of parameters in the model. It can be seen that the variation is not appreciable when one goes from two to six parameters (first order to fifth order model) but the computation time is increased almost seven times in going from six to seven parameters (fifth order to sixth order model). Table 7 also shows how the sum of squares of residuals changes as we increase the number of parameters in the model. It can be noticed that the sum of squares of residuals for the fourth, fifth and sixth order models are all very small. They are almost the same for sixth and seventh order models. Figure 13 indicates the variation of function and derivative evaluation in the estimation procedures as the number of parameters in the model increase. It can be seen from Table 8 that in

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*
The data on the tower system using the flow model was taken by Dr. Arthur E. Humphrey and his associates at the Biochemical Engineering Laboratory University of Pennsylvania, Philadephia, Pennsylvania 19104.

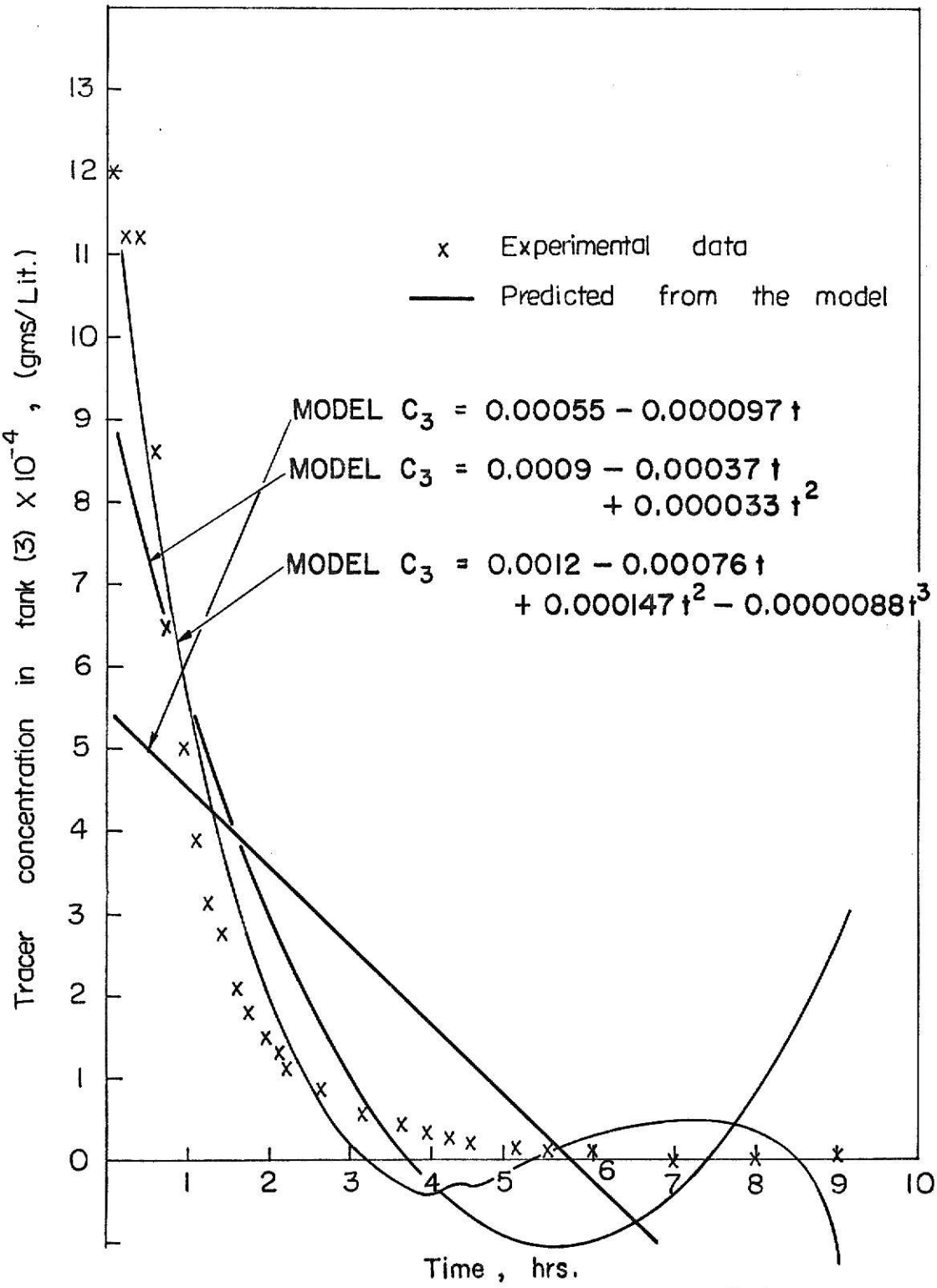

Fig.8. Tracer concentration in tank (3) vs. time.

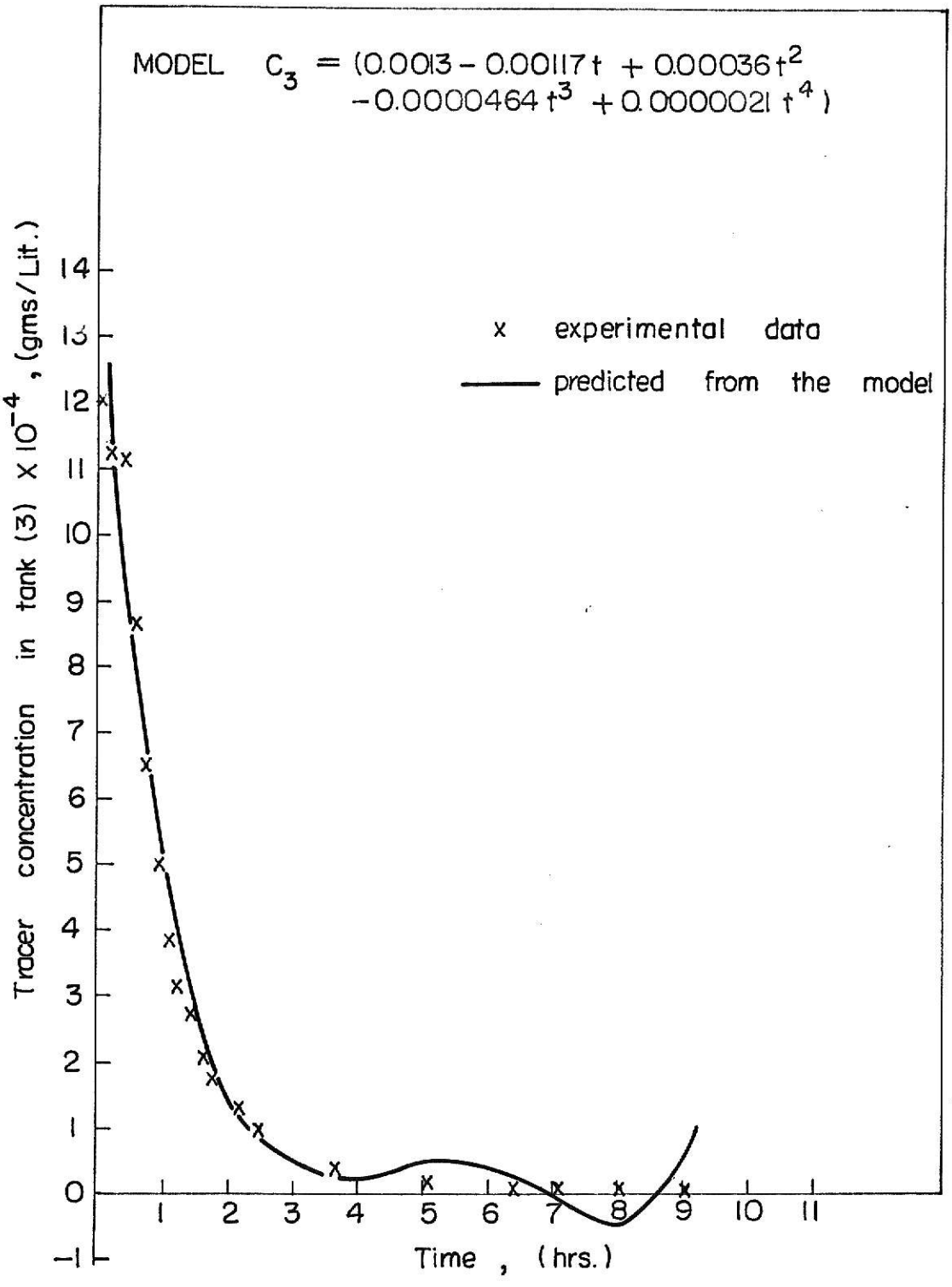

Fig. 9. Tracer concentration in tank (3) vs time.

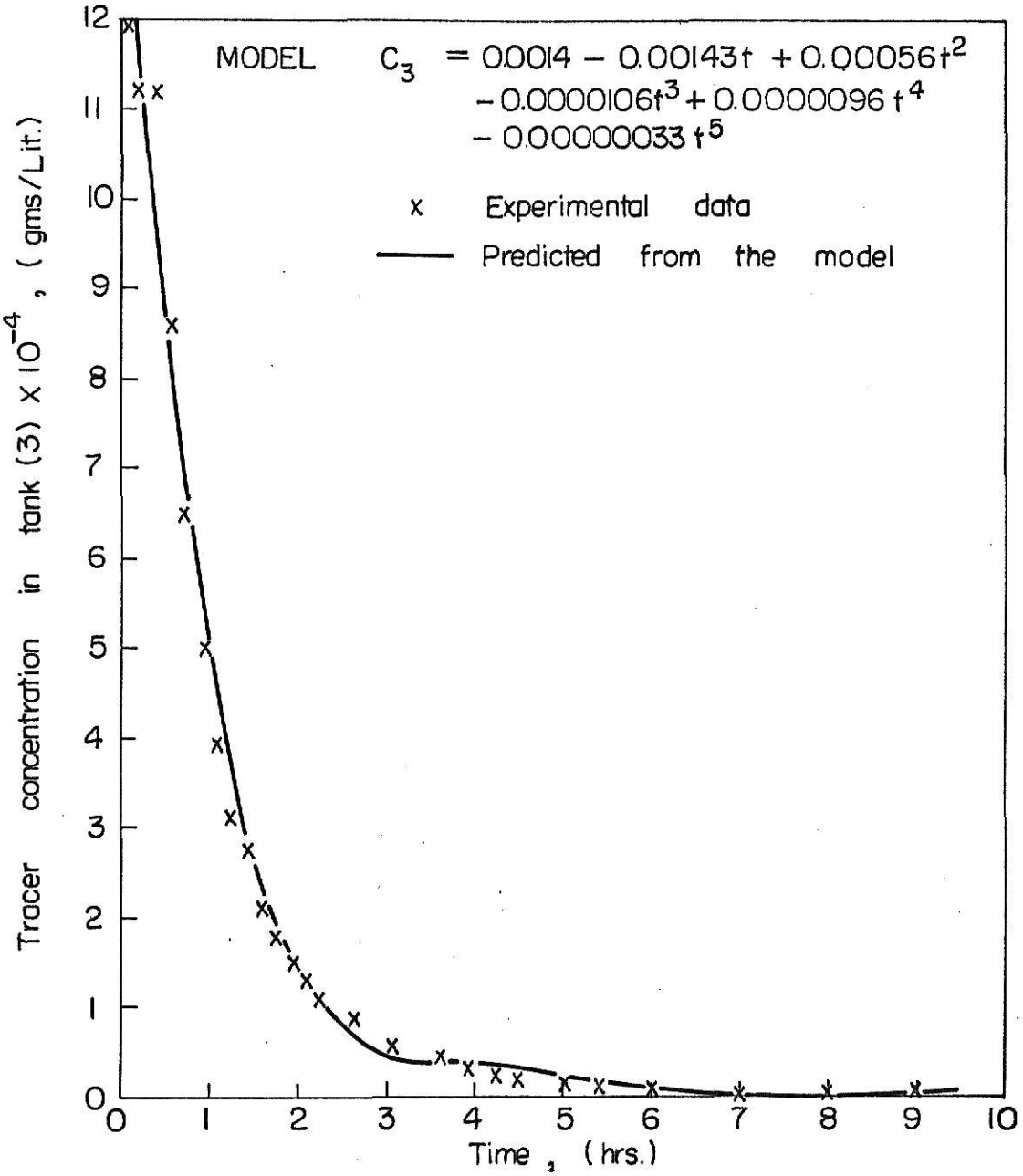

Fig. 10. Tracer concentration in tank (3) vs time.

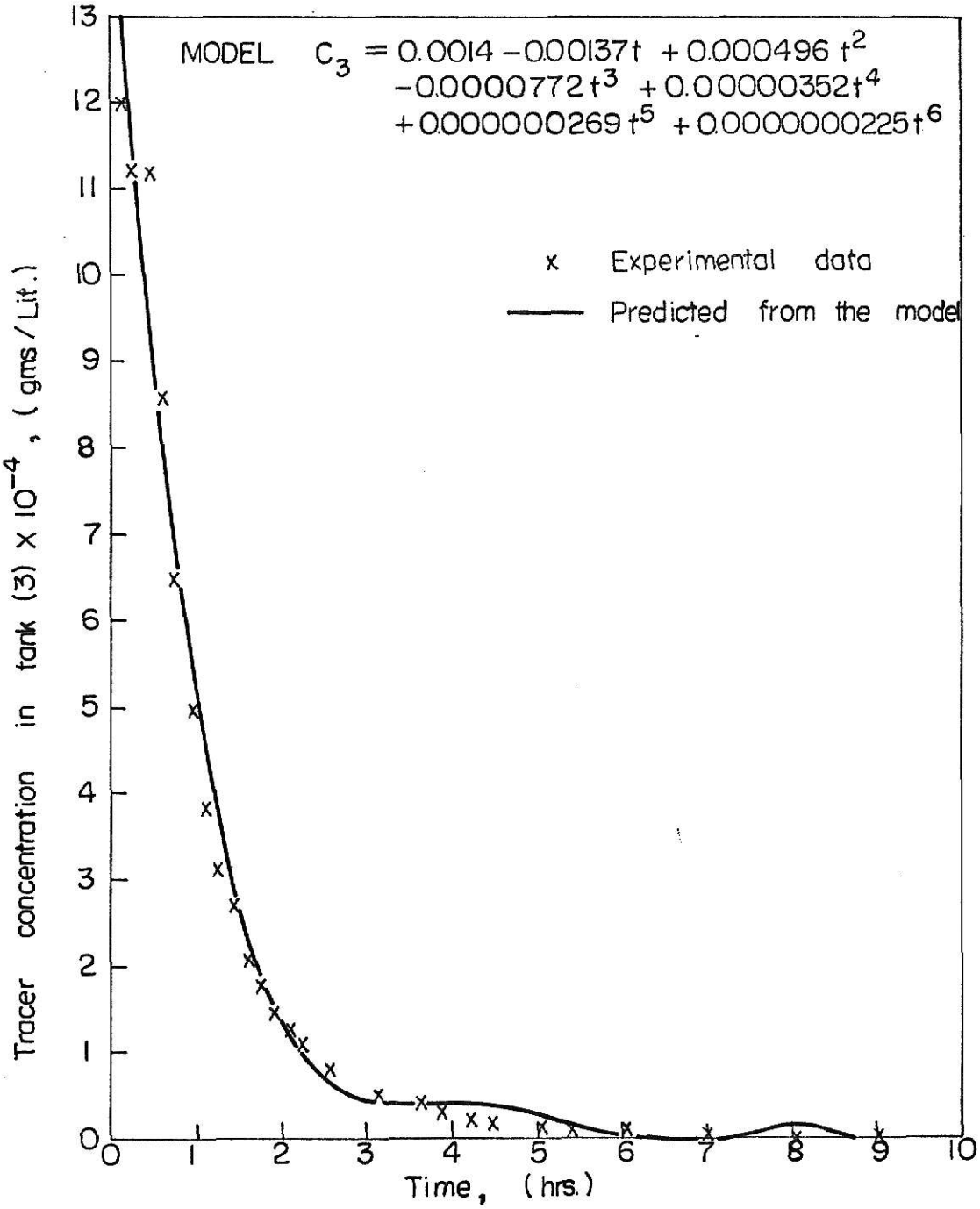

Fig. 11. Tracer concentration in tank (3) vs time.

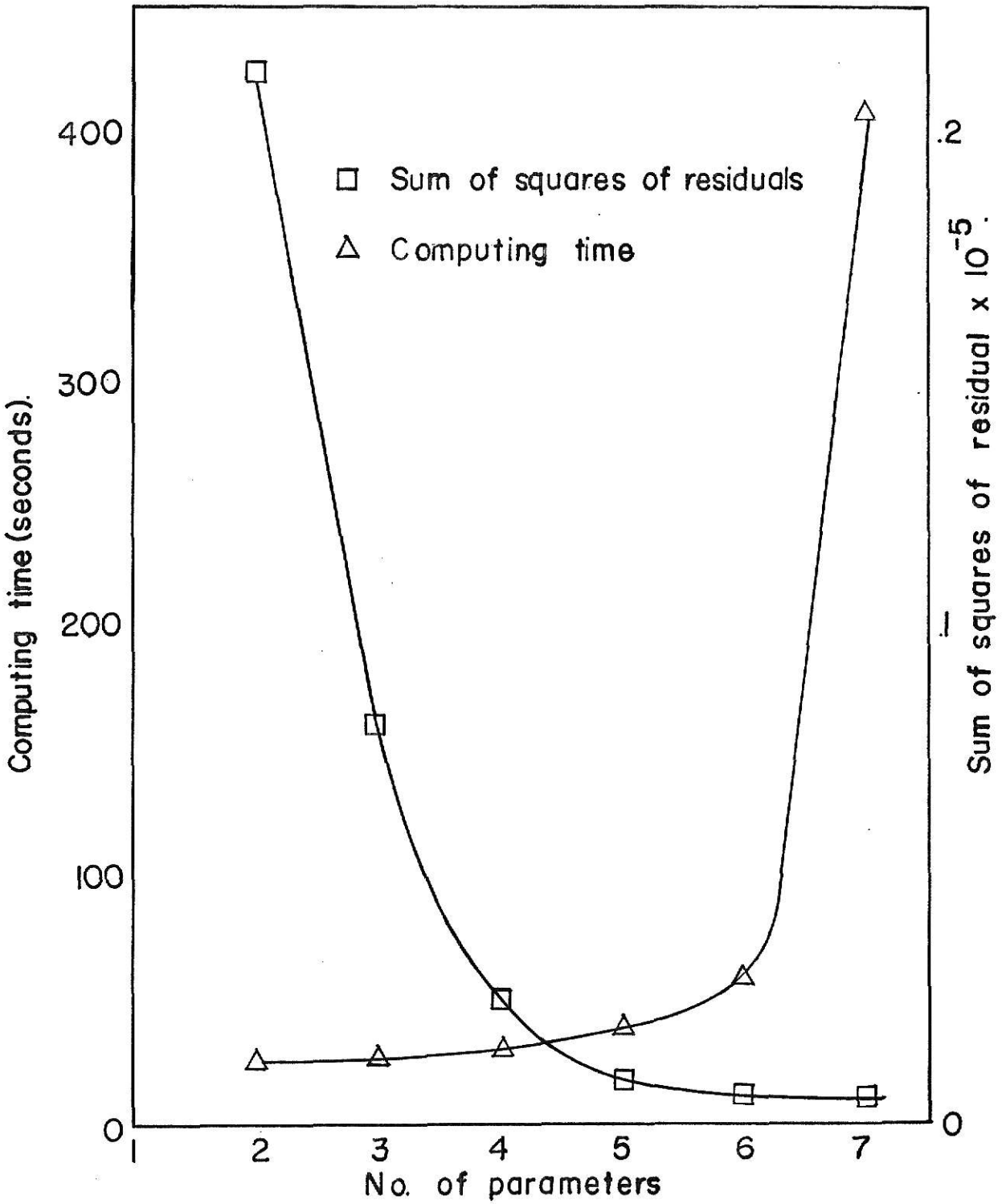

Fig. 12. Computing time and sum of squares of residuals vs. number of parameters in the model.

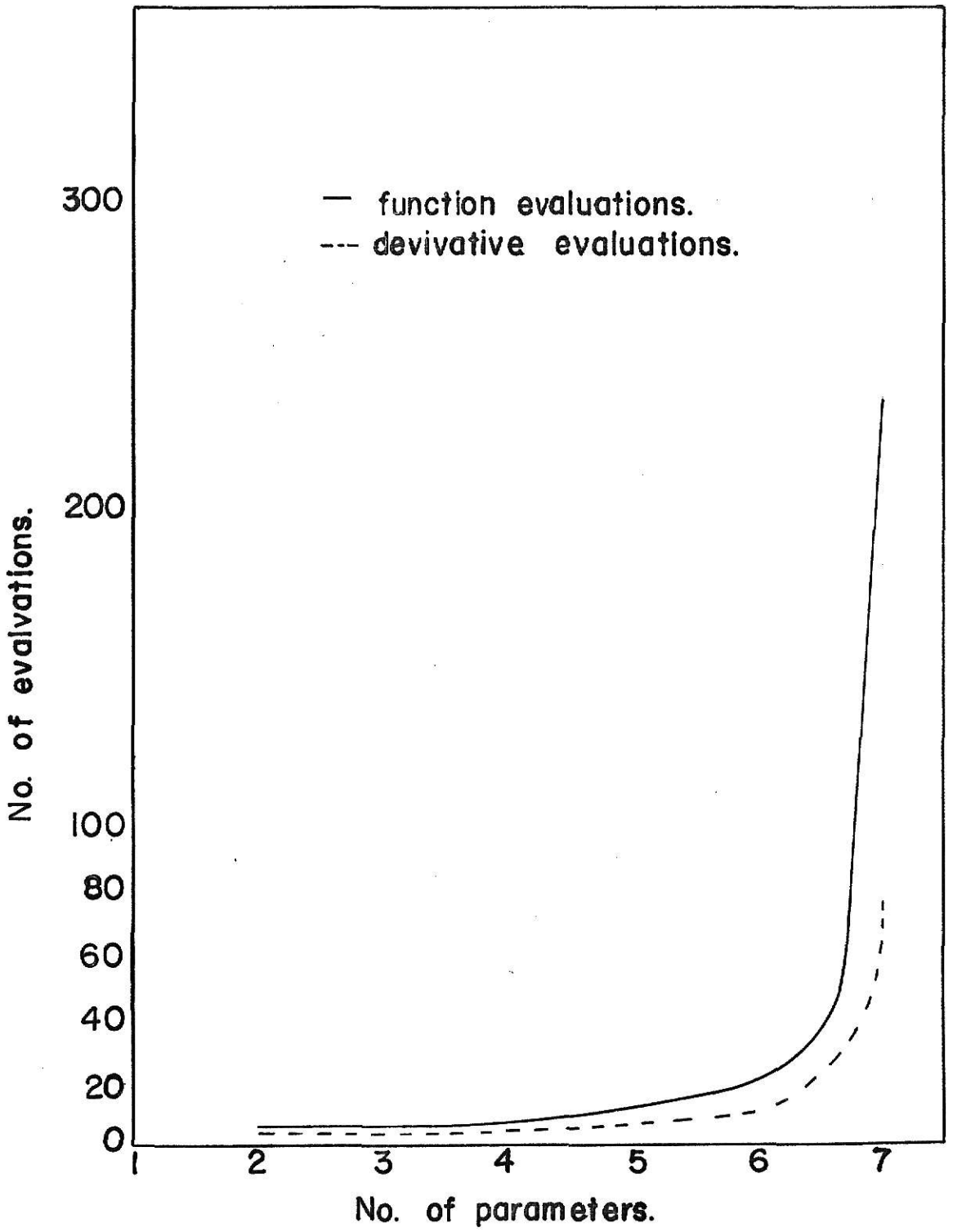

Fig. 13. Number of evaluations vs. number of parameters in the model.

going from the fifth order model to the sixth order model (six parameters to seven parameters) the function and derivative evaluations increase appreciably. However, this increase is comparatively inappreciable when the number of parameters are increased from two to five.

