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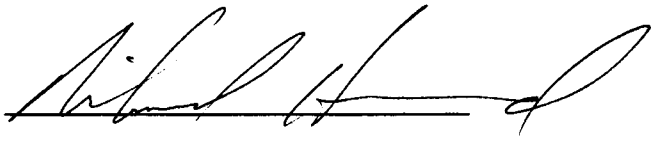
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CALIBRATION OF THE SURVIVAL OF SALMONELLA BACTERIUM TO RADIATION USING BAYESIAN PROCEDURES

A Thesis

Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Michael Edward Howard

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ABSTRACT

Calibration of food irradiation (i.e. foodborne bacterial pathogen control) uses data from a calibration experiment. This data consists of pairs (x_i, y_i) where x_i are the doses (kGy) at which the chicken meat is irradiated and y_i are the corresponding bacteria populations (log CFU/g), where CFU stands for colony forming units. In order to describe the relationship between dose and population reduction the Student t-distribution is used to describe the predictive and calibrative densities:

By using Bayes theorem and incorporating a uniform prior distribution, the expression for the posterior density is obtained. Integrating the product of the likelihood of a new observation and the posterior density yields the predictive density. The predictive density acts like a new likelihood in the subsequent derivation of the calibrative density.

By incorporating a uniform prior distribution, the relationship between the calibrative and predictive density is simplified. The calibrative density describes the uncertainty of a new unknown dose x_n given a specific bacteria population y_n and the calibration data set $\underline{z} = (x_i, y_i)$. This type of analysis provides a quantitative description of the uncertainty about the dose. In addition, this approach allows factors such as temperature and atmosphere to be analyzed qualitatively. The effect of these factors on the resistance of Salmonella to radiation is examined. The Bayesian approach is used in the analysis of two research experiments.

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CHAPTER 1

INTRODUCTION

1.0 Background

Microorganisms such as Salmonella cause thousands of people to become ill each year in the United States [Urbain, 1986]. Even with the United States Department of Agriculture (USDA) meat inspections, this bacterium cannot be detected with methods presently used. Estimates suggest that as many as 35% of poultry carcasses contain the Salmonella bacterium [IAEA, 1978]. With this level of contamination other methods should be employed in order to reduce the risk of food related illness. One such promising method is food irradiation with gamma rays. Opponents of food irradiation often argue that with proper handling and thorough cooking the potential for illness is greatly reduced. However, it is widely known that poultry is frequently undercooked and mishandled.

The process of irradiating food with gamma radiation has been in practice for more than 30 years by the U.S. military and other countries. In May of 1990, the Food and Drug Administration (FDA) granted federal approval for the irradiation of poultry. The process of food irradiation is simple by design. Food (i.e. chicken, beef or fruit) is placed in a radiation field for a given period of time to control the amount of radiation the food is exposed to. The radiation source is either Cesium-137 or Cobalt-60, both of which are gamma emitters. As the gamma rays pass through the food they interact with the bacteria present. The level of bacterium reduction is dependent upon the time during which the food is in the radiation field, the activity of the radiation source, presence of oxygen and temperature during irradiation [ICRPF, 1978].

Currently, the federal guidelines for poultry irradiation permit doses from 1.5 to 3.0 kGy (1 kGy = 100 krad). It is important to note that doses in this range do not sterilize the

food but rather reduce the bacterial count to significantly lower levels. Also, as with chemical processes, free radicals are produced. However, repeated studies have shown these free radicals to be no significant health threat to the consumer [Urbain, 1986].

The objective of this thesis is to use Bayesian methods [Aitchison, 1975] to estimate the dose and its uncertainty associated with a specified bacteria population reduction. The influence of factors such as temperature and atmosphere are analyzed along with dose in order to determine the optimal value of each factor. A calibrative density is derived for the various data sets and subsequently plotted to "show" the probabilistically weighted range of doses associated with a desired population reduction. Also, the corresponding predictive density is derived and it conversely shows the corresponding range in population reduction with a fixed dose. Both the calibrative and predictive distributions are numerically calculated using computer programs developed with the software package Mathematica® [Wolfram, 1992].

1.1 The Bayesian Approach

Below we describe briefly the basic framework of Bayesian calibration and list reasons why a Bayesian approach is taken. First, by utilizing an informative or calibration experiment, it is possible to infer the result of a future experiment by relating a specified value for a variable to information gathered in the informative experiment. Hence, the name calibration experiment. The predictive and calibrative densities derived "show" the estimated range of values for a fixed dose and bacterium population respectively. The mode of these densities is the most probable dose or population size. In addition these densities give a quantitative description of the uncertainty about the dose or population size. Second, Bayesian statistics incorporates a prior distribution into the calibrative density. This prior distribution can reduce the uncertainty by incorporating information other than

the data used in the likelihood. This information for instance can be gathered from independent experiments and separate studies.

1.2 Thesis Outline

This thesis is organized into five different chapters. The first section of chapter 1 is intended to familiarize the reader with the motivation behind food irradiation. The next section explains the reasons for taking a Bayesian statistical approach to solve the problem and gives the reader a basic understanding of the Bayesian viewpoint. The final part of chapter 1 is a broad outline of the contents of the rest of the thesis.

Chapter 2 discusses the data. This discussion gives background information about the research data used for this thesis and an explanation why certain data sets are chosen. Experiments performed by Thayer and Boyd of the USDA are the primary studies used [Thayer and Boyd, 1991]. The assumptions for this analysis along with the experimental procedures used by Thayer and Boyd are discussed in detail.

In chapter 3 we derive the calibrative and predictive densities respectively. The relationship between the predictive and the calibrative density is explained. This chapter gives the reader a more detailed description of the underlying principles of Bayesian statistical analysis. Also discussed in chapter 3 is the Student t-distribution and all of the parameters associated with this distribution.

Chapter 4 discusses the results of the analysis. Included are the methods with which the parameters of the Student t-distribution are determined. The predictive and calibrative densities are shown for the corresponding data sets. Differences of the response with variations in temperature, atmosphere and dose are addressed. Chapter 5 contains conclusions and suggestions for future work. Finally, the appendices include relevant data sets and calculations along with computer programs written in Mathematica®.

CHAPTER 2

DATA AND EXPERIMENTAL PROCEDURES

2.0 Description of the Data

Two studies are used for this thesis. Both studies were performed by Thayer and Boyd for the Food Safety Research Unit of the USDA [Thayer and Boyd, 1991]. In order to differentiate between studies, the USDA assigns each study a four digit code. When referencing each study this code is given. The first study [5564, 1991] analyzed mechanically deboned chicken meat that is sterilized prior to bacterial inoculation. The sterilization process insures that competing microflora will not influence the determination of Salmonella typhimurium growth and response to radiation. The chicken meat received a dose of 42 kGy of gamma radiation for sterilization prior to the experimental procedure.

Three variables were used in the procedure. The first variable, temperature, is varied between -20 and +20 °C. The temperature is kept constant during the irradiation process. This is necessary in order to determine the effect that a certain temperature will have on the reduction of a bacterial population.

The second variable is atmosphere. The experiment is replicated with the chicken meat irradiated in the presence of oxygen and in the absence of oxygen. Both of these situations are analyzed here.

The third variable is dose. The dose ranges from 0 kGy to 3.6 kGy for all experimental setups.

The second study [5588, 1991], refers also to mechanically deboned chicken meat. The primary difference between the studies is that for the second study the meat is not sterilized prior to inoculation. This experiment is performed to determine if prior sterilization has an observable affect on the sensitivity of Salmonella typhimurium to

radiation. This is determined by the reduction in bacteria populations with respect to dose. The results of the study indicate no such effect. Thus, the results from 5588 are assumed to be equivalent to 5564. All aforementioned variables and ranges are again used in the performance of study 5588.

2.1 Evaluation of the Data

One assumption used for this analysis regards the replication of the experiments from both studies. In the first study (5564), each experiment is replicated three times. The average of the three replicates is taken. This average is reported. Along with this mean the standard deviation is also given. This standard deviation is calculated using the three data points that make up the mean. Since the "raw" data could not be obtained, the mean values were used. In order to simplify the computations, the variance is assumed to be constant. This is a reasonable assumption since the estimates of the variances are not significantly different.

2.2 Quantification of the Reduction of the Bacterium Population

There are two methods of quantifying bacteria reduction in food irradiation. These methods are referred to as the D-value and reduction of a bacterium population to a specified level. These two methods are not equivalent. The D-value estimates the reduction in a bacterium population per kGy [Hermann, 1970]. Death is defined as the inability of the cell to reproduce. The reduction of a bacterium population to a specified level is defined by setting a numerical limit. This limit represents the maximum number of bacterium present in a food item post irradiation.

2.2.1 The D-value

In this thesis the D-value is not estimated. However, the method for obtaining the D-value needs to be understood in order to determine a parameter in the Student t-distribution. An experimental data set consisting of dose and bacteria populations values is used in determining the D-value. It is important to note that the size of the bacteria populations is expressed in the units of log CFU/g. The abbreviation CFU stands for colony forming units [Doyle, 1989]. The populations are expressed in log form due to the large range in the number of CFU of bacteria present. The dose is the independent variable and the log population is the dependent variable. If a linear relationship between the two variables is assumed the graph of log CFU/g vs. dose results in a straight line. See Figure 2.1. The straight line is described by the following equation:

$$y = mx + b \tag{2.1}$$

where m is the slope of the line and b is the y-intercept. The negative reciprocal of the slope yields the D-value:

$$D = -\frac{1}{m} \tag{2.2}$$

The parameters m and b are used in the Student t-distribution. Figure 2.1 shows a typical plot for determination of the D-value. A linear function is fitted to the data points. The slope is the negative reciprocal of the D-value. Alternatively, the negative reciprocal of the slope is the D-value. It is given in units of kGy^{-1}. The D-value is defined as reduction in bacterium population per kGy.

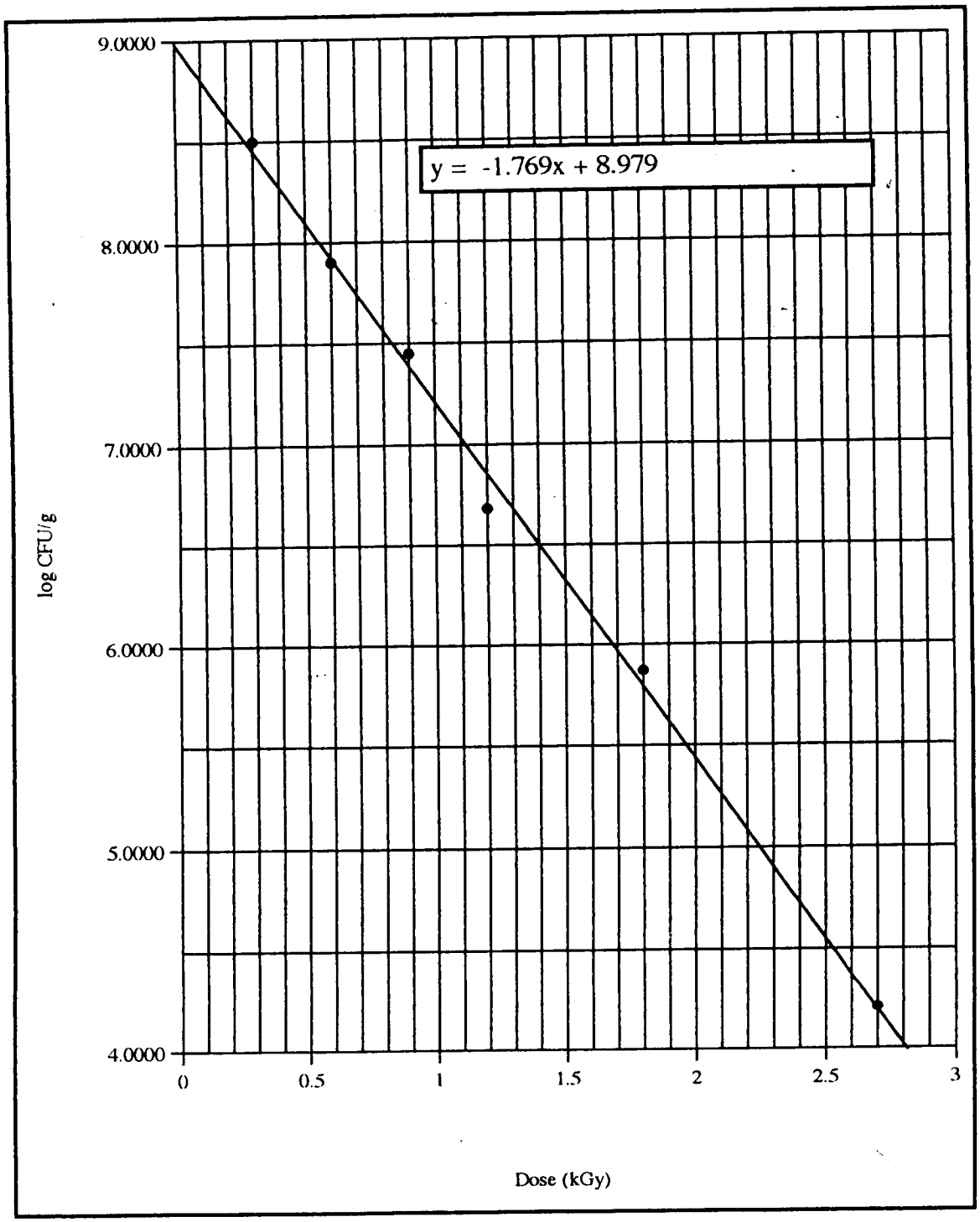


Figure 2.1 Example of scatter plot for a sample data set

2.2.2 Reduction of a Bacterium Population to a Specified Level

The other method of quantifying population reduction is determining the number of CFUs of surviving bacteria after a given dose. Federal standards and guidelines will use this method in the future. However, there are two key problems with this method. First, since Salmonella will continue to grow after irradiation, proper handling is vital in order to keep these populations to acceptable levels. Secondly, determining safe levels of Salmonella contamination is controversial. However, by estimating a numerical limit it is possible to set guidelines for the amount of dose in order to achieve acceptable levels of bacteria populations. We later show calibrative densities that give the range of dose associated with a specific population reduction. As in Figure 2.1 a plot of bacterium population vs. dose shows a straight line fit.

CHAPTER 3

BAYESIAN METHODS FOR CALIBRATION

3.0 Bayesian Calibration

Calibration is defined as the process where the scale of a measuring instrument is determined using an experiment in which an agent induces a response from the measuring instrument. The calibration experiment usually consists of several trials. These trials measure the response of the instrument to a known substance. An example of such an experiment consists of calibrating the response of a pressure gauge to various pressures. If the gauge is not scaled then measuring the response of the gauge at separate pressures will scale the gauge. It is assumed however that each pressure response is indicative of only one reading on the gauge. The problem with this assumption is that error is inherently built in to such processes. In order to account for such variations a calibrative density is needed. This density shows the properly weighted range of doses associated with a given response and characterizes the uncertainty about the dose.

There are two distinct types of calibration experiments. They are referred to as the controlled and natural calibration experiments [Aitchison, 1975]. In the natural calibration experiment, the independent variable provides information about future values of the independent variable. For example, assume that there are two instruments with one being more accurate than the other. The response of each instrument is representative of the independent variable. Since the independent variable is naturally occurring the experimenter has no control over it. Thus, assuming that any future responses arise in the same manner as previous responses then the calibration experiment will provide some type of indication about the pattern. The response of the accurate instrument can be compared to the response of the less accurate instrument.

In a controlled calibration experiment the values of the independent variable are fixed and give no information on future values of the independent variable. For example, instead of using two instruments to measure the response, only one instrument is used. The independent variable has pre-selected values. Future responses of the independent variable are not dependent on previous selected values. The techniques used in food irradiation take the form of a controlled calibration experiment.

3.1 The Predictive Density

At first a definition of the predictive density is given. The basic premise of the predictive density is given a specific x (independent variable), what can be said about a future y (dependent variable). This prediction is arrived at by multiplying the likelihood for a future y by the posterior density and integrating. The predictive density can be described by the following equation.

$$p(y|x) = \int_{\Theta} p(y|\theta)p(\theta|x)d\theta \tag{3.1}$$

The variable θ represents the unknown index on the indexing set Θ which is the statistical basis for the prediction problem. There are several steps in deriving the predictive density. First, the likelihood function is multiplied by the prior distribution. Bayes theorem is used in this process. The following equation describes how the posterior density in the integral of equation 3.1 is obtained.

$$p(\theta|x) = \frac{p(\theta)p(x|\theta)}{p(x)} \tag{3.2}$$

In equation 3.2 the term p(x) represents the probability of a given x. This term can be represented in a different form as follows:

$$p(x) = \int_{\theta} p(x|\theta)p(\theta)d\theta \tag{3.3}$$

The substitution of equations 3.2 and 3.3 into equation 3.1 yields the predictive density as described below [Aitchison, 1975].

$$p(y|x) = \frac{\int_{\theta} p(y|\theta)p(x|\theta)p(\theta)d\theta}{\int_{\theta} p(x|\theta)p(\theta)d\theta} \tag{3.4}$$

The data can be described by the variables x and y. Specifically, x represents the doses (kGy) used in the informative experiment. The variable y represents the surviving bacteria populations (log CFU/g) in the informative experiment. In order to simplify this notation, the variables x and y can be written as $\underline{z}$. The following equations show this method.

$$y = (y_1, \ldots, y_n): \; x = (x_1, \ldots, x_n) \tag{3.5}$$
$$\underline{z} = (x, y)$$

By incorporating the variables for the regression model in equation 3.5, the predictive density can be described in the following manner.

$$p(y|x, \underline{z}) = \int_{\theta} p(y|x, \theta)p(\theta|\underline{z})d\theta \tag{3.6}$$

For the regression model, the predictive density takes the form of the Student t-distribution. Section 3.2 defines the Student t-distribution in more detail along with methods used in obtaining the parameters. In equation 3.7 it is important to note that y is the dependent variable defined by the number of bacterium (log CFU/g).

$$St(k, b, c) = \frac{1}{B\left(\frac{1}{2}, \frac{1}{2}k\right)(kc)^{\frac{1}{2}}\left\{1 + (kc)^{-1}(y-b)^2\right\}^{\frac{k+1}{2}}} \tag{3.7}$$

3.2 The Student t-distribution

The Student t-distribution has three parameters: k, b and c. A definition of each is given below.

The first parameter k is simply the number of observations minus two. In this case the independent variable is dose (kGy). With the number of observations equal to n we have;

$$k = n - 2 \tag{3.8}$$

The next parameter is b. The parater b can best be defined by a linear equation as shown in equation 3.9. Then, the coefficient of regression and the y-intercept can be calculated using the data from each study [Ott, 1988]. This method is often referred to as least squares estimate. The equations used in estimating the regression coefficient are:

$$y = \beta_0 + \beta_1 u \tag{3.9}$$

$$\hat{\beta}_1 = \frac{S_{xy}}{S_{xx}} \tag{3.10}$$

$$S_{xx} = \sum_{i=1}^{n} \left(x_i - \bar{x}\right)^2 \tag{3.11}$$

$$S_{xy} = \sum_{i=1}^{n} \left(x_i - \bar{x}\right)\left(y_i - \bar{y}\right) \tag{3.12}$$

The variable u in equation 3.9 represents the unknown dose (kGy). In equations 3.11 and 3.12 the variable x_i represents the ith radiation dose (kGy) and $\bar{x}$ represents the mean radiation dose (kGy). The variable y_i represents the ith bacterium population (log CFU/g) and $\bar{y}$ represents the mean bacterium population (log CFU/g). The method used in determining the y-intercept is dependent upon solving equation 3.10. This can be seen in the following equation.

$$\hat{\beta}_0 = \bar{y} - \hat{\beta}_1 \bar{x} \tag{3.13}$$

The final parameter of the Student t-distribution is c. This parameter is calculated from the following equation.

$$c = \left(1 + \frac{1}{h}\right)\frac{\upsilon}{\nu} \tag{3.14}$$

The variable h in equation 3.14 is described below. Equation (3.15) represents the reciprocal of h and consequently can be directly substituted into equation 3.14.

$$\frac{1}{h} = \frac{1}{n} + \frac{\left(u - \bar{x}\right)^2}{S_{xx}} \tag{3.15}$$

The variable u represents the dose (kGy). The variable υ is defined as the residual sum of squares [Ott, 1988]. The mathematical formula defining υ is as follows:

$$v = \sum_{i=1}^{n} \left(y_i - \bar{y} - \hat{\beta}_1 \left(x_i - \bar{x} \right) \right)^2 \tag{3.16}$$

The variable y in equation 3.16 is the dependent variable bacterium population (log CFU/g) and the variable x is dose (kGy). The remaining variable ν is given by equation 3.17.

$$\nu = n - 2 \tag{3.17}$$

The next section of chapter 3 deals with the prior distribution. In the subsequent section the calibrative density is analyzed. The Student t-distribution is used for the predictive and the calibrative density. However, the variables that are fixed differ. The predictive density specifies a fixed dose (kGy). The calibrative density specifies a fixed bacterium population (log CFU/g). The similarities and differences are discussed.

3.3 The Prior Distribution

The next step in determining the calibrative density is assessing a plausible prior distribution. There are two approaches analyzed, theses include combining data sets and uniform priors.

Two separate research studies are used to determine the response of Salmonella typhimurium to varying doses of radiation. Combining data sets from independent studies will theoretically lead to better results (i.e. sharper peak of the calibration density). However, there is a fundamental problem with this premise. The problem is that in order for the peak to become sharper, the model must support both data sets. If this is not the case, the distribution will widen and thus increase the uncertainty.

The second approach is to use a uniform prior distribution. This method is easy to use but ignores information that might be available. If the prior information is not

significant, a constant prior distribution is justified. The derivation of the relationship between the predictive and calibrative density is simplified. If a uniform prior is chosen, the predictive and calibrative densities are in effect given by the same mathematical formula and only the independent variables in the Student t-distribution are different. This is explained in more detail in section 3.4.

3.4 The Calibrative Density

In section 3.1 the predictive density was derived. It is a Student t-distribution. The same mathematical equation also describes the calibrative density. In order to obtain the calibrative density, the predictive density is multiplied by the prior distribution p(u):

$$p(u|y,\underline{z}) \propto p(u)p(y|u,\underline{z}) \tag{3.18}$$

If a uniform prior distribution [p(u) = constant] is used in equation 3.18, the calculation of the calibrative density is simplified. The key difference between the predictive and calibrative densities is the independent variable of interest. The variable u, dose (kGy) is fixed in the predictive density and conversely, the variable y, bacterium population (log CFU/g) is fixed in the calibrative density. The Student t-distribution is given by:

$$St(k,b,c) = \frac{1}{B\left(\frac{1}{2},\frac{1}{2}k\right)(kc)^{\frac{1}{2}}\left\{1+(kc)^{-1}(y-b)^2\right\}^{\frac{k+1}{2}}} \tag{3.19}$$

Equation 3.19 contains three parameters. Only k is a constant. The parameters b and c are functions of u. The variable u represents the dose in units of kGy. Below we use a numerical example in order to more clearly illustrate the difference between the

calibrative and the predictive density. The following equations are typical for the relationships between the parameters in equation 3.19.

$$k = 4 \tag{3.20}$$

$$b = 3.257 - 1.127u \tag{3.21}$$

$$c = 0.0428 + 0.008739(u - 2.3)^2 \tag{3.22}$$

To obtain the predictive density the dose is fixed. The chosen dose value is substituted into equations 3.21 - 3.22. Thus, the parameters in equation 3.19 become numerical constants. The y in equation 3.19 represents bacterium population (log CFU/g). By plotting equation 3.19 with population of surviving bacteria on the abscissa, the predictive density is shown.

In order to obtain the calibrative density the bacterium population (log CFU/g) is fixed and the dose (kGy) varies. When a specific bacterium population is chosen it is then substituted into equation 3.19 in place of y. Equations 3.20 - 3.22 are then subsequently substituted into equation 3.19 with u representing the variable dose (kGy).

CHAPTER 4

DATA RESULTS AND ANALYSIS

4.0 Background

Two studies are analyzed [Thayer and Boyd, 1991]. Each study contains six separate data sets. They are defined by the values of variables. These variables include temperature, dose and atmosphere. The three corresponding temperatures are: 20, 0 and (-)20 °C. At each temperature the irradiation is performed in air and in vacuum.

In chapter 4 the predictive and calibrative densities are calculated for study (5564). The analysis of the second study (5588) is shown in appendix II. The predictive density is calculated for a fixed dose of 1.5 kGy. The calibrative density is analyzed at two bacterium populations of 100 CFU/g and 1 CFU/g respectively. Transforming these populations into the units of log CFU/g yields 2 log CFU/g and 0 log CFU/g respectively.

4.1 Calculation of the Student t-distribution Parameters

The Student t-distribution contains three parameters as described in section 3.2. These parameters are calculated for each data set prior to analysis. In order to demonstrate the calculations used, the parameters are calculated in detail using one of the data sets in study 5564. Calculation of the parameters for all other data sets is done by the same method. Table 4.1 gives the data for mechanically deboned chicken meat irradiated at 20 °C and in the presence of oxygen.

Table 4.1 Data set for chicken meat irradiated at 20 °C and in oxygen

Dose (kGy)	Population (log CFU/g)
0.30	8.12
0.60	7.66
0.90	7.09
1.20	6.40
1.80	5.14
2.70	3.63

The first parameter in the Student t-distribution is *k*. As previously stated in equation 3.8, this is represented by the number of observations minus two. There are six doses and corresponding bacteria populations and hence k is equal to four.

The second parameter is *b*. This parameter is described in chapter 3 by equations 3.10 through 3.13. Equations 3.11 and 3.12 represent the sum of squares for the independent variable dose (kGy) and the dependent variable bacterium population (logCFU/g) respectively. In order to calculate equations 3.11 and 3.12, the mean dose and mean bacterium population are estimated. These values are computed to be 1.25 kGy and 6.34 CFU/g respectively. The respective values calculated using equations 3.10 through 3.13 are given in Table 4.2.

Table 4.2 Numerical solutions for the variables in parameter *b* of the Student t-distribution

Variable	Numerical Value
β_1	-1.92
S_{yy}	14.3
S_{xy}	-7.40
β_0	8.74

The values in table 4.2 are simple computations obtained by substitutions of the data in table 4.1 into the respective equations 3.10 through 3.13. The two betas in table 4.2 represent the slope (β_1) and y-intercept (β_0) respectively. These estimates determine a regression line for a scatter plot of data. The dose is plotted on the x-axis and the bacterium population is plotted on the y-axis. Both methods give equivalent results. In order to obtain the final form for the Student t-distribution, the coefficients in table 4.2 are put into equation 3.9. In the following equation the variable u represents dose (kGy).

$$y = -1.92u + 8.74 \tag{4.1}$$

The final parameter in the Student t-distribution is c. Equations 3.14 through 3.17 are used in computing the numerical value of c. In calculating c, dose (kGy) is the independent variable x and bacterium population (log CFU/g) is the dependent variable y. The computed values of all three variables in equation 3.14 are shown in Table 4.3:

Table 4.3 Numerical values for the parameter c in the Student t-distribution

Variable	Numerical Value
1/k	$0.167 + (u-1.25)^2/ 14.3$
S_{xx}	3.85
v	0.0397
ν	4

The numerical value of 1/k contains an additional variable u. As previously stated this variable represents dose (kGy). Combining the variables in table 4.3 and substituting them into equation 3.14 yields the numerical value of c.

$$c = 0.0116 + 0.00257(u - 1.25)^2 \tag{4.2}$$

By combining all three parameters and substituting them into the Student t-distribution, the following form of equation 3.7 is obtained.

$$St\left(4, -1.92u + 8.74, 0.0116 + 0.00257\{u - 1.25\}^2\right) \qquad (4.3)$$

Section 4.2 analyzes the predictive and calibrative densities for mechanically deboned chicken meat at 20 °C using equation 4.3.

4.2 Analysis of Irradiated Chicken Meat at 20 °C

There are two data sets analyzed below for the 20 °C temperature experiment. One experiment was conducted in the presence of oxygen and the other in the absence of oxygen (i.e. vacuum). Table 4.4 lists the data used in the analysis.

Table 4.4 Data set for chicken meat irradiated at 20 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.22	9.10
0.30	8.12	8.93
0.60	7.66	8.69
0.90	7.09	7.97
1.20	6.40	7.38
1.80	5.14	6.11
2.70	3.63	4.57

There are several assumptions concerning the data sets analyzed for the 20 °C temperature setting. The first assumption concerns the number of data points used in the

analysis. In order to reduce shouldering, the initial data point is not used. The initial data point is defined as the bacterium population (log CFU/g) at a dose of 0 kGy. There are two motivations for this assumption. First, we are only interested in the linear portion of the scatter plot. Secondly, this assumption is consistent with the previous analysis [Thayer and Boyd, 1991]. The second assumption for this data set is that the population values in table 4.4 are averages of populations of repeated experiments. Since the raw data was unobtainable, these means are treated as actual measurements of bacterium populations. Figures 4.1 and 4.2 show the corresponding plots of bacteria populations (log CFU/g) versus dose (kGy) with the difference being in the presence or absence of oxygen.