Figure 2 shows the overall plot of the residuals for the fifth order model. To facilitate the judgment of this plot, a plot of random normal deviates is made as shown in Figure 1. Comparison of overall residual plots with Figure 1 suggests that the plot obtained from the fifth order model has almost the same trend as this. This gives us an indication that the fifth order model is more adequate and correct as compared to other models for predictive purposes. Figure 5 shows the time sequence plot of residuals for the fifth and sixth order models. The criterion for judging these plots also indicates that most of the residuals lie within a horizontal strip which tends to confirm that the fifth order model is reasonable. The same criterion is used for judging the remaining plots also. Figure 6 shows the plots of fitted values of the tracer concentration in tank (3) vs. residuals for the fifth order model. Time has been plotted vs. residuals in Figure 7. Observation of this plot also shows that in the case of the fifth order model most of the residuals lie within a horizontal strip, which tends to indicate that the basic assumptions which were made about the errors (residuals) are adequate.

Hence it can be concluded from the graphical examination of residuals that one has sufficient reason to believe that the fifth order model is best among the various models considered. In other words, available experimental data fits the fifth order model best and no further improvement is obtained if the order of the model is increased.

4.10 NOTATION

a'_0, a'_1 = constants in Equation (8)

a_μ = μ^{th} independent variable

b_0, b_1, b_2 = constants in Equation (9)

C = constants in Equation (6)

C_3 = tracer concentration in tank (3), (gms/c.c.)

C' = normalizing constant ($\int P(Y_\mu|\theta)\ P_0(\theta)d\theta$)

$Cov(e_i, e_j)$ = covariance of i^{th} and j^{th} residual

d_0, d_1, d_2, d_3, d_4 = constants in Equation (11)

e'_0, e'_1, e'_2, e'_3, e'_4, e'_5 = constants in Equation (12)

e_i = i^{th} residual

$F(U_\mu)$ = selected criterion function

$G(\theta)$ = F-function when independent and dependent variables are known

h_0, h_1, h_2, h_3, h_4, h_5, h_6 = constants in Equation (13)

n = number of observations

$P(U,\phi)$ = joint probability density function

$P(Y_\mu|\theta)$ = probability density of Y_μ given θ

$P_0(\theta)$ = prior distribution of θ

Q_{ij} = positive definite matrix $(-\frac{\partial^2 G}{\partial\theta_i\,\partial\theta_j})$

R = any positive definite matrix

t = time (hrs.)

T_i = objective function value for i^{th} order regression model

U_μ = μ^{th} residual

V_{ik} = i^{th} element of the k^{th} eigenvector of Q

V_{jk} = j^{th} element of the k^{th} eigenvector of Q

Y_i = i^{th} observation

y_i = fitted value obtained by fitted regression model

Greek Letters

α_i = lower bounds on θ_i

β_i = upper bounds on θ_i

$\Delta\theta_i$ = direction vector

θ_i = i^{th} parameter

$\theta_i^{(0)}$ = initial estimate of θ_i

$\theta_i^{(1)}$ = value of θ_i after first iteration

λ = step size

ρ_{ij} = correlation coefficient between i^{th} and j^{th} residual

μ' = mean

μ_k = k^{th} eigen value of Q

σ = variance

ϕ = additional unknown parameter

Subscripts

B = baysian estimate

L.S. = least squares estimate

M.L. = maximum likelihood estimate

W.L.S. = weighted least squares estimate

4.11 REFERENCES

1. Anscombe, F. J., and J. W. Tukey, "The Examination and Analysis of Residuals," Technometrics, 5, 141-160 (1963).

2. Draper, N. R., and H. Smith, "Applied Regression Analysis," John Wiley, New York, N. Y., (1962).

3. Anscombe, F. J., "Rejection of Outliers," Technometrics, 2, 123-147 (1960).

4. Jeffreys, H., "Theory of Probability," University Press, Oxford (1948).

5. Bard, Y., IBM New York Scientific Centre Report No. 320-2902 (1967).

6. Carroll, C. W., "The Created Response Surface Technique for Optimizing Nonlinear, Restrained Systems," Operations Research, 9, (2), 169-184 (1961).

7. Eisenpress, H., and J. Greenstadt, "The Estimation of Nonlinear Econometric Systems," IBM New York Scientific Centre Report No. 322-0910 (1966).

8. Cornfield, J., "Bayes Theorem," Review of the International Statistical Institute, 35, (1), 34-49 (1967).

9. Fletcher, R., and M. J. D. Powell, "A Rapidly Convergent Descent Method for Minimization," The Comp. J., 6, (2), 163-168 (1963).

10. Rand Corporation, "A Million Random Digits with 10,000 Normal Deviates," The Free Press, Glencoe, Illinois, (1955).

11. Daniel, C., "Use of Half-Normal Plots in Interpreting Factorial Two Level Experiments," Technometrics, 1, 311-341 (1959).

12. Anscombe, F. J., "Examination of Residuals," Proc. of Fourth Berkeley Symposium on Mathematical Statistics and Probability, 1, 1-36 (1960).

13. Marquardt, D. W., "An Algorithm for Least Squares Estimation of Nonlinear Parameters," J. Soc. Industrial Appl. Math., 11, (2), 431-441 (1963).

APPENDIX I

Overall plots of residuals for various regression models

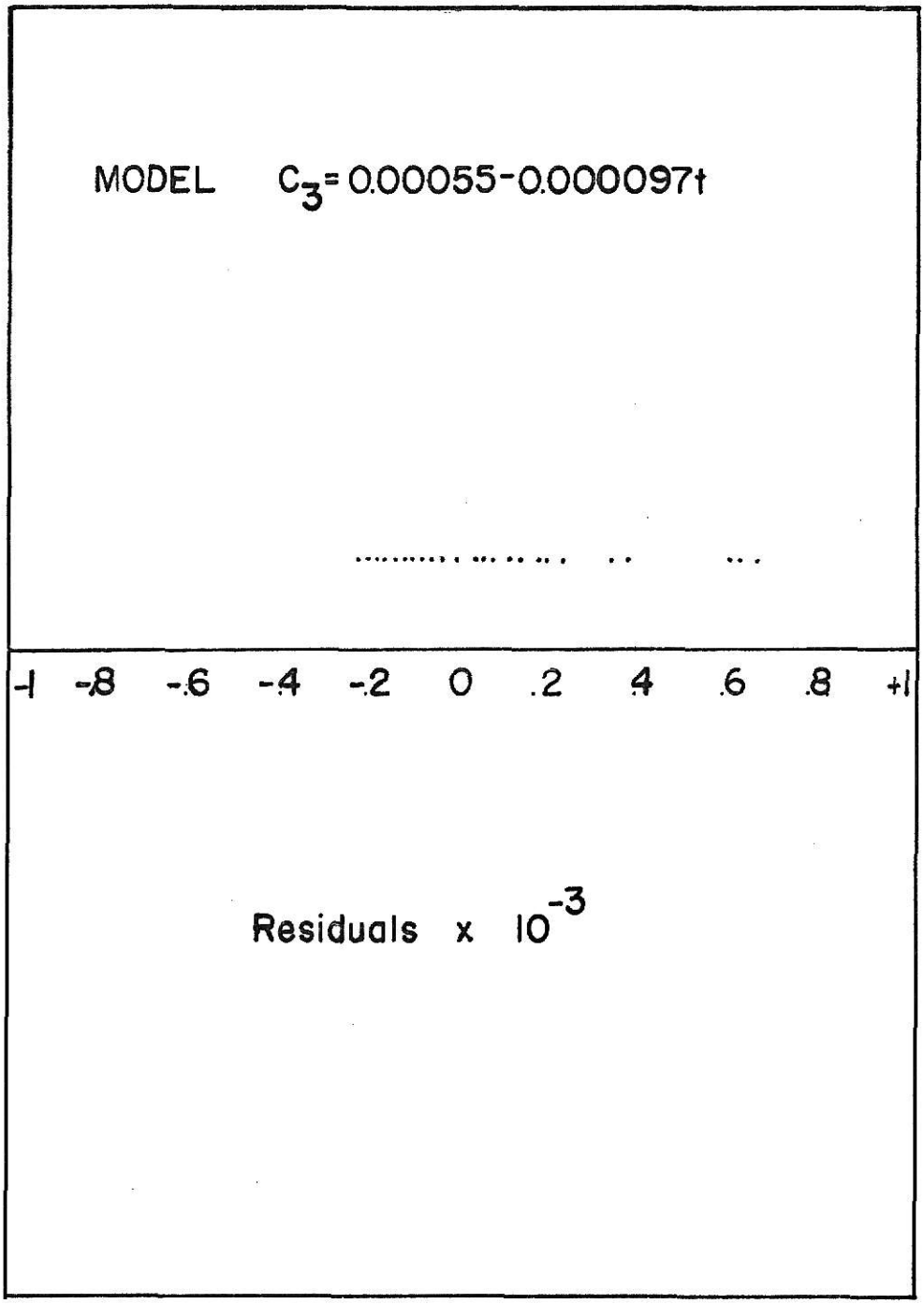

Fig.I-1. Overall plot of residuals.

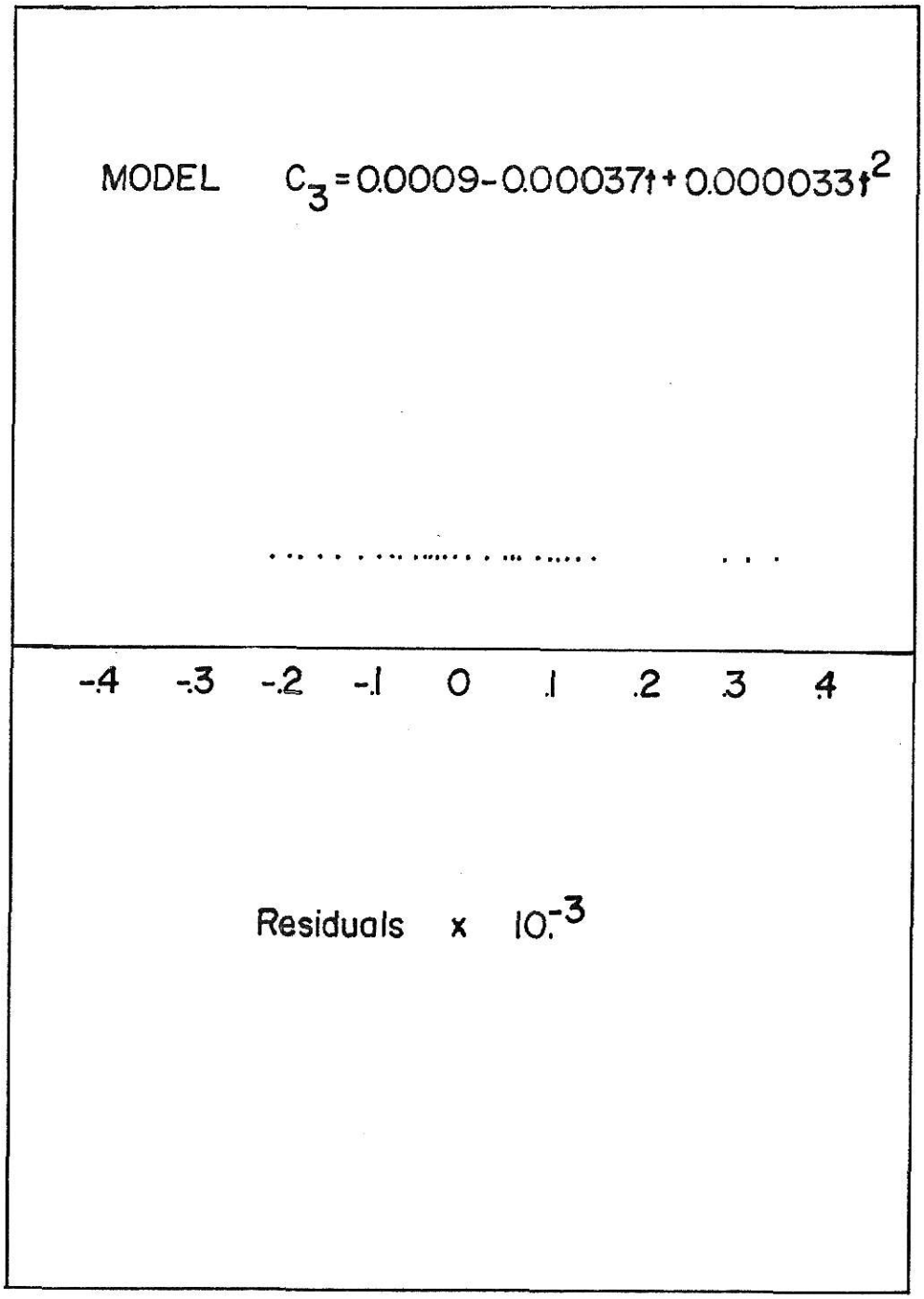

Fig.I-2. Overall plot of residuals.

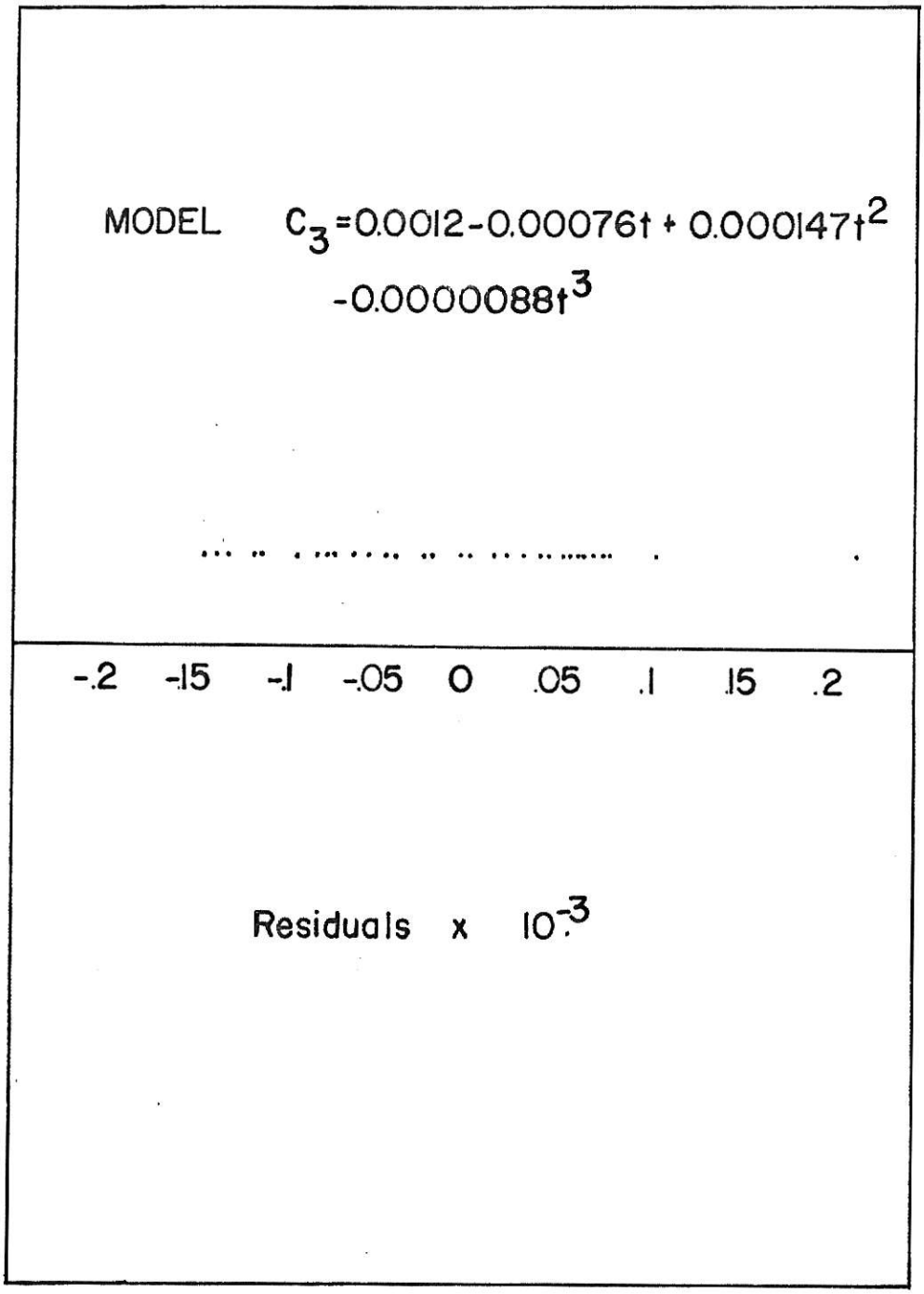

Fig. I-3. Overall plot of residuals.

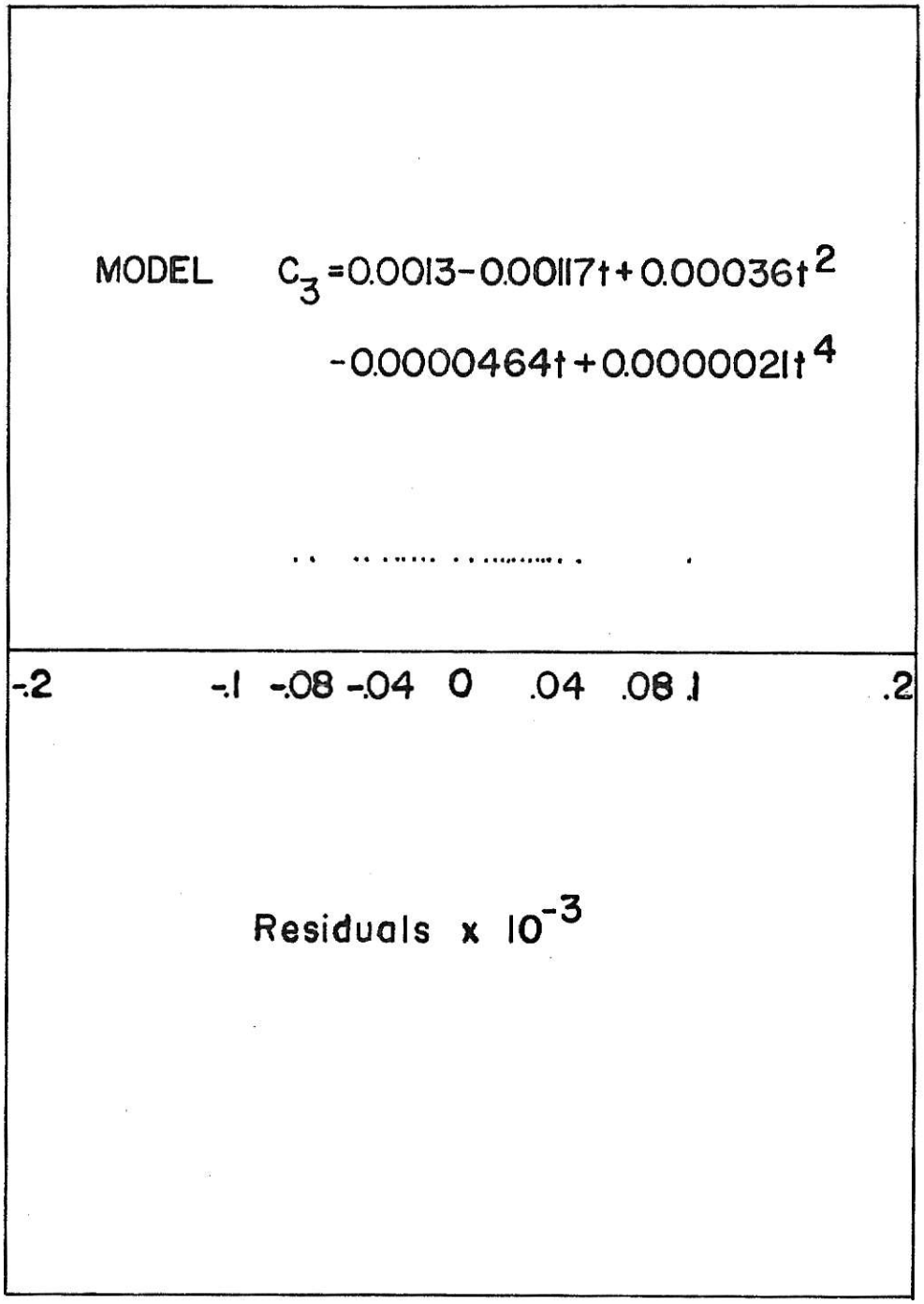

Fig.I-4. Overall plot of residuals.