4.2.1 The Predictive Density

The predictive density is analyzed for a fixed dose of 1.5 kGy. This dose was chosen for two reasons. The first reason is that it represents a dose that is well within the range of doses in the experiment. Secondly, it is the lowest approved dose level for the irradiation of chicken [FSIS, 1992]. Figure 4.3 represents the predictive density for the chicken meat irradiated in the presence of oxygen. The mode of the predictive density is 5.97 log CFU/g. This represents a 64.7% survival fraction of bacterium. In figure 4.4, the predictive density for irradiated chicken in vacuum is shown. The mode of this function occurs at 6.75 log CFU/g. This indicates that 74.2% of the bacterium survived this dose. Thus, initially it can be inferred that the presence of oxygen leads to greater reduction in surviving bacterium. It is also important to note that the width of the densities is different. This can be explained by the difference in the linear regression fit of the data in the respective scatter plots. It is important to note that the slopes of the regressions lines are not as expected. Generally, the slope of the vacuum data is not as steep as the air data slope [Bacq, 1961].

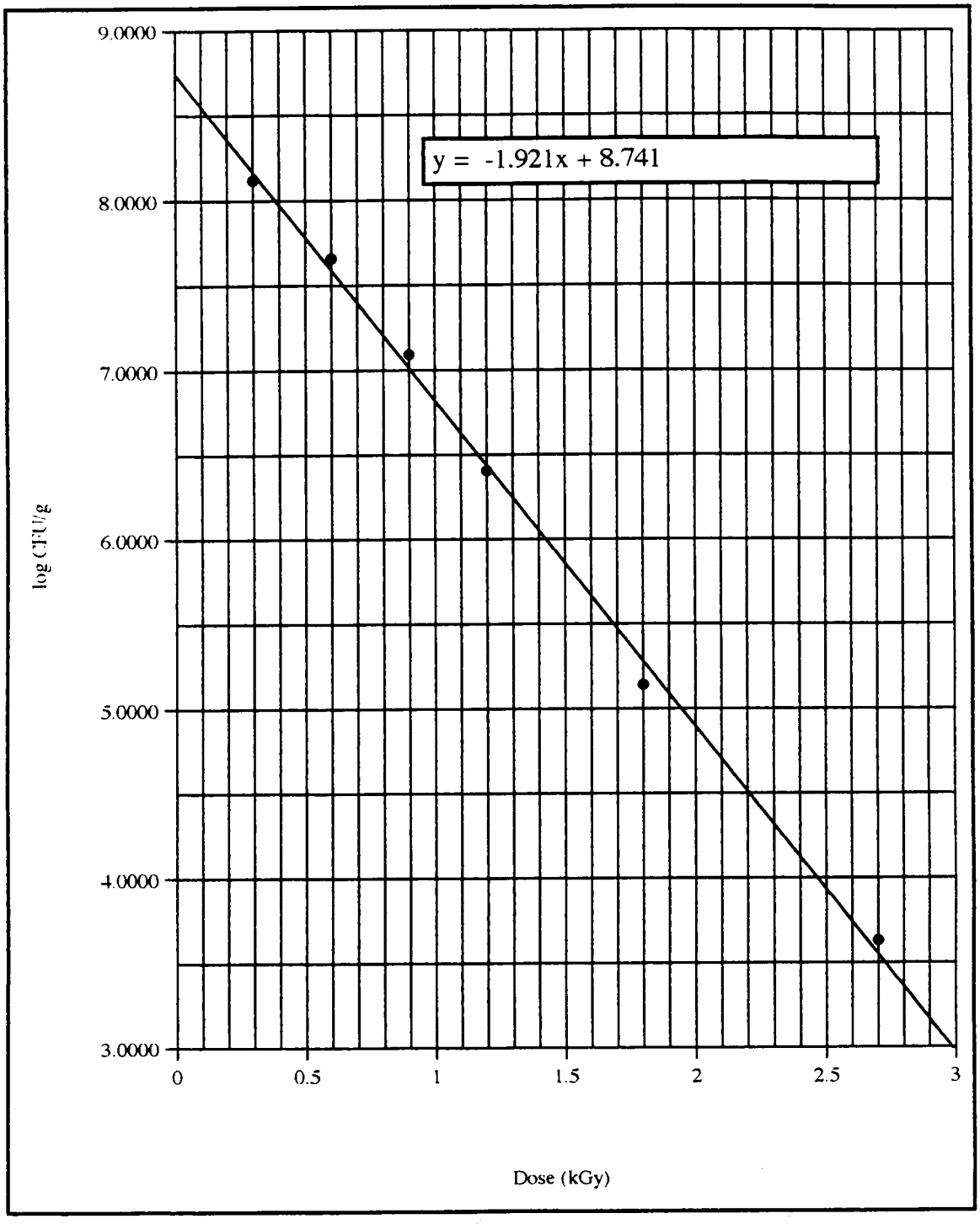


Figure 4.1 Scatter plot for mechanically deboned chicken meat irradiated at 20 °C and in the presence of air

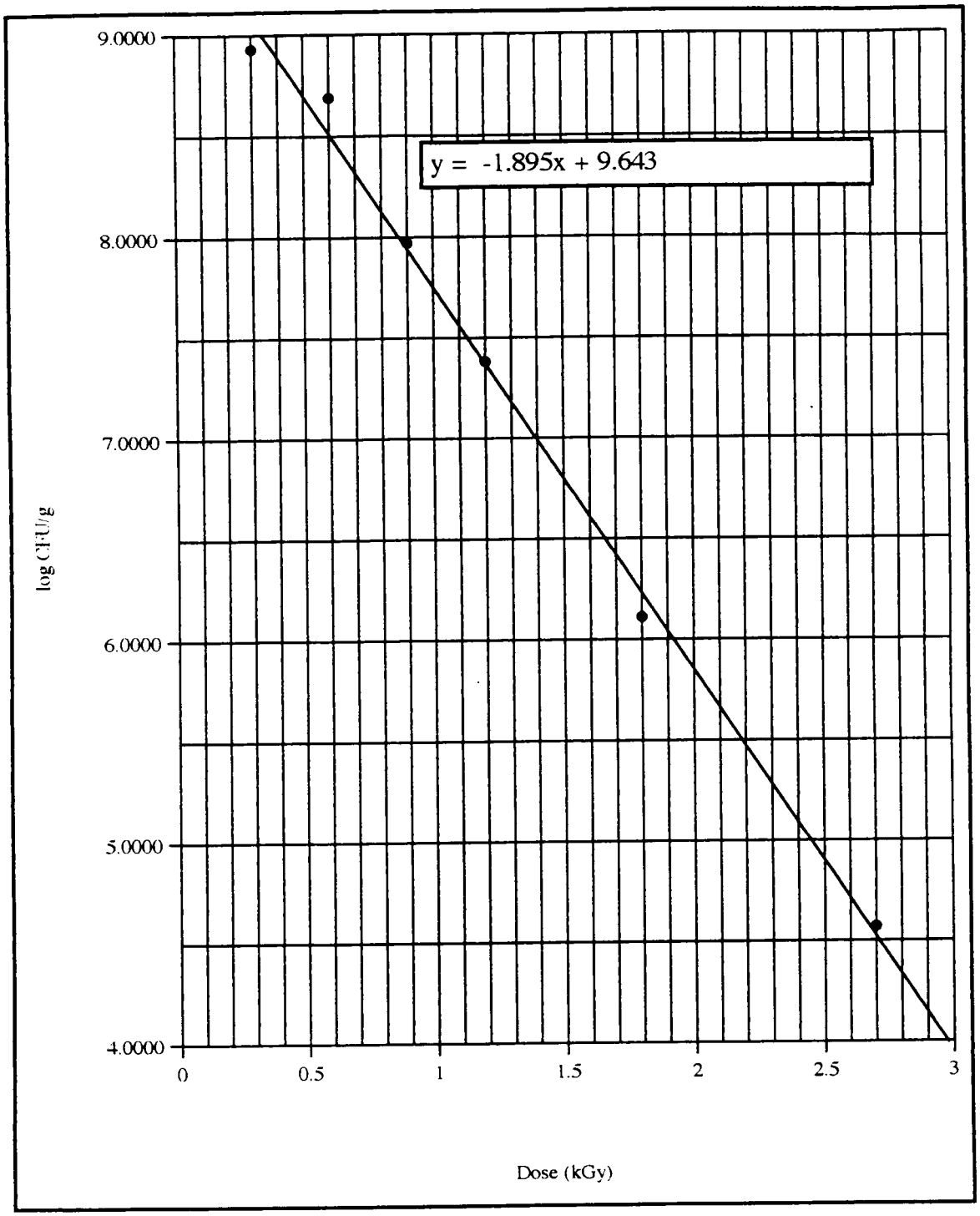


Figure 4.2 Scatter plot for mechanically deboned chicken meat irradiated at 20 °C and in the absence of air

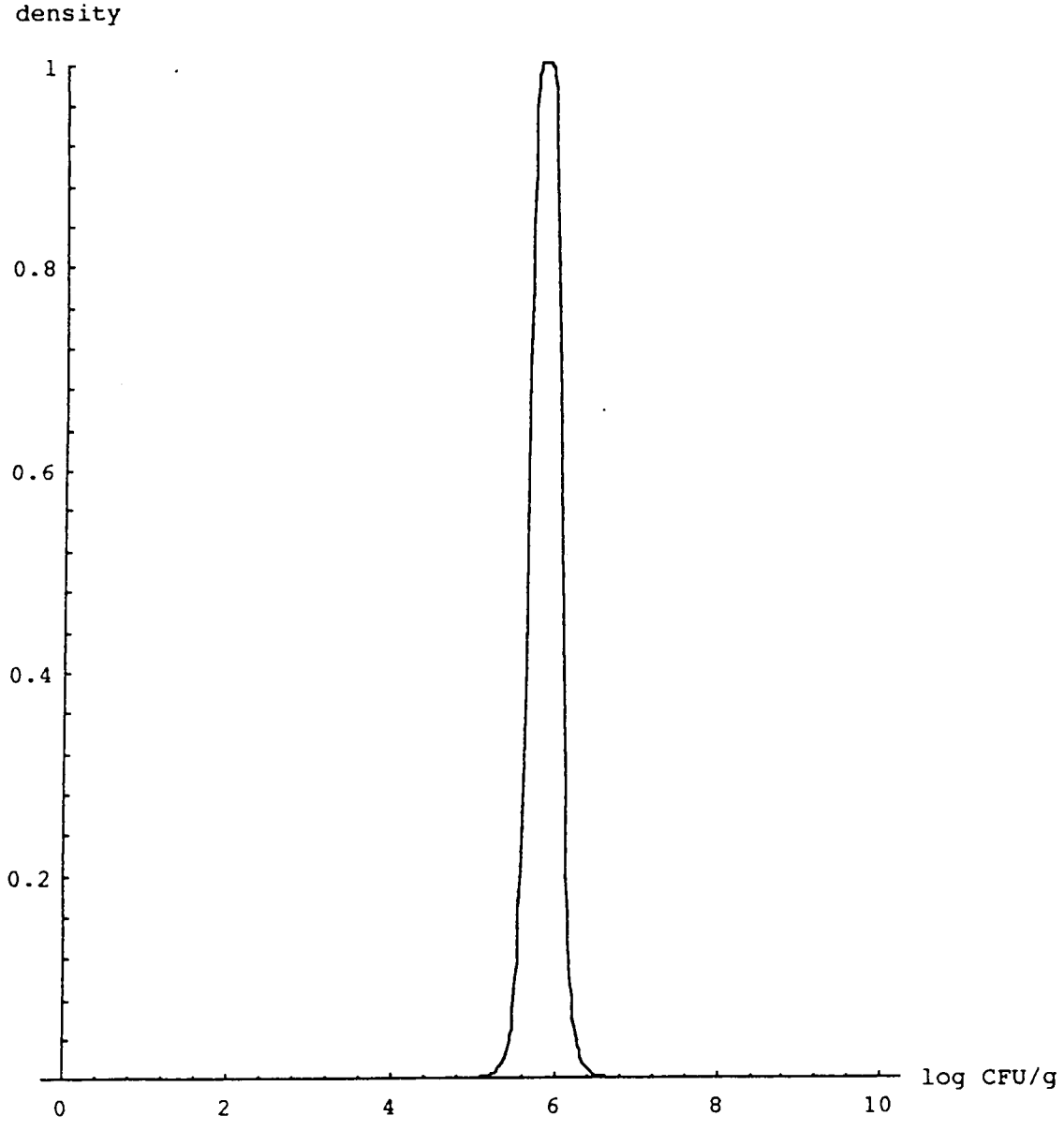


Figure 4.3 The predictive density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed dose of 1.5 kGy

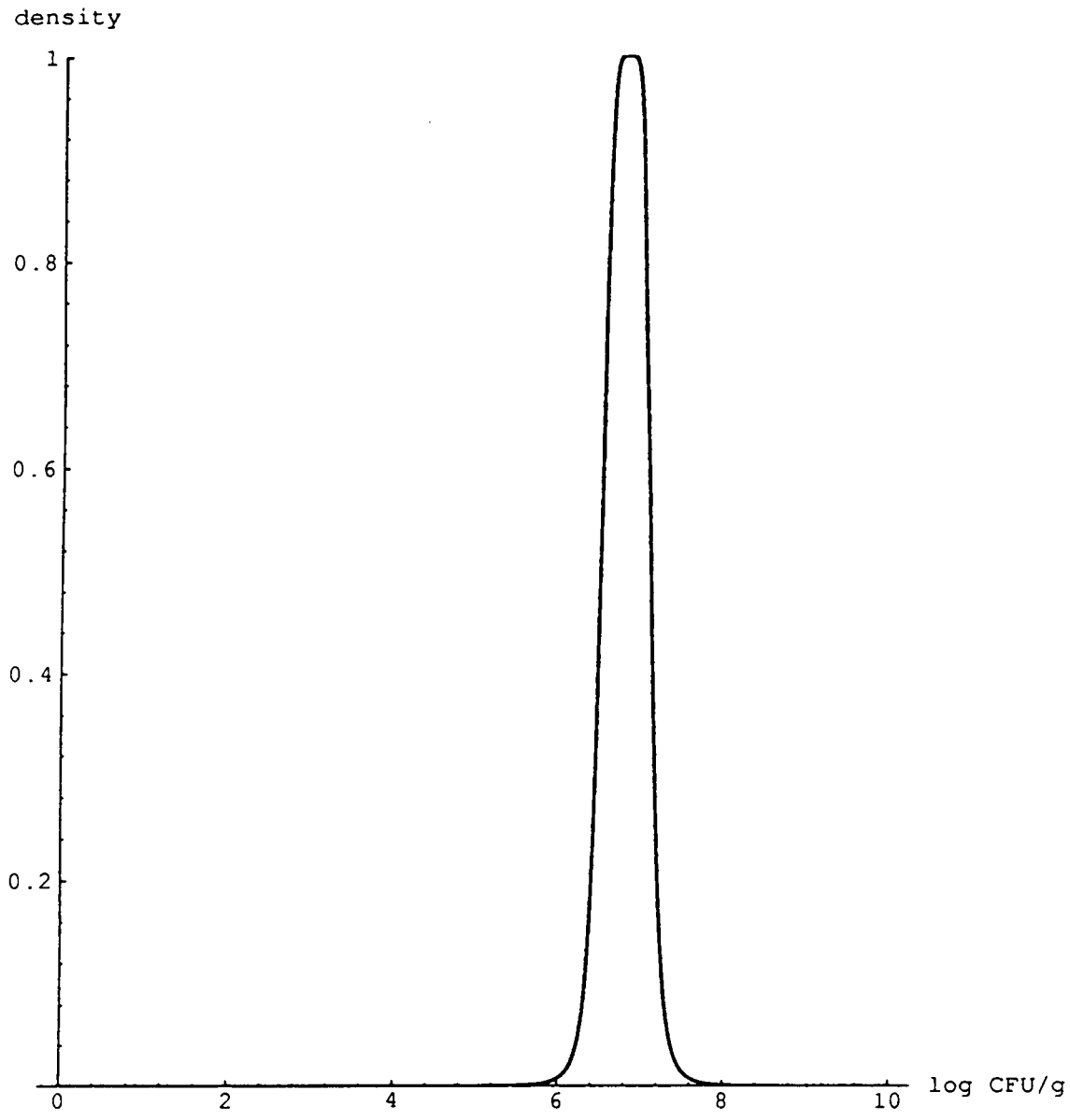


Figure 4.4 The predictive density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed dose of 1.5 kGy

4.2.2 The Calibrative Density

The calibrative density is calculated for two different bacterium populations. These populations are 100 CFU/g and 1 CFU/g respectively. Theses values were chosen for one main reason. These bacterium populations are within the data range for the 100 CFU/g limit and outside the range for the 1 CFU/g limit. This gives us the chance to see if the uncertainty substantially changes with respect to the data range. Figures 4.5 and 4.6 show the calibrative density for chicken meat irradiated in the presence of oxygen. Figure 4.5 implies that a dose of 3.75 kGy is the most likely to reduce the bacterium population to 100 CFU/g. However, in figure 4.6 the calibrative density implies that a dose of 4.25 kGy is needed in order to reduce the bacterium population to 1 CFU/g. These results are expected but it is important to note that the density widens slightly when the density is analyzed for data outside the range.

Figures 4.7 and 4.8 show the calibrative density for chicken meat irradiated without the presence of oxygen. The calibrative density in figure 4.7 estimates that a dose of 3.97 kGy is required to achieve 100 CFU/g. Figure 4.8 estimates that a dose of 5.0 kGy is needed to reduce the bacterium population to 1 CFU/g. In both cases, the dose needed to reduce the bacterium population to specific levels is higher in the case of the vacuum data set. Also, the vacuum data set started with less bacteria, 9.10 log CFU/g as compared to 9.22 log CFU/g for the air data set. This supports the idea that the presence of oxygen increases the amount of reduction in a bacterium population for the same fixed dose.

4.3 Analysis of Irradiated Chicken Meat at 0 °C

As with the 20 °C temperature data sets, there are two data sets analyzed here at the 0 °C temperature setting. Table 4.5 gives the respective values of the dose and corresponding bacteria populations associated with the experiment.

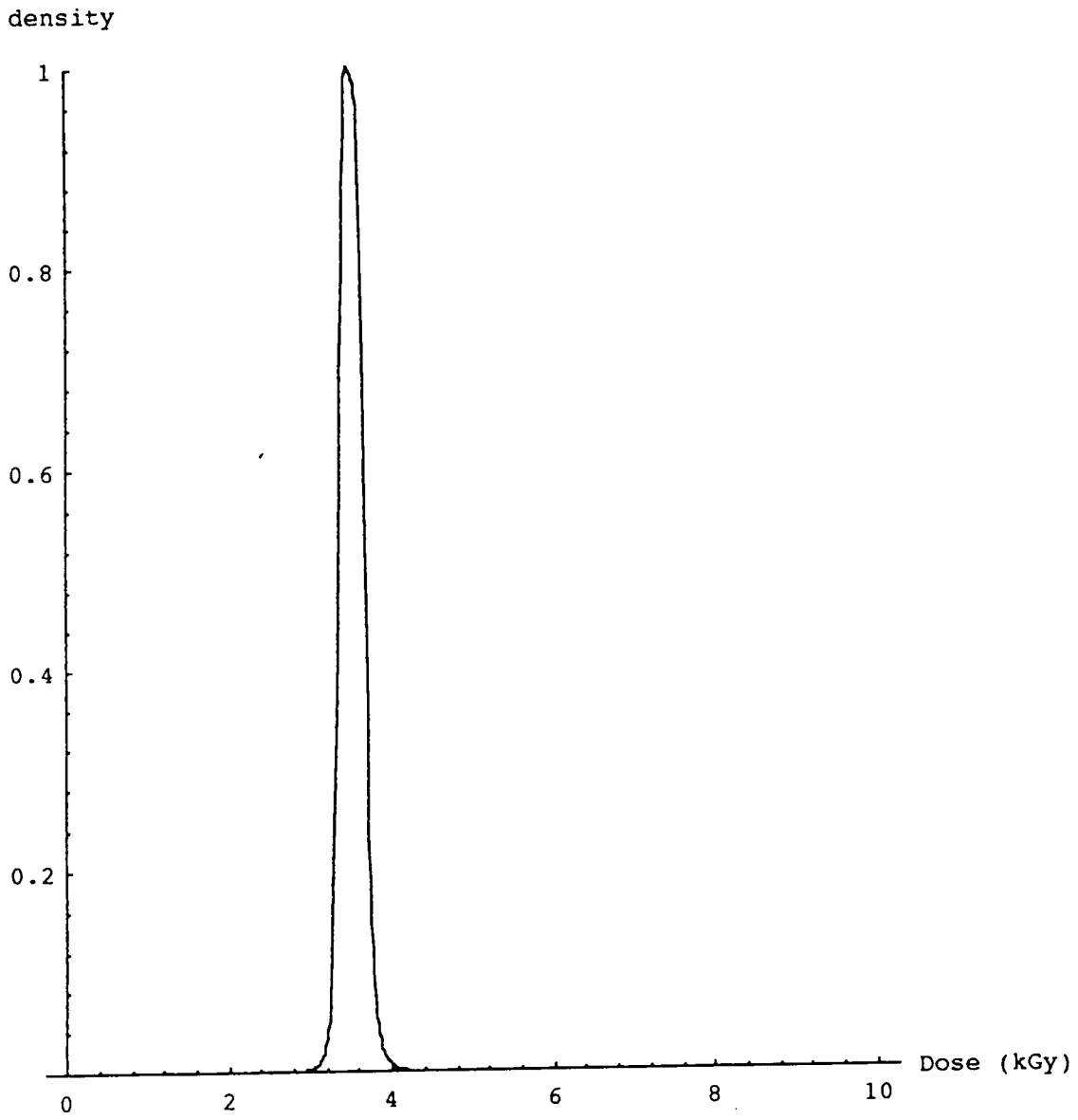


Figure 4.5 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed bacteria population of 100 CFU/g .

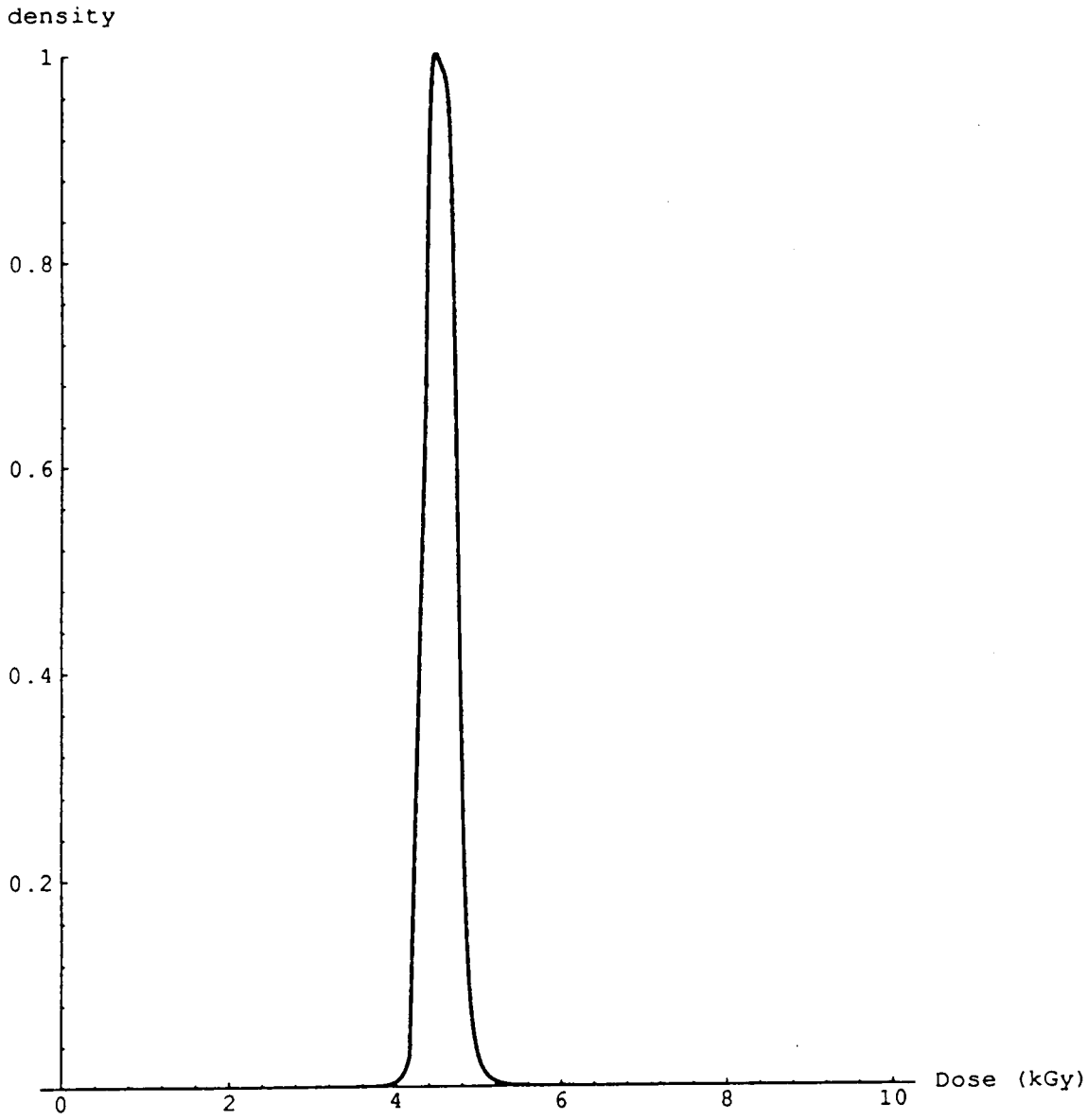


Figure 4.6 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

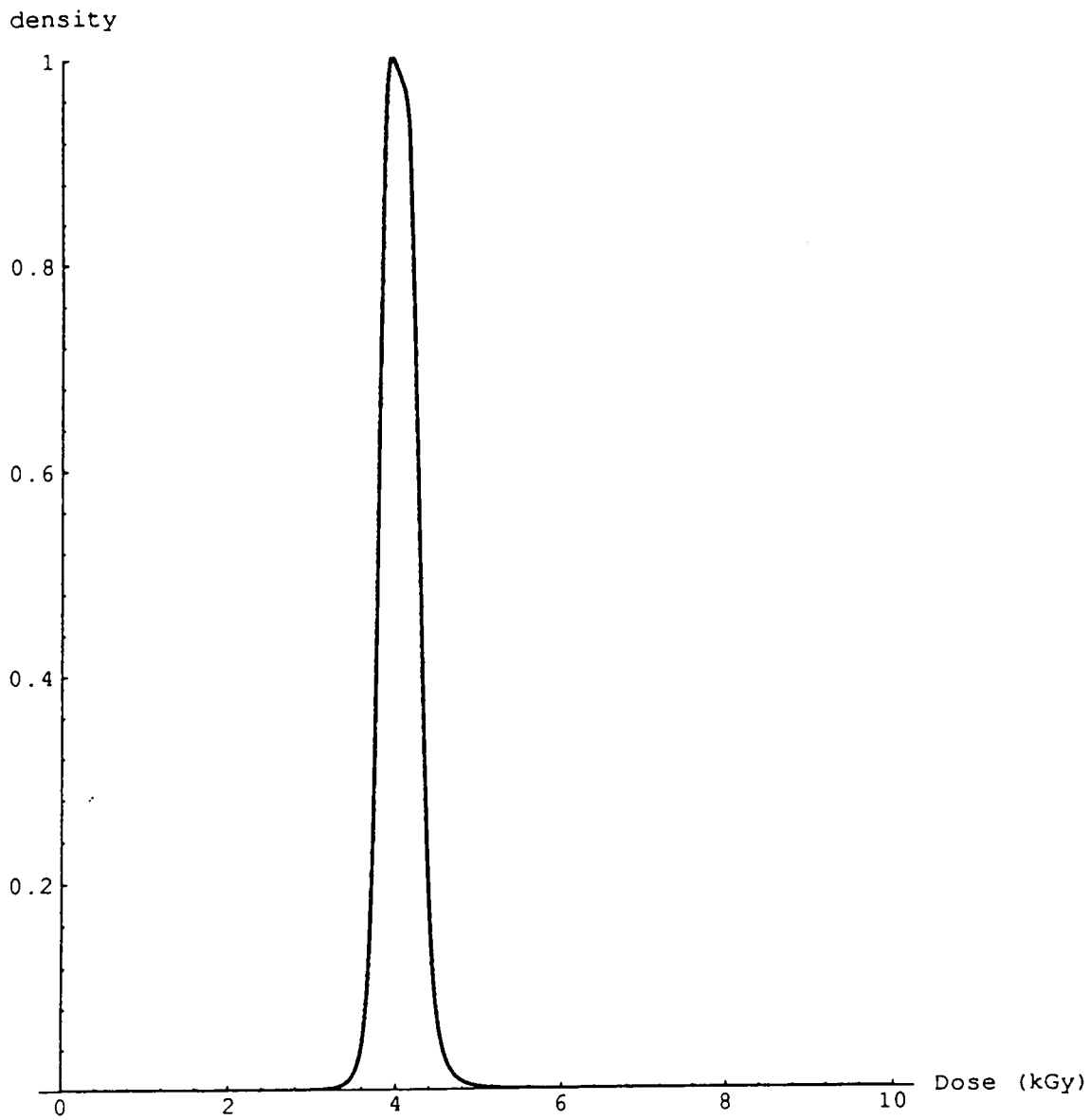


Figure 4.7 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed bacteria population of 100 CFU/g