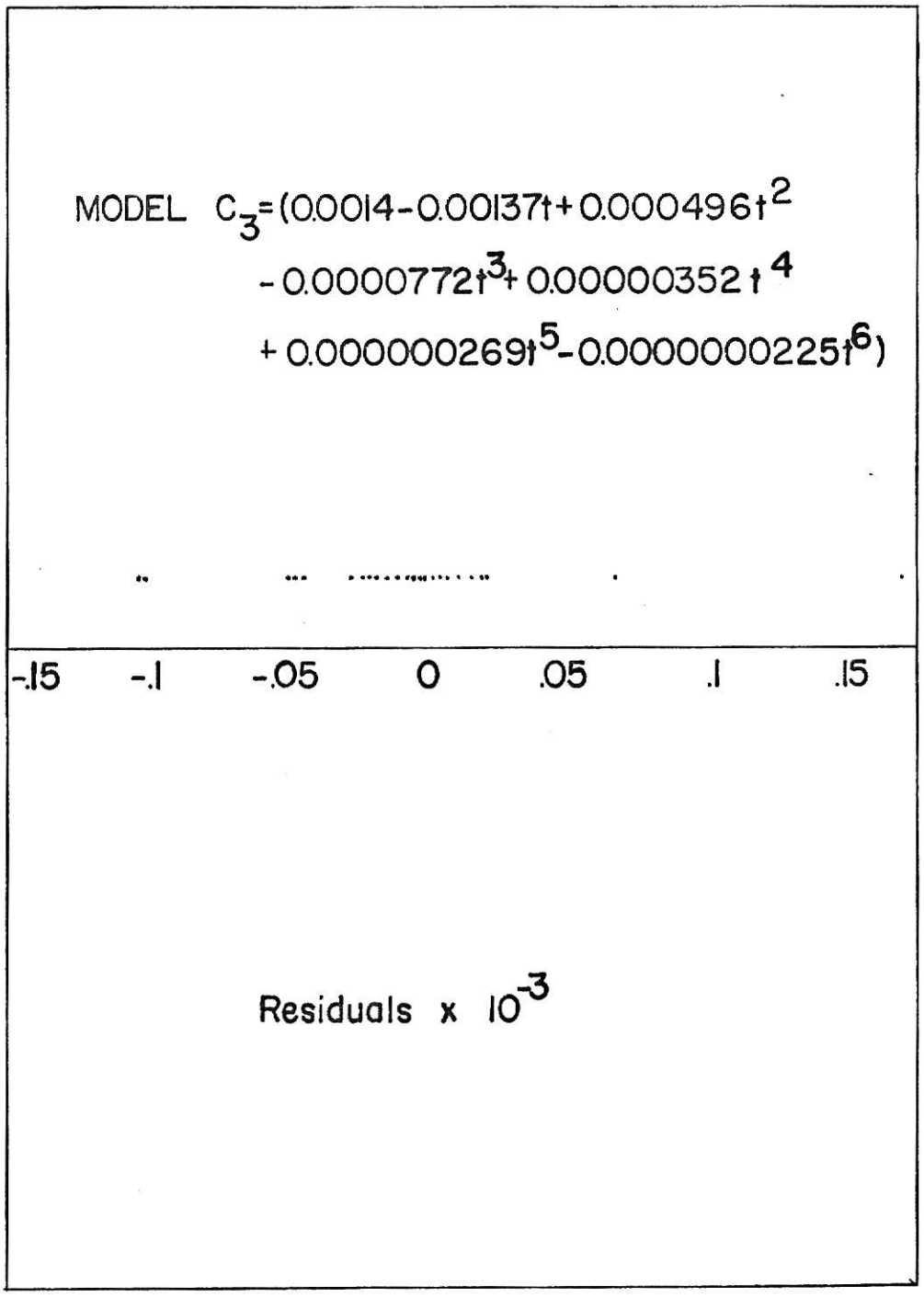

Fig. I-5. Overall plot of residuals.

APPENDIX II

Time sequence plot of residuals for various regression models

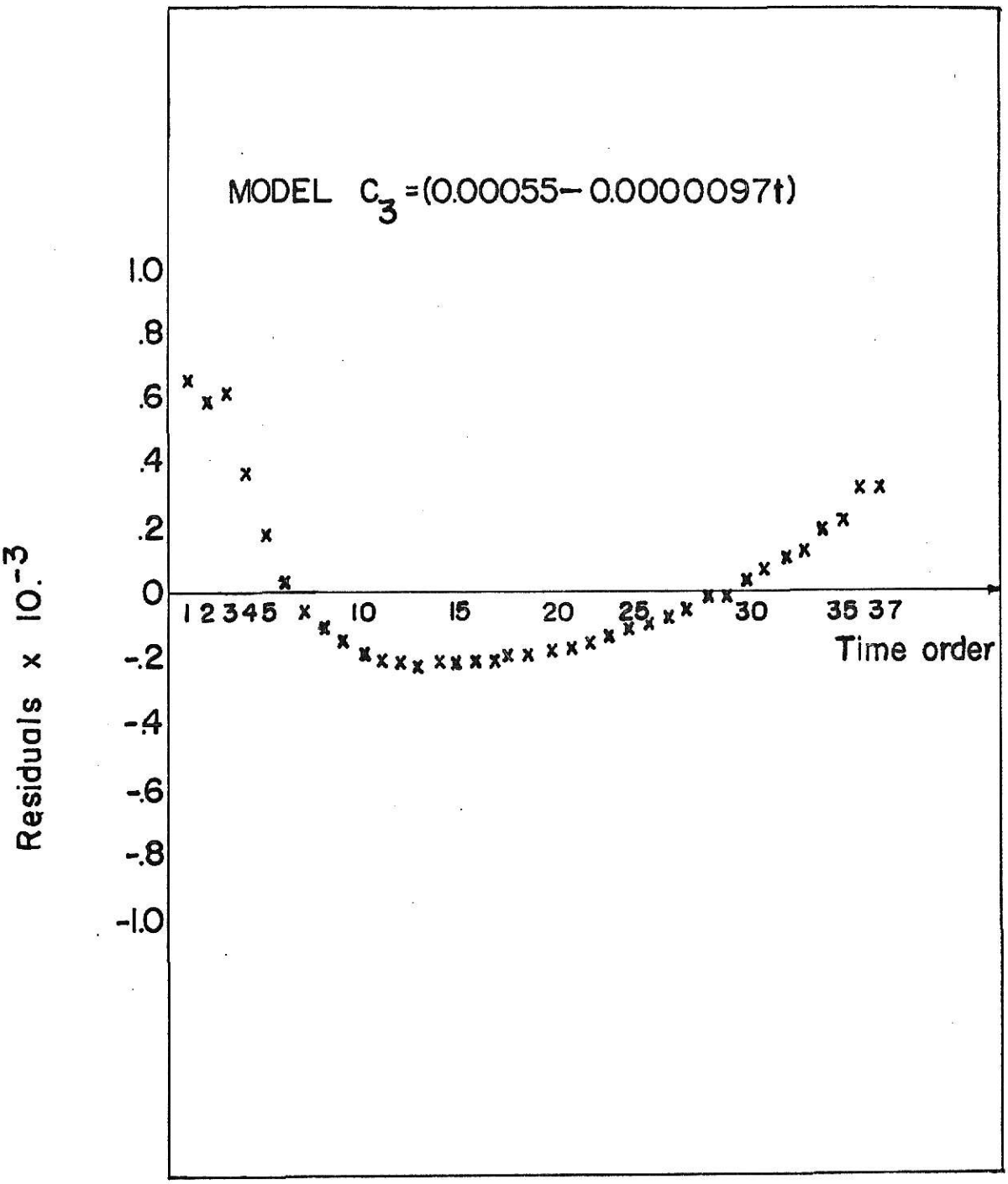

Fig. II-1. Time sequence plot of residuals.

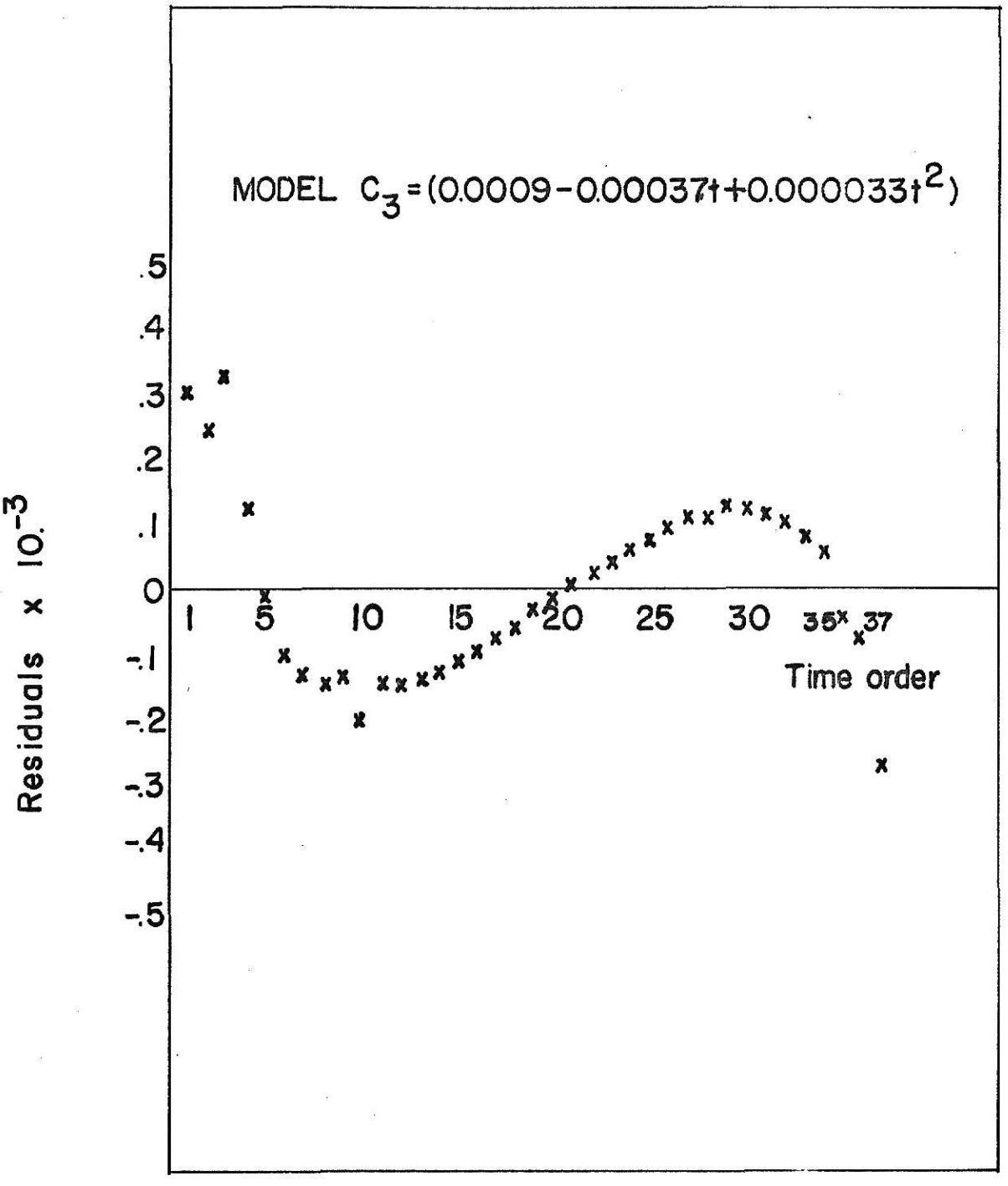

Fig. II-2. Time sequence plot of residuals.

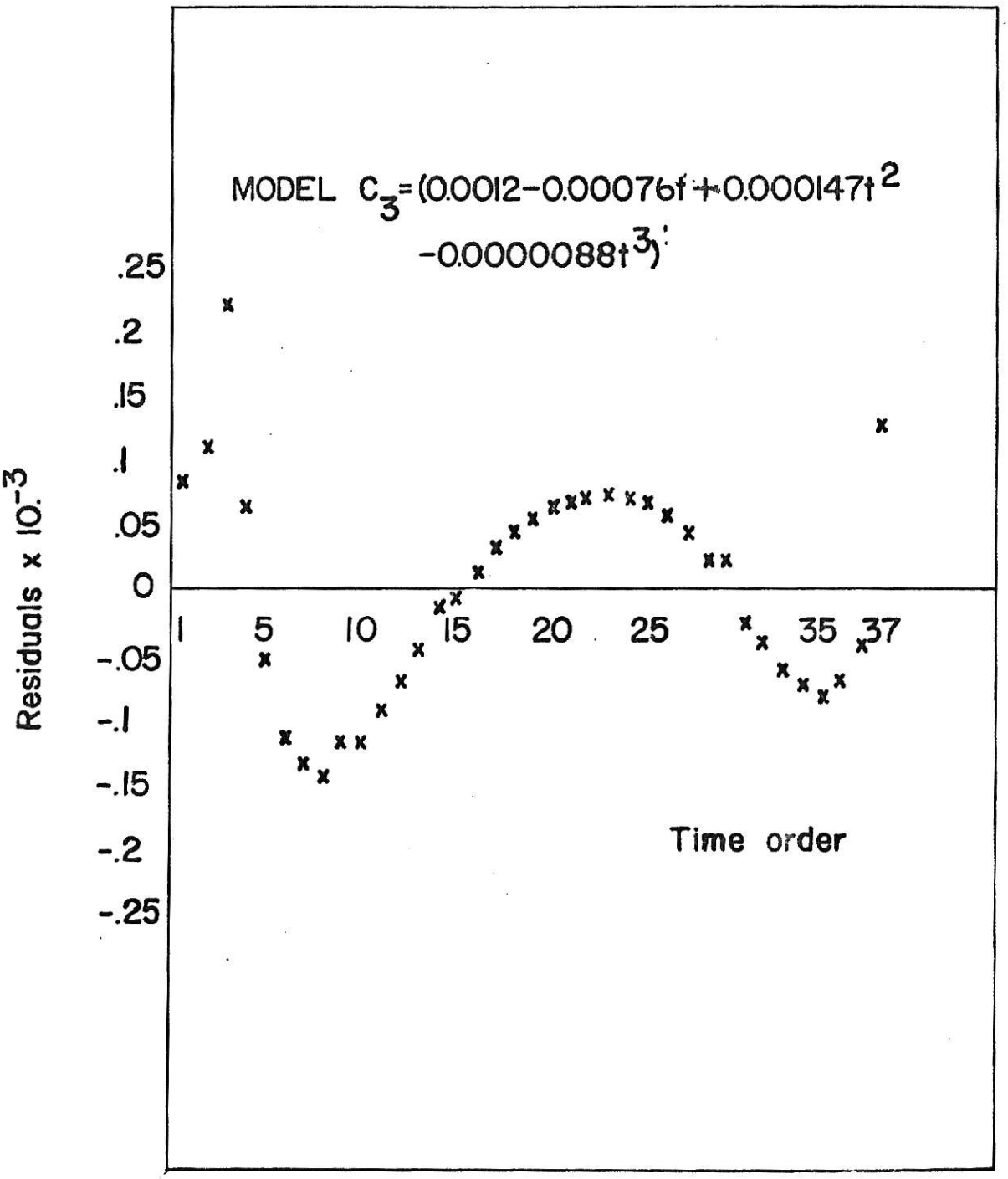

Fig. II-3. Time sequence plot of residuals.

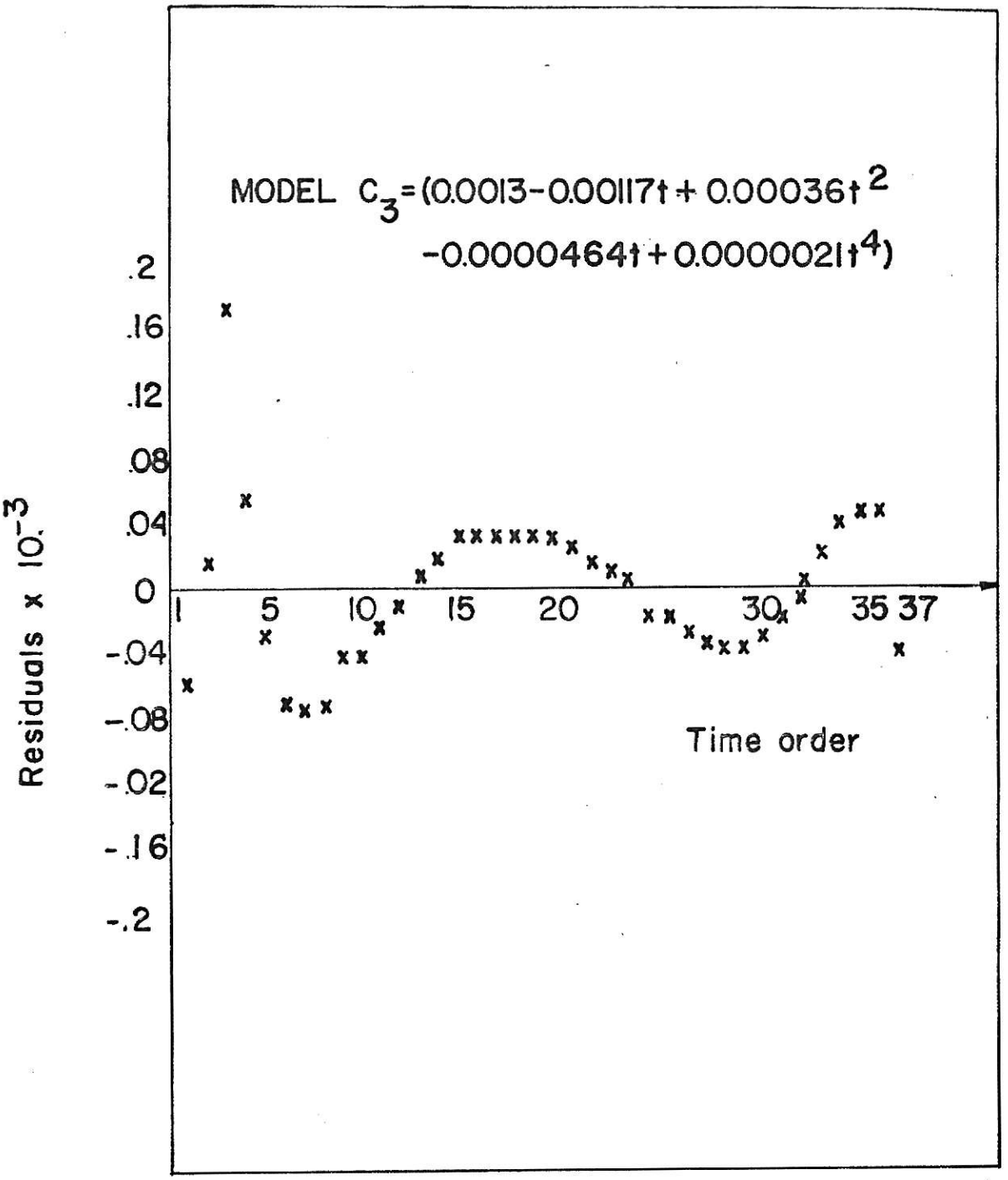

Fig. II-4. Time sequence plot of residuals.

APPENDIX III

Plots of residuals against predicted values of C_3 for various regression models

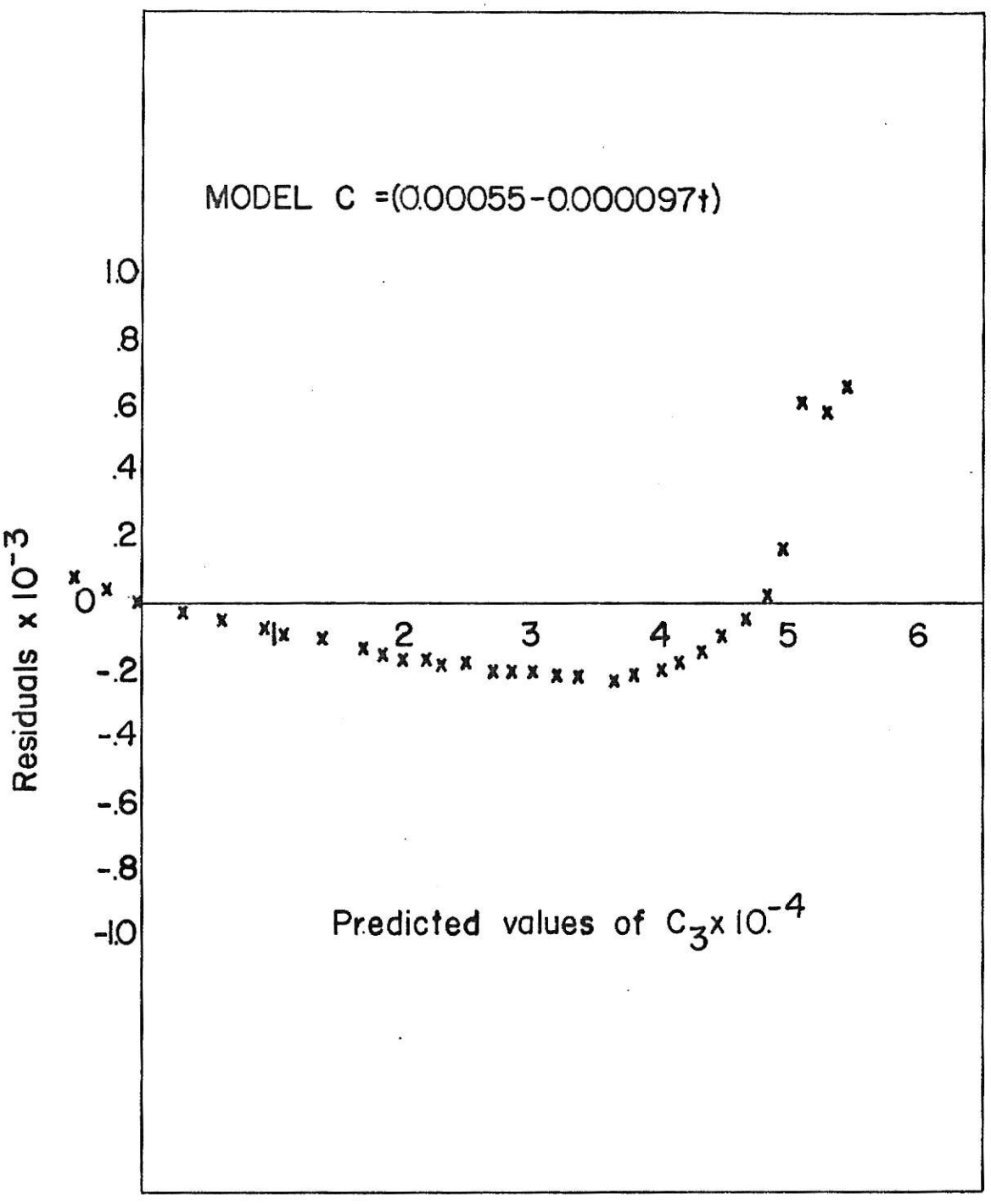

Fig.III-1. Plot of residuals against predicted values of C_3.