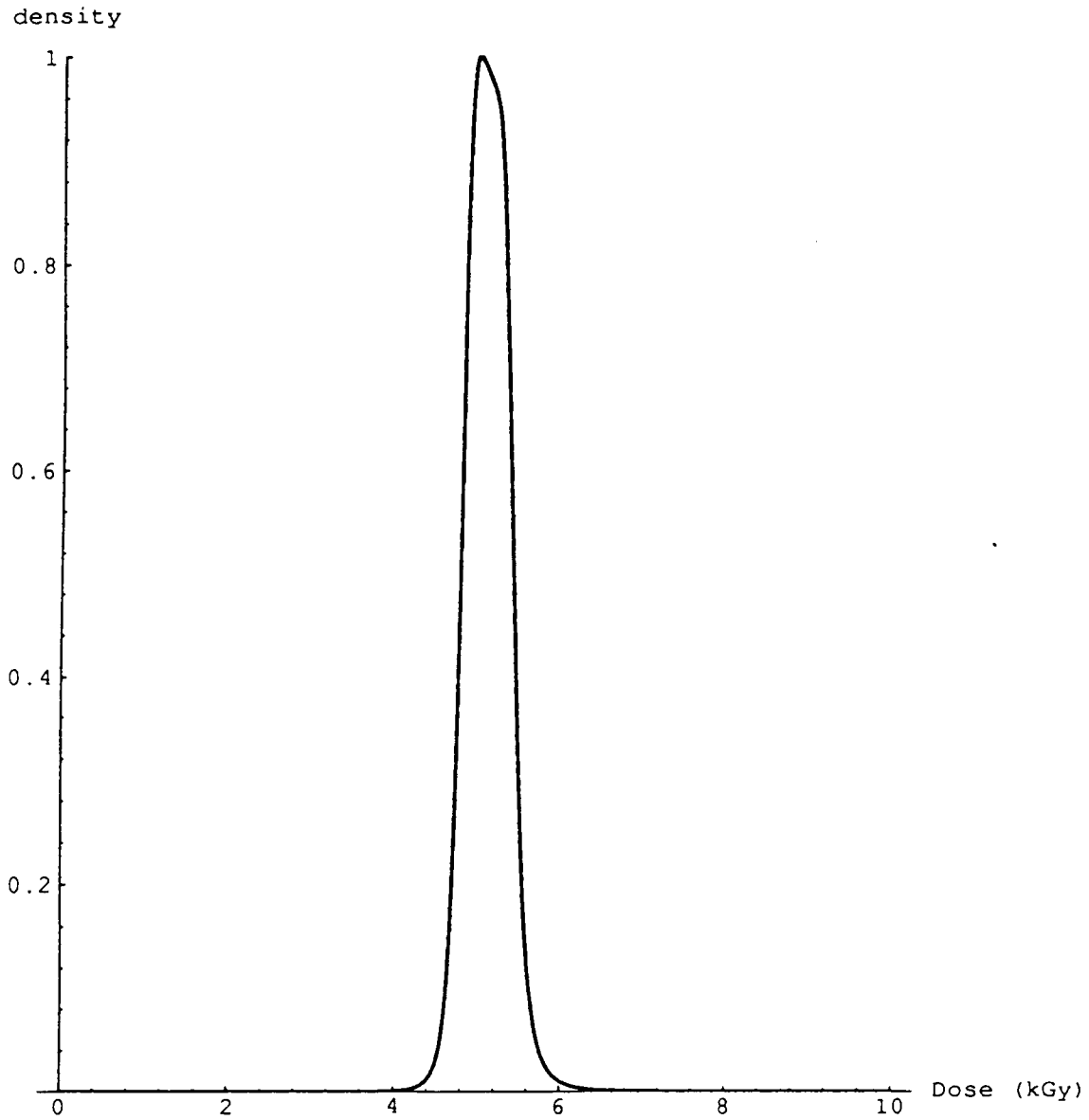


Figure 4.8 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

Table 4.5 Data set for chicken meat irradiated at 0 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.44	9.26
0.30	8.74	8.91
0.60	8.22	8.22
0.90	8.00	8.26
1.20	7.17	7.26

The 0 °C experiment only analyzes doses up to 1.2 kGy. The reason for this is not clear. The experimental analysis used this range in its D-value calculations [Thayer and Boyd, 1991]. There was no reason mentioned for this approach. In order to maintain consistency with the research, data points up to 1.2 kGy were used in developing the predictive and calibrative densities. The scatter plots of the data sets in table 4.5 are shown in figures 4.9 and 4.10.

4.3.1 The Predictive Density

The predictive density is again calculated at a fixed dose of 1.5 kGy. Figure 4.11 shows the predictive density for chicken meat irradiated in the presence of oxygen. The mode of the density occurs at 6.00 log CFU/g. This results in a 65.5% survival rate of Salmonella bacterium. In figure 4.12 the predictive density for chicken meat irradiated in vacuum is graphed. The maximum of this density is at 6.92 log CFU/g. This represents a 74.7% survival rate of bacterium. As stated in section 4.2.1 for the 20 °C temperature setting, the presence of air has a significant effect on the percentage of bacterium surviving a fixed dose. Another aspect that is apparent is temperature dependence. It appears that as the temperature decreases, the amount of bacteria reduction decreases. Also, it is evident

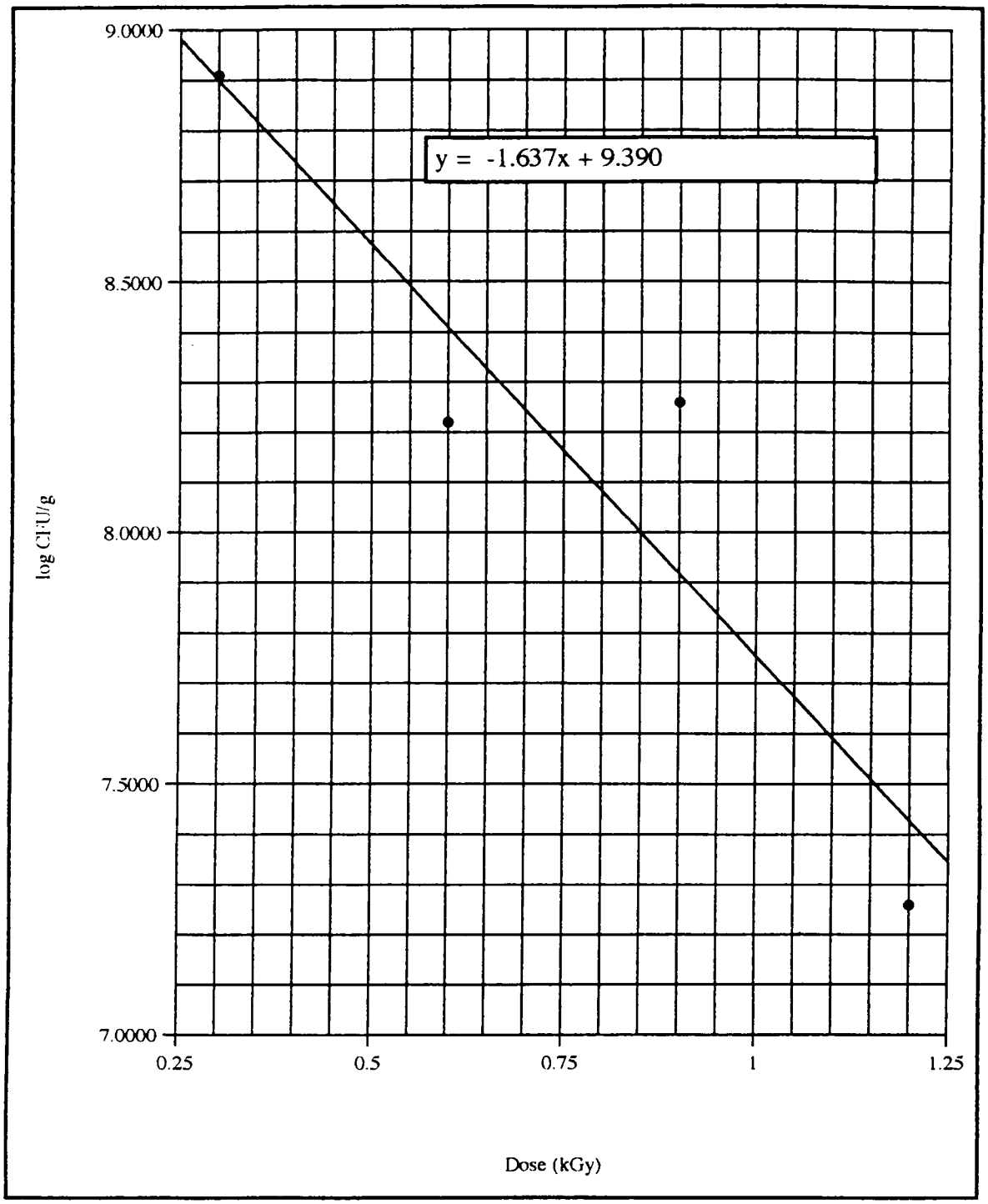


Figure 4.9 Scatter plot for mechanically deboned chicken meat irradiated at 0 °C and in the presence of air

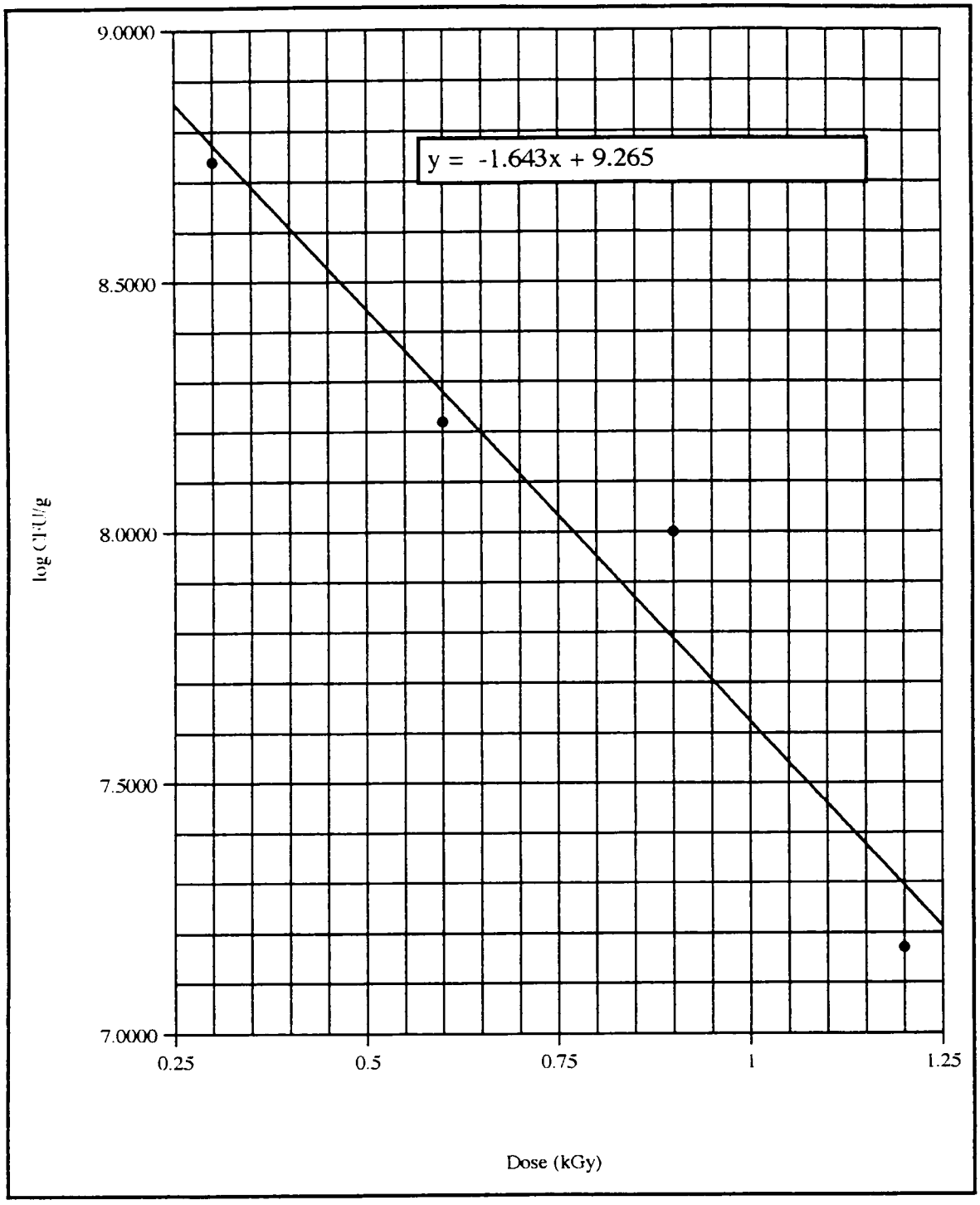


Figure 4.10 Scatter plot for mechanically deboned chicken meat irradiated at 0 °C and in the absence of air

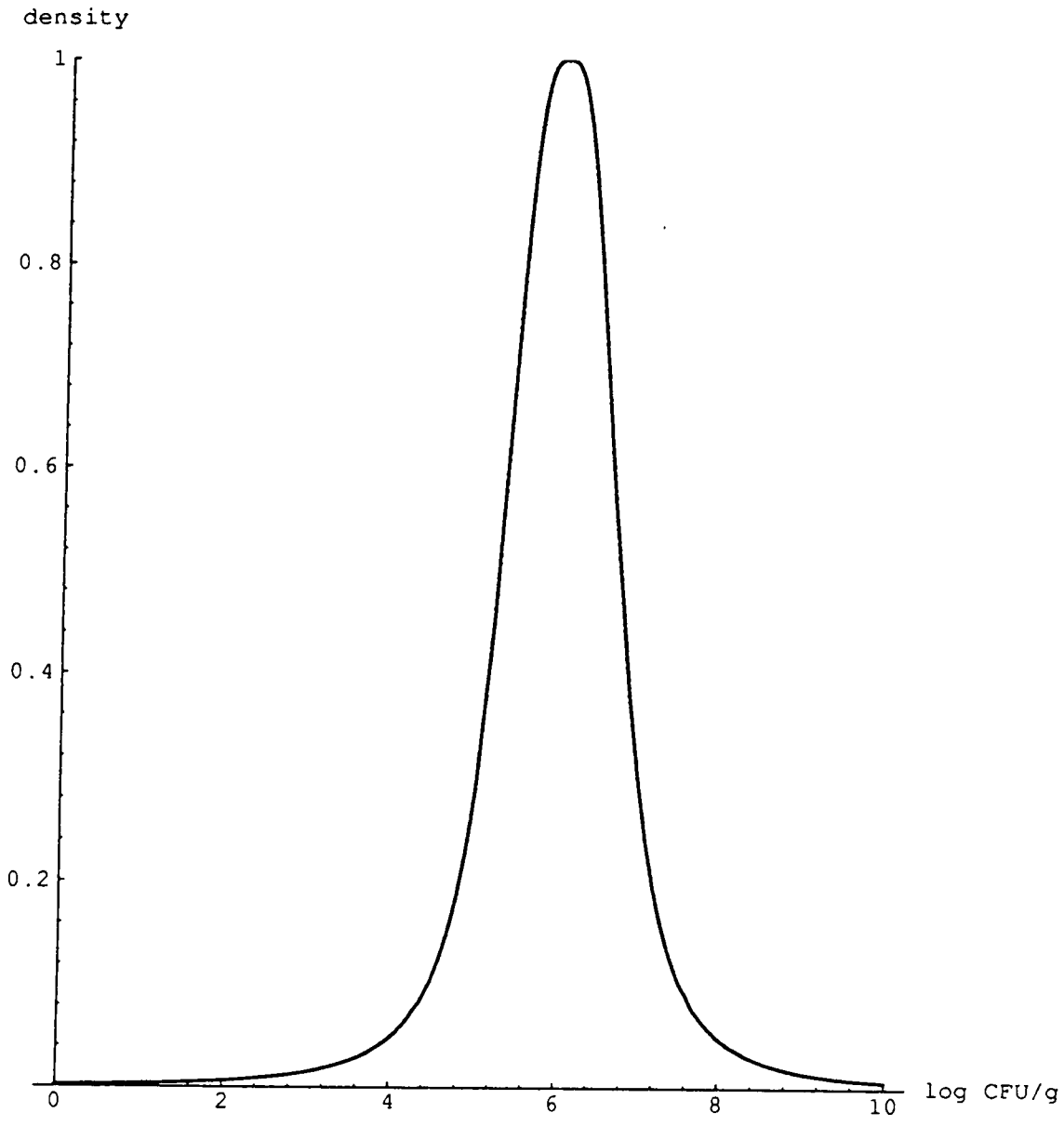


Figure 4.11 The predictive density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed dose of 1.5 kGy

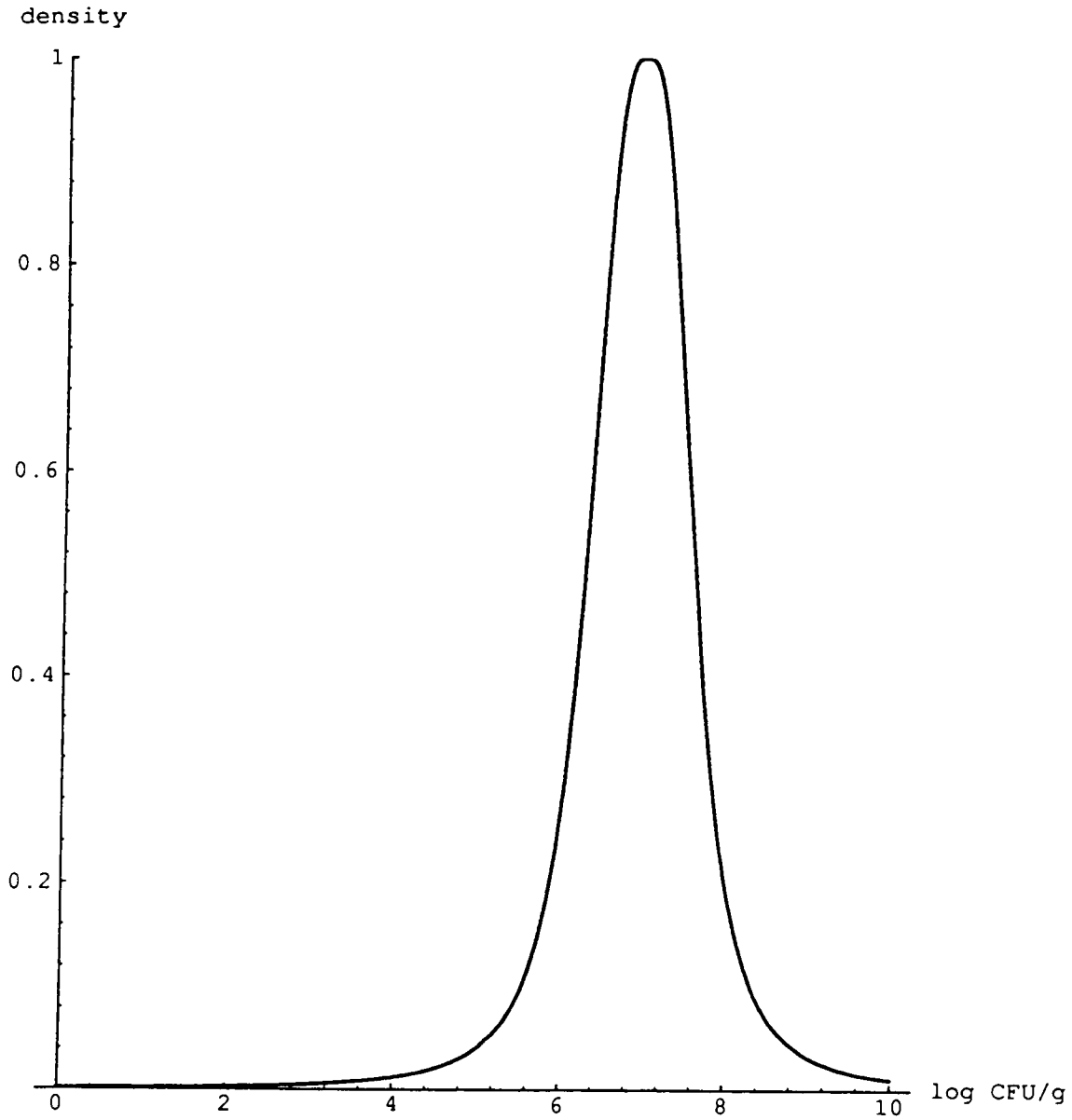


Figure 4.12 The predictive density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed dose of 1.5 kGy

the densities are significantly wider for the 0 °C experiment. This is explained by the fact that the regression fit of the data in figures 4.9 and 4.10 is not very close.

4.3.2 The Calibrative Density

As stated in section 4.2.2, the calibrative density is analyzed for two different bacteria populations. Figures 4.13 and 4.14 show the calibrative density for chicken meat irradiated in the presence of oxygen. There is more uncertainty in these calibrative densities as opposed to the densities in figures 4.5 and 4.6. However, the required doses for specific bacteria populations are higher. This can be attributed to two reasons. The first is that the initial bacteria populations are slightly higher in the 20 °C experiment. Secondly, there is an apparent dependence of bacteria survival corresponding to temperature during irradiation (i.e. as temperature increases bacteria resistance to radiation decreases) .

Figures 4.15 and 4.16 show the calibrative density for chicken meat irradiated in the absence of air. Even though the dose levels are not significantly different, it should be noted that the vacuum data set started out with a smaller bacterium population than the air data set. Thus, it would be natural to assume given the supporting data that the absence of air leads to less bacteria reduction at a fixed dose. The uncertainty is much greater in the vacuum data set. The reasons attributed are that the data does not fit the model very well and there are fewer data points.

4.4 Analysis of Irradiated Chicken Meat at (-)20 °C

For the (-) 20 °C temperature experiment, two data sets are analyzed. As in the previous two sections, the data sets consist of irradiation in air and vacuum. Table 4.6 gives the corresponding values.

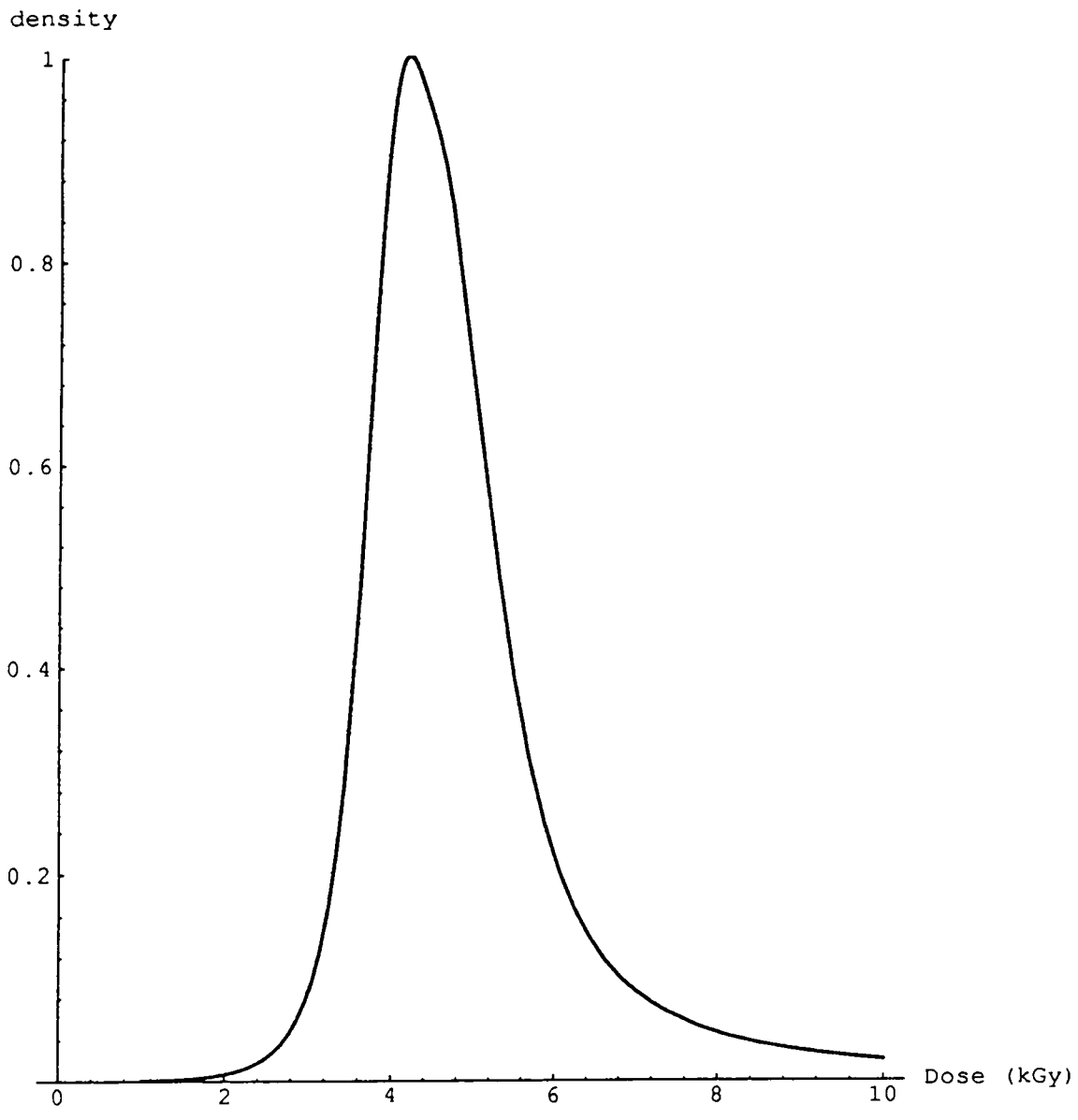


Figure 4.13 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed bacteria population of 100 CFU/g

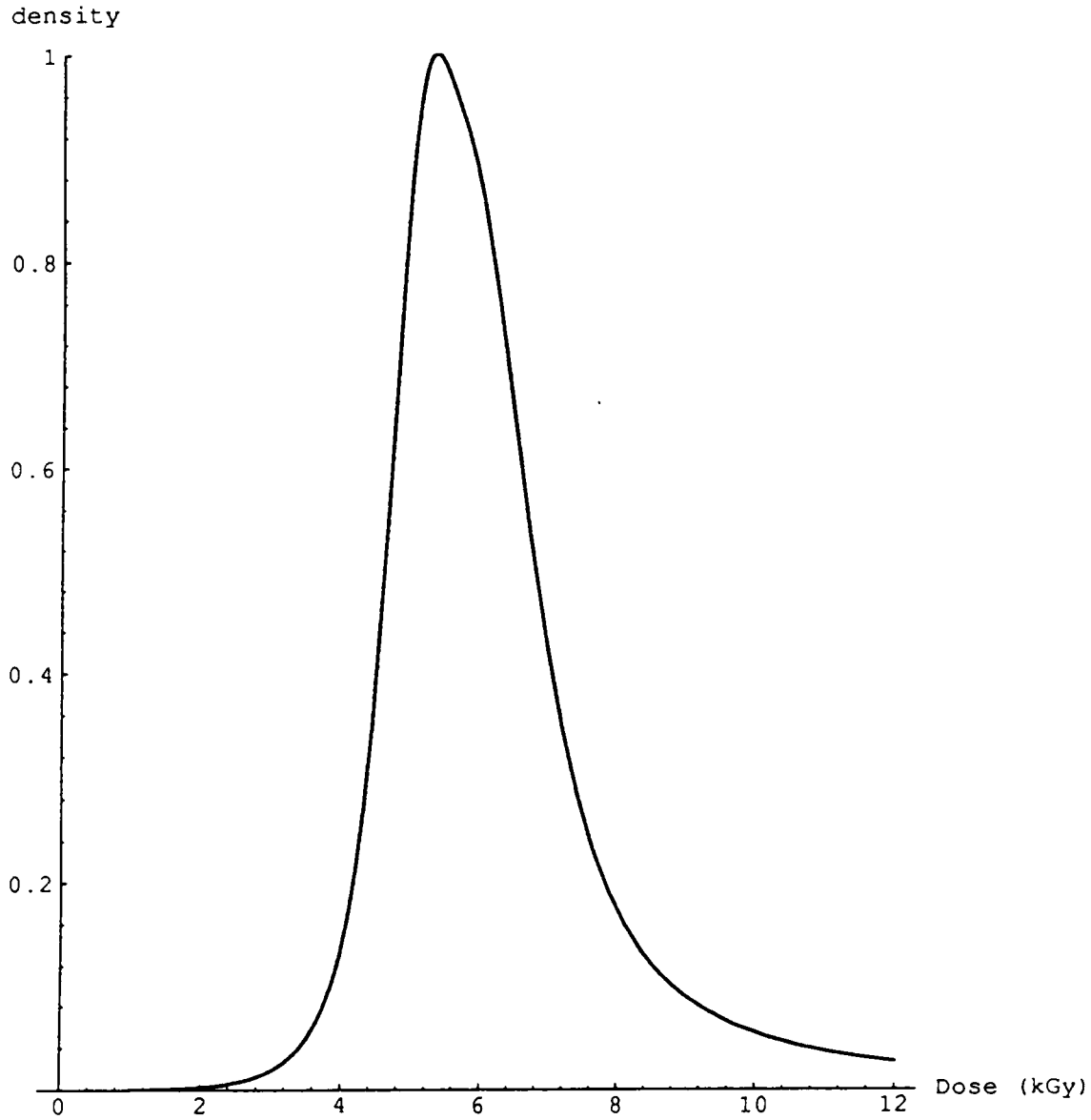


Figure 4.14 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

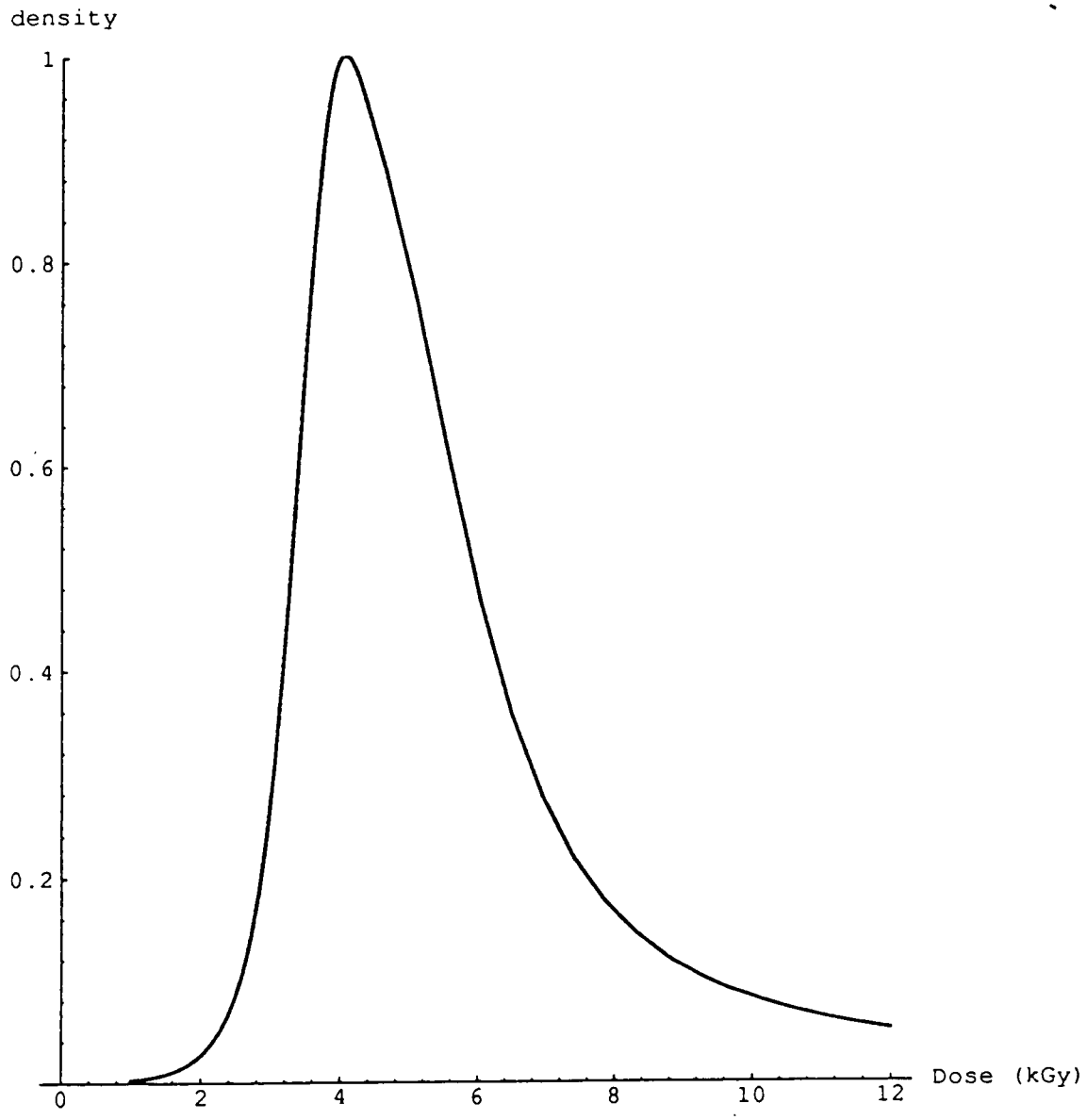


Figure 4.15 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed bacteria population of 100 CFU/g.

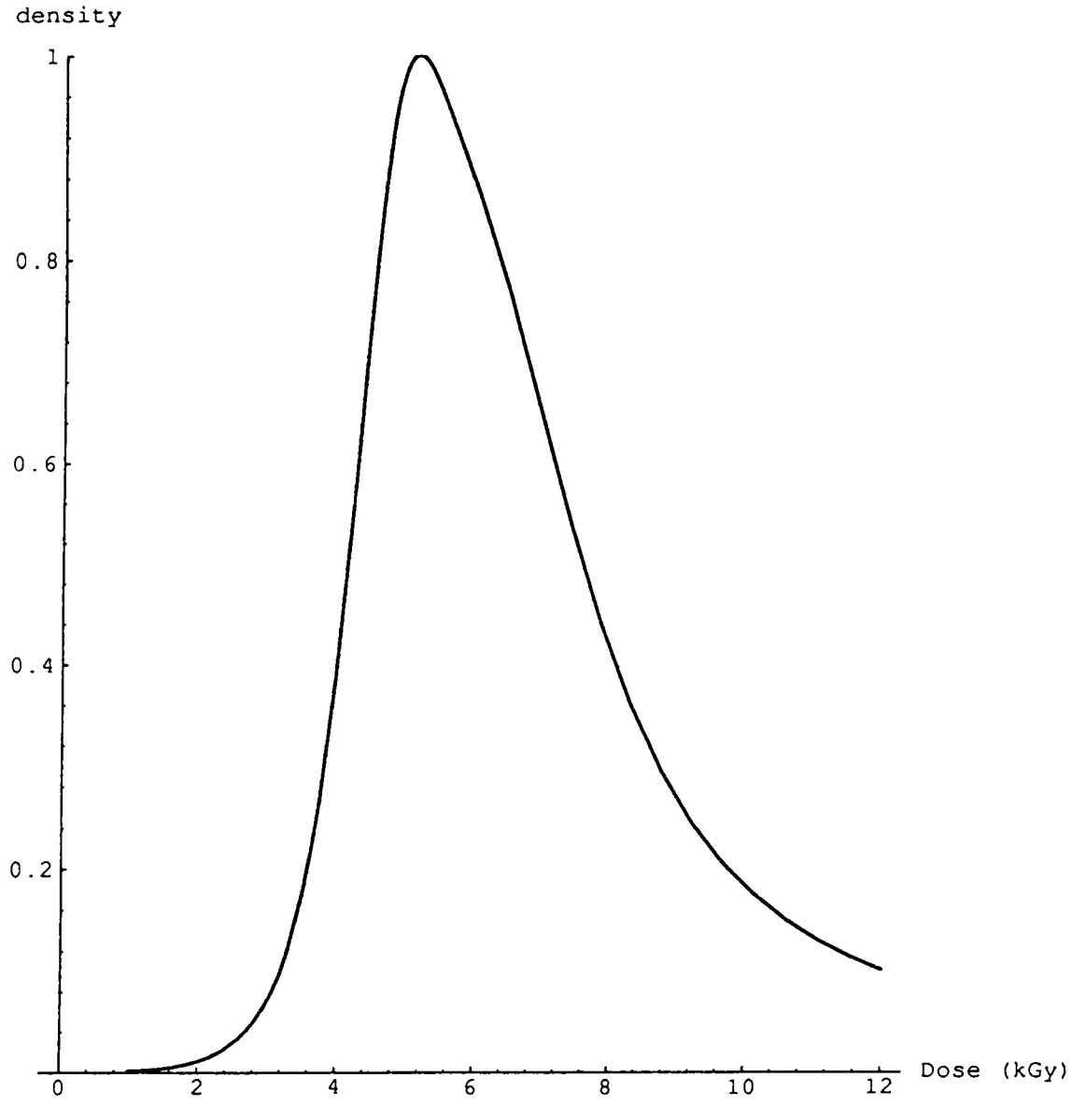


Figure 4.16 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

Table 4.6 Data set for chicken meat irradiated at (-)20 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.09	9.14
0.30	8.64	8.73
0.90	7.79	7.93
1.20	6.57	6.77
1.80	6.66	7.13
2.70	5.14	5.56

The key difference between the data sets in table 4.6 and table 4.4 is the lack of a 0.60 kGy data point. Thus, there are five data points that make up the calibration data set instead of six. The corresponding scatter plots can be seen in figures 4.17 and 4.18. Note that the slopes again are unexpectedly similiar in steepness [Bacq, 1961].

4.4.1 The Predictive Density

For a fixed dose of 1.5 kGy, the predictive density is shown in figures 4.19 and 4.20. The maximum of the predictive density for chicken meat irradiated in air occurs at 6.95 log CFU/g. This corresponds to a 76.4% survival rate of Salmonella. This is a significantly higher survival rate than previous temperature settings. The mode of the predictive density in figure 4.20 is 7.00. This represents a 76.6% survival rate of the bacterium population. As expected theses survival rates are higher than the previous two experimental settings (i.e. sections 4.2 and 4.3). It is apparent that as the temperature during irradiation decreases, the survival of Salmonella increases. Also, there appears to be a flattening of the densities in figures 4.19 and 4.20. This spreads out the maximum due to the scale on the ordinate of the plot.

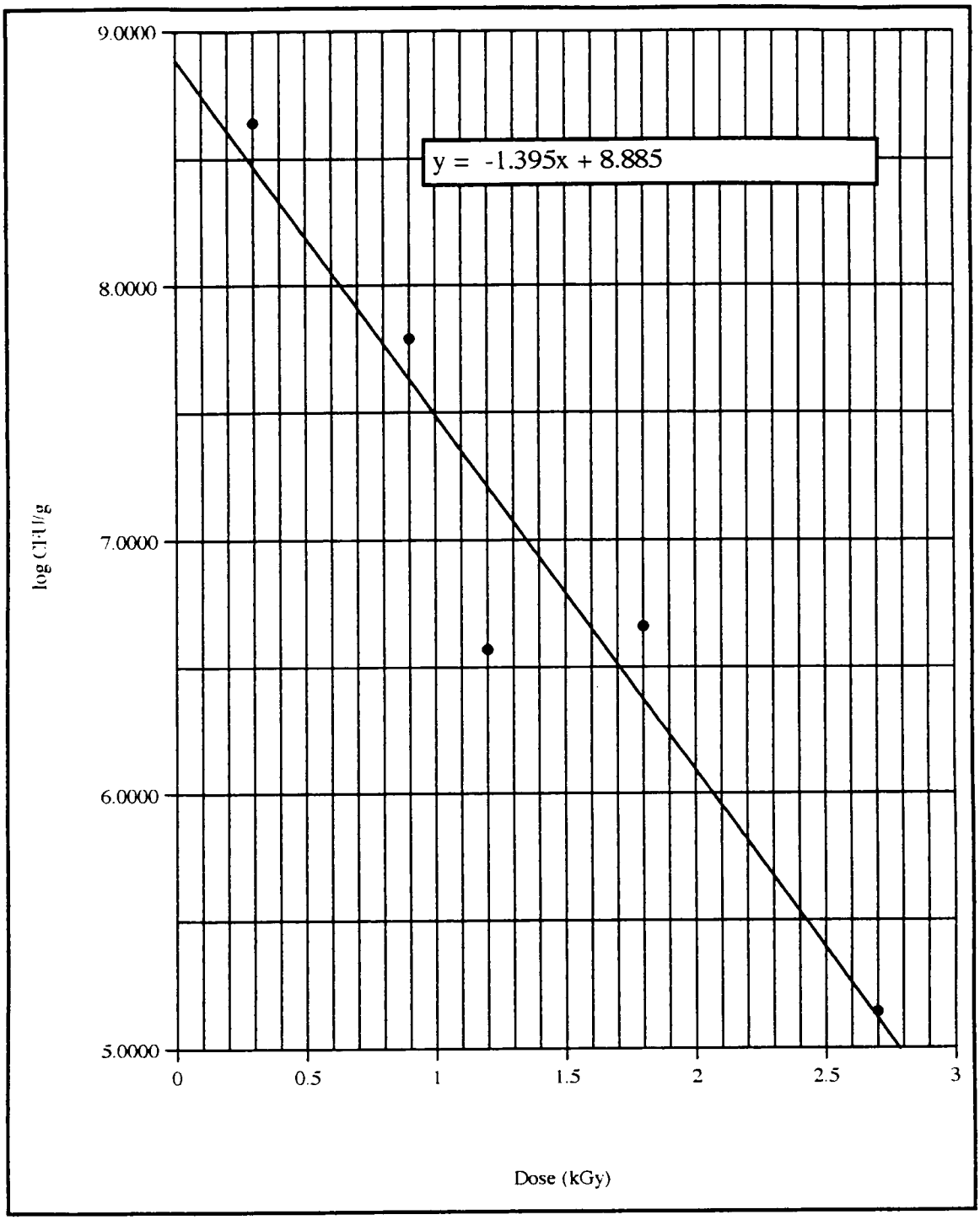


Figure 4.17 Scatter plot for mechanically deboned chicken meat irradiated at (-)20 °C and in the presence of air

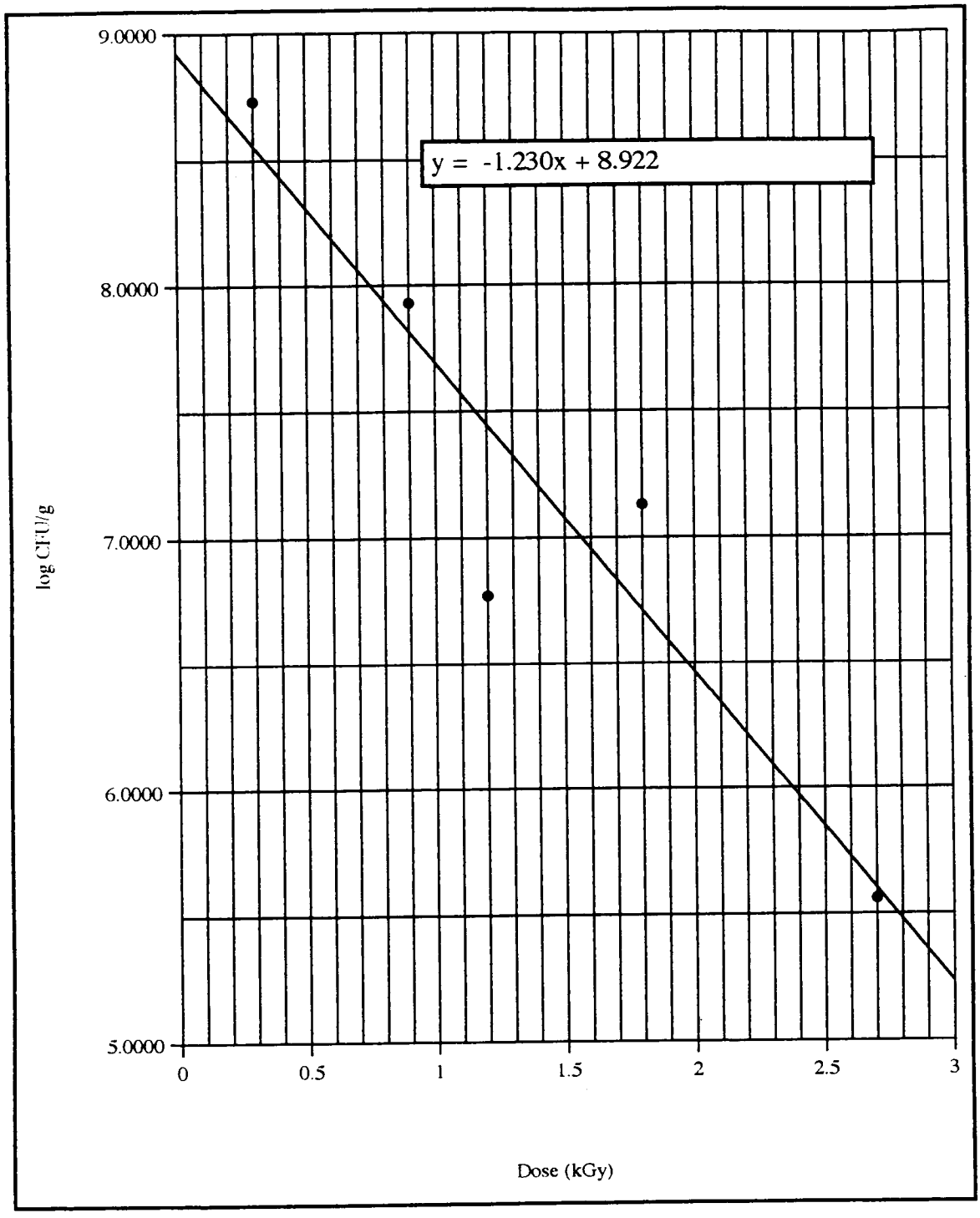


Figure 4.18 Scatter plot for mechanically deboned chicken meat irradiated at (-)20 °C and in the absence of air

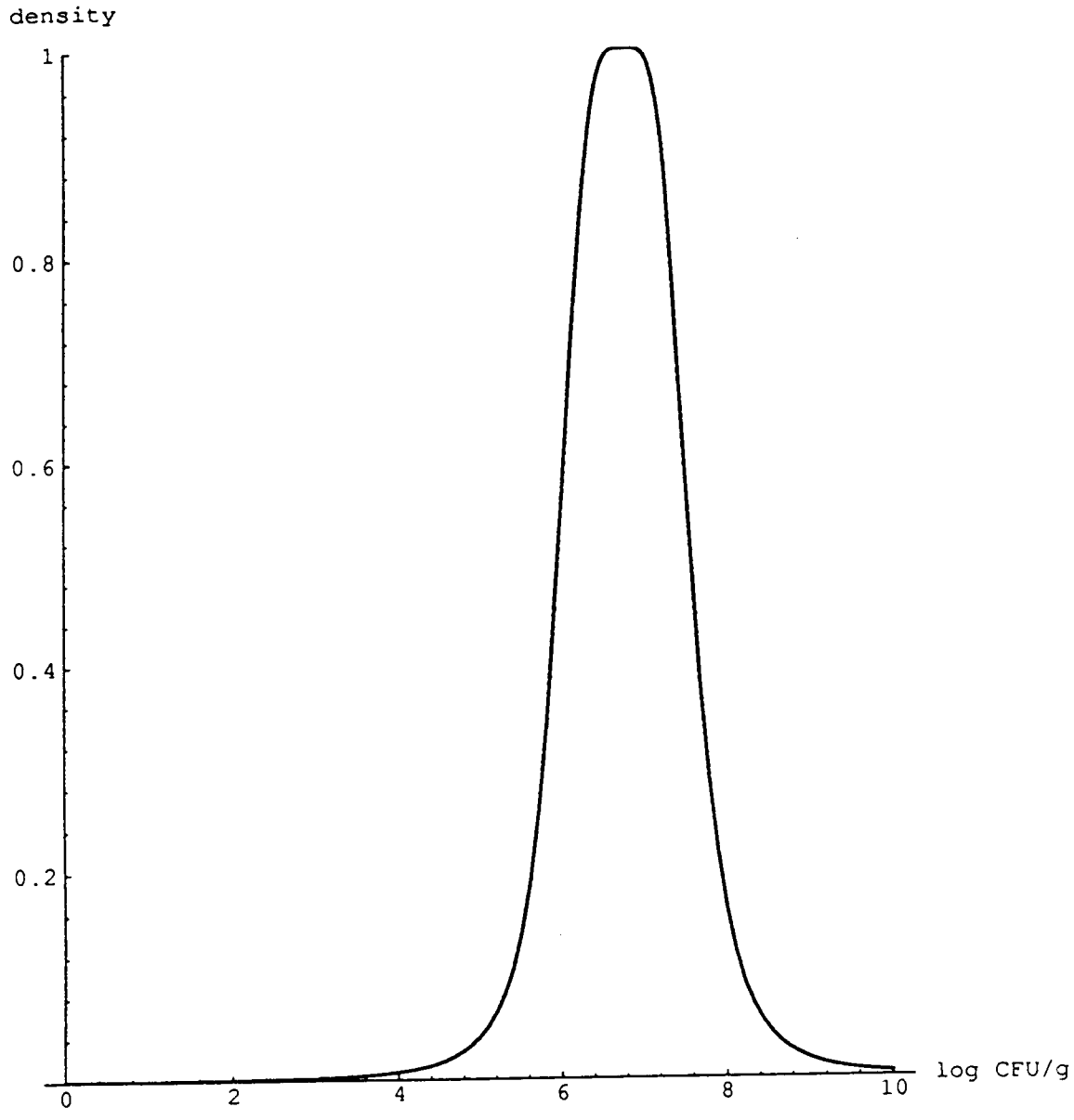


Figure 4.19 The predictive density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed dose of 1.5 kGy

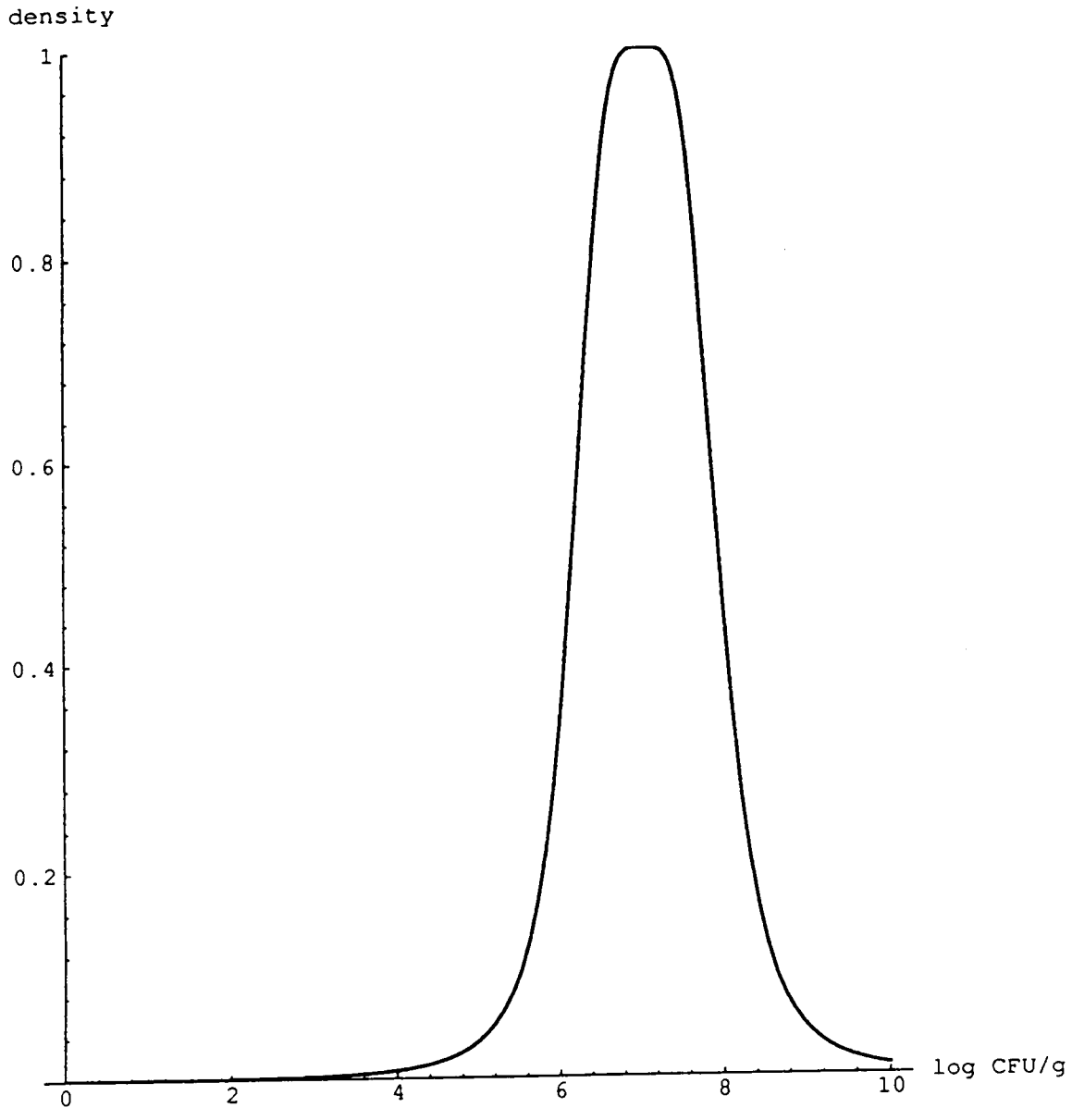


Figure 4.20 The predictive density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed dose of 1.5 kGy

4.4.2 The Calibrative Density

As in both previous temperature settings, the calibrative density is calculated at two specific bacteria populations. Figures 4.21 and 4.22 show the calibrative density for chicken meat irradiated in the presence of oxygen. In figure 4.21, a dose of 4.52 kGy is lowest dose with the highest probability of reducing the Salmonella to a population of 100 CFU/g. Figure 4.22 shows that a dose of 5.84 kGy has the highest probability of reducing the bacterium population to 1 CFU/g. It is important to note that the uncertainty (i.e. width of the density) is larger than the other two temperature settings. The reason behind this is that the model does not fit the data as well as the other data sets.

Figures 4.23 and 4.24 show the calibrative density for mechanically deboned chicken meat irradiated in the absence of air. A dose of 4.83 kGy has the highest probability of reducing the bacterium population to 100 CFU/g. In order to reduce the bacterium population to 1 CFU/g then a dose of 5.93 kGy is needed. The uncertainty in the vacuum data set appears to be similar to the oxygen data set (i.e. width of density).