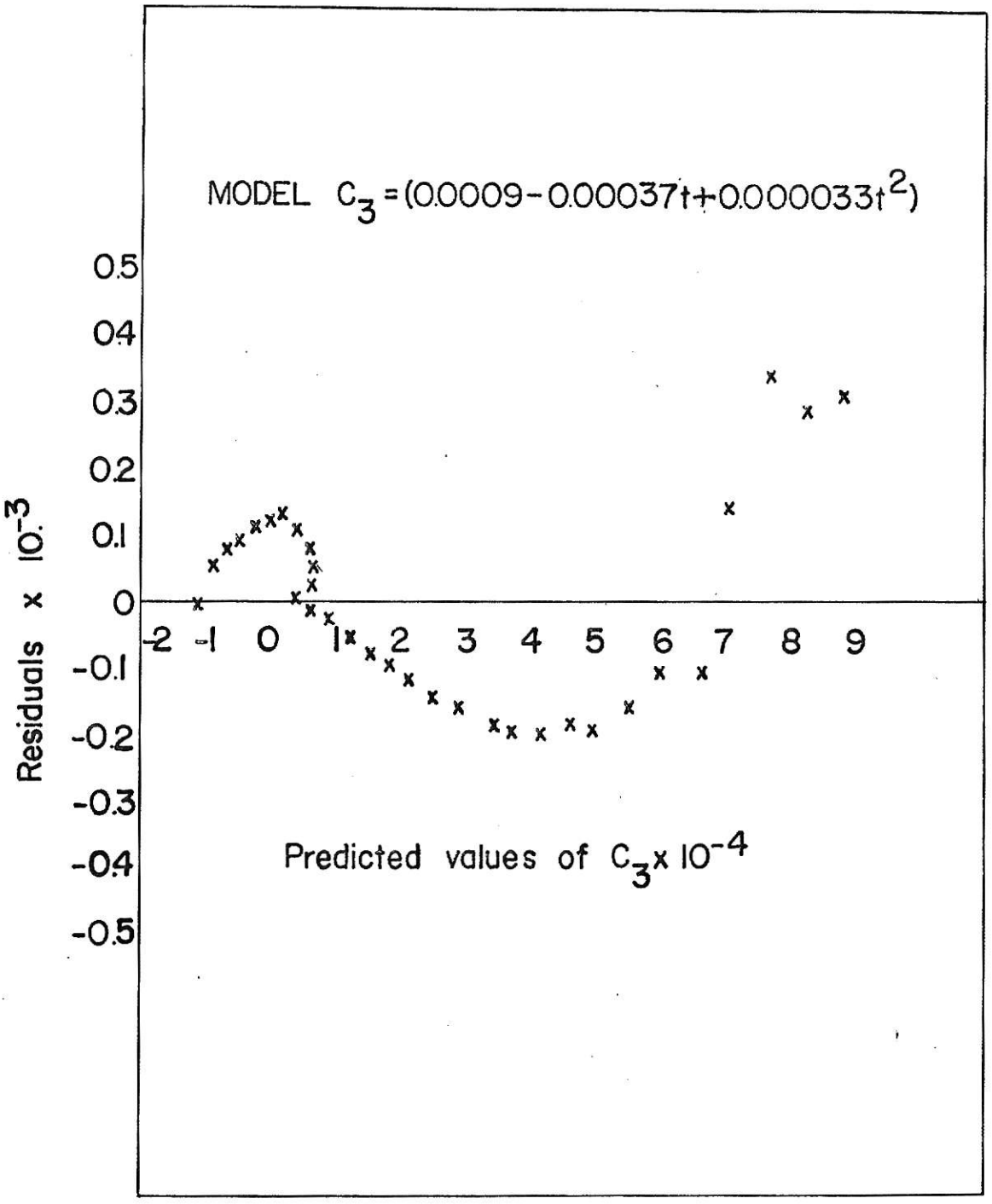

Fig. III-2. Plot of residuals against predicted values of C_3.

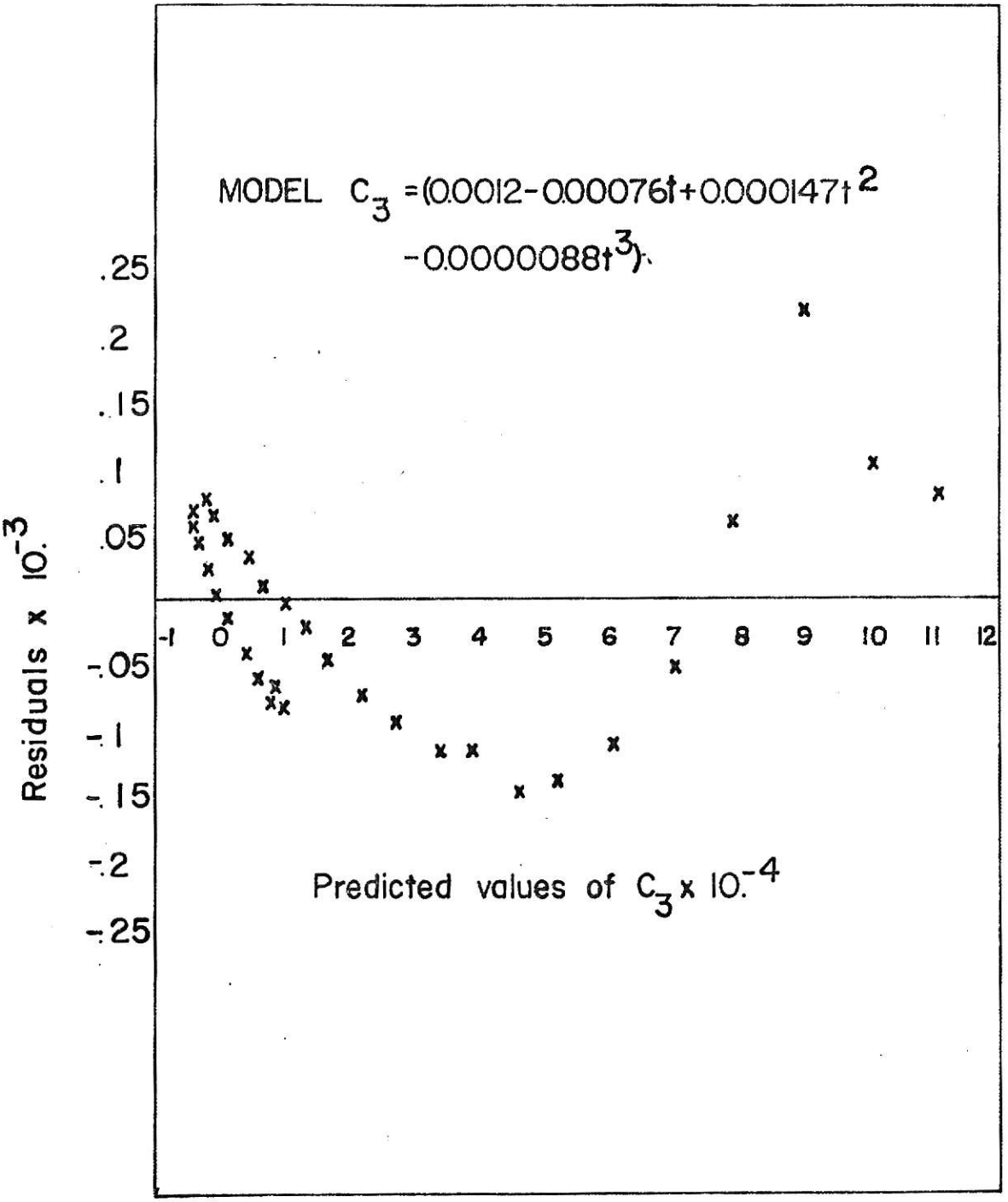

Fig. III-3. Plot of residuals against predicted values of C_3.

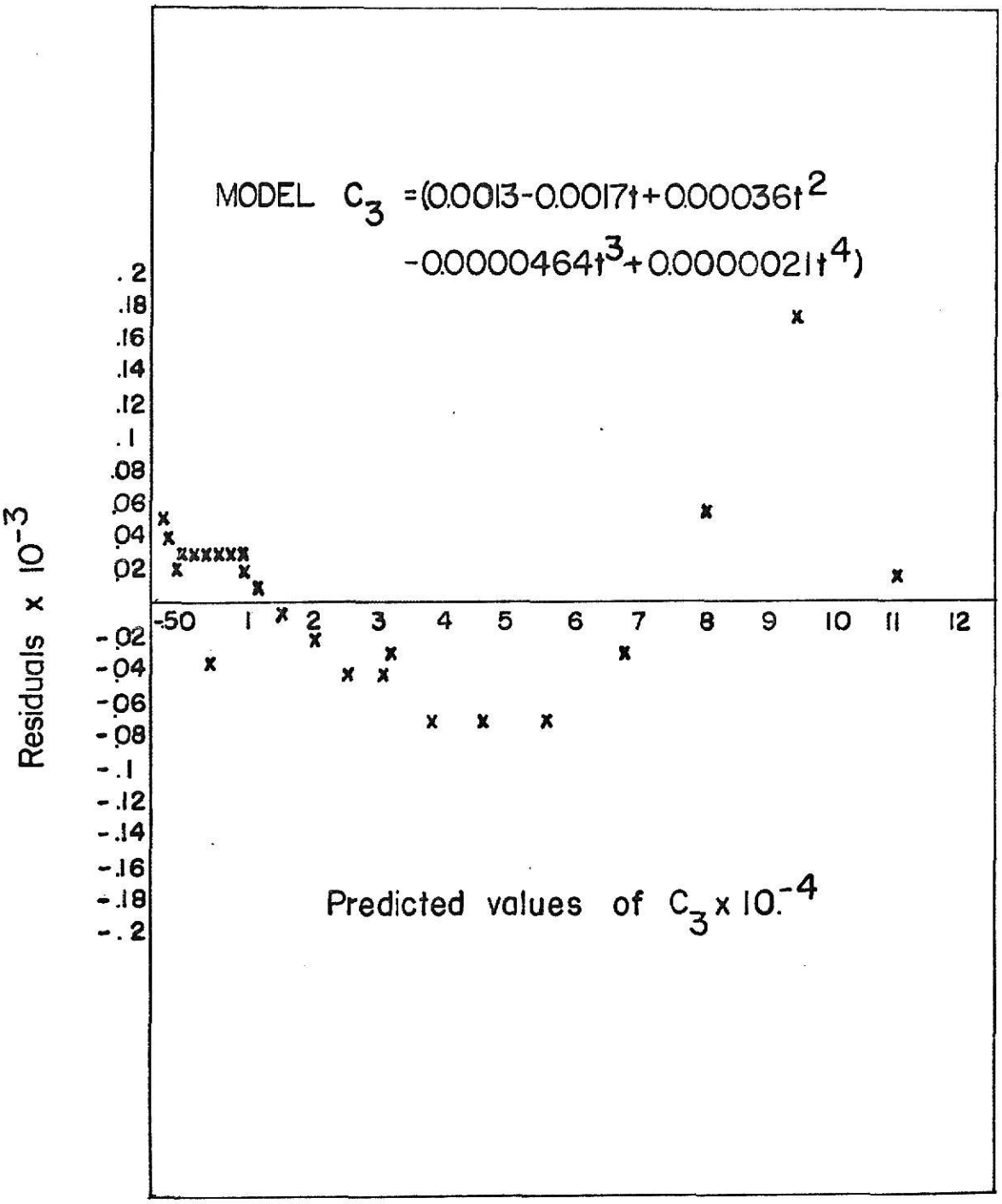

Fig. III-4. Plot of residuals against predicted values of C_3.

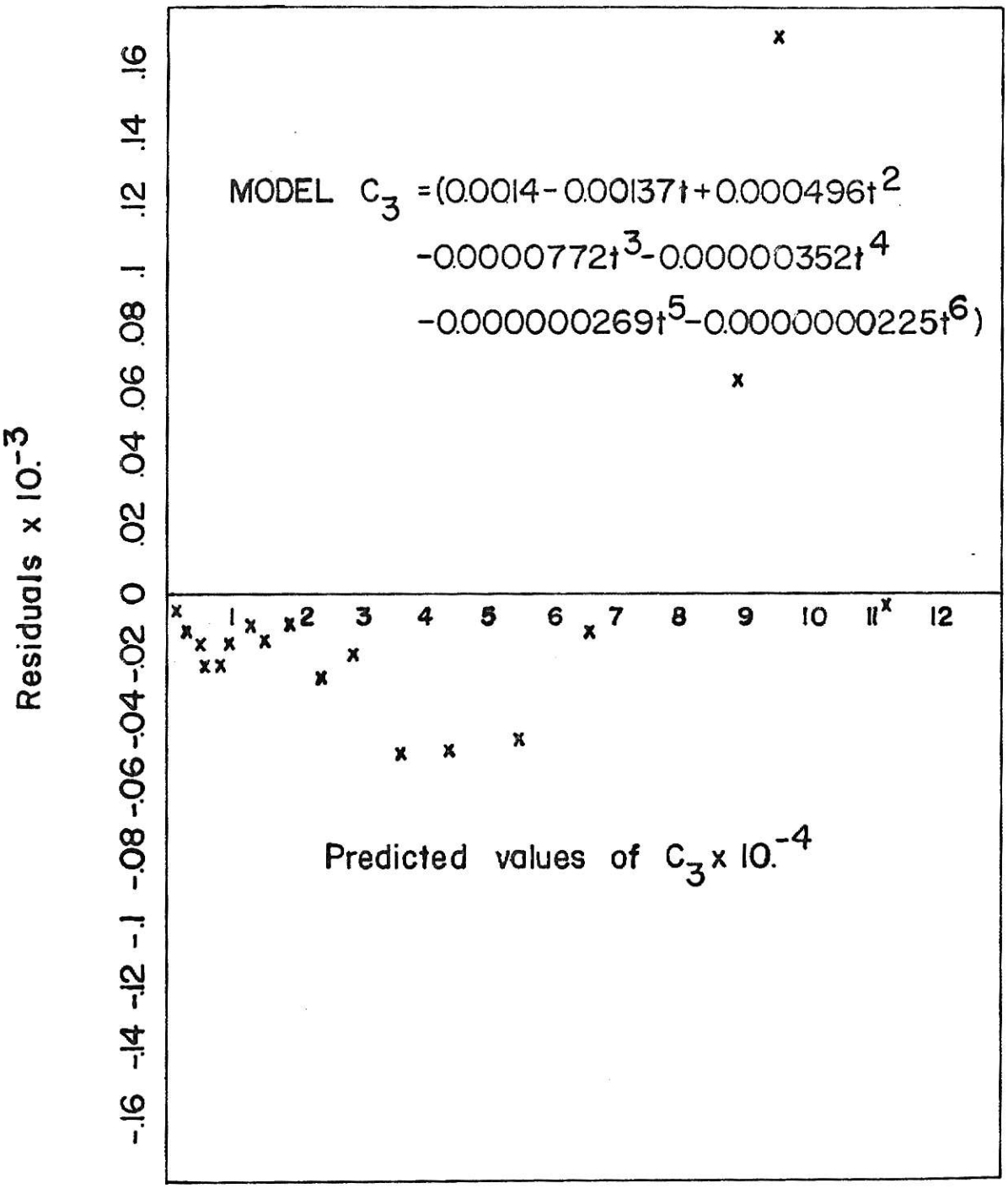

Fig.Ⅲ-5. Predicted values of C_3 vs residuals.

APPENDIX IV

Plots of residuals against the independent variable for various regression models

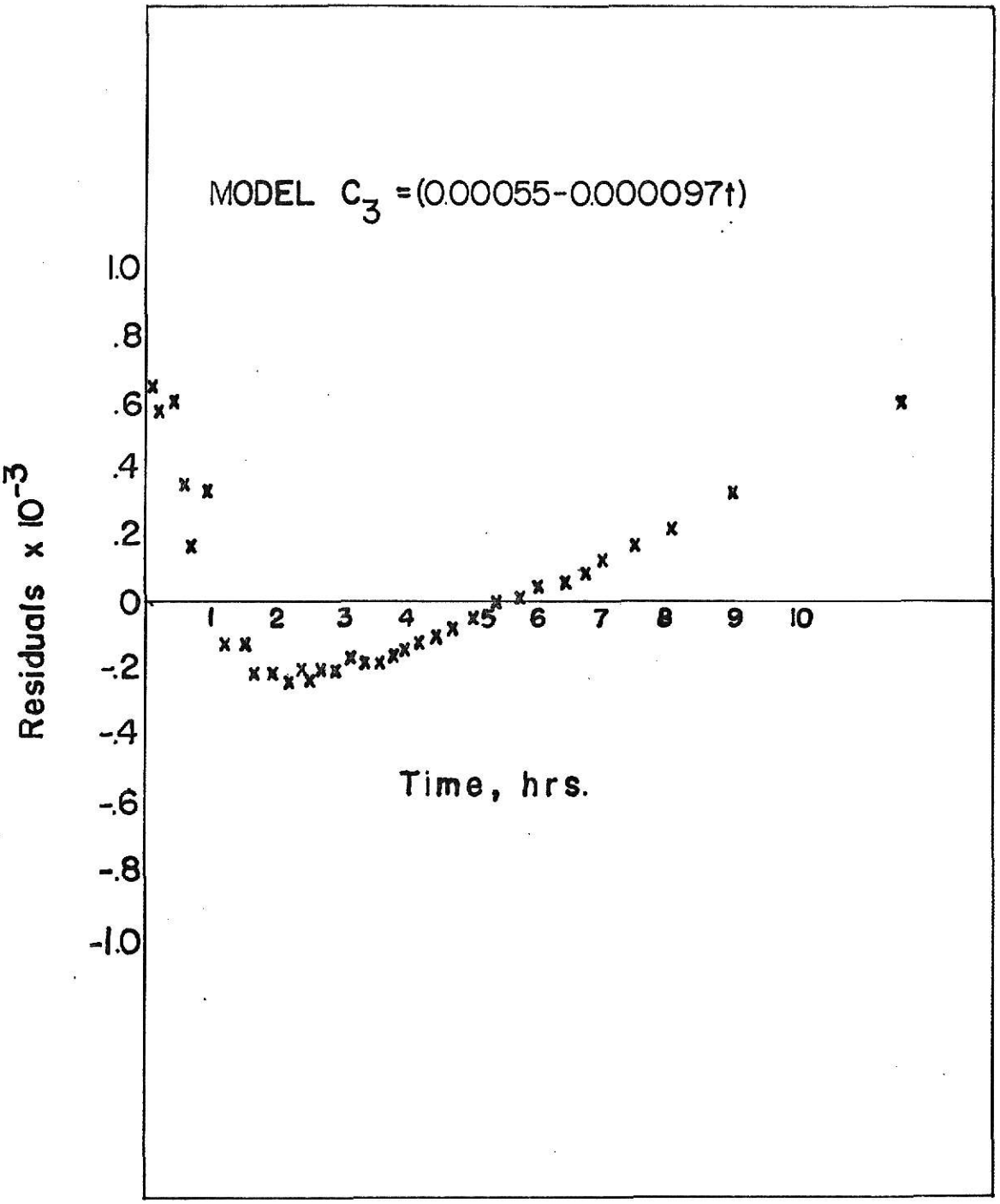

Fig. IV-1. Time vs residuals.

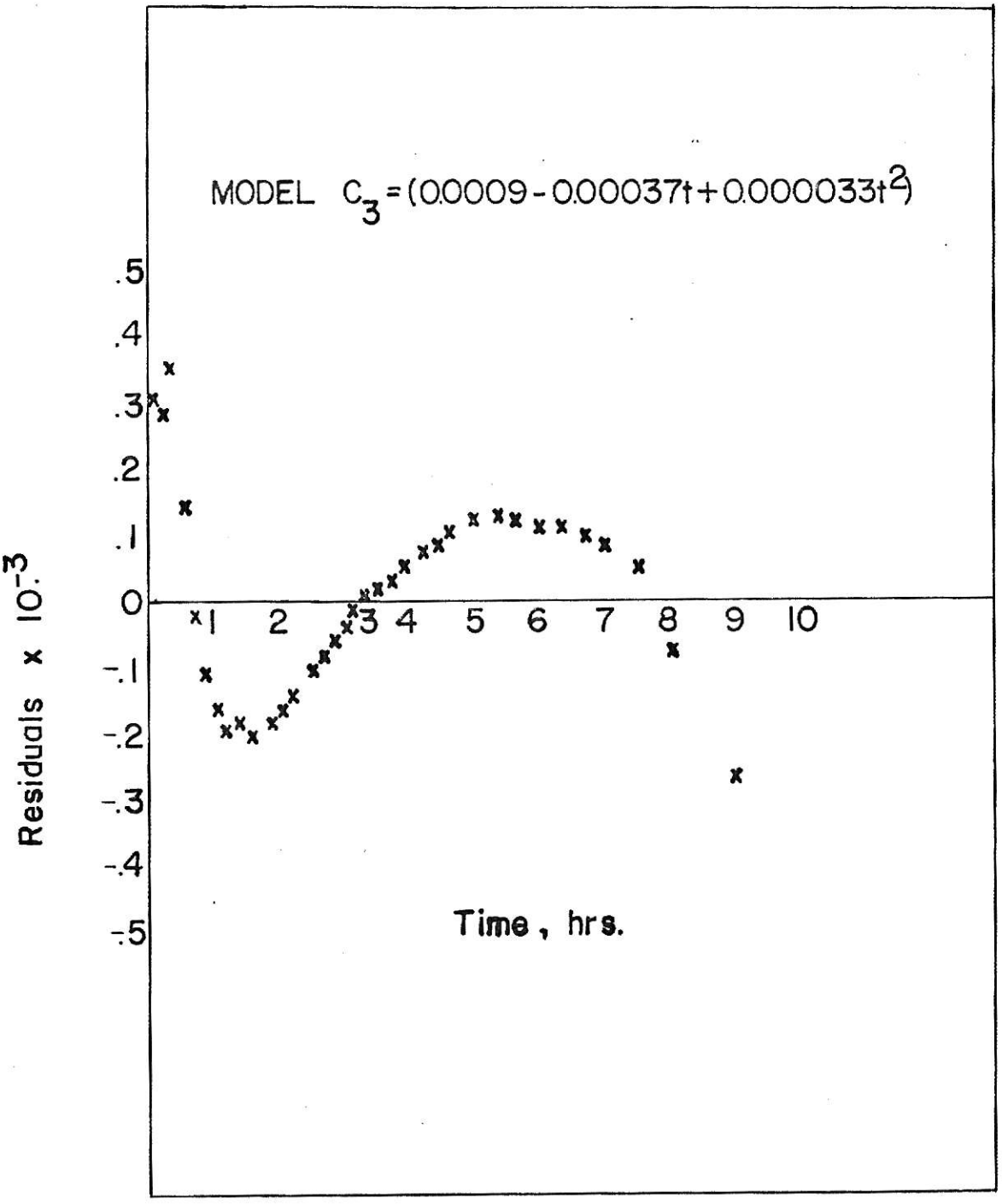

Fig. IV-2. Time vs. residuals.