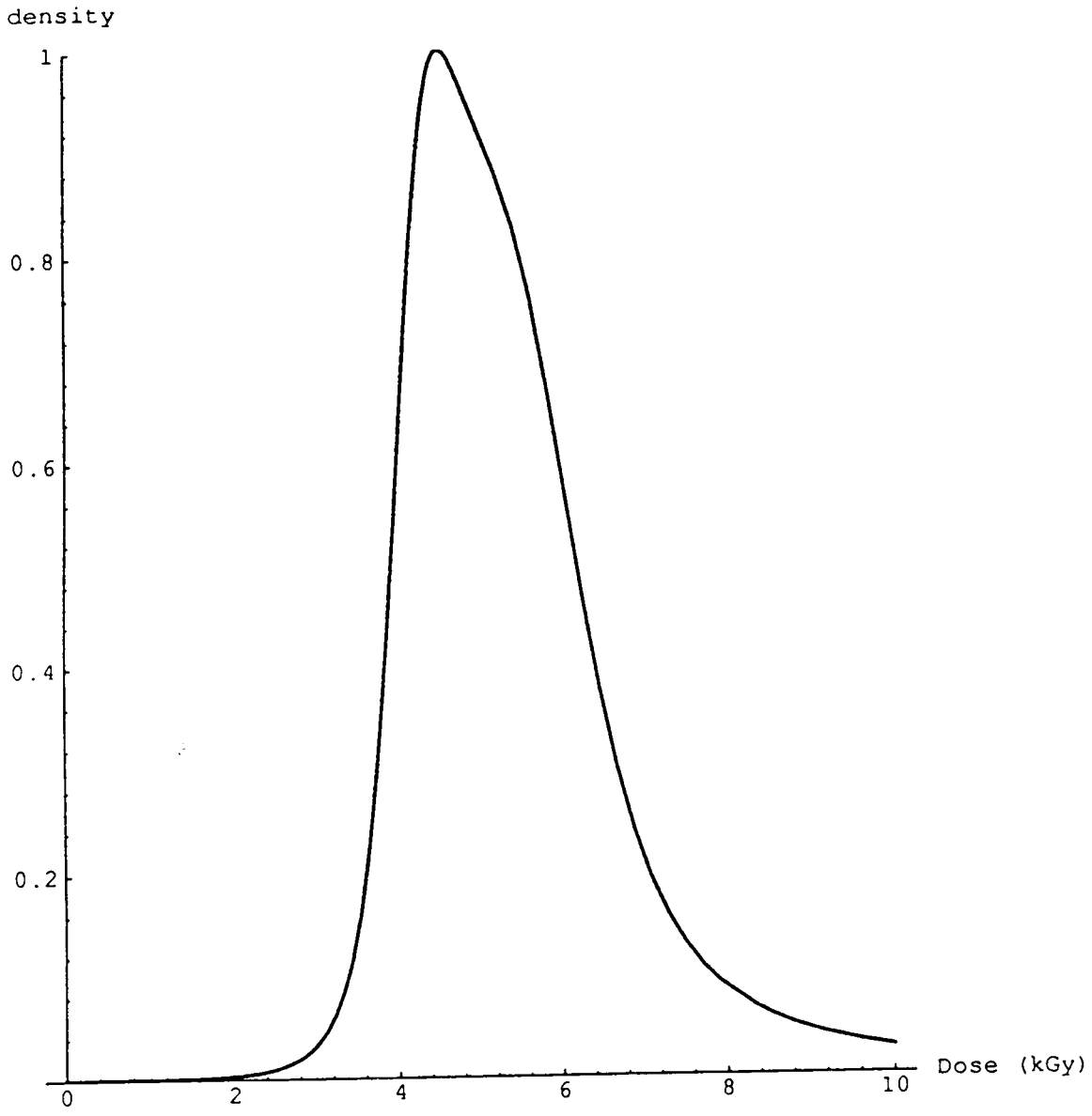


Figure 4.21 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed bacteria population of 100 CFU/g .

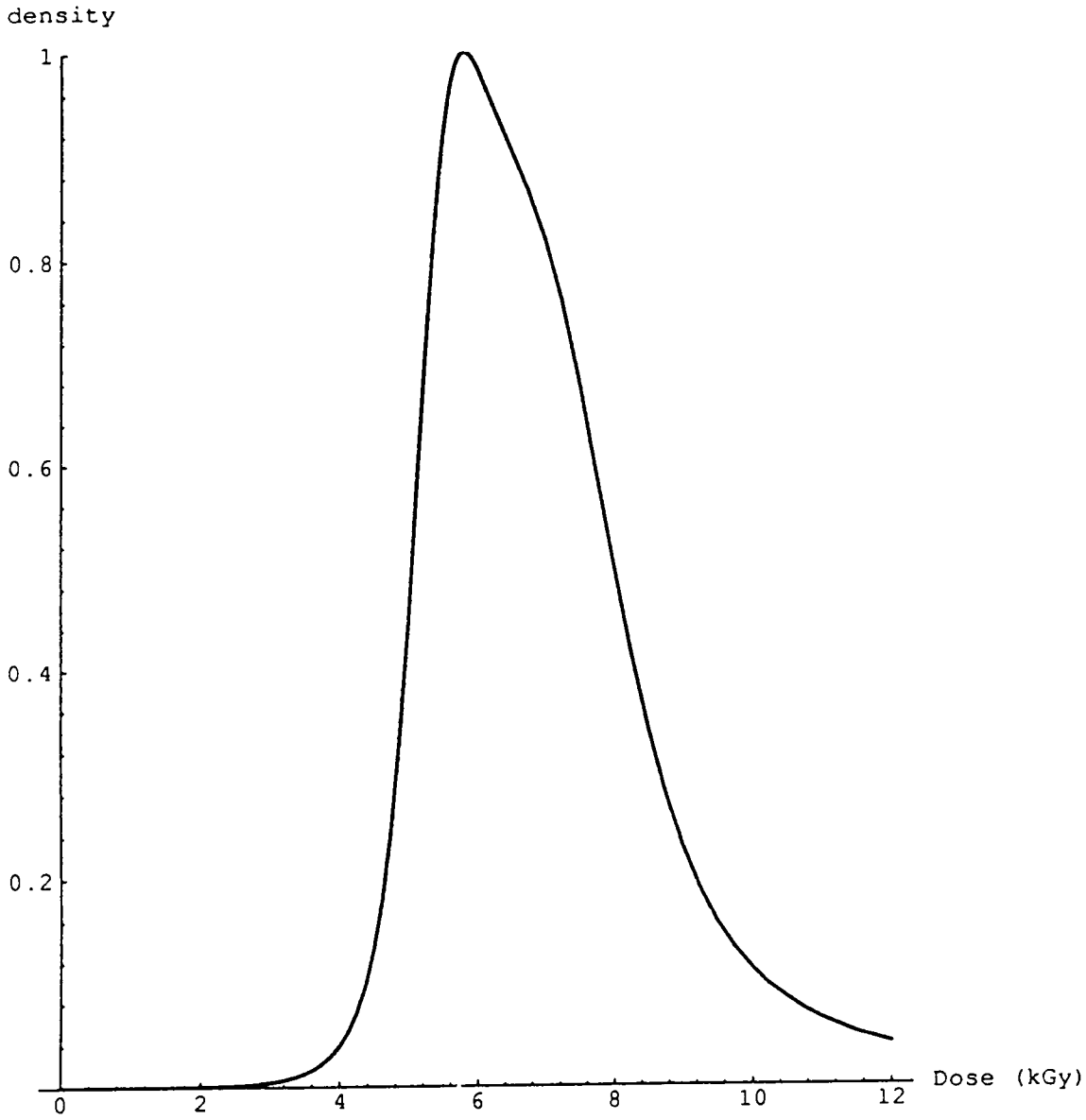


Figure 4.22 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

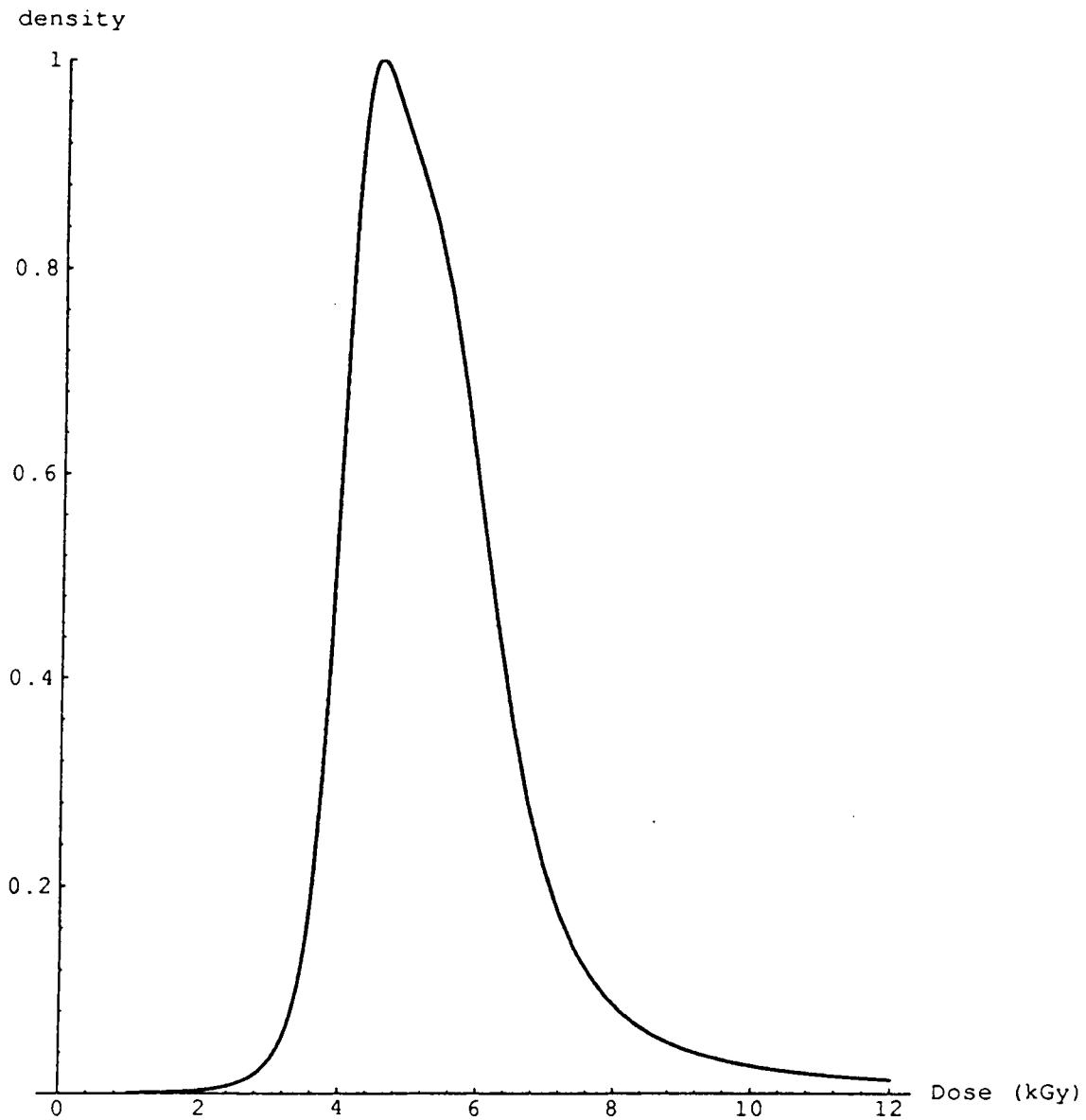


Figure 4.23 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed bacteria population of 100 CFU/g

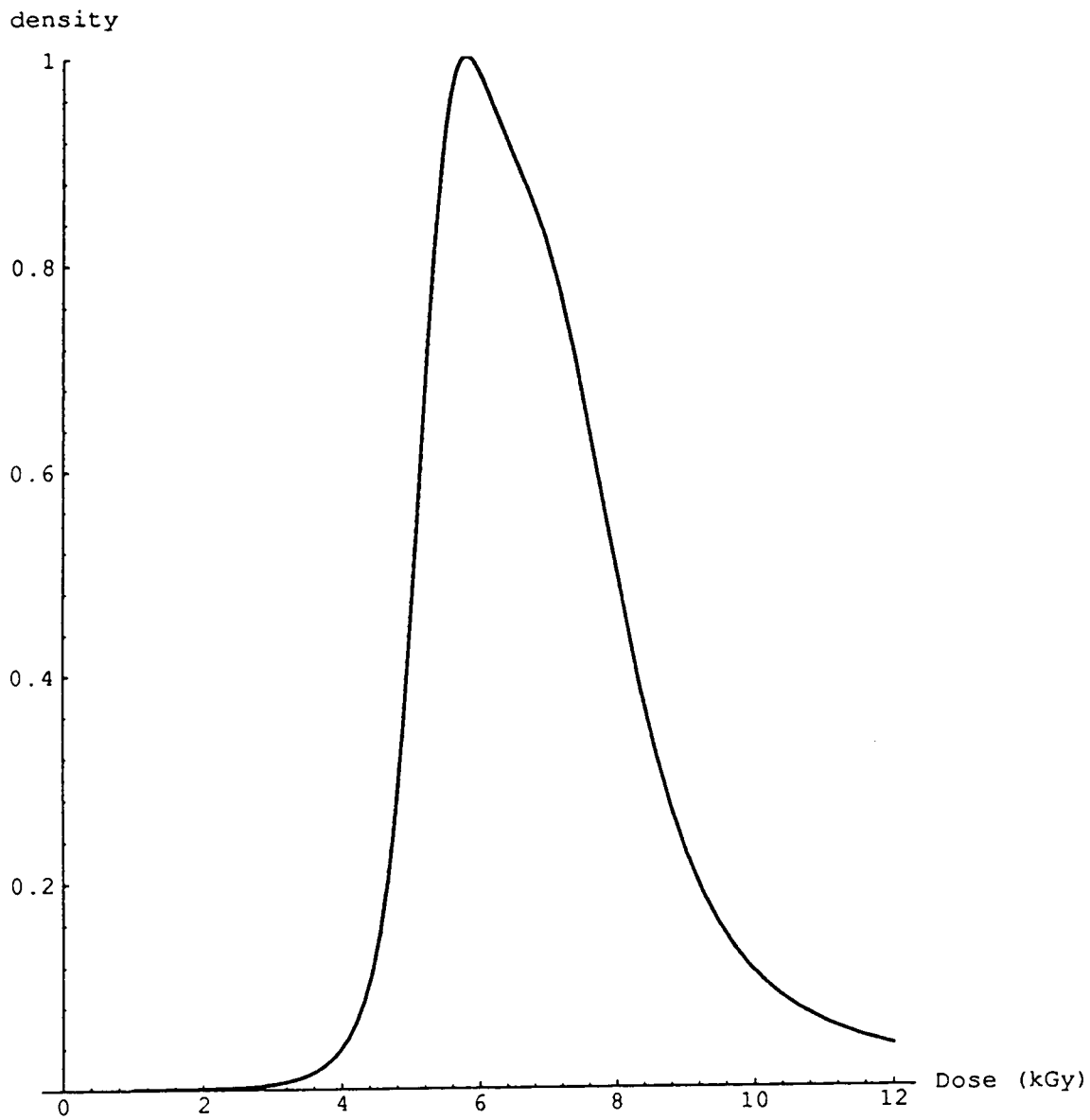


Figure 4.24 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

5.0 Summary of Thesis

This thesis analyzed the calibration of the response of bacteria to radiation. It provides the first quantitative description of the uncertainty in dose given a specific bacteria population. This uncertainty can be related to a specific bacterium population and thus quantitatively describes the dose necessary to achieve a certain population reduction.

First, we provided background on the process of food irradiation. Included in this discussion was the bacterium Salmonella and the need for its control. In addition, federal guidelines and approved radiation levels were discussed.

Second, we discussed in detail the aspects of the two studies used for analysis. A brief description of the data sets in each study was given. The data points used, omitted and modified were addressed. Then, definitions of the D-value and other methods of bacteria population reduction were given.

Next, we described how Bayesian methods can be used in food irradiation to give a quantitative description of the uncertainty. This description included the derivation of the predictive and calibrative densities. An overview of the prior distribution and how it was obtained is also discussed. In addition, the Student t-distribution was analyzed. This discussion included the description of the three parameters and their numerical values using a sample data set.

Finally, the results of the analysis for one study (5564) were given and comparisons between factors such as temperature, atmosphere and dose were discussed. This study was chosen because it used a larger number of data points for its experimental

procedure. The second study was also analyzed to check for reproducibility and its results are included in Appendix II.

5.1 Future Work

The results of this analysis give a description of the uncertainty in dose given a specific bacterium population for fixed experimental conditions. In order to describe the dependence on the other variables (e.g. temperature), a multivariate calibration approach would have to be used. For instance, models for temperature dependence and atmosphere would have to be derived. This was considered beyond the scope of this thesis because of the inherent mathematical complexities.

It is important to note that Bayesian methodology for calibration can be useful in other areas such as external dosimetry. Advantages such as a quantitative description of the uncertainty and incorporating other information into the analysis, make Bayesian calibration an excellent choice over conventional methods.

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APPENDICES

APPENDIX I

Combination of data sets for 20 °C and in air

The combining of two data sets is researched for the 20 °C temperature setting and in the presence of air. It can be inferred that the results of other data sets is comparable. This analysis was run for 100 CFU/g, 1 CFU/g and a fixed dose of 1.5 kGy. It is evident that the distributions widen in all scenarios. Even more notable is the flattening of the distribution in Figure 2 for a fixed dose of 1.5 kGy. The reasons behind this is not very clear.

Table 1. Numerical values for combined data sets

Dose (kGy)	Air Pop (log CFU/g)
0.0	9.62
0.30	8.12
0.60	7.16
0.90	7.09
1.20	6.40
1.80	5.14
1.80	4.78
2.7	3.63
3.6	1.58

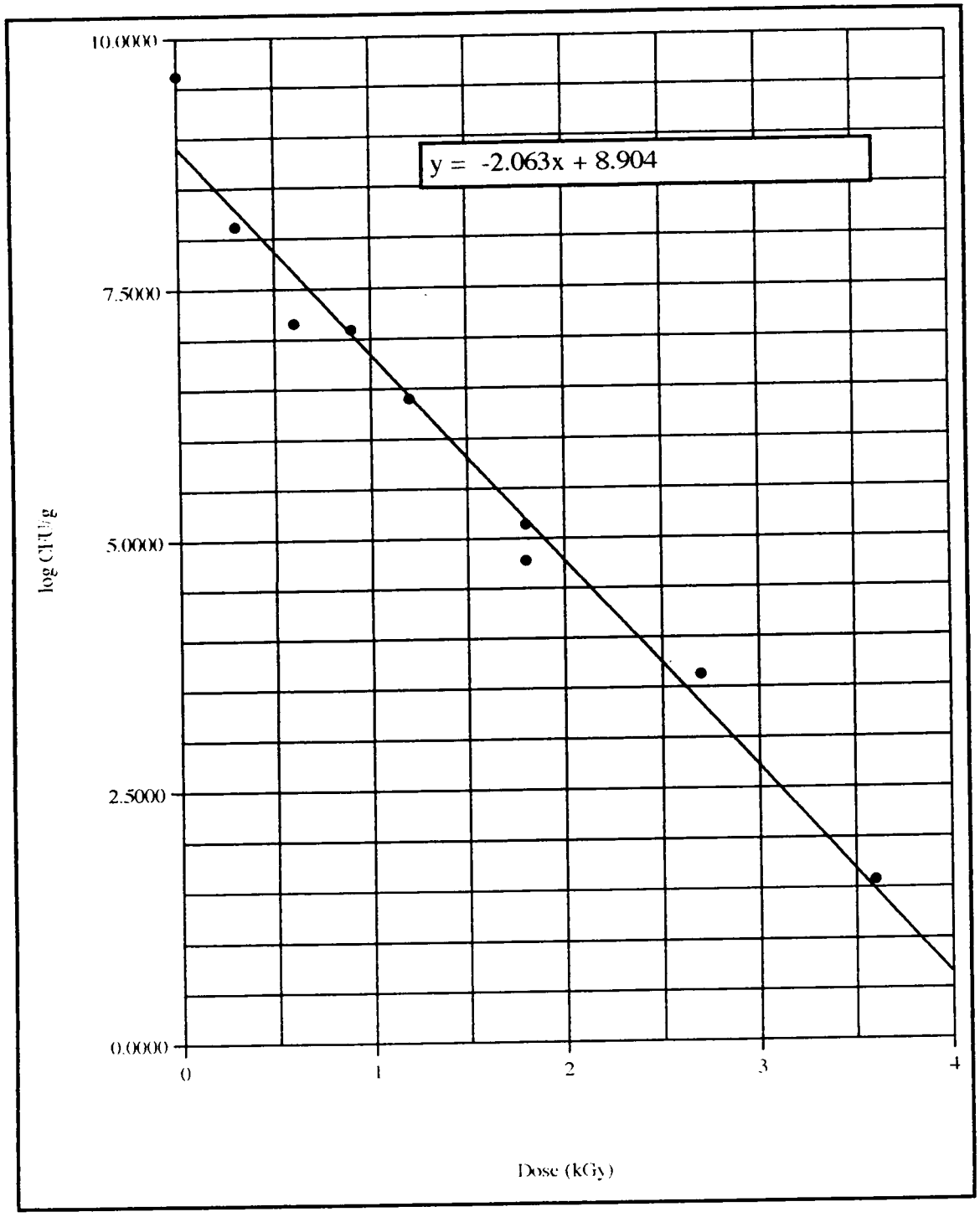


Figure 1. Scatter plot for combined data sets irradiated at 20 °C and in the presence of air

```
f[st_]:=   (1/((Beta[.5,3.5])*(Sqrt[7*0.121])*(1+((u-5.81)^2
/(7*0.121))^4))/1.1)
Plot[f[st_],   {u,0,10},PlotRange->{0,1.1},AspectRatio->1.2,
AxesLabel->{"log  CFU/g","density"}]
```

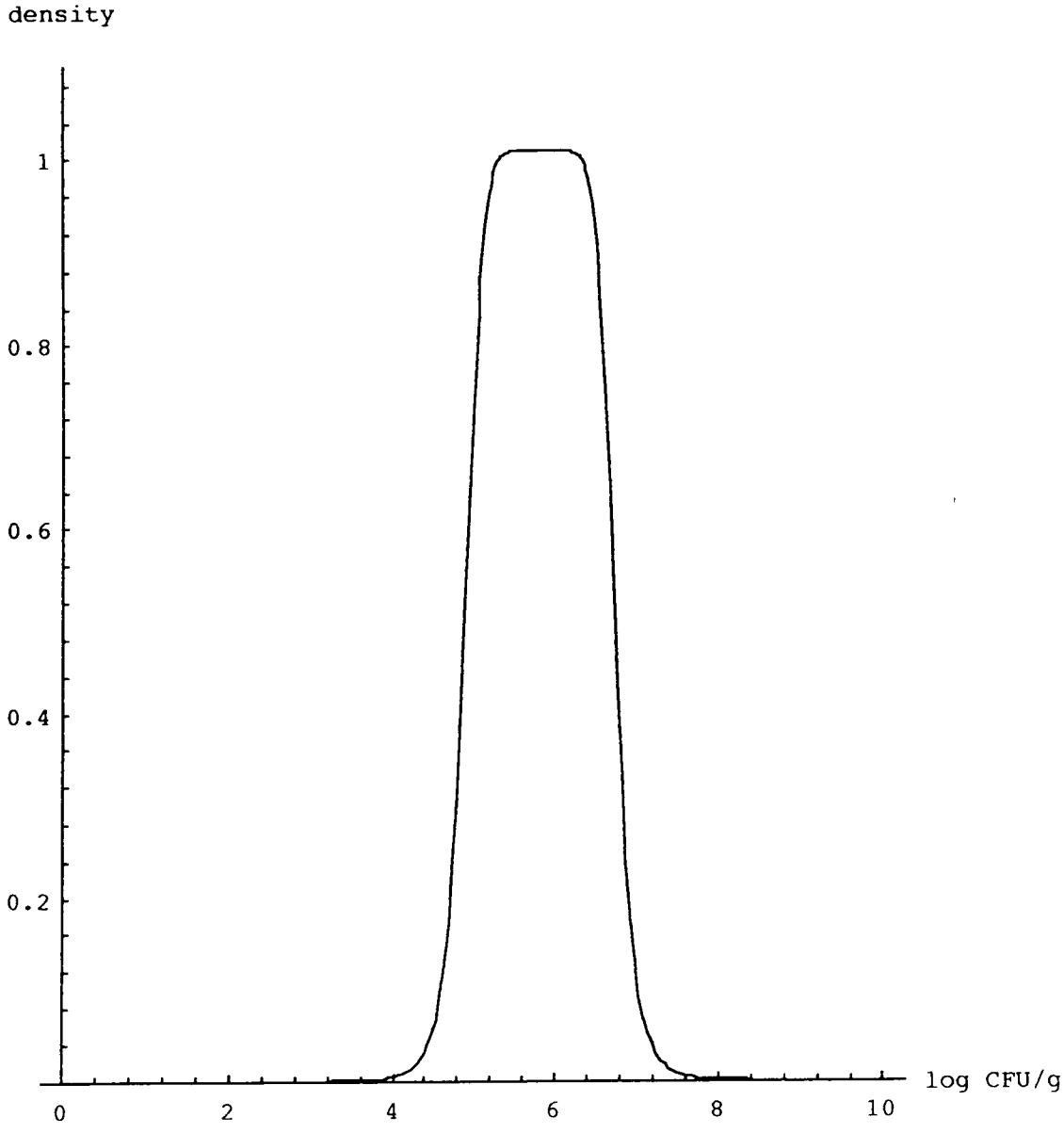


Figure 2. The predictive density for combined data sets irradiated at 20 °C, in the presence of air and a fixed dose of 1.5 kGy

```
f[st_]:=   (1/((Beta[.5,3.5])*(Sqrt[7*(.121+(.00999*(u-1.43)^2))])
*(1+((2-((-2.093*u)+8.904))^2/(7*(.121+(.00999*(u-1.43)^2))))
^4))/.99889)
Plot[f[st_],  {u,0,10},AxesLabel->{"Dose  (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```

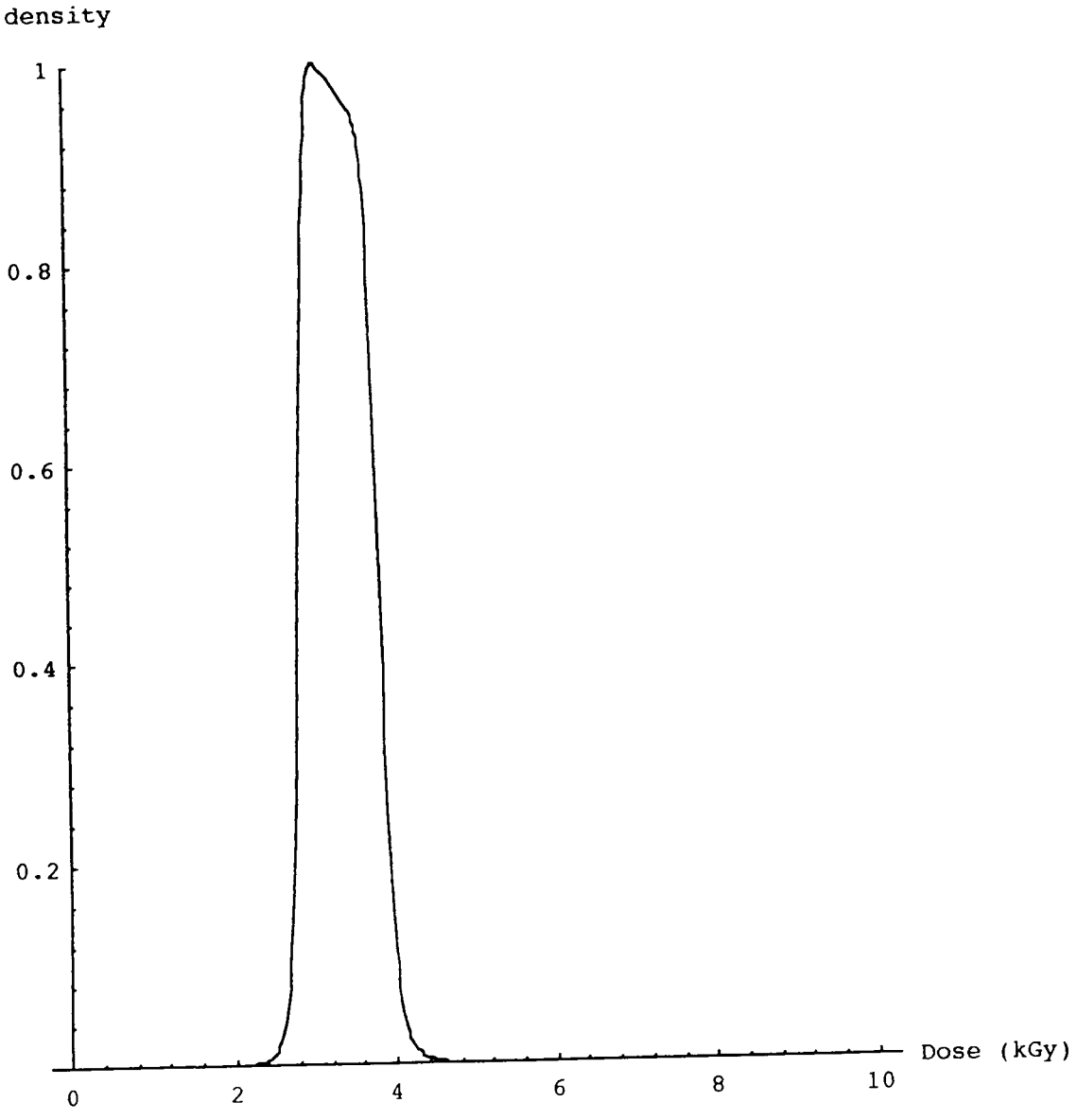


Figure 3. The calibrative density for combined data sets irradiated at 20 °C, in the presence of air and a fixed bacteria population of 100 CFU/g

```
f[st_]:=  (1/((Beta[.5,3.5])*(Sqrt[7*(.121+(.00999*(u-1.43)^2))
*(1+((0-((-2.093*u)+8.904))^2/(7*(.121+(.00999*(u-1.43)^2))))
^4))/.889132)
Plot[f[st_],  {u,0,10},AxesLabel->{"Dose  (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```

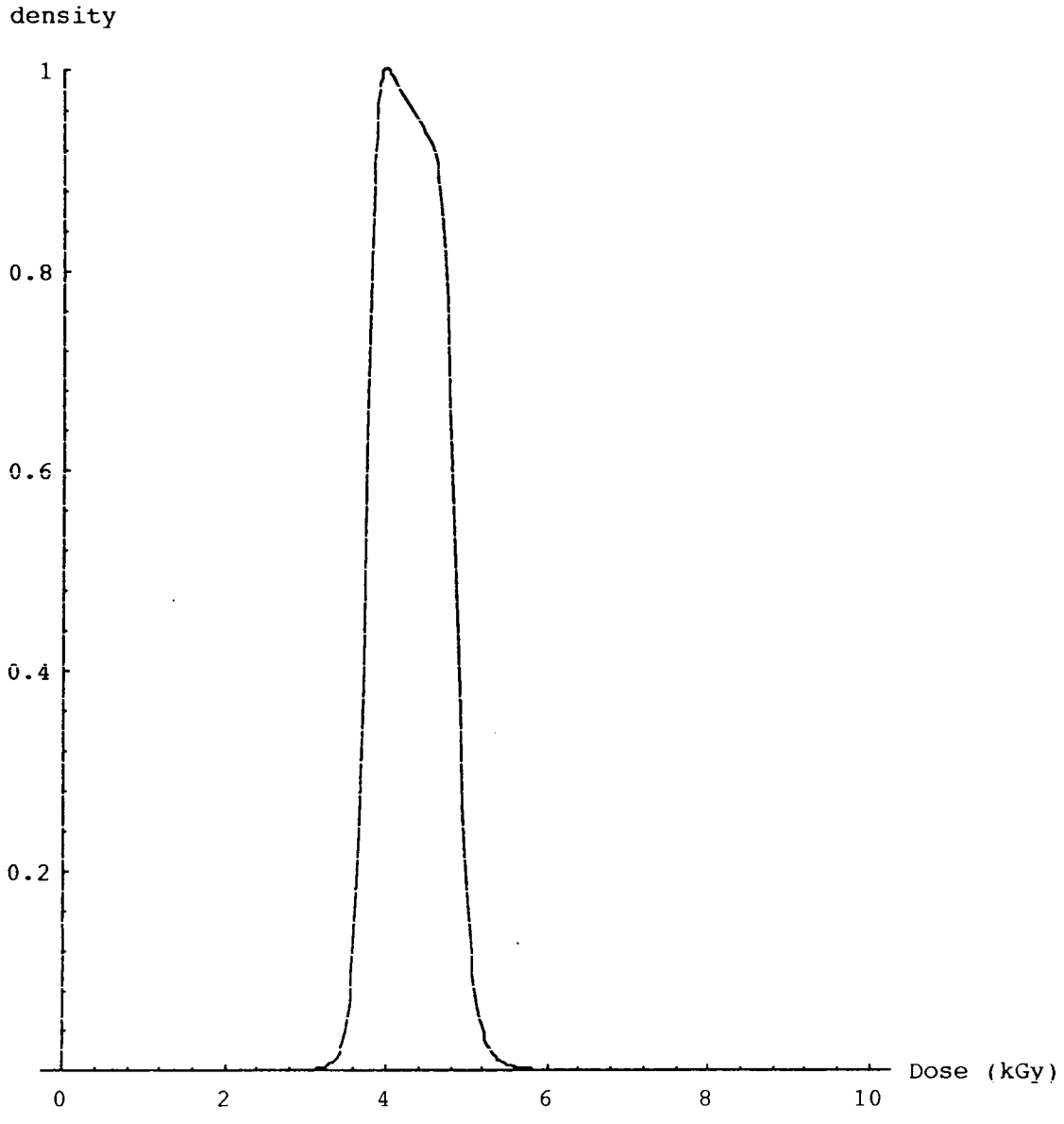


Figure 4. The calibrative density for combined data sets irradiated at 20 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

APPENDIX II

Calculation of the Predictive and Calibrative densities for study 5588

Tables 1 through 3 list the data obtained in study 5588. This data was used in calculating the predictive and calibrative densities. The following figures show the corresponding scatter plots as well as the densities obtained using Bayesian techniques. The same methods used in study 5564 were used in obtaining the results of study 5588.

Table 1 Data set for chicken meat irradiated at 20 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.62	9.78
1.80	4.78	5.20
3.60	1.58	1.52

Table 2 Data set for chicken meat irradiated at 0 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.74	9.57
0.90	7.73	8.19
3.60	1.74	1.88

Table 3 Data set for chicken meat irradiated at (-)20 °C in oxygen and in vacuum

Dose (kGy)	Air Pop (log CFU/g)	Vac. Pop (log CFU/g)
0.0	9.62	9.62
1.80	7.37	7.83
3.60	3.62	3.83

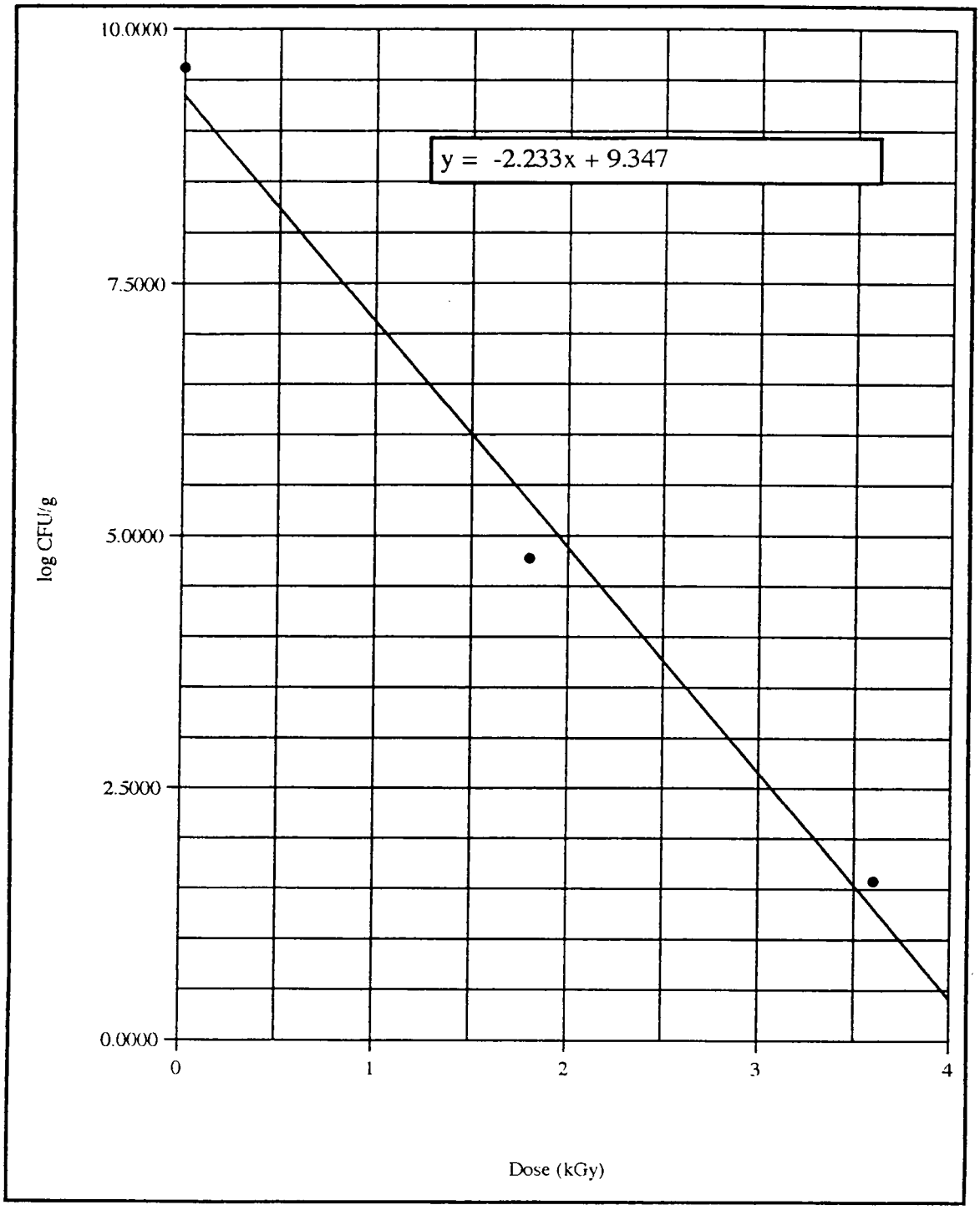


Figure 1 Scatter plot for mechanically deboned chicken meat irradiated at 20 °C and in the presence of air

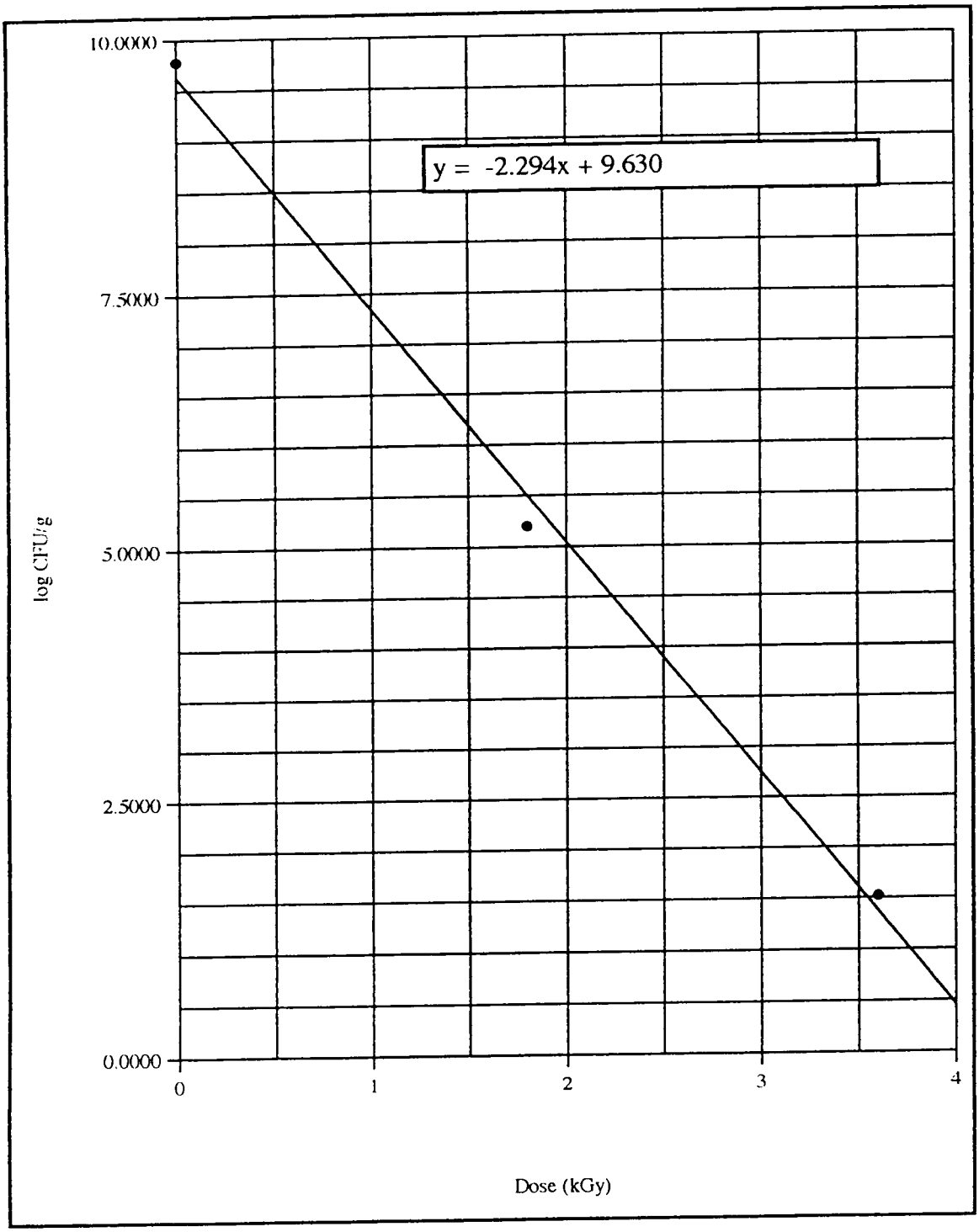


Figure 2 Scatter plot for mechanically deboned chicken meat irradiated at 20 °C and in the absence of air

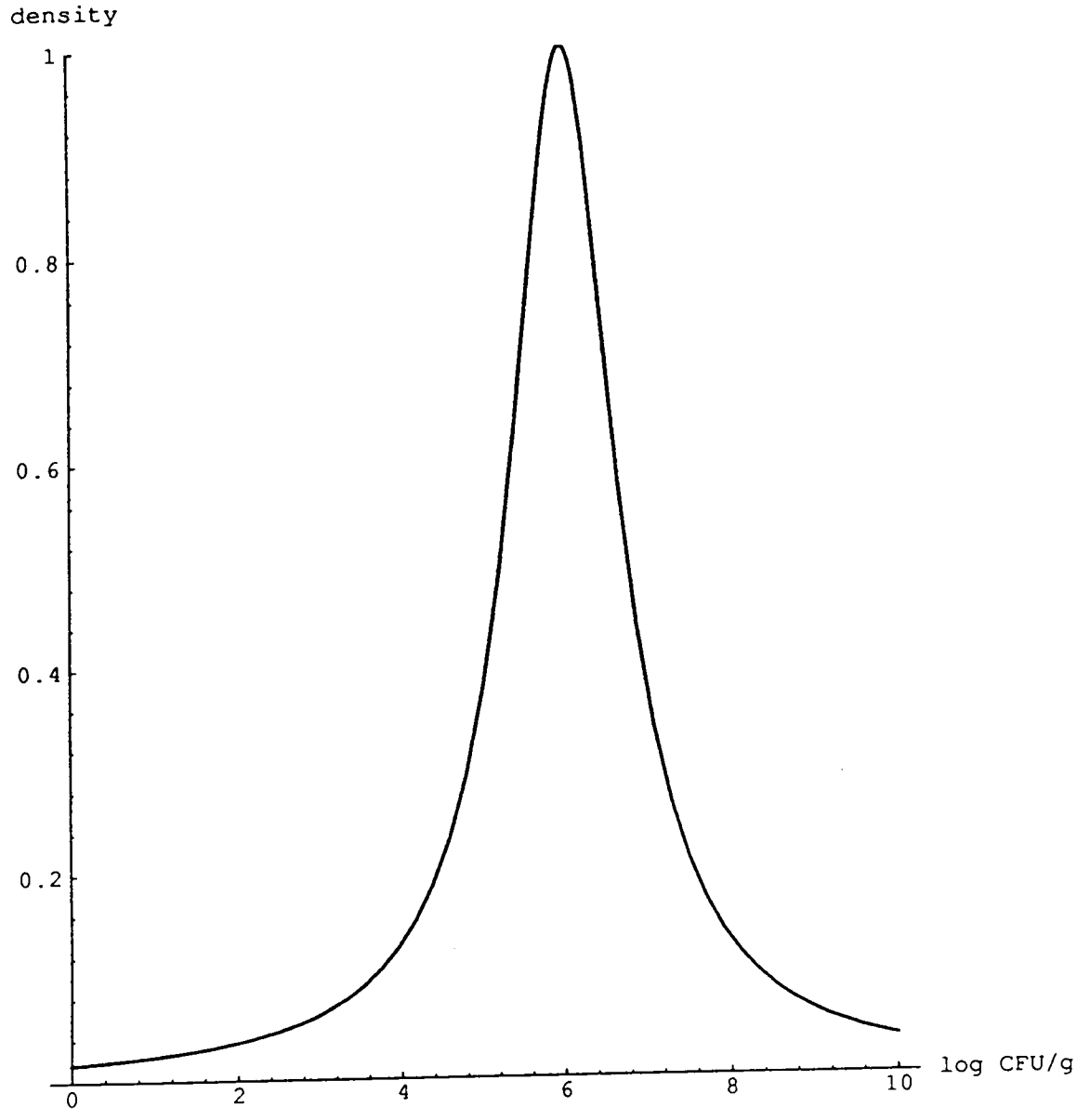


Figure 3 The predictive density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed dose of 1.5 kGy

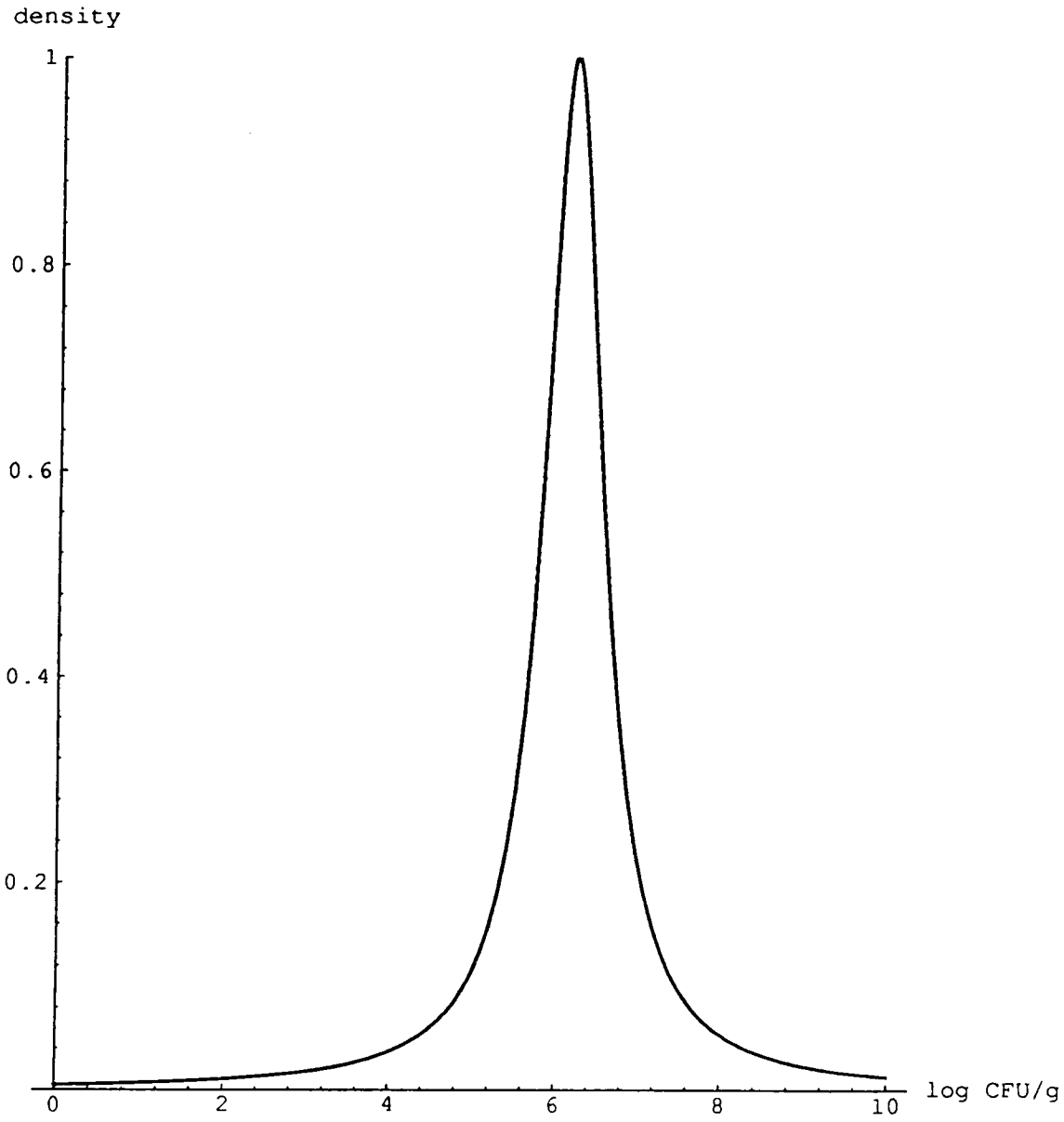


Figure 4 The predictive density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed dose of 1.5 kGy

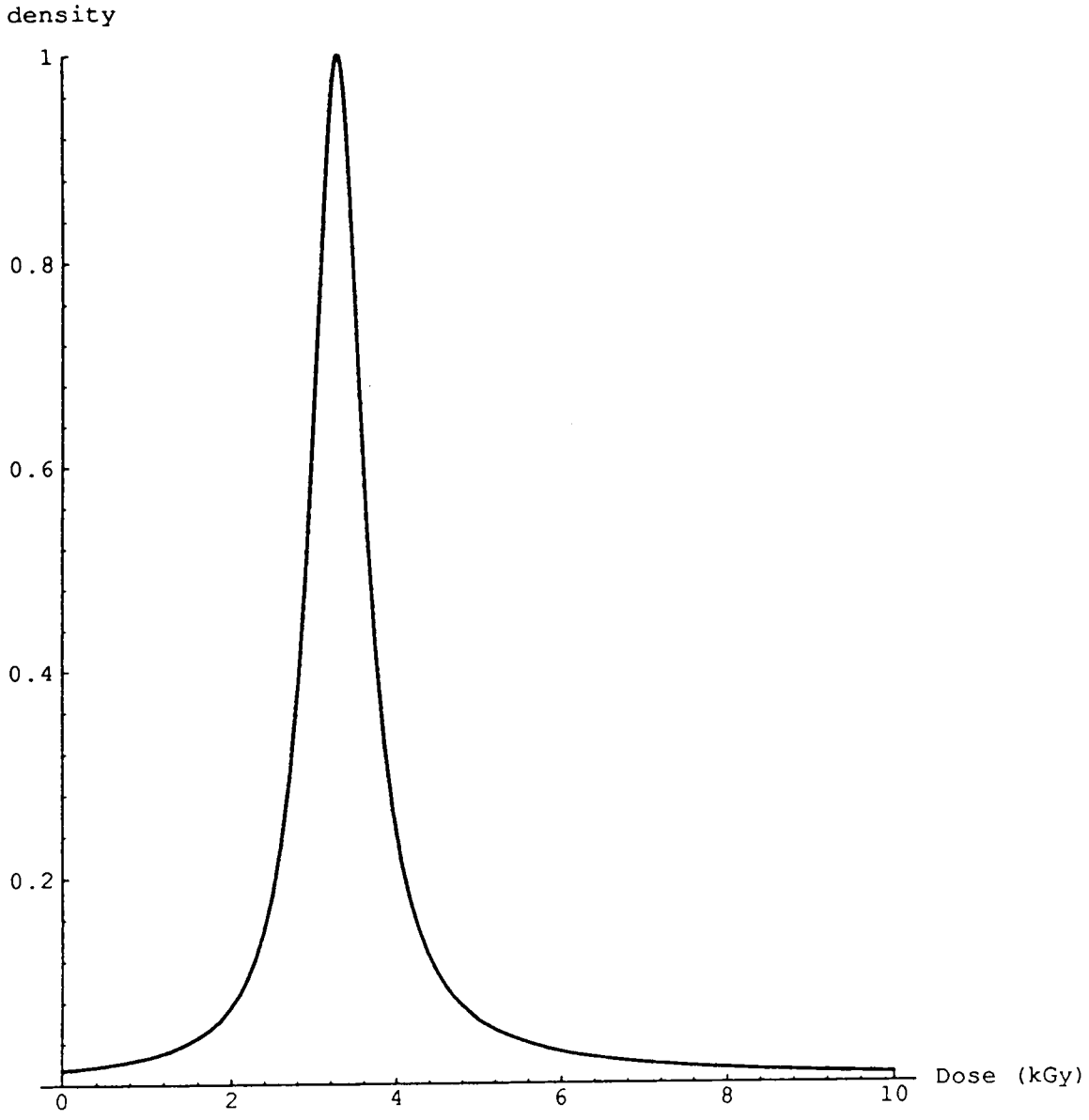


Figure 5 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed bacteria population of 100 CFU/g

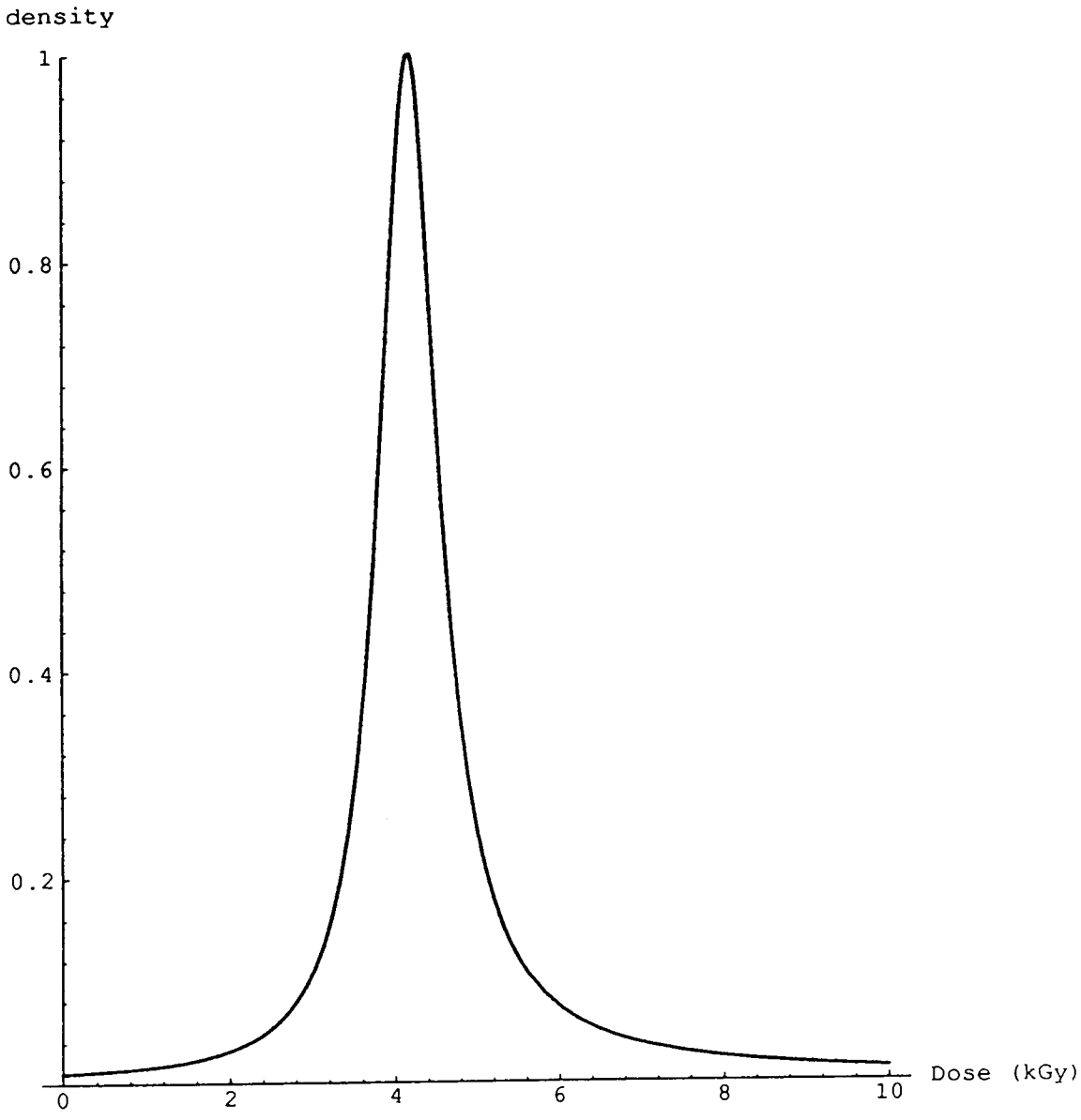


Figure 6 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

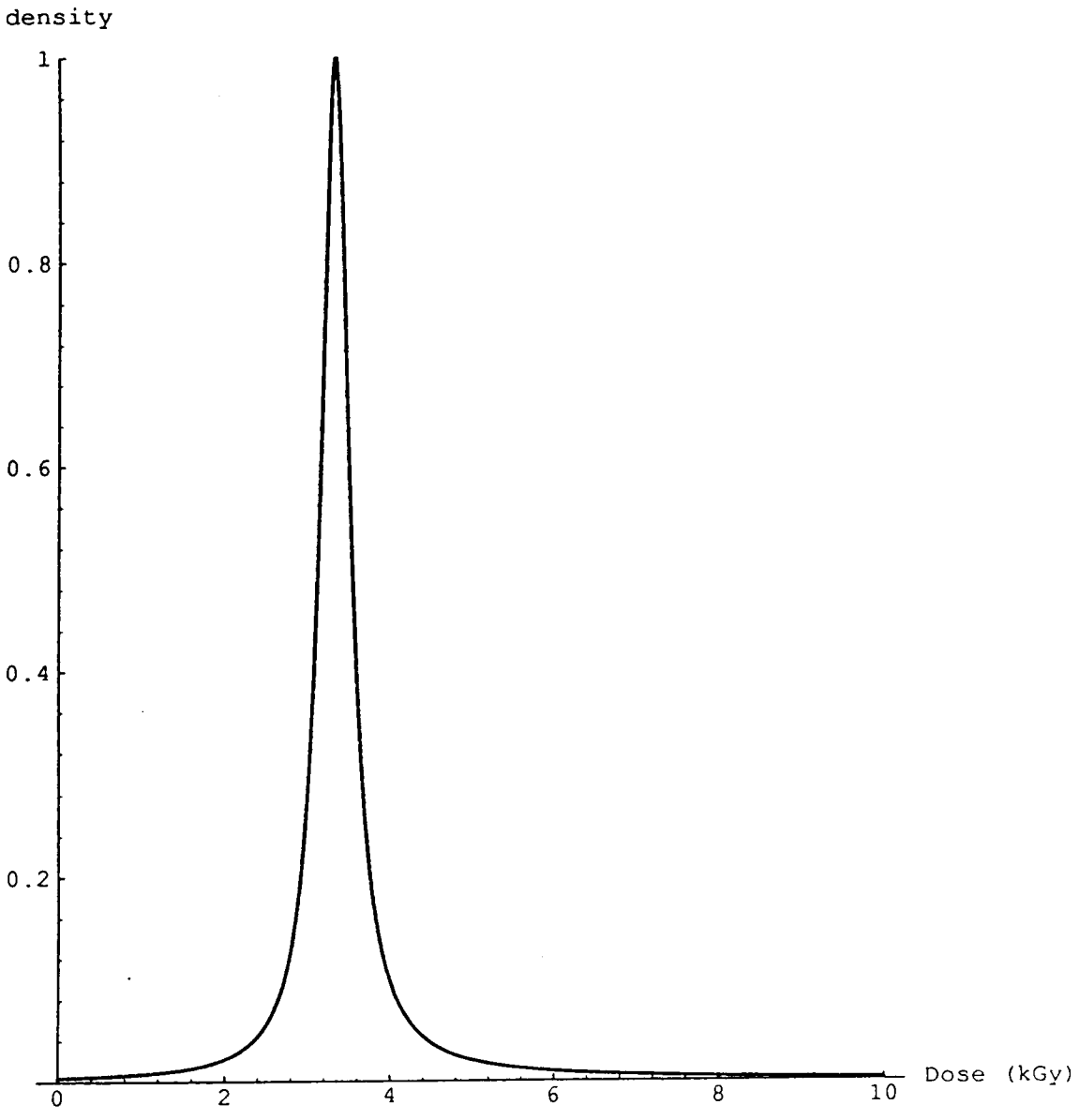


Figure 7 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed bacteria population of 100 CFU/g