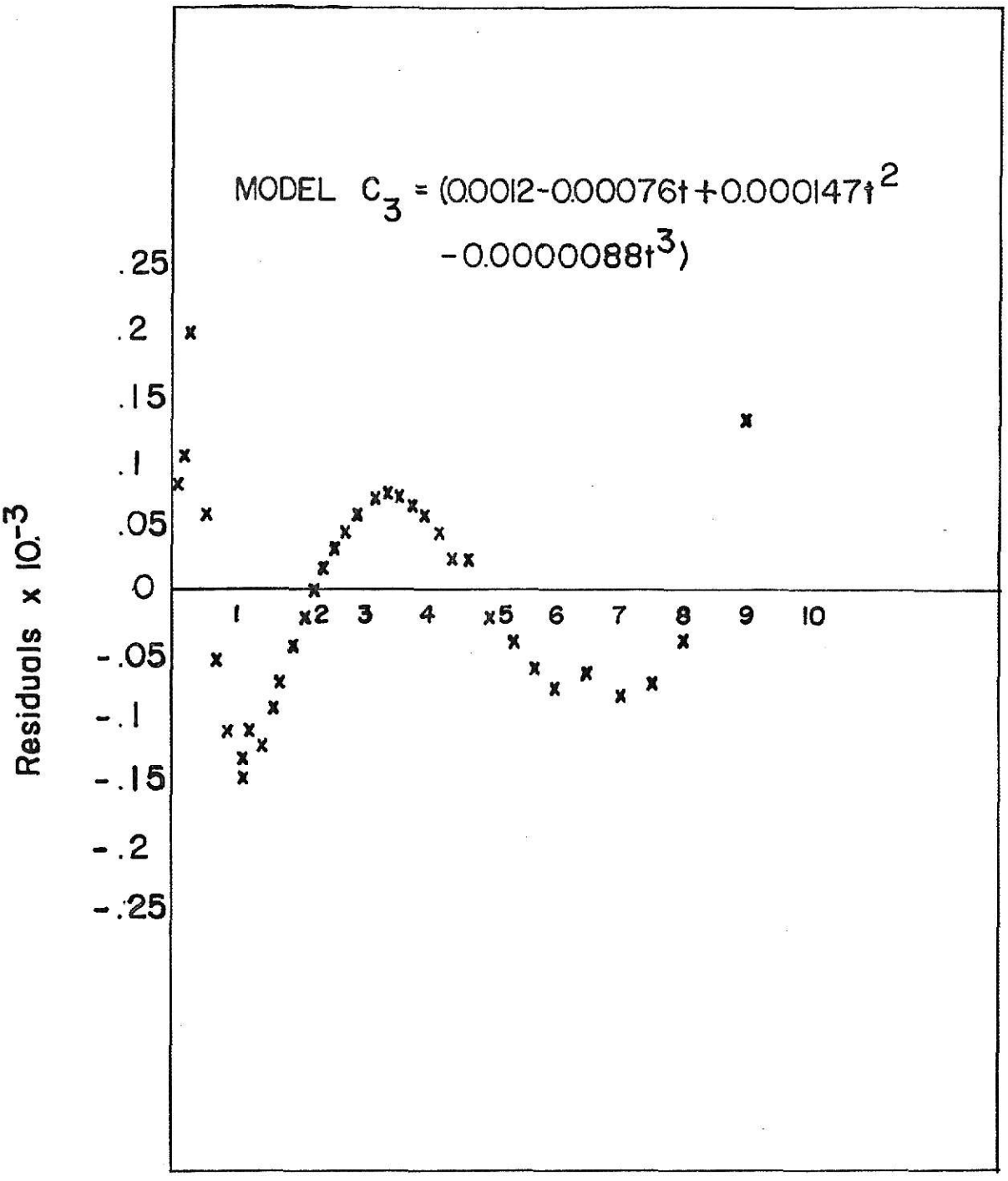

Fig. IV-3. Time vs residuals.

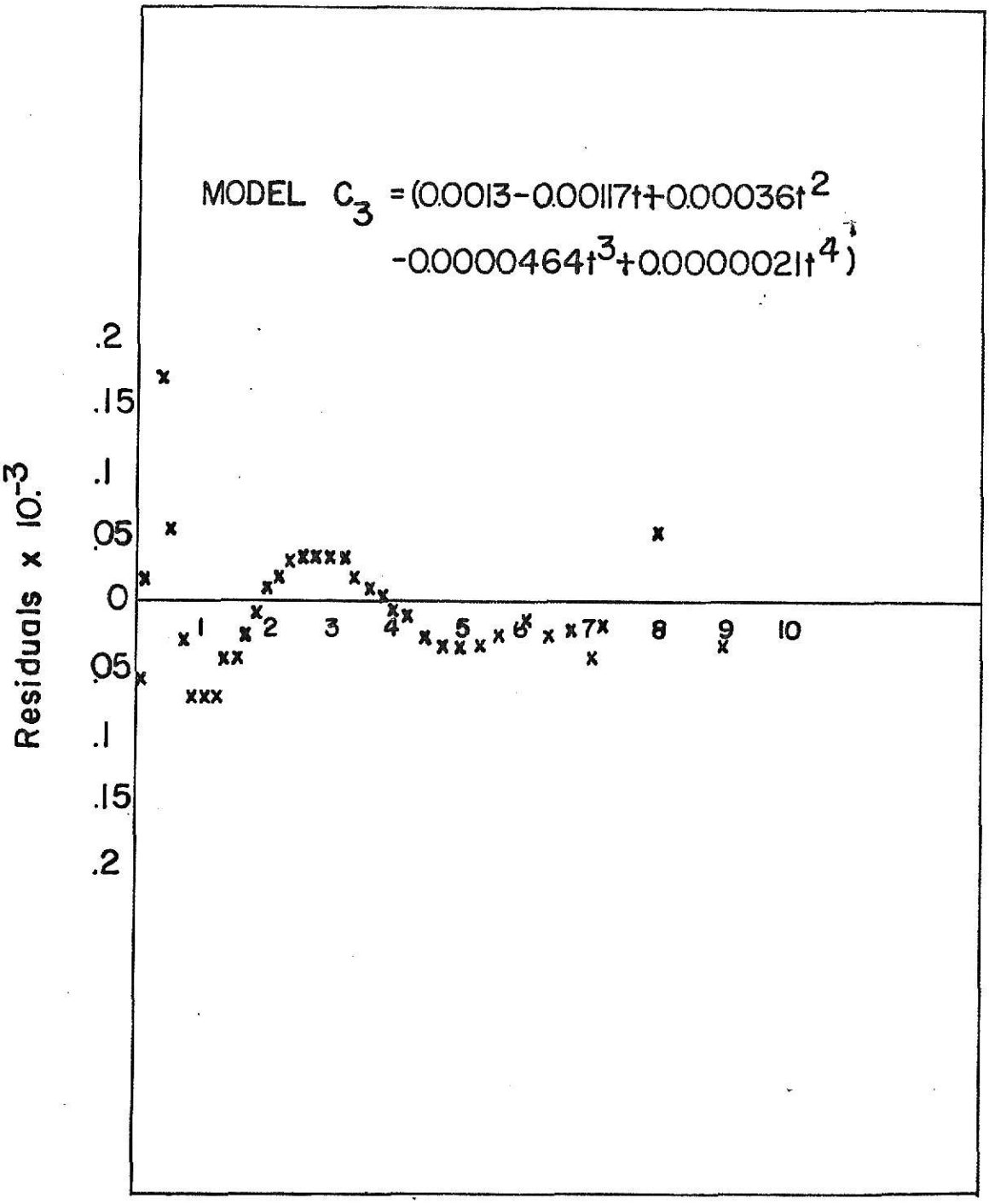

Fig. IV-4. Time vs residuals.

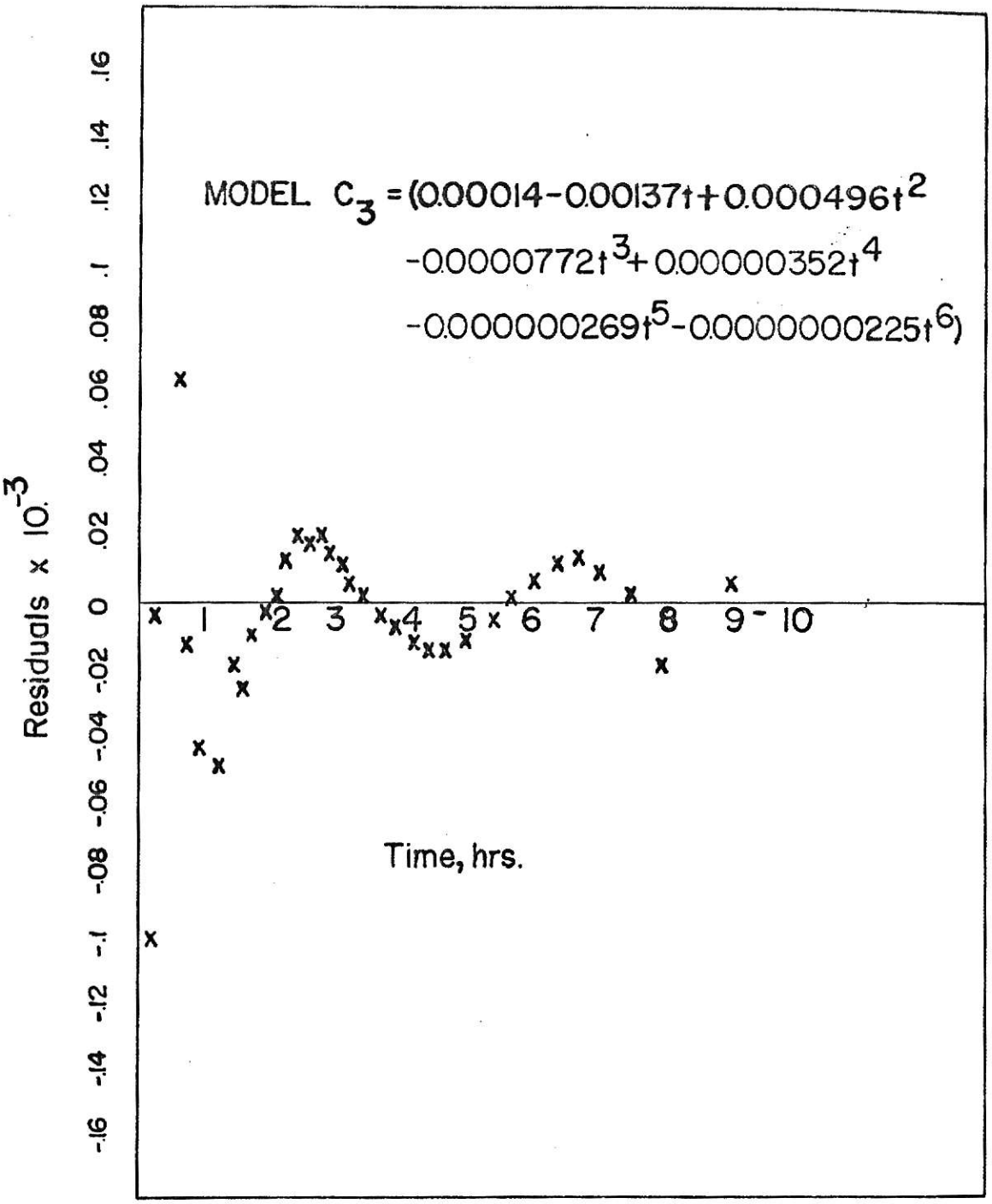

Fig. IV-5. Time vs residuals.

APPENDIX V

Modified Gauss-Newton Method for maximization of a selected criterion function is briefly described in this appendix.

The Gauss-Newton method, with modification by Greenstadt-Eisenpress [7], Bard [5], and Carroll [6] was used for maximizing the selected criterion function. This method is fully described in reference [9]. A brief description taken from [9] is given below.

The maximization of the $G(\theta)$ proceeds in an iterative fashion. At each iteration we start with an initial guess $\theta^{(0)}$ for the vector θ, and proceed to find a new guess $\theta^{(1)}$ such that $G(\theta^{(1)}) > G(\theta^{(0)})$. This now becomes the initial guess for the next iteration. In the course of iteration two things must be determined: a direction $\Delta\theta$ to proceed in, and length λ of the step to be taken along this direction, so that $\theta^{(1)} = (\theta^{(0)} + \lambda\ \Delta\theta)$. To be acceptable, the direction must be such that as one proceeds along it from $\theta^{(0)}$ the value of G - function increases at least initially, i.e., for sufficiently small λ. The value of λ is ideally such that as to take us near the peak of G along the chosen direction. The other requirement being that, λ must be small enough to insure that $\theta^{(1)}$satisfies all bounds and constraints on θ.

(i) Choice of Direction

Let P be the vector of partial derivatives $\frac{\partial G}{\partial \theta}$ at $\theta=\theta^{(0)}$. If R is any positive definite matrix, the direction $\Delta\theta = R_P$ is easily shown to be acceptable. If $\theta^{(0)}$ is sufficiently near a maximum of G, the matrix $Q_{ij} = -\frac{\partial^2 G}{\partial\theta_i \partial\theta_j}$ is positive (or at least non-negative) definite, and it can be shown that Q^{-1} is the most efficient choice of R. Using $R = Q^{-1}$ at all points constitutes the Newton-Raphson method. In regions where Q is not

positive definite this method is likely to yield non-acceptable directions, and hence fail to converge.

This problem can be overcome according to Greenstadt [7] if one represents the matrix Q by means of its eigenvalues and eigenvectors:

$$Q_{ij} = \sum_k V_{ik} V_{jk} \mu_k \qquad \text{(V-1)}$$

where

V_{ik} = i^{th} element of the k^{th} eigenvector of Q

V_{jk} = j^{th} element of the k^{th} eigenvector of Q

μ_k = k^{th} eigenvalue of Q

Now setting:

$$R_{ij} = \sum_k V_{ik} V_{jk} |\mu_k|^{-1} \qquad \text{(V-2)}$$

This R is always positive definite, and coincides with Q where the latter is itself positive definite, i.e., around the maximum. It is important to note that if some μ_k is zero, this value is replaced by a small, non-zero number.

(ii) Choice of Step Size

As stated above, we computed $\Delta\theta = R_p$, with R as defined by Equation (V-2). Let λ_o be a positive number such that $(\theta^{(0)} + \lambda_o \Delta\theta)$ is in the acceptable region. Now set $\lambda_2 = \min(\lambda_o, 1)$ and $\theta^{(2)} = (\theta^{(0)} + \lambda_2 \Delta\theta)$.

Define $\Gamma(\lambda) = G[\theta^{(0)} + \lambda\ \Delta\theta]$. Then $\Gamma(0) = G(\theta^{(0)})$; $\Gamma(\lambda_2) = G(\theta^{(2)})$; and

$$\frac{d\Gamma}{d\lambda}\Big|_{\lambda=0} = \frac{\partial G}{\partial \theta}\Big|_{\theta=\theta(0)} \cdot \Delta\theta = p \cdot \Delta\theta = \sum_i p_i \Delta\theta_i$$

$\Gamma(\lambda)$ can be approximated by a parabola $(a + b\lambda + c\lambda^2)$ matching the values of the parabola at $\lambda = 0$ and $\lambda = \lambda_2$, and its derivatives at $\lambda = 0$ with corresponding known quantities of the function $\Gamma(\lambda)$. Now values of λ are computed, say λ_3, which maximize this parabola. If the parabola has no maximum, λ_3 is set equal to λ_2. If $(\theta^{(0)} + \lambda_3\Delta\theta)$ is infeasible, λ_3 is truncated approximately. The following cases are considered:

(i) $\Gamma(\lambda_2) > \Gamma(0)$. If $\frac{\lambda_3\lambda_2}{\lambda_0} < 0.1$, set $\lambda = \lambda_2$ or $\lambda = \lambda_3$ and proceed to the next iteration depending upon whether $\Gamma(\lambda_2)$ is or is not greater than $\Gamma(\lambda_3)$.

(ii) $\Gamma(\lambda_2) \leq \Gamma(0)$.

Set $\lambda_4 = \max(\lambda_3, \frac{1}{4}\lambda_2)$ and compute $\Gamma(\lambda_4)$. If $\Gamma(\lambda_4) > \Gamma(0)$ set $\lambda = \lambda_4$ and proceed to next iteration. Otherwise replace λ_2 with λ_4, draw a parabola and proceed as before.

(iii) Convergence and Termination

In principle the method converges to a stationary point of the objective function, provided the latter possesses continuous first derivatives [5]. The process is terminated when each component of the vector $\lambda\Delta\theta$ is so small as to satisfy the inequality:

$$|\lambda\Delta\theta_i \leq 0.0001\ (0.001 + |\theta_i^{(0)}|) \qquad (i=1, 2, \ldots,\ell) \qquad (V\text{-}3)$$

This criterion was suggested by Marquardt [13]. However it does not guarantee that the maximum has been attained, but seems to work in most practical situations.

(iv) Penalty Functions

It was stated above that objective function must have continuous first derivatives. When bounds or complex constraints are placed on θ the distribution function $p_0(\theta)$ may be discontinuous; that is it may be zero outside the acceptable region and finite inside it. Bard [5] has suggested the following device to smooth out this discontinuity.

Suppose the restrictions placed on θ are stated in the form of inequalities:

$$Z_i(\theta) \leq 0 \qquad (k = 1, 2, \ldots, r) \qquad (V\text{-}4)$$

where the Z_i are specified functions. For instance, in the case of bounds $\alpha_i \leq \theta_i \leq \beta_i$ the corresponding Z_i would be:

$$Z_{2i-1}(\theta) = (\alpha_i - \theta_i) \leq 0 \qquad (V\text{-}5)$$

$$Z_{2i}(\theta) = (\theta_i - \beta_i) \leq 0 \qquad (V\text{-}6)$$

Now the prior distribution $p_o(\theta)$ is replaced by the function $p_o^*(\theta)$ $= p_o(\theta) \exp \sum_{i=1} \frac{\nu_i}{Z_i(\theta)}$, where ν_i are preassigned constants. If ν_i are sufficiently small, p_o^* will not differ appreciably from p_o in the interior

of the acceptable region, where Z_i are significantly different from zero. As one approaches the boundry of the region, however, at least one Z_i approaches zero from the negative side and hence p_o^* also approaches zero. Since p_o enters G through its logarithm, replacing p_o is equivalent to adding the penalty function $\sum_{i=1} \frac{\nu_i}{Z_i(\theta)}$ to $G(\theta)$, as suggested by Carroll [6].

The maximization proceeds in several phases: first the maximum of G and the penalty function is found. If at this point the value of penalty function is not negligible, the ν_i are multiplied by 0.1, and the maximum of the resulting function is found. If necessary, this process is repeated once more. Finally, the true maximum of G is found. If the maximum of G is an interior point of the admissible region, all the maxima will practically coincide [5]. Letting

$$Z = \sum_{i=1}^{r} \frac{\nu_i}{Z_i(\theta)} \tag{V-7}$$

The required derivatives can be found by differentiation.

$$\frac{\partial Z}{\partial \theta_\alpha} = - \sum_{i=1}^{r} \frac{\nu_i}{Z_i^2(\theta)} \frac{\partial Z_i}{\partial \theta_\alpha} \tag{V-8}$$

$$\frac{\partial^2 Z}{\partial \theta_\alpha \partial \theta_\beta} = \sum_{i=1}^{r} \frac{\nu_i}{Z_i^3} \left[2 \frac{\partial Z_i}{\partial \theta_\alpha} \frac{\partial Z_i}{\partial \theta_\beta} - Z_i \frac{\partial^2 Z_i}{\partial \theta_\alpha \partial \theta_\beta}\right]$$

The important thing to note here is that when the constraints are linear (in particular, in the case of upper and lower bound), we have

$$\frac{\partial^2 Z_i}{\partial\theta_\alpha \partial\theta_\beta} = 0 \qquad \text{(V-10)}$$

Chapter 5

ANALYSIS OF FLOW BEHAVIOR IN A CONTINUOUS, MULTISTAGE TOWER FERMENTOR: EFFECT OF DESIGN VARIABLES ON THE EXTENT OF BACKFLOW

5.1 INTRODUCTION

In continuous fermentation processes, there are applications where mixing patterns other than those of the chemostat (complete mixing) are desired. Plug flow conditions, although very difficult to attain, offer an opportunity to immediately apply knowledge gained in batch growth to continuous processing. The desire to develop an economical fermentor that could approximate plug flow conditions led to the development and construction of the tower fermentor employed in this study [1]. In the design of sieve plate columns such as the fermentor, control of liquid backflow is an important factor. The need to develop techniques to measure and control the backflow rate in a cocurrent tower fermentor provided the incentive for this study. One important goal was to show that the backflow rate could be reduced to near zero so that plug flow conditions could be approximated. In the tower fermentor introduced by Prokop et. al. [1] and the somewhat different tower fermentor of Falch and Gaden [2, 3], a non-zero backflow rate is common, and zero backflow appears not to have been encountered. Since the backflow rate may be influenced by hole size, hole void area, plate thickness, air flow rate, and liquid flow rate, an investigation to determine the extent of liquid backflow under various conditions was undertaken. Three variables, hole void area, air flow rate, and liquid flow

rate are varied in this study to determine their effect on liquid backflow.

A tracer study is employed to investigate the flow behavior of the liquid phase in the tower fermentor. A flow model for the liquid phase is developed and used to investigate the extent of backflow in the tower system.

5.2 EXPERIMENTAL PROCEDURE

Flow behavior studies were carried out in an aerated column, consisting of eight compartments, separated by sieve plates, with feed to the second stage. Water and air flow were cocurrent from the bottom to the top of the column. The column fermentor employed in the tracer study has been described previously [1]. Tracer (salt) was injected to the second stage and its concentration was recorded in the first three stages and the eighth stage at different instants of time. Sieve plates with hole area per plate of 3.43 and 15 percent were employed. Air flow rates of 4, 8, and 15.5 lit./min and water flow rates of 2, 4, and 8.55 lit./hr were used.

5.3 MATHEMATICAL MODEL

An eight stage tower fermentor with feed introduced at the second stage was used for tracer study. Experimental measurements were made using salt as a tracer for the first three stages and the eighth stage only. The flow model of the tower fermentor is shown in Figure 1. A dotted boundary is used to designate the partial system consisting of the first three stages which is used in estimating the parameters. Complete mixing is assumed in each compartment and backflow from each stage is included in the model.