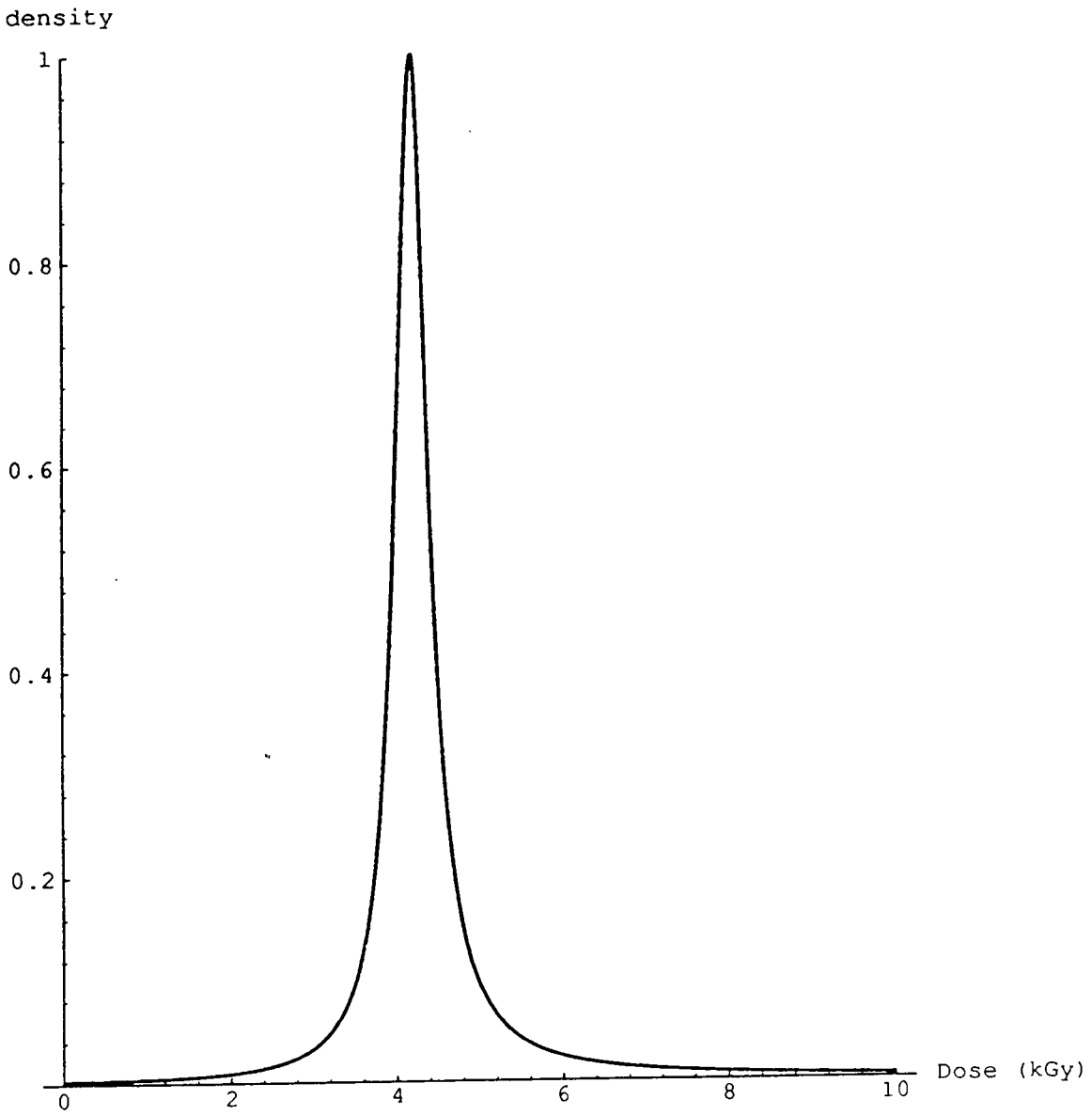


Figure 8 The calibrative density for mechanically deboned chicken meat irradiated at 20 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

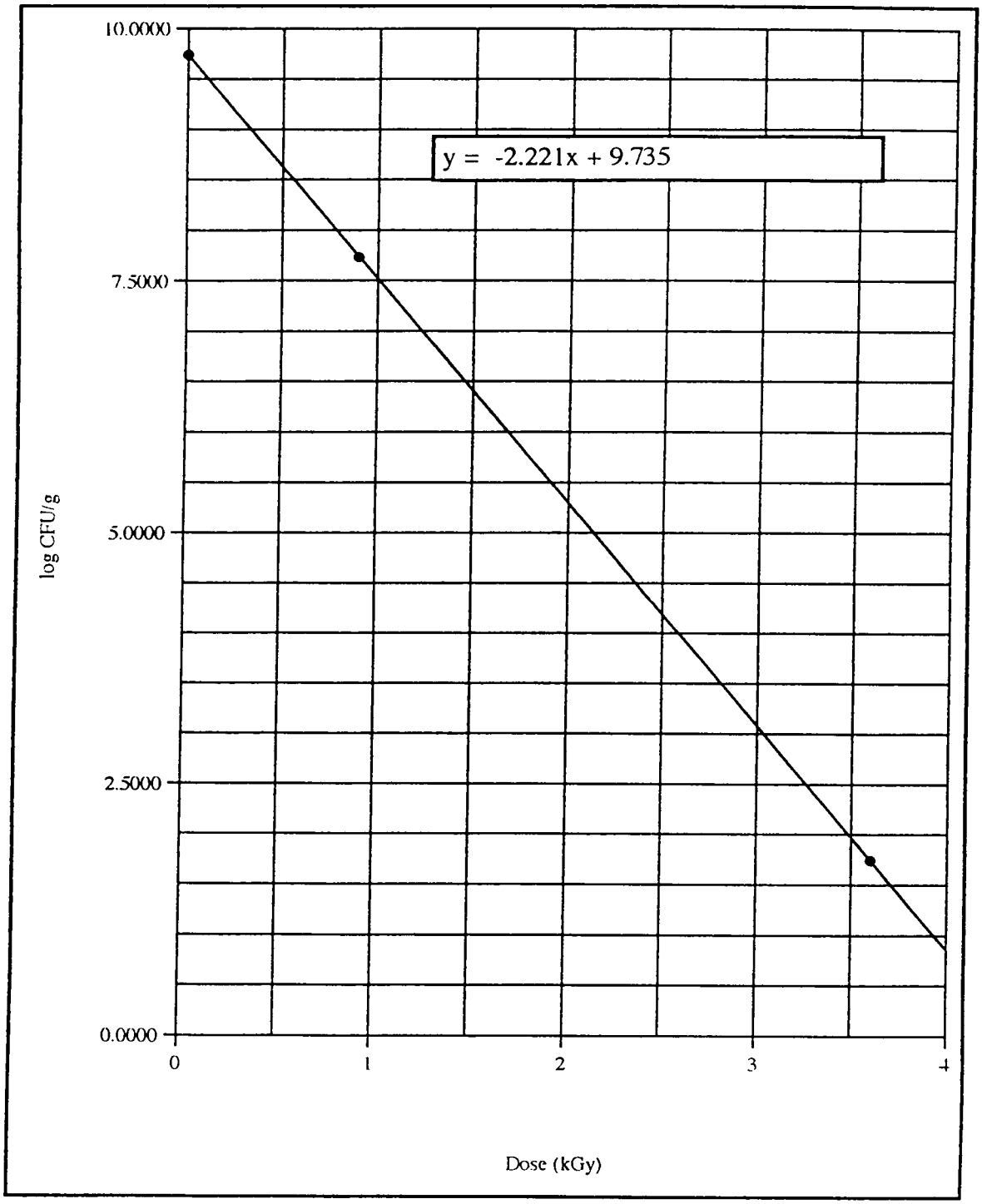


Figure 9 Scatter plot for mechanically deboned chicken meat irradiated at 0 °C and in the presence of air

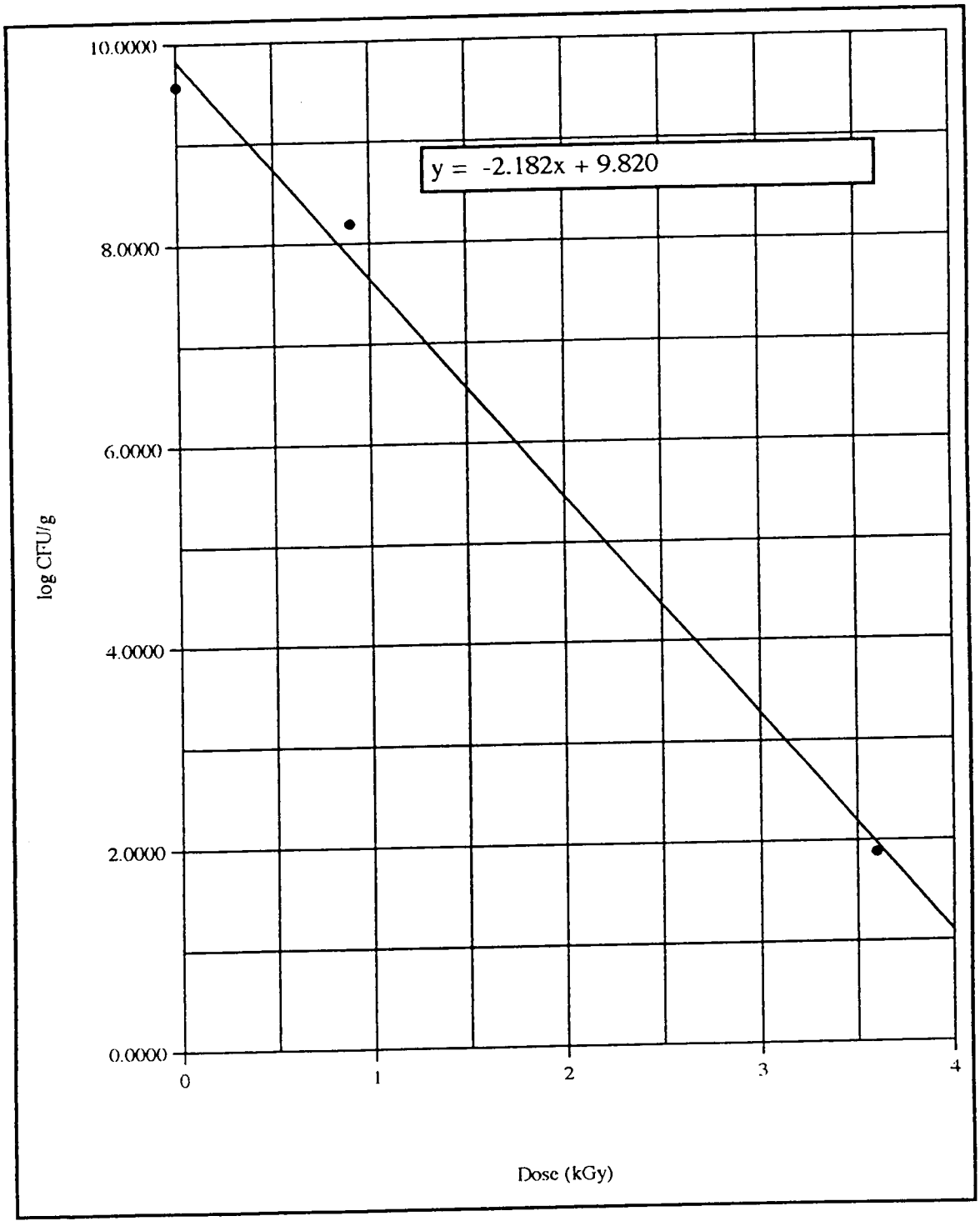


Figure 10 Scatter plot for mechanically deboned chicken meat irradiated at 0 °C and in the absence of air

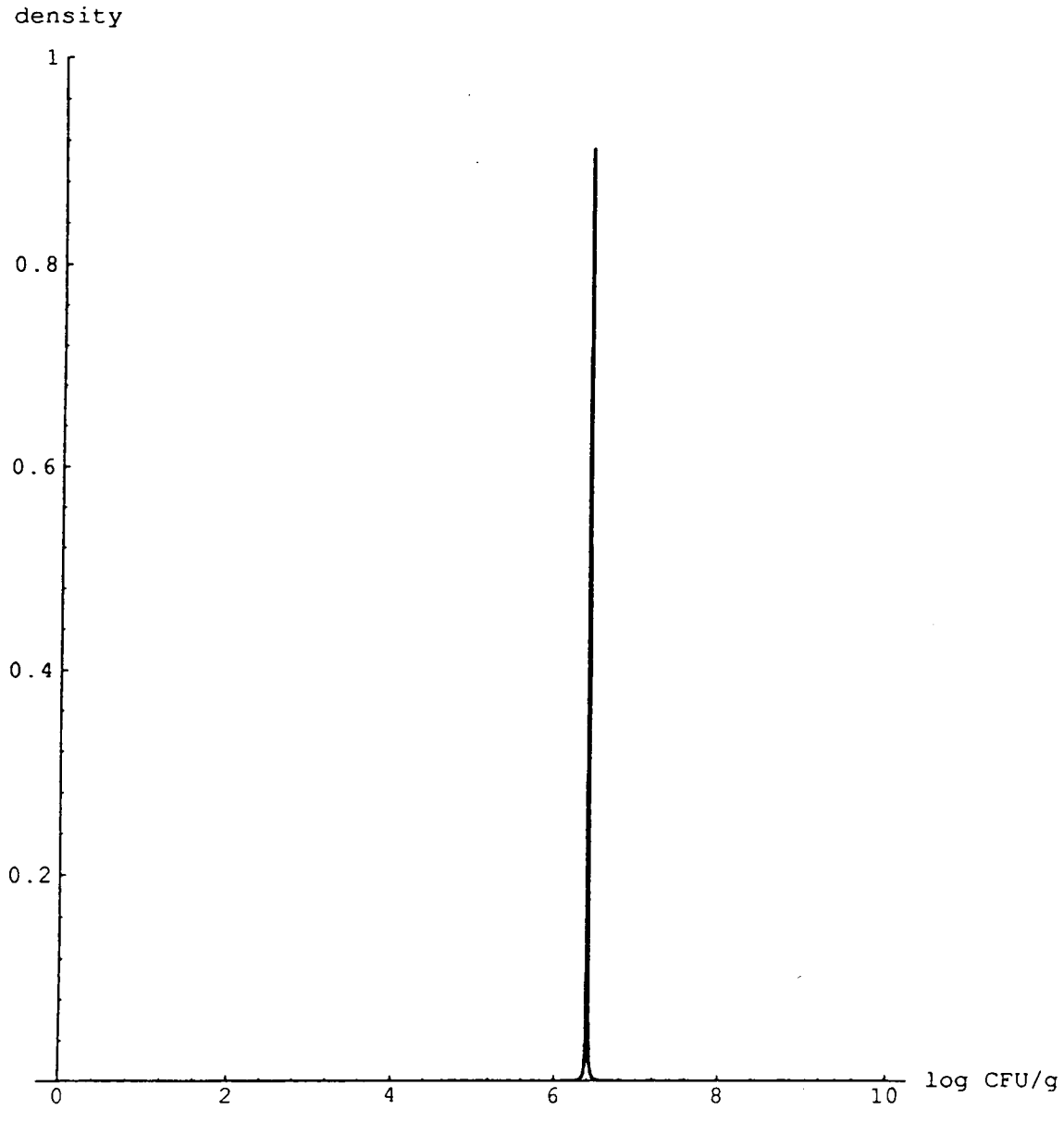


Figure 11 The predictive density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed dose of 1.5 kGy

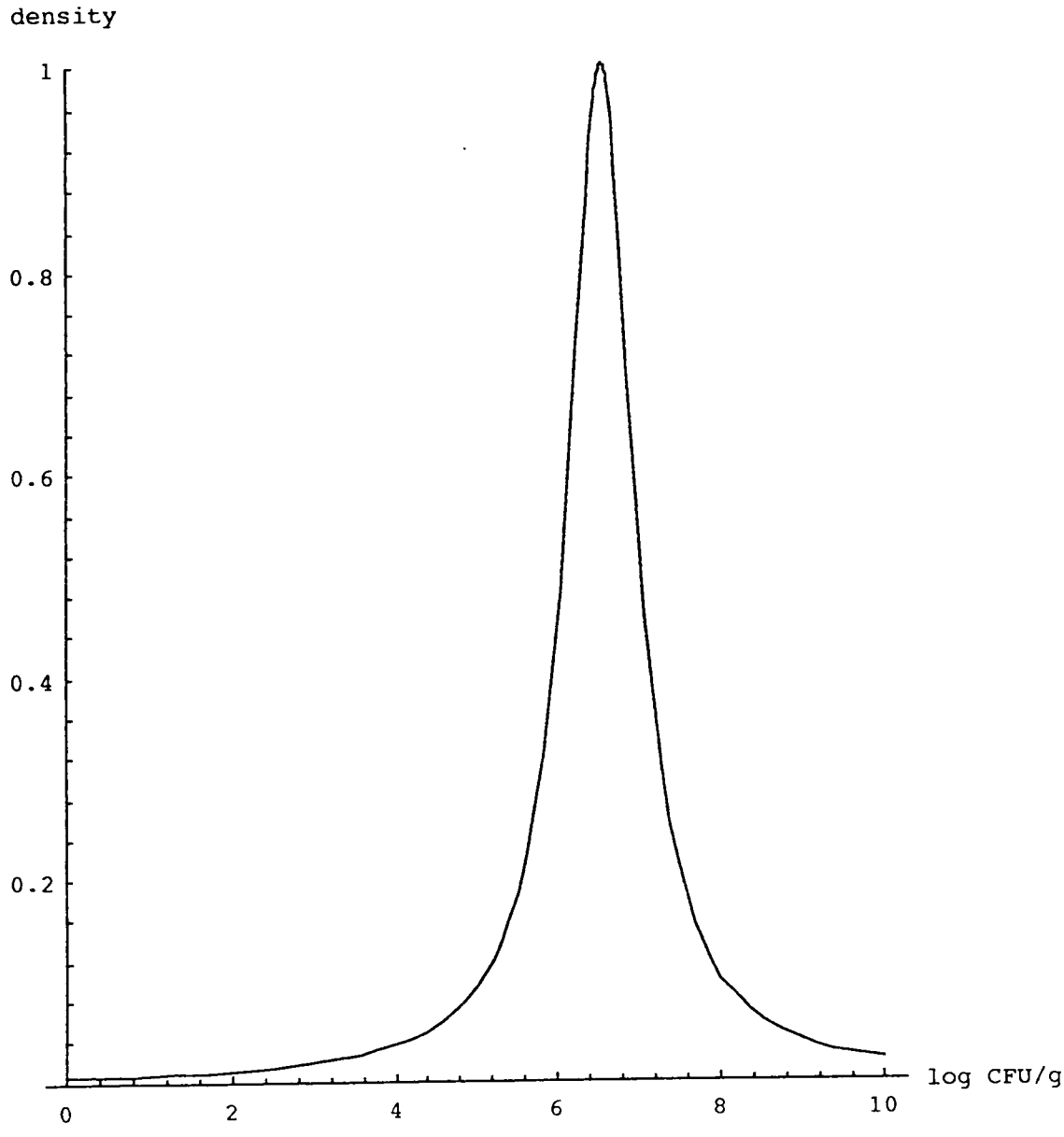


Figure 12 The predictive density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed dose of 1.5 kGy

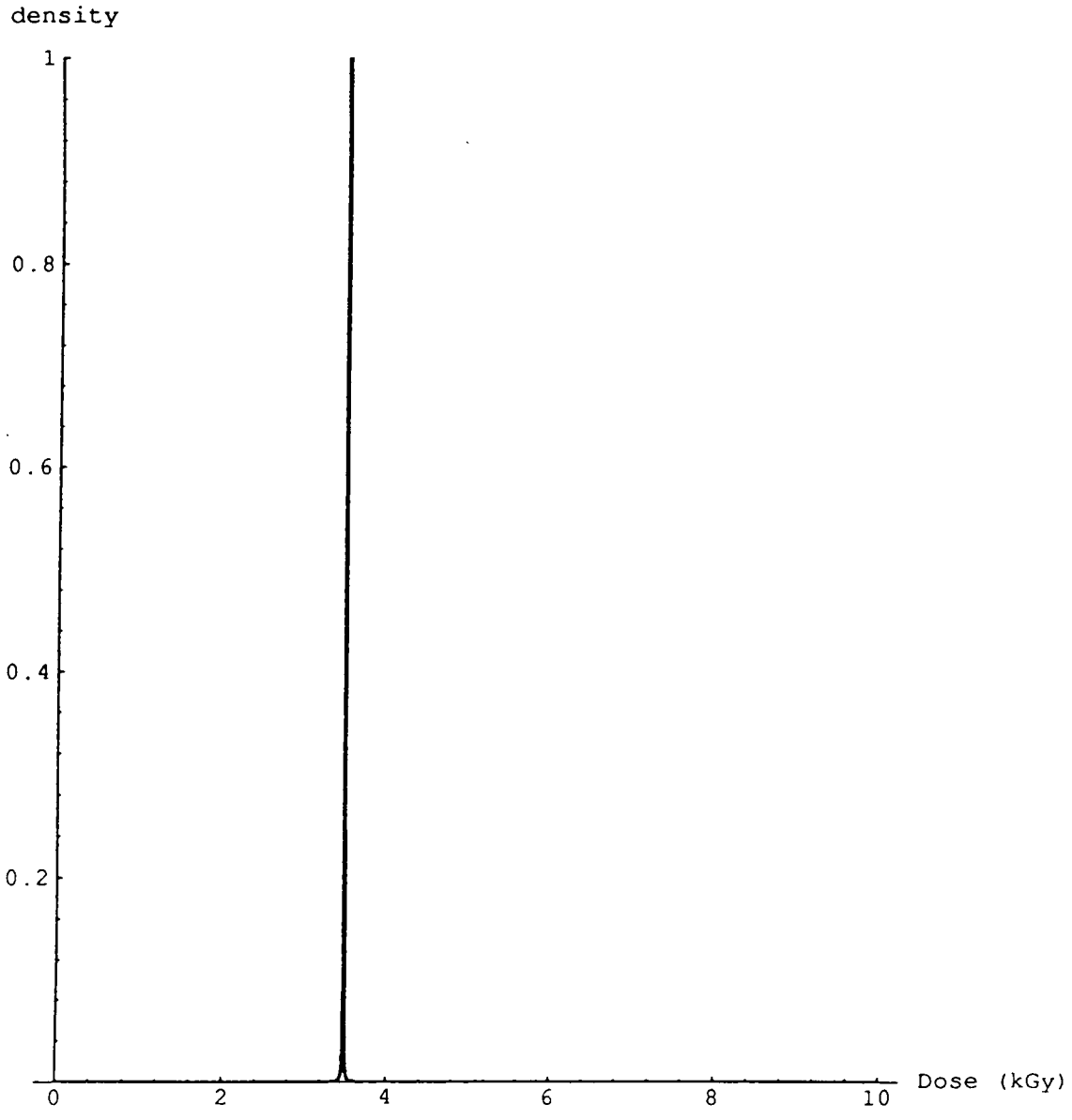


Figure 13 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed bacteria population of 100 CFU/g