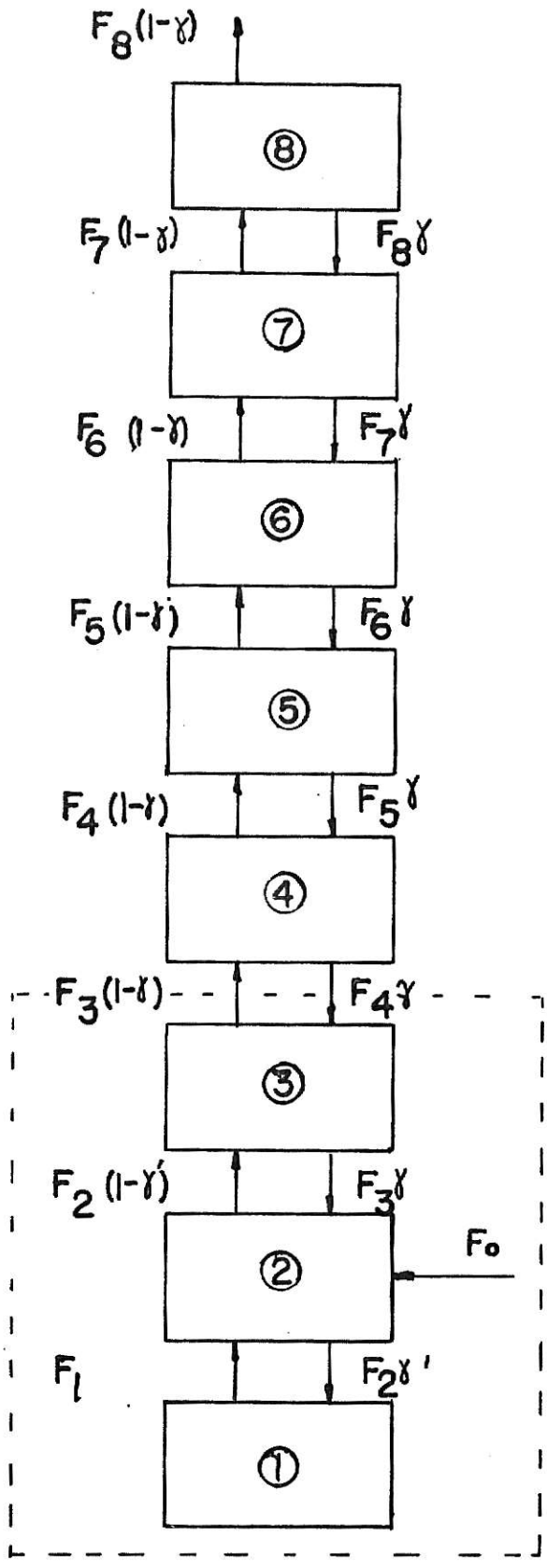

Fig. 1. Flow model of the tower fermentor.

One of the important factors considered is the rate of backflow, and this is represented by $F_i\gamma_i$, where γ_i is the fraction of the total flow F_i from the ith stage which is backflow.

The unsteady state tracer material balances for each stage of the tower fermentor are as follows;

First stage: $$V_1 \frac{dC_1}{dt} = F_2\gamma_2C_2 - F_1C_1 \quad (1)$$

Second stage: $$V_2 \frac{dC_2}{dt} = F_1C_1 + F_3\gamma_3C_3 - F_2C_2 \quad (2)$$

Stages 3 to 7: $$V_i \frac{dC_i}{dt} = F_{i-1}(1 - \gamma_{i-1})C_{i-1} + F_{i+1}\gamma_{i+1}C_{i+1} - F_iC_i,$$

$$i=3, 4, 5, 6, 7 \quad (3)$$

Eighth stage: $$V_8 \frac{dC_8}{dt} = F_7(1 - \gamma_7)C_7 - F_8C_8 \quad (4)$$

The initial conditions are

$C_i = 0$ at $t=0$, $i=1, 3, 4, 5, 6, 7, 8$

$C_2 =$ finite at $t=0$

During the tracer experiment, the liquid flow rate is constant. The following steady state material balances are used in this study:

Overall balance: $$F_0 = F_8(1 - \gamma_8) \quad (5)$$

Stage 1: $$F_2\gamma_2 - F_1 = 0 \quad (6)$$

Stage 2: $F_1 + F_3\gamma_3 - F_2 = - F_0$ (7)

Stages 3 to 7: $F_{i-1}(1 - \gamma_{i-1}) + F_{i+1}\gamma_{i+1} - F_i = 0$, i=3, 4, 5, 6, 7 (8)

Stage 8: $F_7(1 - \gamma_7) - F_8 = 0$ (9)

where

V_i = volume of the ith stage, (lit.), i = 1, 2, ..., 8

C_i = tracer concentration in the ith stage, (gm./cc), i = 1,2,...,8

F_i = flow out of stage i, (lit./hr.), i = 1, 2, ..., 8

F_0 = feed flow rate to the second stage, (lit./hr.)

t = time, (hrs.)

γ_i = backflow coefficient for ith stage, (i = 2, 3, ..., 8)

Two simplifications are made in the above mathematical model. First of all the individual stage volumes are assumed to be equal i.e. $V_1 = V_2 = V_3 = \ldots = V_8 = V$. The backflow coefficients for stages three through eight are assumed to be equal i.e. $\gamma_3 = \gamma_4 = \gamma_5 = \ldots = \gamma_8 = \gamma$. The backflow coefficient for stage two may differ from γ because the feed is introduced to the second stage: γ_2 will be represented by γ'.

5.4 FORMULATION OF THE PROBLEM

Experimental measurements were made for tracer concentrations in the first three stages and eighth stage. Two distinct approaches may be used to identify the unknown parameters. In the first one (Method 1), the differential mathematical model consisting of Equations (1) through (4) may be

considered and backflow parameters can be estimated using the usual identification techniques for differential models. However an alternative approach would be to consider the partial model indicated by the dotted boundary in Figure 1. The performance equations for the first two stages can be solved to obtain the tracer concentrations C_1 and C_2 as a function of time and unknown parameters, if the tracer concentration in the third tank is approximated by a polynomial. The backflow parameters can then be estimated using a direct search procedure.

Both of these approaches (Method 1 and Method 2) were employed to estimate the extent of backflow using one set of data from the experimental study (Experiment 1). Comparison of the optimal results indicated that Method 2 gave a better fit for the experimental data [Chapter 3]. Hence Method 2 was used to identify the backflow parameters in the rest of the experiments (Experiments 2 through 11).

Equations (1) and (2) were solved to obtain tracer concentrations in the first and second tank as a function of time and unknown parameters. Detailed description of these techniques and complete analytical solution of the partial system has been presented elsewhere [Chapter 3]. The solution of Equations (1) and (2) can be written as follows:

$$C_1 = \frac{1}{(\lambda_1 - \lambda_2)} \left[\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right] e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left(\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right) + K_0\right] e^{\lambda_2 t} \qquad (10)$$

$$+ K_0 + K_1 t + K_2 t^2 + K_3 t^3 + K_4 t^4$$

$$C_2 = \frac{1}{(\lambda_1 - \lambda_2)} \left[\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right] e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\} + K_0\right] e^{\lambda_2 t}$$

$$+ \frac{V}{F_1} \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\}\right] \lambda_1 e^{\lambda_1 t}$$

$$- \left[\frac{1}{(\lambda_1 - \lambda_2)} \left\{\frac{C_2(0)F_1}{V} + (\lambda_2 K_0 - K_1)\right\} + K_0\right] \lambda_2 e^{\lambda_2 t}$$

$$+ \frac{V}{F_1} \left[K_1 + 2K_2 t + 3K_3 t^2 + 4K_4 t^3\right] \tag{11}$$

where

$$\lambda_1 = \frac{- \left(1 + \frac{F_2}{F_1}\right) + \sqrt{\left(1 + \frac{F_2}{F_1}\right)^2 - 4 \frac{V}{F_1} \left(\frac{F_2 - F_1}{V}\right)}}{2 \frac{V}{F_1}} \tag{12}$$

$$\lambda_2 = \frac{- \left(1 + \frac{F_2}{F_1}\right) - \sqrt{\left(1 + \frac{F_2}{F_1}\right)^2 - 4 \frac{V}{F_1} \left(\frac{F_2 - F_1}{V}\right)}}{2 \frac{V}{F_1}} \tag{13}$$

$$K_0 = \left[\frac{mA_0}{g} - \frac{2amA_2}{g^2} + \frac{24a^2 mA_4}{g^3} + \frac{6abmA_3}{g^3} - \frac{24ab^2 mA_4}{g^4} - \frac{bmA_1}{g^2}\right.$$

$$+ \frac{6abmA_3}{g^3} - \frac{24ab^2mA_4}{g^4} + \frac{2b^2mA_4}{g^3} - \frac{24ab^2mA_4}{g^4} - \frac{6b^3mA_3}{g^4} + \frac{24b^4mA_4}{g^5}] \quad (14)$$

$$K_1 = [\frac{mA_1}{g} - \frac{6amA_3}{g^2} + \frac{24abmA_4}{g^3} - \frac{2bmA_2}{g^2} + \frac{24abmA_4}{g^3} + \frac{6b^2mA_3}{g^3} - \frac{24b^3mA_4}{g^4}] \quad (15)$$

$$K_2 = [\frac{mA_2}{g} - \frac{12amA_4}{g^2} - \frac{3bmA_3}{g^2} + \frac{12b^2mA_4}{g^3}] \quad (16)$$

$$K_3 = [\frac{mA_3}{g} - \frac{4bmA_4}{g^2}] \quad (17)$$

$$K_4 = [\frac{mA_4}{g}] \quad (18)$$

$$a = \frac{V}{F_1}$$

$$b = (1 + \frac{F_2}{F_1})$$

$$g = \frac{(F_2 - F_1)}{V}$$

$$m = \frac{(F_2 - F_1 - F_0)}{V}$$

A fifth order polynomial

$$C_3 = A_0 + A_1 t + A_2 t^2 + A_3 t^3 + A_4 t^4 + A_5 t^5,$$

where A_0, A_1, A_2, A_3, A_4, and A_5 are constants, was employed to approximate the tracer response curve in stage 3.

The criterion function employed in this investigation was of the form

$$S = \sum_{i=1}^{N} |EC_1^i - C_1^i| + \sum_{i=1}^{N} 0.5 \, |EC_2^i - C_2^i|$$

$$+ \sum_{i=1}^{N} 0.5 \, (EC_2^i - C_2^i)^2 \qquad (19)$$

where EC_1 and EC_2 are experimental values of tracer concentrations in stage 1 and 2 respectively. As described in Chapter 3, a modified sequential simplex pattern search [4] is used to estimate the backflow parameters by minimizing the criterion function using Equations (10) and (11). The statistical techniques which were used to select the degree of the polynomial are described in Chapter 4. Bard's method [5] for "Single Equation Least Squares" was used to estimate the parameters in the polynomial.

5.5 RESULTS AND DISCUSSION

Backflow parameters were estimated for the 11 tracer experiments described in Table 1. Table 2 shows the parameter estimates, optimum function value, and total number of data points (N) for each experiment. Reasonable fit was obtained for all the experiments. Three parameters $C_2(0)$, γ and

Table 1. Experimental Conditions for Tracer Experiments 1 through 11.

Variable	Experiment Number										
	1	2	3	4	5	6	7	8	9	10	11
Hole size, m.m.	2	2	2	2	2	2	2	2	2	2	2
Hole area/plate %	3.43	3.43	3.43	3.43	3.43	3.43	3.43	3.43	3.43	7.62	15
Liquid flow rate F_0, lit./hr.	2.0	2.0	2.0	4.0	4.0	4.0	8.55	8.55	8.55	2.06	2.02
Air flow rate lit./min.	4.0	8.0	15.5	4.0	8.0	15.5	4.0	8.0	15.5	4.0	4.0
Liquid volume lit./stage	0.633	0.522	0.495	0.633	0.522	0.496	0.495	0.522	0.633	0.633	0.633

Table 2. Parameter Estimation Results for Tracer Experiments.

Parameter	Experiment Number										
	1	2	3	4	5	6	7	8	9	10	11
$C_2(0) \times 10^{-3}$	3.73	4.807	5.303	4.475	4.862	6.227	5.543	5.491	3.876	4.41	4.45
γ	0.289	0.0	0.0	0.159	0.0	0.0	0.0	0.0	0.0	0.0	0.0
$'\gamma$	.089	.019	.052	.068	.023	.026	.022	.021	.078	.119	.237
$S \times 10^{-3}$	.636	.522	.495	.84	.873	1.097	.444	.563	.358	1.195	1.142
N	74	60	54	62	43	34	32	35	39	59	46
$S/N \times 10^{-5}$	.85	1.3	.80	1.3	2.0	3.2	1.3	1.6	.9	2.0	2.4
$S/N \times 10^{-4}$	3.4	4.8	3.8	4.6	6.8	9.7	6.5	6.7	4.8	5.8	7.3
$A_0 \times 10^{-3}$	1.391	1.836	1.988	1.819	2.199	2.962	3.135	2.293	2.125	1.467	1.454
$A_1 \times 10^{-3}$	-1.179	-1.750	-2.036	-3.600	-4.629	-8.311	-19.663	-9.949	-9.466	-1.292	-1.098
$A_2 \times 10^{-3}$	.362	.611	.685	2.815	3.699	8.862	47.943	16.373	15.723	.451	.289
$A_3 \times 10^{-3}$	- .046	- .097	- .084	-1.064	-1.403	-4.492	-56.656	-12.788	-12.110	- .075	- .027
$A_4 \times 10^{-4}$	.090	.070	.015	1.934	2.540	10.883	324.775	47.535	43.572	.061	- .0006
$A_5 \times 10^{-5}$	*	- .018	.023	-1.355	-1.766	-10.131	-724.058	-67.471	-59.122	- .018	.0088

* A fourth order polynomial was used in Experiment 1.

γ' are estimated using the tracer data from stages 1, 2, and 3. Quantities S/N and $\sqrt{S/N}$, which give an approximate measure of the mean error (or deviation) were computed and used to compare the goodness of the fit for different experiments. Two typical experiments (3 and 6) have been chosen to illustrate the agreement between the predicted and experimental tracer concentrations in different stages (see Figures 2 through 5). Good agreement between experimental and predicted tracer concentrations was obtained as shown in the figures. Based on the value of S/N, experiment 3 illustrates the best fit obtained while experiment 6 illustrates the poorest fit. Because of the nature of the criterion function employed, S/N under estimates the mean error, while $\sqrt{S/N}$ over estimates the mean error.

The sieve plate design is the same for the first nine experiments; however, for experiments 10 and 11 the hole void area is increased from 3.43% to 7.62% and 15% respectively. Figures 6 through 9 show the extent of backflow from each stage as a function of air and medium (water) flow rates and per cent hole void area. The symbols denote the rate of backflow at each stage. The lines connecting these points have no physical significance.

The analysis of different experiments indicates that one can adjust the backflow rate by changing either liquid or air flow rates or the hole void area. The backflow rate increases as the water flow rate (or hole void area) decreases (Figure 2 through 4). Lee [6] found that a comparitively wide range of operation can be obtained in a multistage tower fermentor if the dilution rate (or backflow rate) can be controlled. If the hole void area, hole size, and flow rates can be adjusted to obtain any

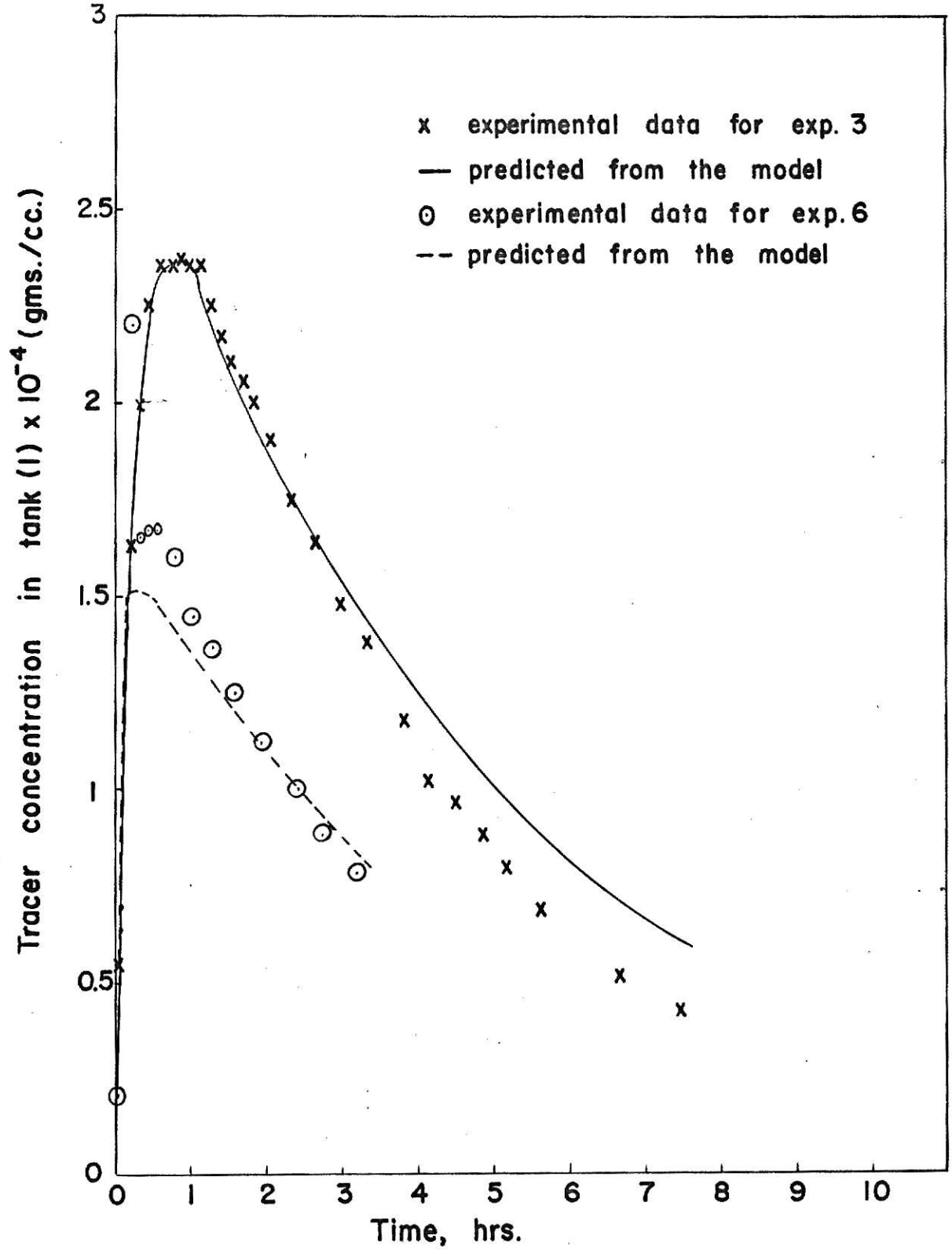

Fig. 2. Tracer concentration in tank (1) vs. time.

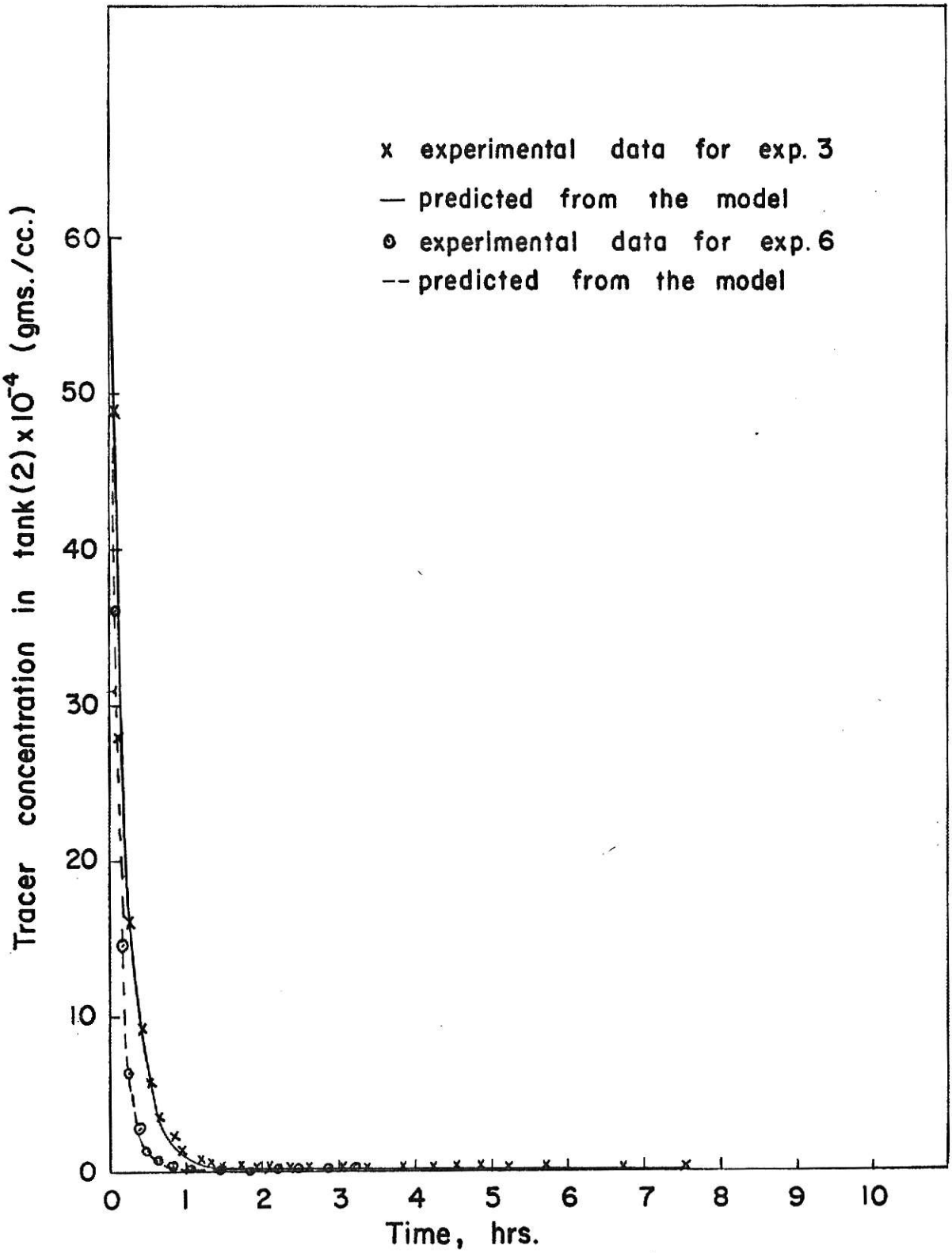

Fig. 3. Tracer concentration in tank (2) vs. time.

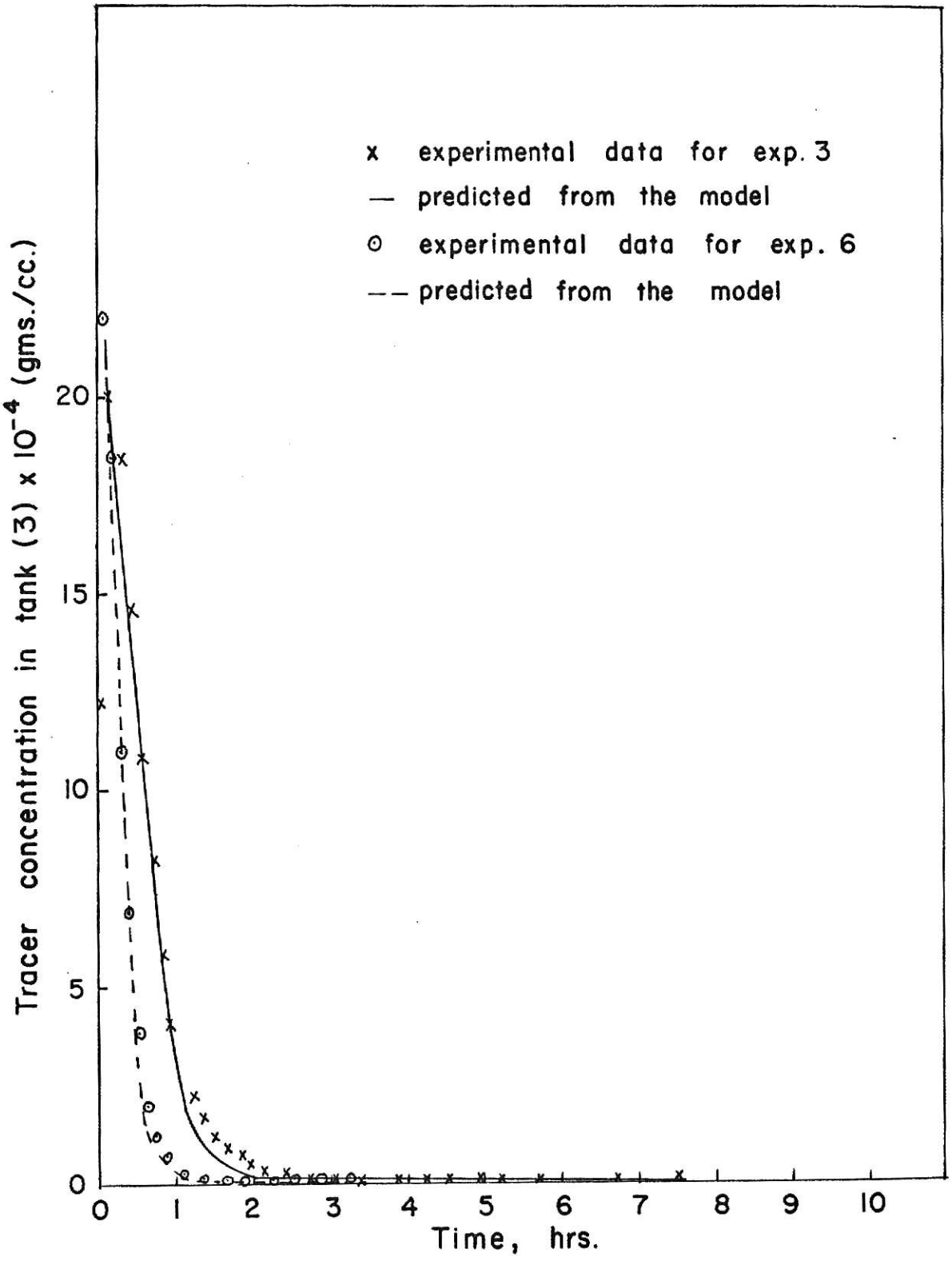

Fig. 4. Tracer concentration in tank (3) vs. time.

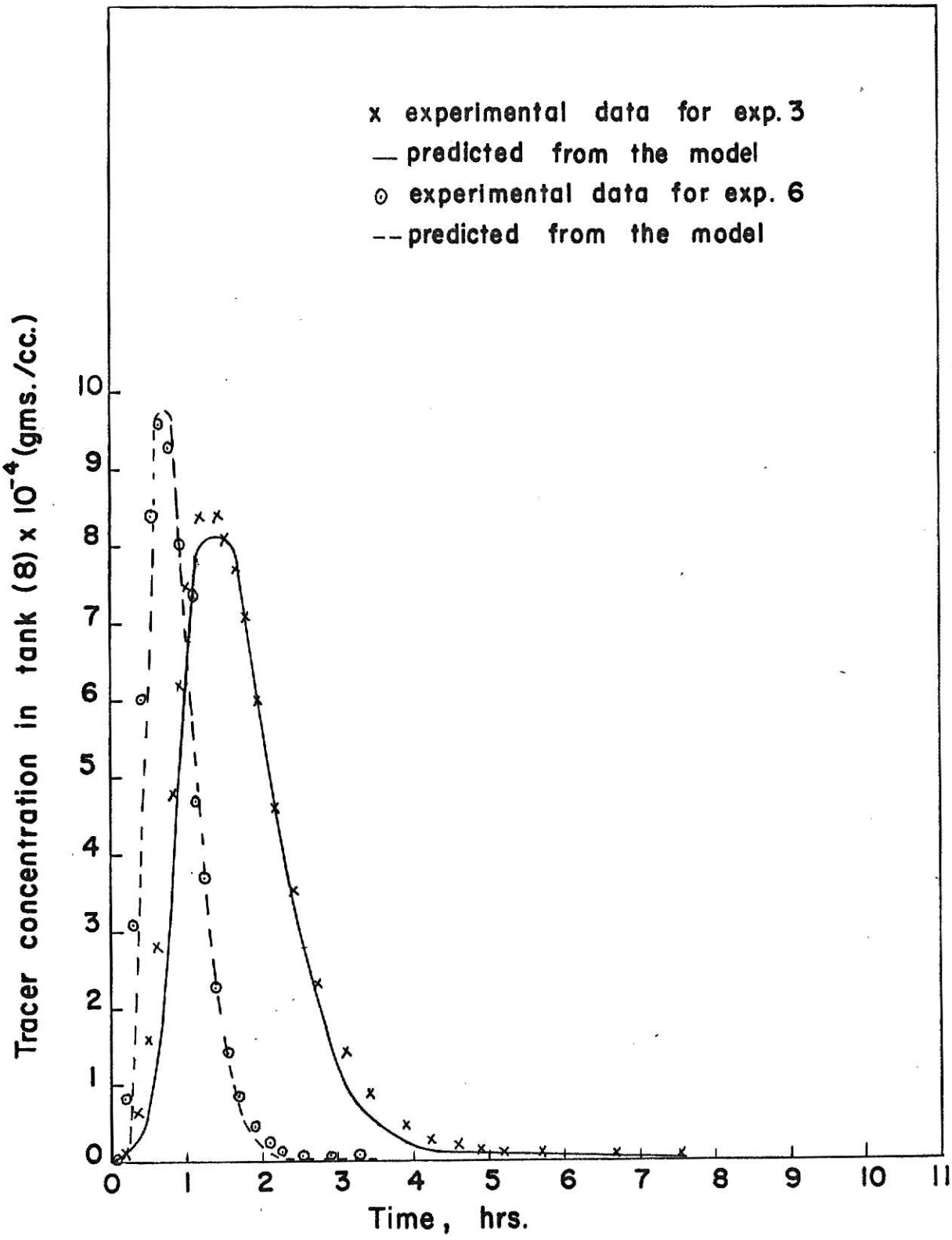

Fig. 5. Tracer concentration in tank (8) vs. time.

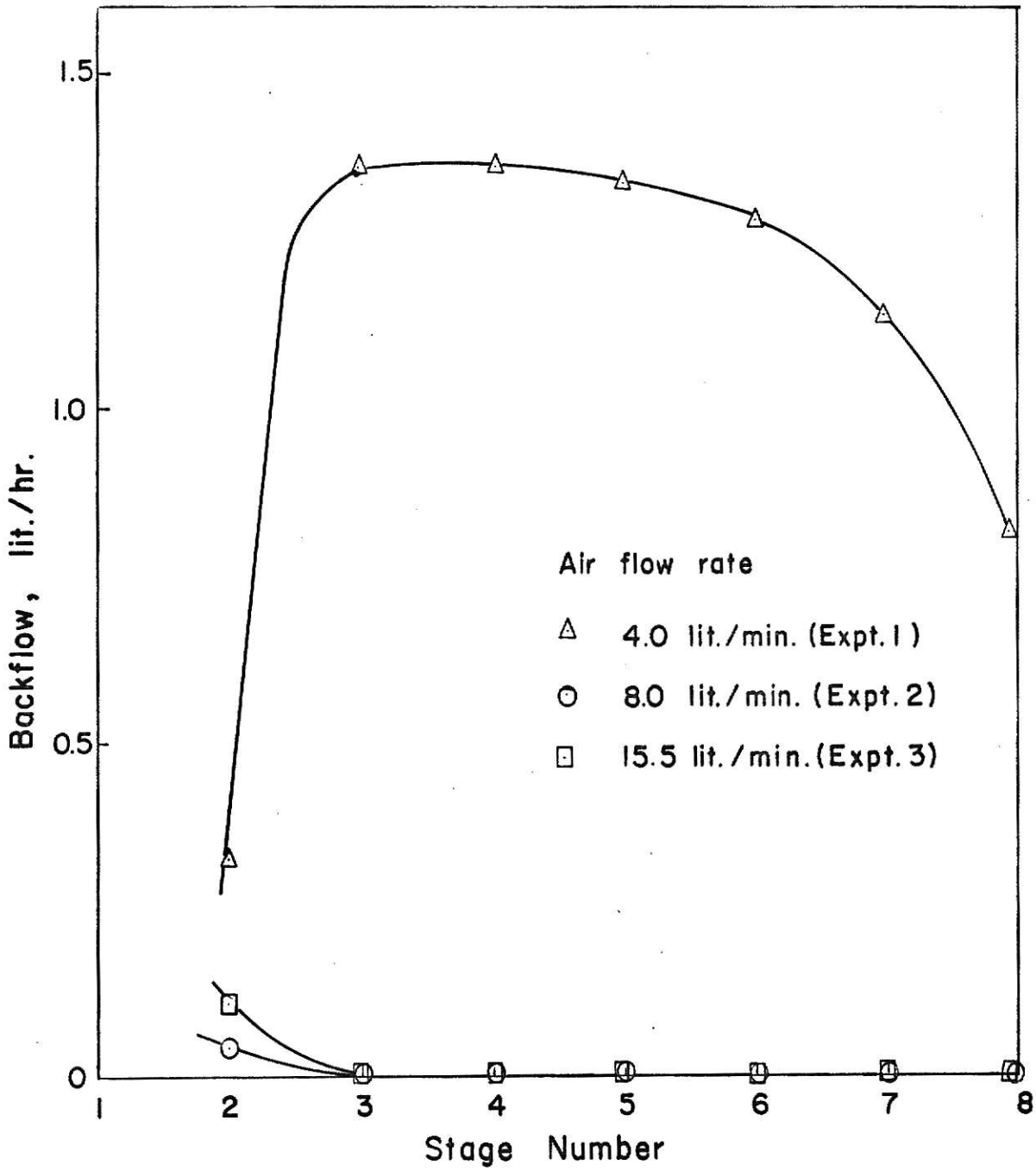

Fig. 6. Backflow rate vs. stage number for hole area per plate = 3.43% and water flow rate = 2.0 lit./hr. with air flow rate as parameter.

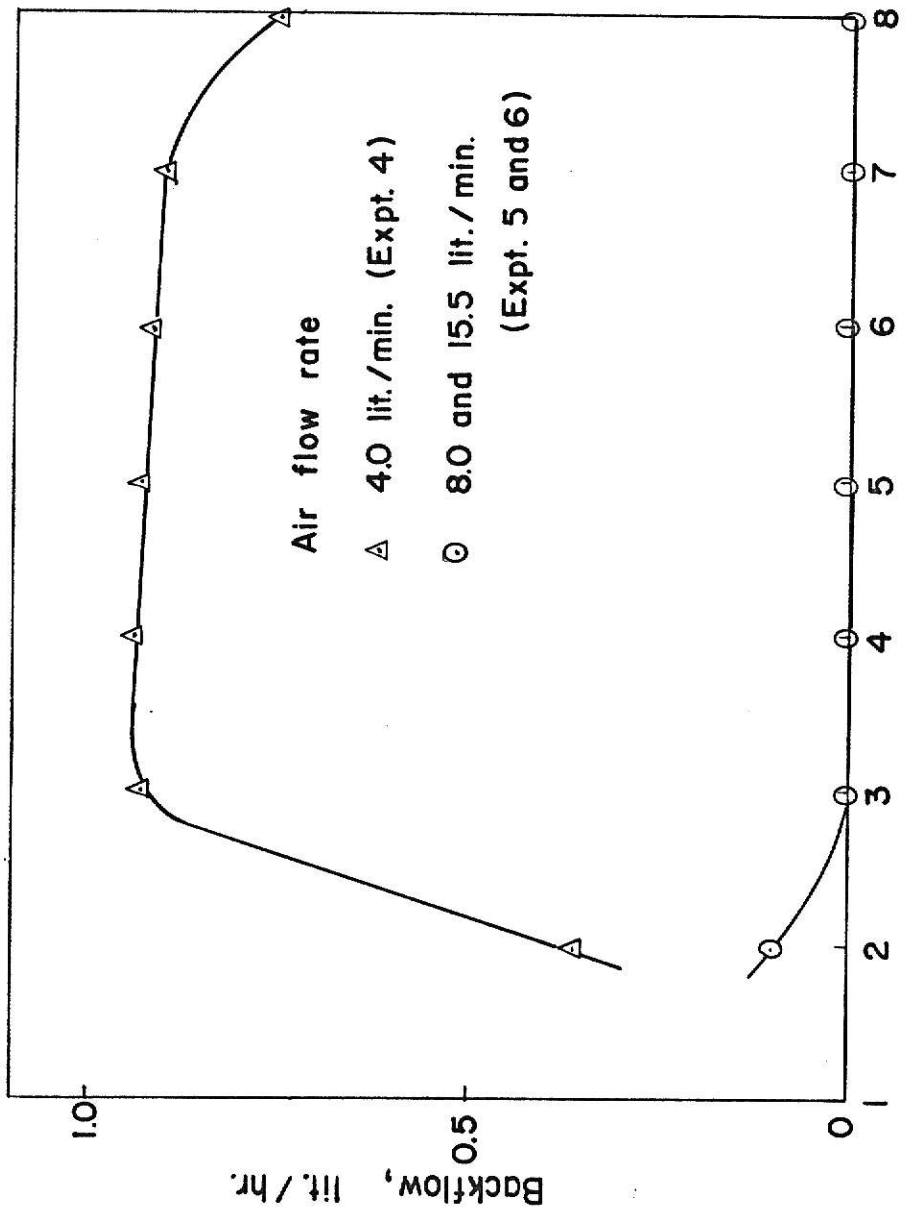

Fig. 7. Backflow rate vs. stage number for hole area/plate =3.43% and water flow rate = 4.0 lit./hr. with air flow rate as parameter.

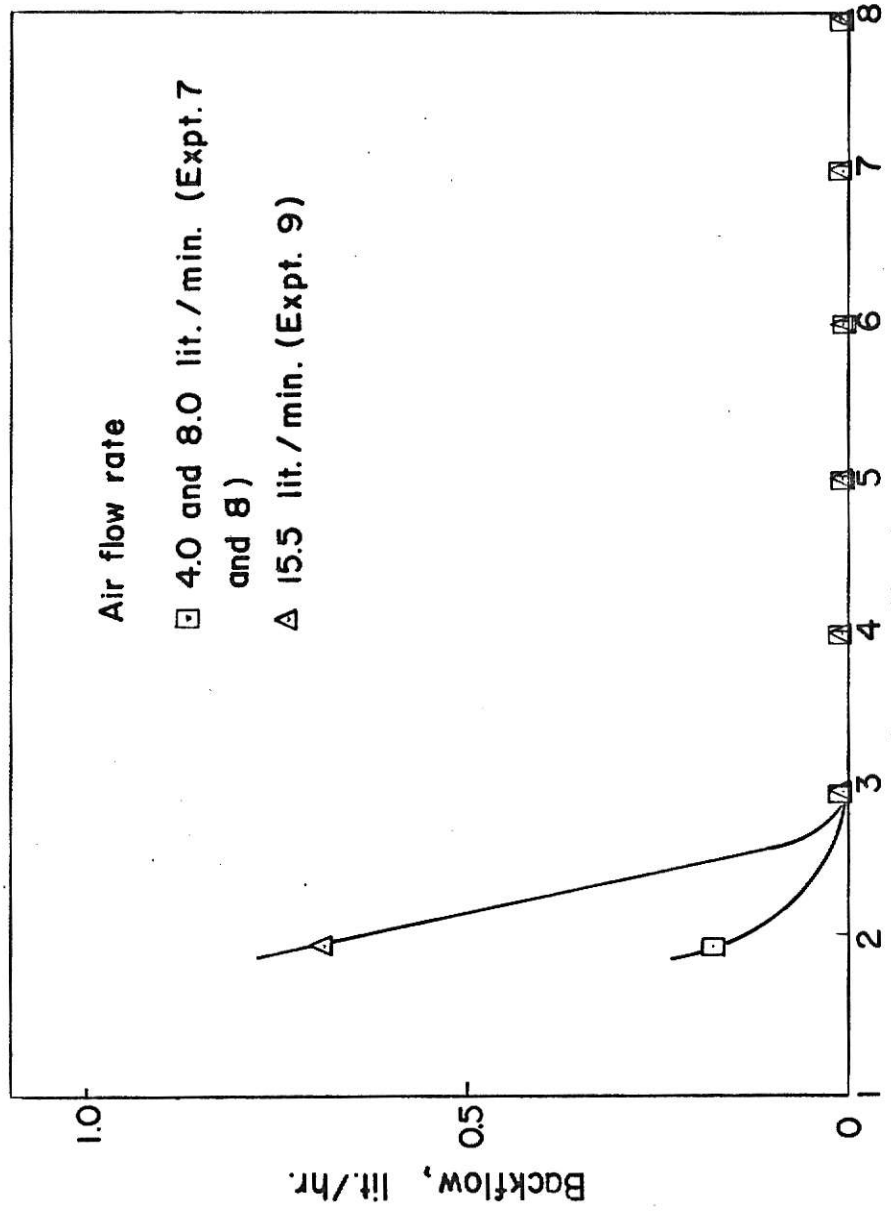

Fig. 8. Backflow rate vs. stage number for hole area per plate = 3.43% and water flow rate = 8.55 lit./hr. with air flow rate as parameter.

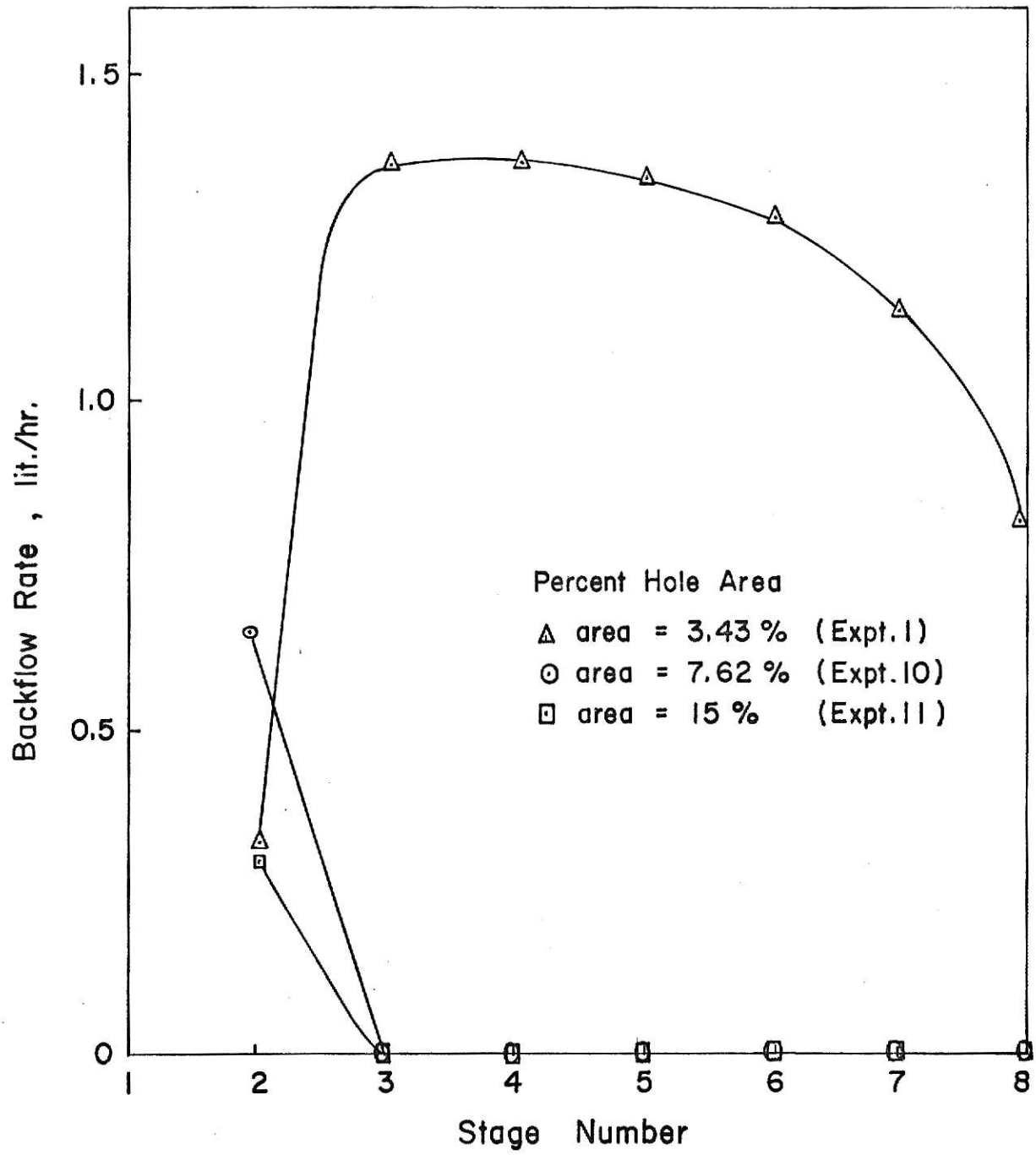

Fig. 9 Backflow rate vs. stage number for air flow rate = 4.0 lit./min. and water flow rate = 2.0 lit./min. with percent hole area as parameter.

desired extent of backflow, the tower fermentor can be very advantageously employed. This work shows that conditions in which the backflow is negligibly small can be found.

To illustrate the behavior of the tower fermentor as an approximation to plug flow behavior, the results for experiment 3 were used. Simulation of biological growth for these flow conditions with $\mu_{max} = 0.6\ hr^{-1}$, $Y = 0.5$, $S_0 = 10$ gms/lit., $X_0 = 0$, and $K_S = 0.1$ gms/lit., gave the results shown in Figure 10. Because of the small amount of backflow to the first stage, there is exponential growth in a large part of the fermentor. Approximately 21 stages would be required for complete substrate utilization. Continuous production of cells in physiological state ranging from cells in exponential growth to cells growing in a rapidly decreasing substrate environment can be obtained with the tower fermentor.

The results of this investigation show that the data collection procedures and parameter estimation procedures employed in this work give good estimates of the backflow. The results show that the procedure of injecting the tracer at a stage other than the bottom stage allows even small quantities of backflow to be observed. Since some differences have been observed from one type of tracer to the next and also from conditions of no active cell growth [1], it may be desirable to conduct further studies using this procedure.

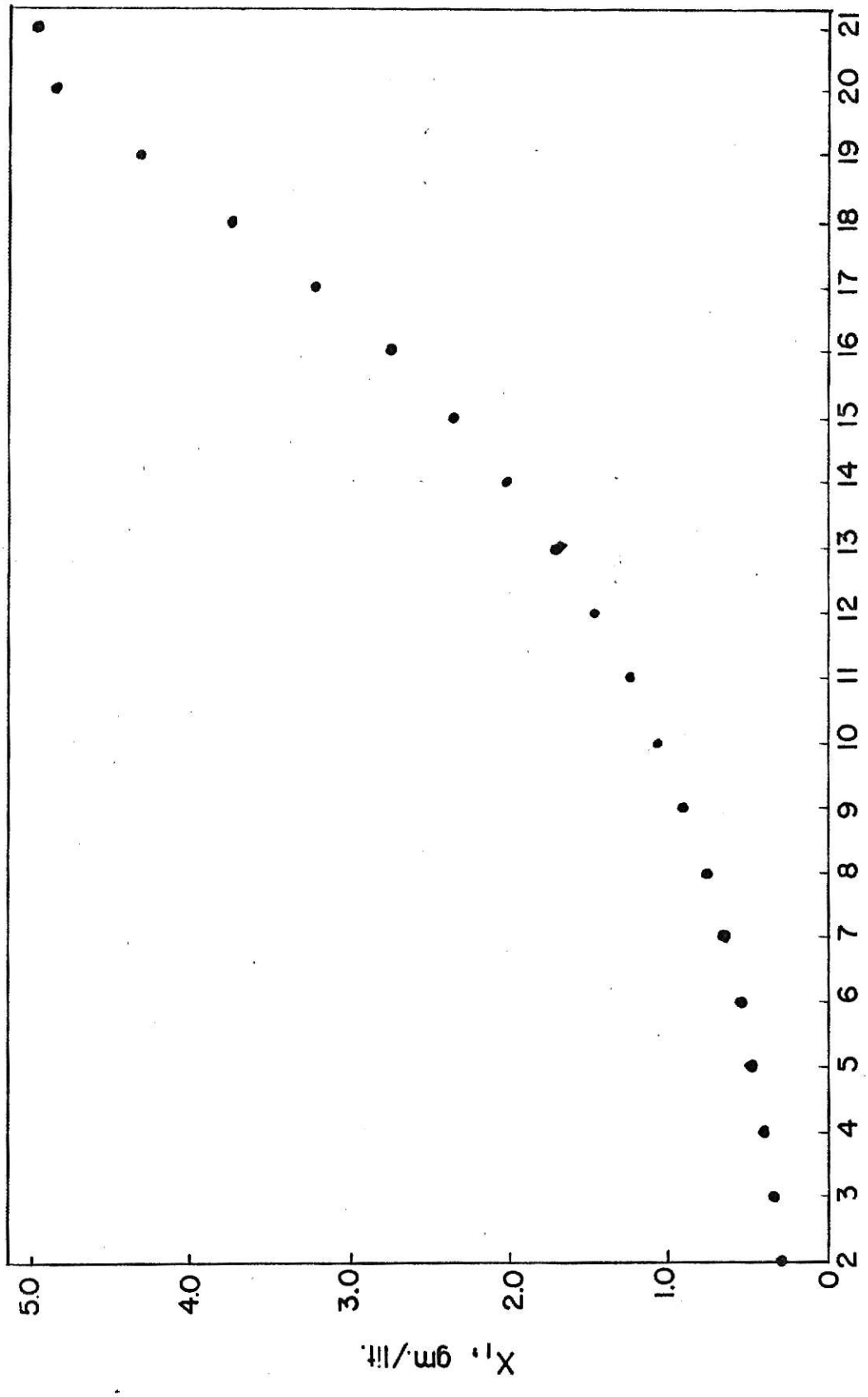

Fig. 10. Cell concentration as a function of stage number for flow conditions of Experiment 3.

5.6 NOTATION

A_0, A_1, A_2, A_3, A_4 = constants in Equations (14) through (18)

C_i = tracer concentration in i^{th} tank, (gm./cc)

$C_i(0)$ = initial tracer concentration in i^{th} tank, (gm./cc)

EC_i = experimental tracer concentration in i^{th} tank, (gm./cc)

D = dilution rate, (hr^{-1})

F_0 = feed flow rate, (lit./hr)

F_i = flow out from the i^{th} stage, (lit./hr)

K_0, K_1, K_2, K_3, K_4 = constants defined by Equations (14) through (18)

N = number of data points

S = objective function value

t = time (hrs.)

V_i = volume of the i^{th} tank, (lit.)

X = cell mass concentration, (mg./ml)

GREEK LETTERS

γ_i = backflow coefficient in i^{th} stage

λ_1, λ_2 = roots defined by Equations (12) and (13)

5.7 REFERENCES

1. Prokop, A., L. E. Erickson, J. Fernandez, and A. E. Humphrey, "Design and Physical Characteristics of a Multistage, Continuous Tower Fermentor," Biotech. and Bioeng., 11, 945-966 (1969).

2. Falch, E. A., and E. L. Gaden, "A Continuous Multistage Tower Fermentor: II. Analysis of Reactor Performance," Biotech. and Bioeng., 12, 465-482 (1970).

3. Falch, E. A., and E. L. Gaden, "A Continuous Multistage Tower Fermentor: I. Design and Performance Tests," Biotech. and Bioeng., 11, 927-943 (1969).

4. Nelder, J. A., and R. Mead, Computer J., 7, 308 (1965).

5. Bard, Y., "A Function Maximization Method with Application to Parameter Estimation," New York Scientific Centre Report 322-0902, IBM (July 1966).

6. Lee, S. S., M. S. Thesis, Kansas State University (1970).

Chapter 6

RECOMMENDATIONS FOR FUTURE RESEARCH

6.1 INTRODUCTION

In this chapter several suggestions for further research are presented. Although considerable research in many areas still needs to be conducted, only suggestions for research closely related to the topic of this thesis are presented in this chapter.

6.2 CONVERGENCE PROBLEMS

Future work needs to be done on the convergence problems of the nonlinear parameter estimation techniques, resulting from poor initial estimates of parameters. Related to Marquardt's approach, it appears desirable to start with search techniques and then as the optimum of the sum of squares is approached to use linearization methods which are efficient in this region.

6.3 NONLINEAR ESTIMATION WITH SIMULATED DATA

Experimental data are often not available in certain chemical or biochemical systems, hence the information which can be gained from the simulation of experimental data to assist in nonlinear parameter estimation seems to merit further consideration. An attempt was made to demonstrate the applicability of a simplex pattern search technique [1] for estimating parameters in continuous multistage tower fermentation systems using unsteady state simulated data, considering various types of experimental errors. The comparison of this technique with other nonlinear

estimation techniques and application of this technique to other systems should form the basis of future work.

6.4 PARAMETER IDENTIFICATION IN DIFFERENTIAL MODELS

In Chapter 3 the performance of a continuous multistage tower fermentor was analyzed using a flow model. An eight stage tower system was considered. Tracer measurements were available in some of the individual stages. The purpose of this investigation was to estimate the backflow parameters and relationship between them. Two distinct approaches were employed. In the first one, the two coupled differential equations, arising from the unsteady state tracer material balances in the first and second stages, were solved analytically, to reduce the differential model to an algebraic model. The other approach was to use the differential model to estimate these backflow parameters. The results obtained from the algebraic model gave a better fit for the experimental data. This was probably due to the assumptions involved in using a differential model. The tracer concentration measurements were available in individual stages at different values of time, which make the independent variable, time, in the differential model different for each equation, and linear interpolation was used to transform the data to get the tracer concentrations at the same intervals of time. The usual parameter estimation techniques developed for differential models are incapable of handling the situation, where the independent variable is different for each differential equation. However, an attempt should be made, in the future, to develop parameter identification techniques which will handle this kind of situation.

6.5 SIMULTANEOUS MEASUREMENT OF TRACER AND KINETIC DATA

Two distinctly different situations have been considered in this work while identifying the system parameters in multistage tower systems. In the first one flow parameters were estimated using available tracer data and a flow model. Next, flow and kinetic parameters were estimated using an unsteady model with reaction, using simulated data. The information that one obtains from the analysis of flow behavior is often useful in analyzing the model with reaction. If such an experiment is carried out under identical experimental conditions, the flow parameters estimated from the flow model could be substituted as constants; this reduces the dimension of the problem and then only kinetic parameters need to be evaluated. This information becomes increasingly important in case of unavailability of a sufficient number of data points. This problem was encountered while identifying system parameters in the steady state model with reaction. Prokop et. al. [1] took experimental measurements on an eight stage tower system with the possibility of backflow and sedimentation. Measurements were available for steady state cell and substrate concentration in various stages and in the exit. An attempt was made to estimate the backflow parameters (γ and γ') and kinetic parameters (μ_{max}, K_S, and β) using two different constrained optimization techniques (1) modified simplex pattern search [2], and (2) modified SUMT [3], and a nonlinear algebraic model. However, reasonable results could not be obtained because of missing observations and a marginal number of data points. Future work needs to be done on this problem. Computer techniques will have to be developed to solve the nonlinear

algebraic system in a very short time, because all the parameter estimation techniques will require the solution of this system at each iteration.

The flow model of the tower fermentor which may be considered is shown in Figure 1.

6.5a Mathematical model

The mathematical model presented here considers the possibility of backflow and cell sedimentation. The following simplifying assumptions are made

(i) complete mixing in each stage.

(ii) equivalent effective liquid volumes in each stage.

(iii) growth kinetics follows the Monod relationship.

(iv) backflow can be expressed as a fraction of the flow leaving a stage.

The steady state cell and substrate balances in each stage of the tower fermentor can be written as follows:

First Stage

$$\text{Substrate balance:} \quad 0 = F_1' S_1' + F_2 \gamma' S_2 - F_1 S_1 - \frac{\mu_{max} S_1 X_1 V_1}{Y(K_S + S_1)} \tag{1}$$

$$\text{Cell balance:} \quad 0 = F_2 \gamma' X_2 - F_1 \beta X_1 + \frac{\mu_{max} S_1 X_1 V_1}{(K_S + S_1)} \tag{2}$$

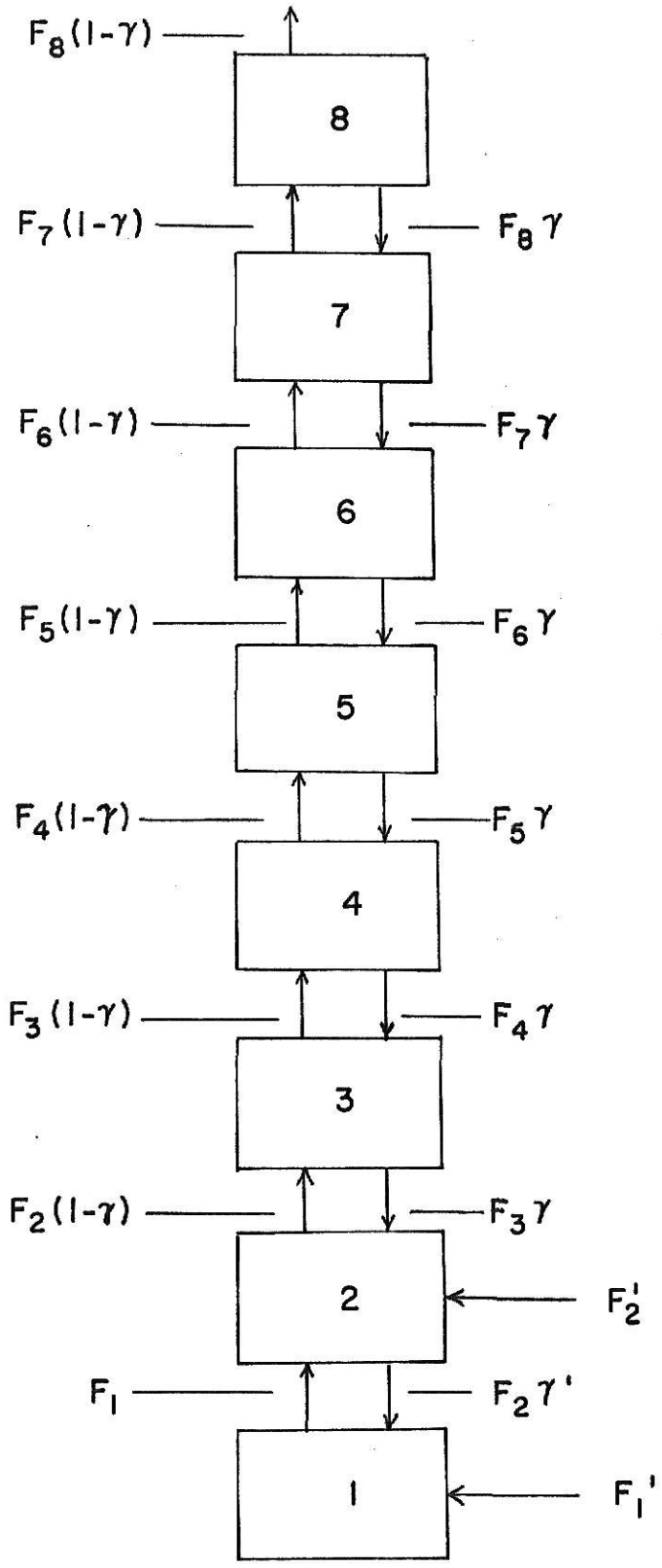

Fig. 1 Flow model of the tower fermentor.

Second Stage

Substrate balance: $$0 = F_2' S_2' + F_3 \gamma S_3 + F_1 S_1 - F_2 \gamma' S_2 - F_2(1-\gamma')S_2 - \frac{\mu_{max} S_2 X_2 V_2}{Y(K_S + S_2)} \quad (3)$$

Cell balance: $$0 = F_1 \beta X_1 + F_3 \gamma X_3 - F_2 \gamma' X_2 - F_2(1-\gamma')\beta X_2 + \frac{\mu_{max} S_2 X_2 V_2}{(K_S + S_2)} \quad (4)$$

Third Stage

Substrate balance: $$0 = F_2 (1-\gamma')S_2 + F_4 \gamma S_4 - F_3 \gamma S_3 - F_3(1-\gamma)S_3 - \frac{\mu_{max} S_3 X_3 V_3}{Y(K_S + S_3)} \quad (5)$$

Cell balance: $$0 = F_2(1-\gamma')X_2 + F_4 \gamma X_4 - F_3 \gamma X_3 - F_3(1-\gamma)\beta X_3 - \frac{\mu_{max} S_3 X_3 V_3}{Y(K_S + S_3)} \quad (6)$$

Stages 4 to 7

Substrate balance:

$$0 = F_{i-1}(1-\gamma)S_{i-1} + F_{i+1}\,\gamma\,S_{i+1} - F_i(1-\gamma)S_i - F_i\,\gamma\,S_i - \frac{\mu_{max}\,S_i\,X_i\,V_i}{Y(K_S + S_i)}\,,\quad i=4,\,5,\,6,\,7 \tag{7}$$

Cell balance:

$$0 = F_{i-1}(1-\gamma)\beta\,X_{i-1} + F_{i+1}\,\gamma\,X_{i+1} - F_i(1-\gamma)\beta\,X_i - F_i\,\gamma\,X_i + \frac{\mu_{max}\,S_i\,X_i\,V_i}{(K_S + S_i)}\,,\quad i=4,\,5,\,6,\,7 \tag{8}$$

Eighth Stage

Substrate balance:

$$0 = F_7(1-\gamma)S_7 - F_8\,\gamma\,S_8 - F_8(1-\gamma)S_8 - \frac{\mu_{max}\,S_8\,X_8\,V_8}{Y(K_S + S_8)} \tag{9}$$

Cell balance:

$$0 = F_7(1-\gamma)\beta\,X_7 - F_8\,\gamma\,X_8 - F_8(1-\gamma)\beta\,X_8 + \frac{\mu_{max}\,S_8\,X_8\,V_8}{(K_S + S_8)} \tag{10}$$

The flow balances in each stage of the tower fermentor are as follows:

Stage 1: $- F_1 + F_2 \gamma' = - F_1'$ (11)

Stage 2: $F_1 - F_2 + F_3 \gamma = - F_2'$ (12)

Stage 3: $F_2(1-\gamma') - F_3 + F_4 \gamma = 0$ (13)

Stage 4 to 7: $F_i(1-\gamma) - F_{i+1} + F_{i+2} \gamma = 0, \quad i=3, 4, 5, 6$ (14)

Stage 8: $F_7(1-\gamma) - F_8 = 0$ (15)

where

S_1' = substrate concentration in the feed to the first stage.

S_2' = substrate concentration in the feed to the second stage.

S_i = substrate concentration in the ith stage.

X_i = cell concentration in the ith stage.

μ_{max} = maximum growth rate.

Y = yield coefficient.

V_i = volume of the ith stage.

β = coefficient for cell sedimentation.

K_S = saturation constant, substrate concentration at which the specific growth rate is one-half the maximum value.

F_i = flow out of ith stage.

F_1', F_2' = feed to the first and second stage, respectively.

γ = backflow coefficient for stages 3 through 8.

γ' = backflow coefficient for stage 2.

However, an alternative approach would be to simultaneously obtain tracer and kinetic data by operating the tower system continuously under prescribed conditions. This may simplify the problem to a great extent from the parameter identification standpoint. It may be possible to obtain tracer data on liquid phase and cell phase, and measure substrate, cell and other concentrations at the same time, for kinetic analysis.

6.6 SELECTION OF THE BEST REGRESSION EQUATION

The basis of selection, of the best regression model for predicting the tracer concentration in one of the individual stages, in Chapter 5, was the examination of residuals obtained from different assumed models. More sophisticated techniques are sometimes required when the choice is to be made among two or more models which are almost equally adequate for predictive purposes, or when one tries to select the variables in regression.

Several statistical techniques have been proposed for accomplishing this which include (1) testing all possible regressions, (2) backward elimination, (3) forward selection, (4) stepwise regression, and (5) stage-wise regression procedures. Testing all possible regressions is rather cumbersome and is quite impossible without high speed computers. The backward elimination method is an improvement on the "all regressions" method in that it attempts to permit the examination not of all regressions

but of only the "best" regression containing a certain number of variables. The backward elimination method begins with the largest regression, using all variables, and subsequently reduces the number of variables in the equation until a decision is reached on the equation to use. The forward selection precedure is an attempt to achieve a similar conclusion working from the other direction, that is, to insert variables in turn until the regression equation is satisfactory. The order of insertion is determined by using a partial correlation coefficient as a measure of the importance of a variable not yet in the equation.

The stepwise regression procedure is an improvement on the forward selection procedure. The improvements involve the re-examination at every stage of the regression of the variables incorporated into the model in previous stages. A variable which may have been the best single variable to enter at an early stage may, at a later stage, be superfluous because of the relationship between it and other variables now in regression. To check on this, the partial F-criterion for each variable in the regression at any stage of calculation is evaluated and compared with a preselected point of the appropriate F-distribution. The stagewise regression procedure does not provide a true least squares solution for the variables included in the final equation. After a regression equation in the x variable most correlated with y has been fitted, the residuals $(y_i-\hat{y}_i)$ are found. These residuals are now considered as response values and regressed against the x (of those which remain) most correlated with this new response. The process is continued to any desired stage. Since at each stage

Response = Fitted Response + (Response - Fitted Response)

the regression equations can be back-substituted stage by stage until the final stagewise equation is attained. A detailed description of all the above mentioned methods for regression analysis can be found in reference [4]. These techniques along with numerical statistical techniques should be applied in the future, to the problem considered in this work and other similar problems, to investigate, if any improvement in the analysis is possible.

6.7 REFERENCES

1. Prokop, A., L. E. Erickson, J. Fernandez, and A. E. Humphrey, "Design and Physical Characteristics of a Multistage, Continuous Tower Fermentor," Biotech. and Bioeng., 11, 945-966 (1969).

2. Nelder, J. A., and R. Mead, Computer J., 7, 308 (1965).

3. Lai, K. C., M.S. Thesis, Kansas State University (1970).

4. Draper, N. R., and H. Smith, "Applied Regression Analysis," John Wiley and Sons, Inc. (1968).

ACKNOWLEDGMENTS

The author wishes to express his sincere gratitude to his major advisor, Dr. L. E. Erickson, for his excellent advice and constant encouragement.

The helpful suggestions and continual interest of Dr. L. T. Fan and Dr. C. L. Hwang are deeply appreciated. Thanks are due to Dr. A. E. Humphrey for making the experimental data available. The author also wishes to thank the staff of the computing center and the secretarial staff of the Chemical Engineering Department for their assistance.

This work was supported by Federal Water Quality Administration project (Project 17090 ELL).

NONLINEAR PARAMETER ESTIMATION IN CONTINUOUS TOWER FERMENTATION SYSTEMS

by

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B. Sc., Banaras Hindu University, 1969

AN ABSTRACT OF MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Chemical Engineering

KANSAS STATE UNIVERSITY

Manhattan, Kansas

1971

This work is concerned with the handling and analysis of experimental data for batch and continuous tower fermentors. Data analysis often plays an important role in research designed to investigate the mechanism and behavior of a process. An analysis of nonlinear parameter estimation techniques for both algebraic and differential models was conducted using both experimental data and simulated data of known error distributions.

Unsteady state, continuous, multistage, tower fermentor performance was analyzed using a four stage tower system. Transient data was simulated using various types of experimental errors including normally distributed error, instrument drift, zero error in the instrument, and various combinations of these. Flow nonidealities like fluid backflow and dispersed (microorganism) phase sedimentation were included in the model.

The second part of this investigation employs tracer data made available by Dr. Humphrey and his associates at the University of Pennsylvania. Experimental measurements of concentration vs. time for the first three stages and the eighth stage of an eight stage tower fermentor with feed to the second stage are employed to determine the extent of backflow in the tower system. Two approaches to the parameter estimation problem are investigated. One method uses the differential model for the eight stage system while the other uses the model for the first two stages together with a polynomial approximation to the tracer response at stage three. Statistical techniques are employed to determine the most adequate and satisfactory regression model for the available tracer data for stage three.