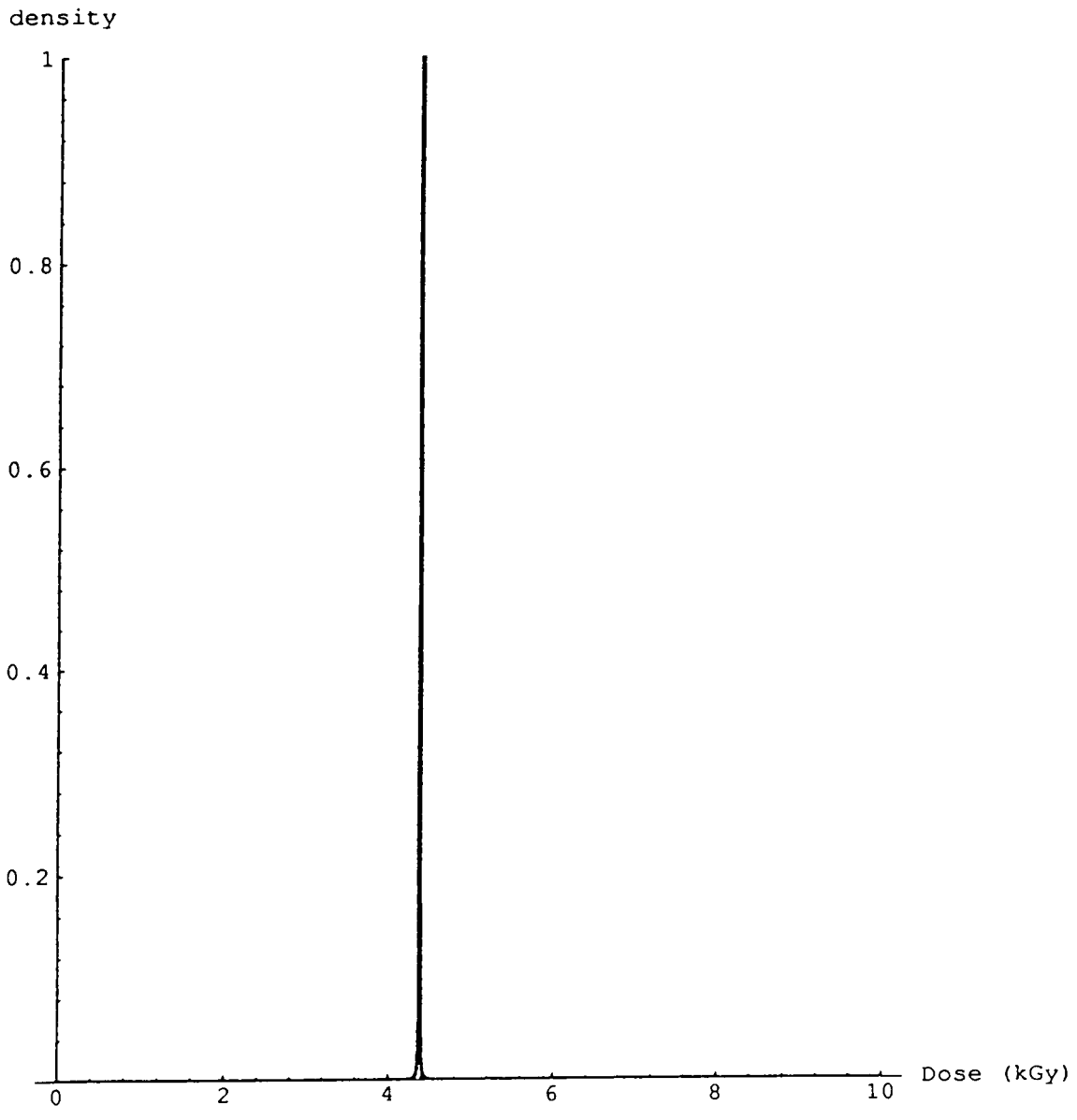


Figure 14 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

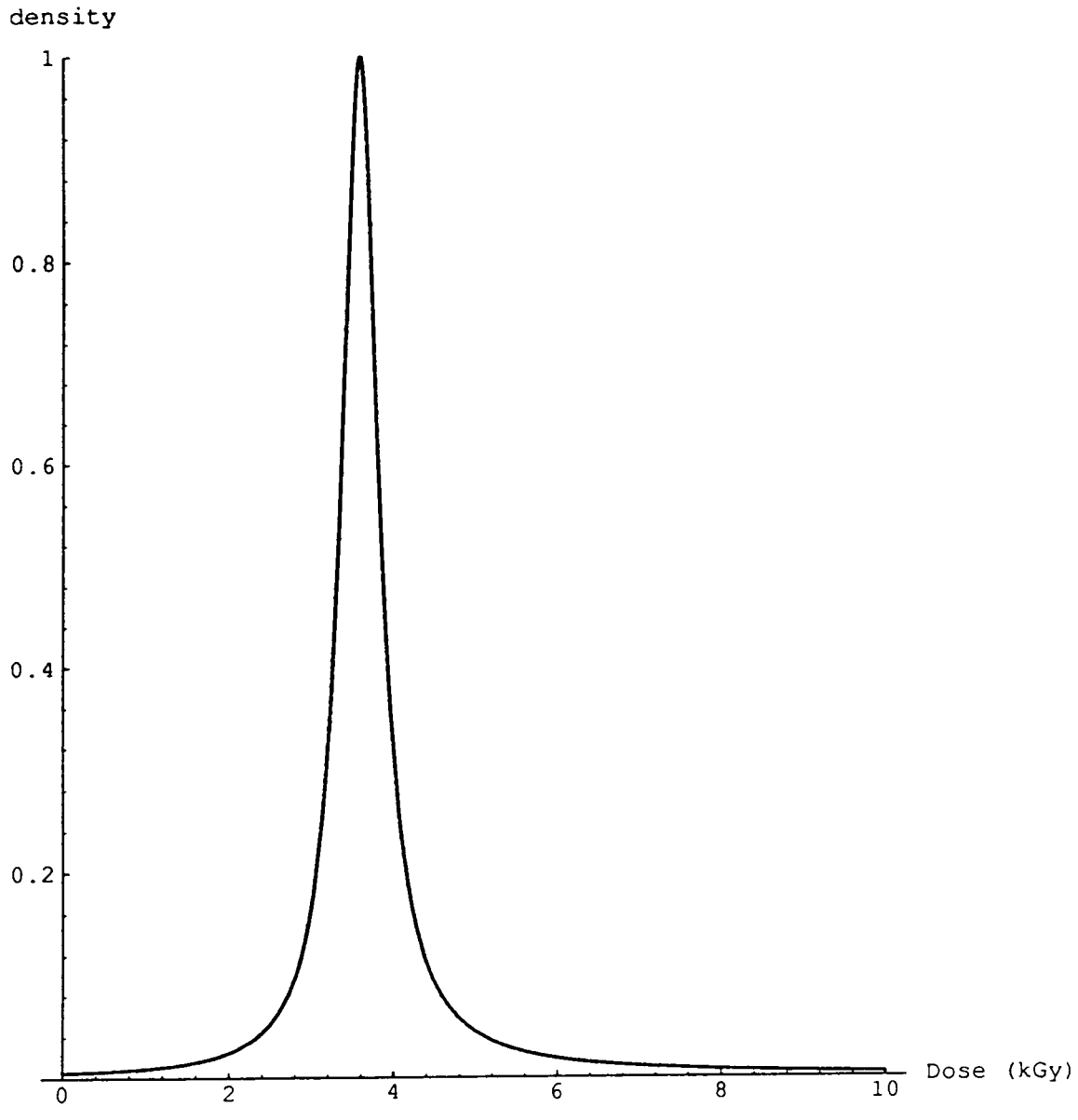


Figure 15 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed bacteria population of 100 CFU/g

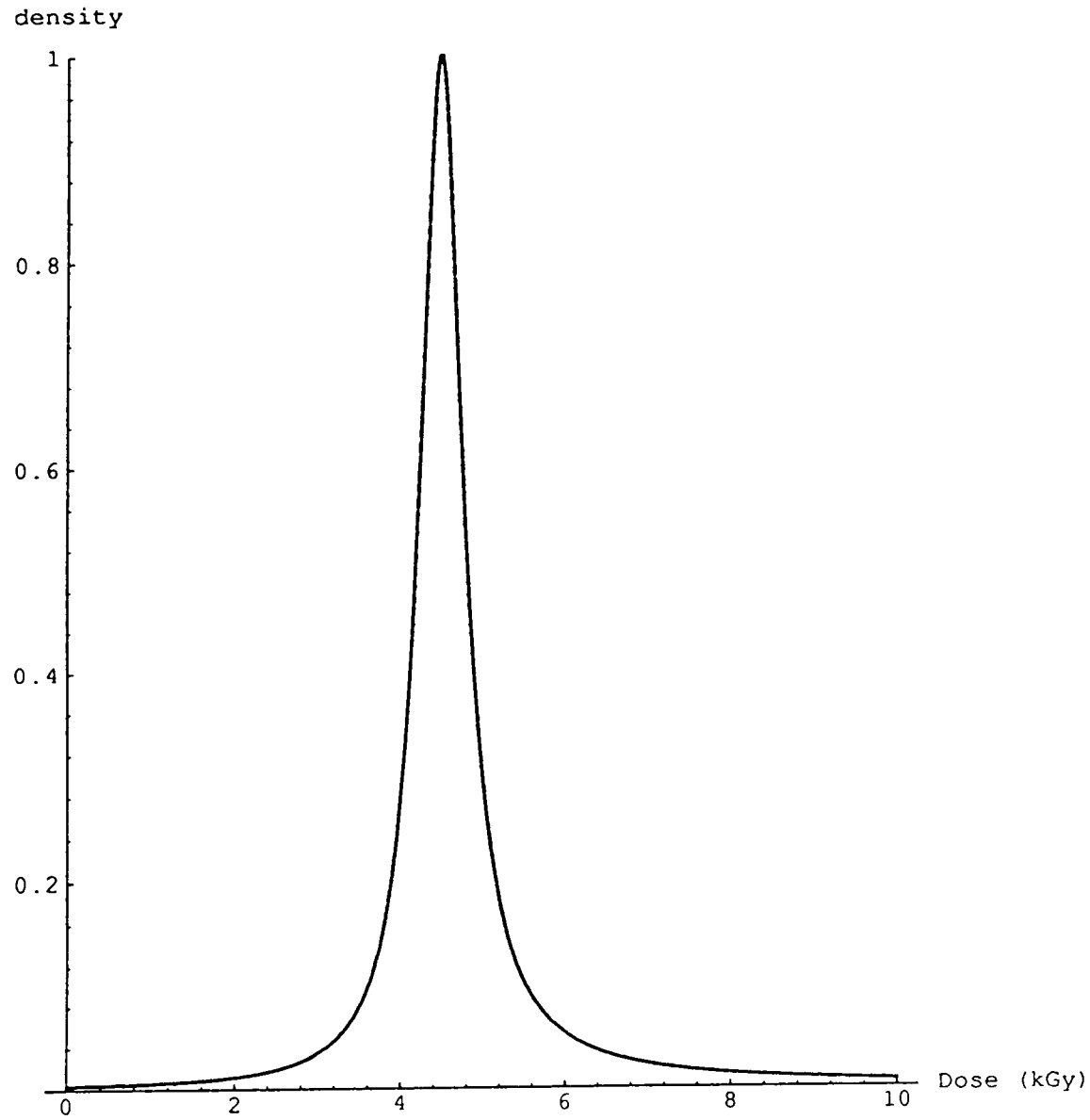


Figure 16 The calibrative density for mechanically deboned chicken meat irradiated at 0 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

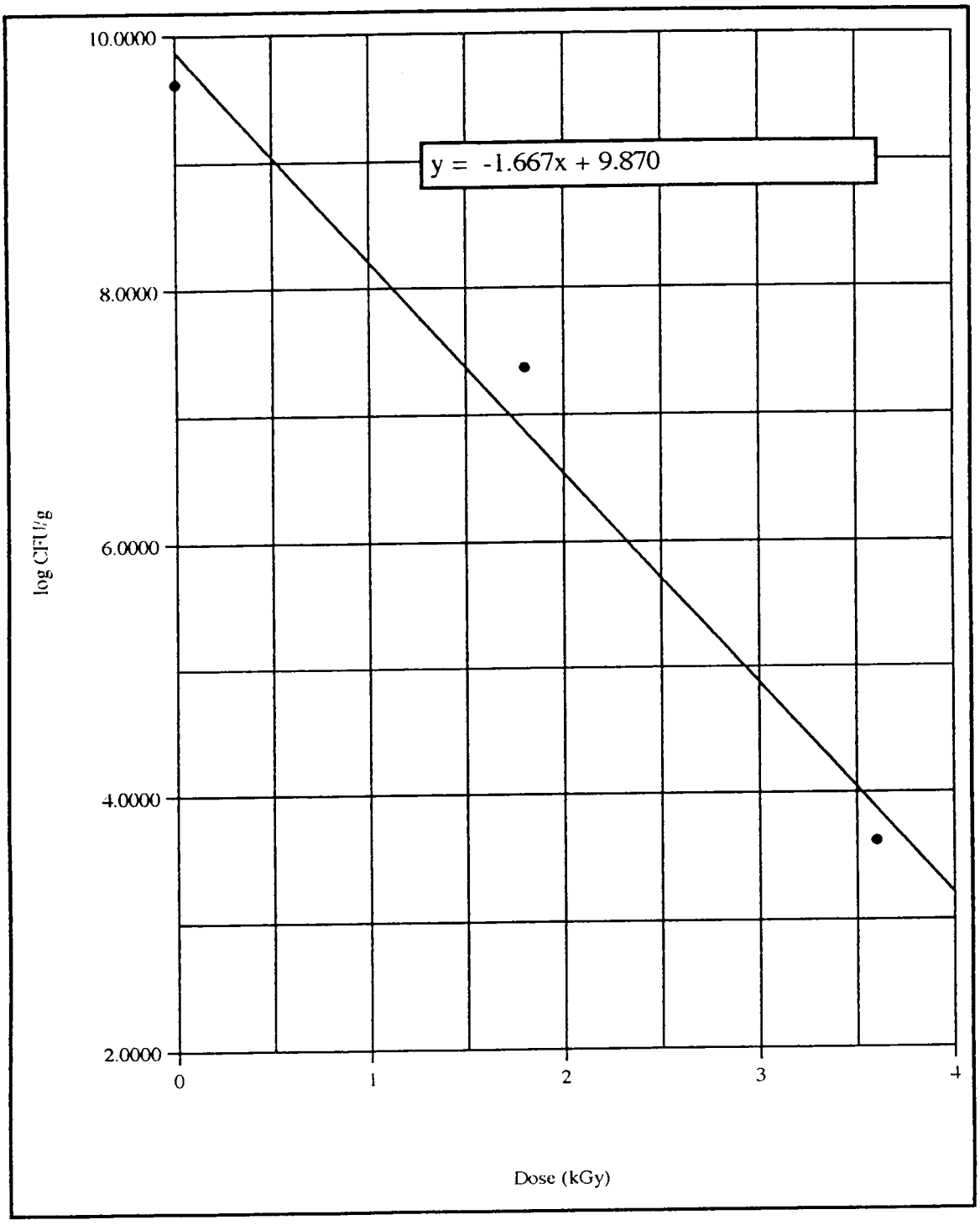


Figure 17 Scatter plot for mechanically deboned chicken meat irradiated at (-)20 °C and in the presence of air

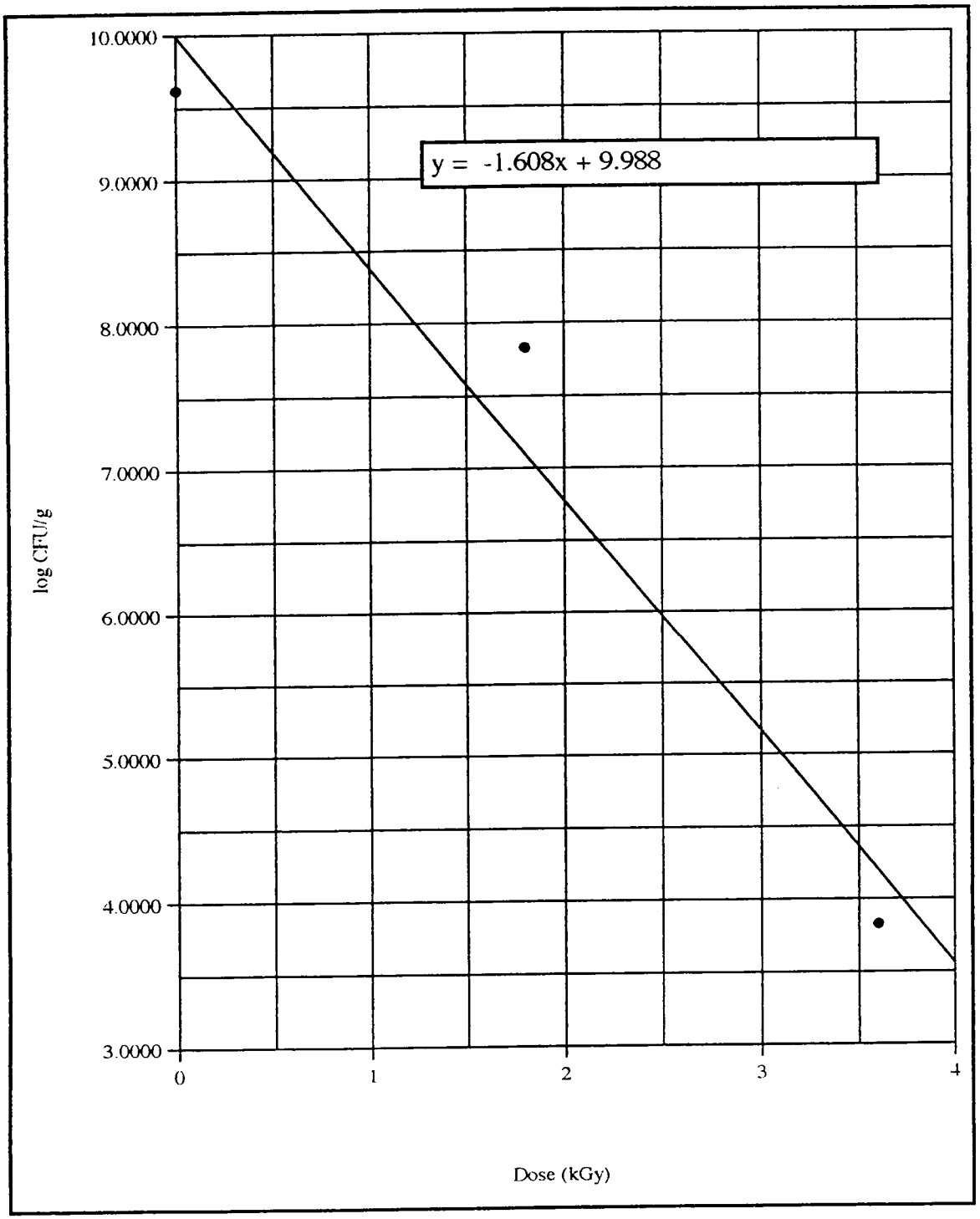


Figure 18 Scatter plot for mechanically deboned chicken meat irradiated at (-)20 °C and in the absence of air

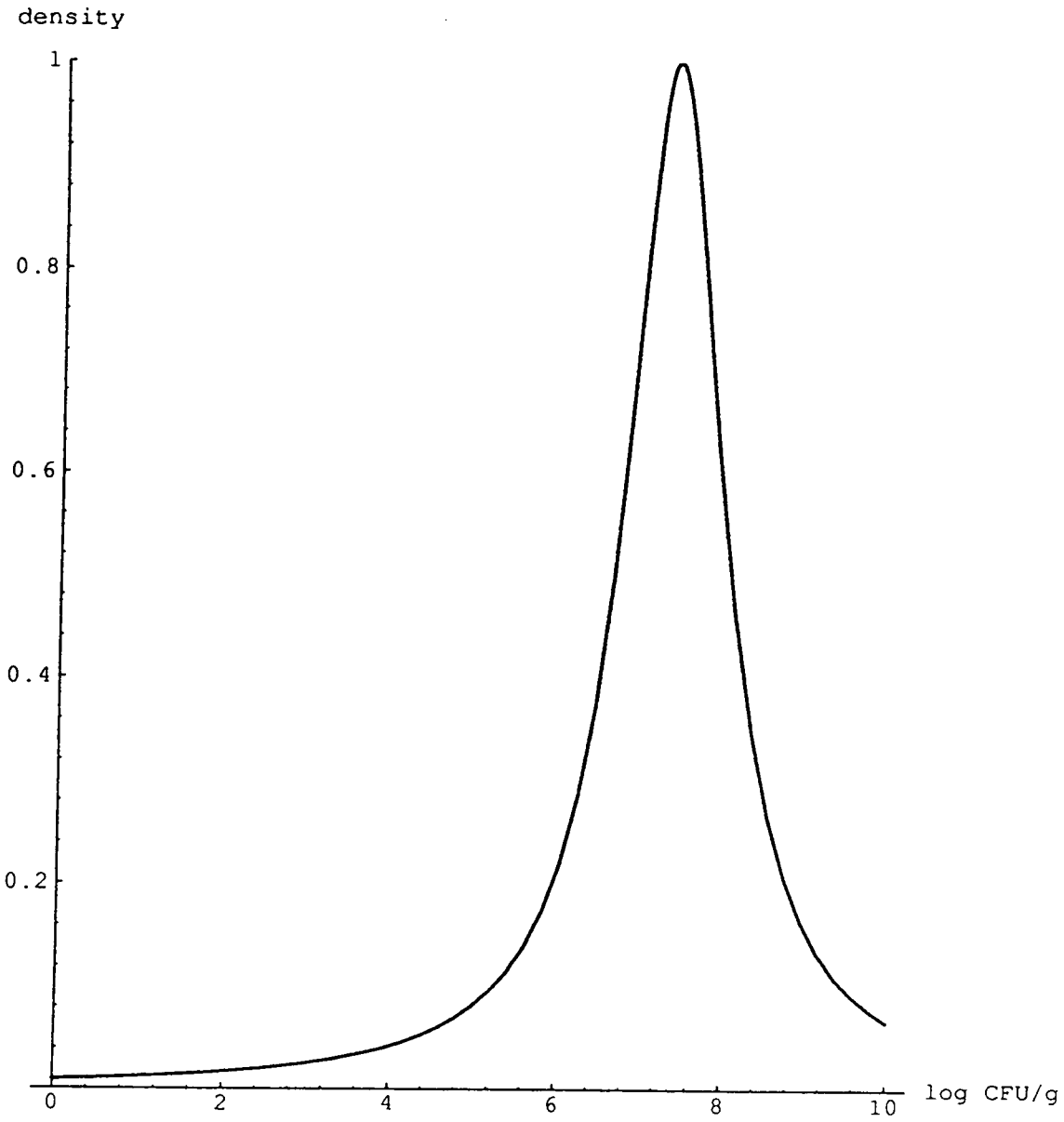


Figure 19 The predictive density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed dose of 1.5 kGy

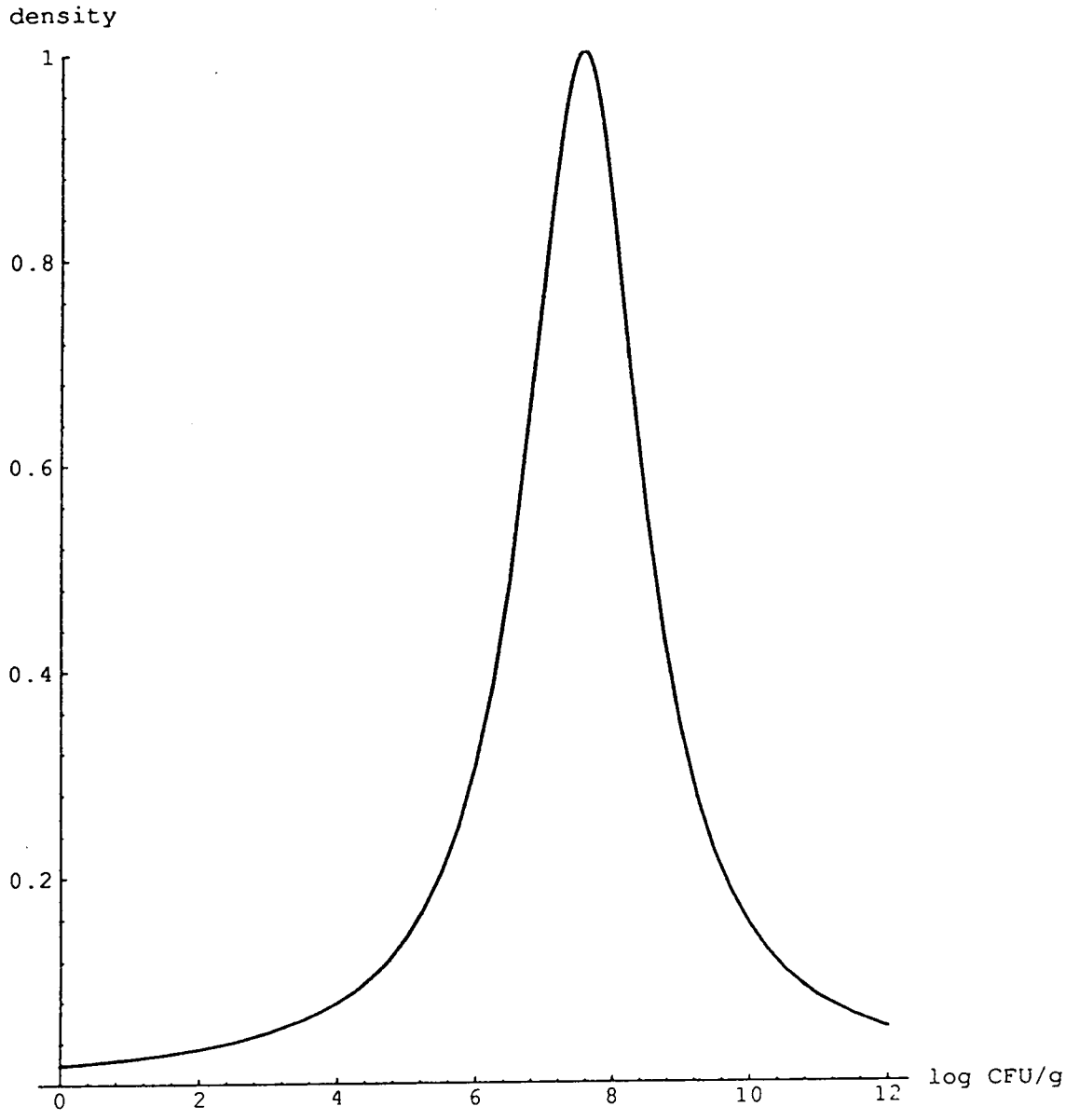


Figure 20 The predictive density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed dose of 1.5 kGy

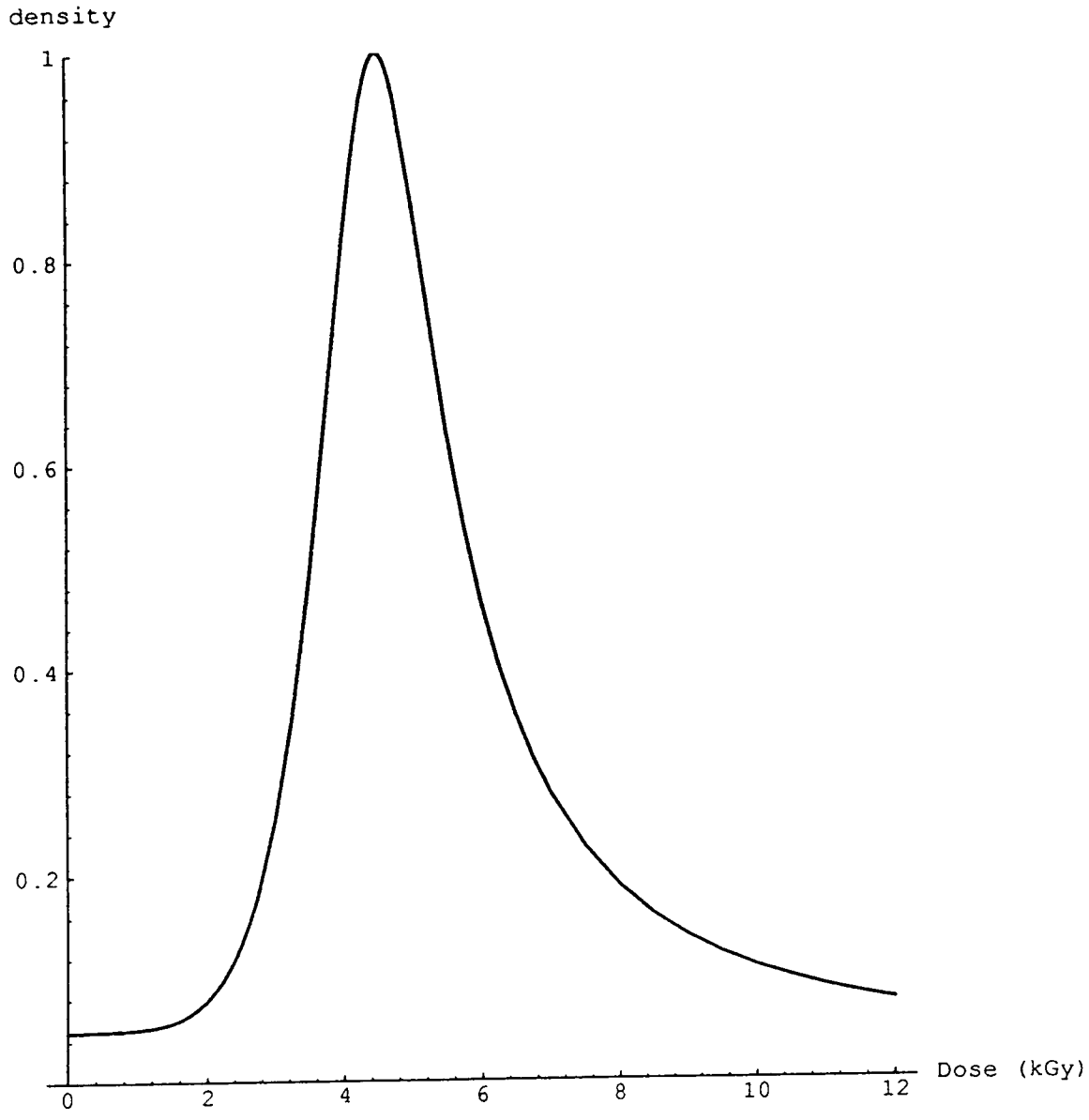


Figure 21 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed bacteria population of 100 CFU/g

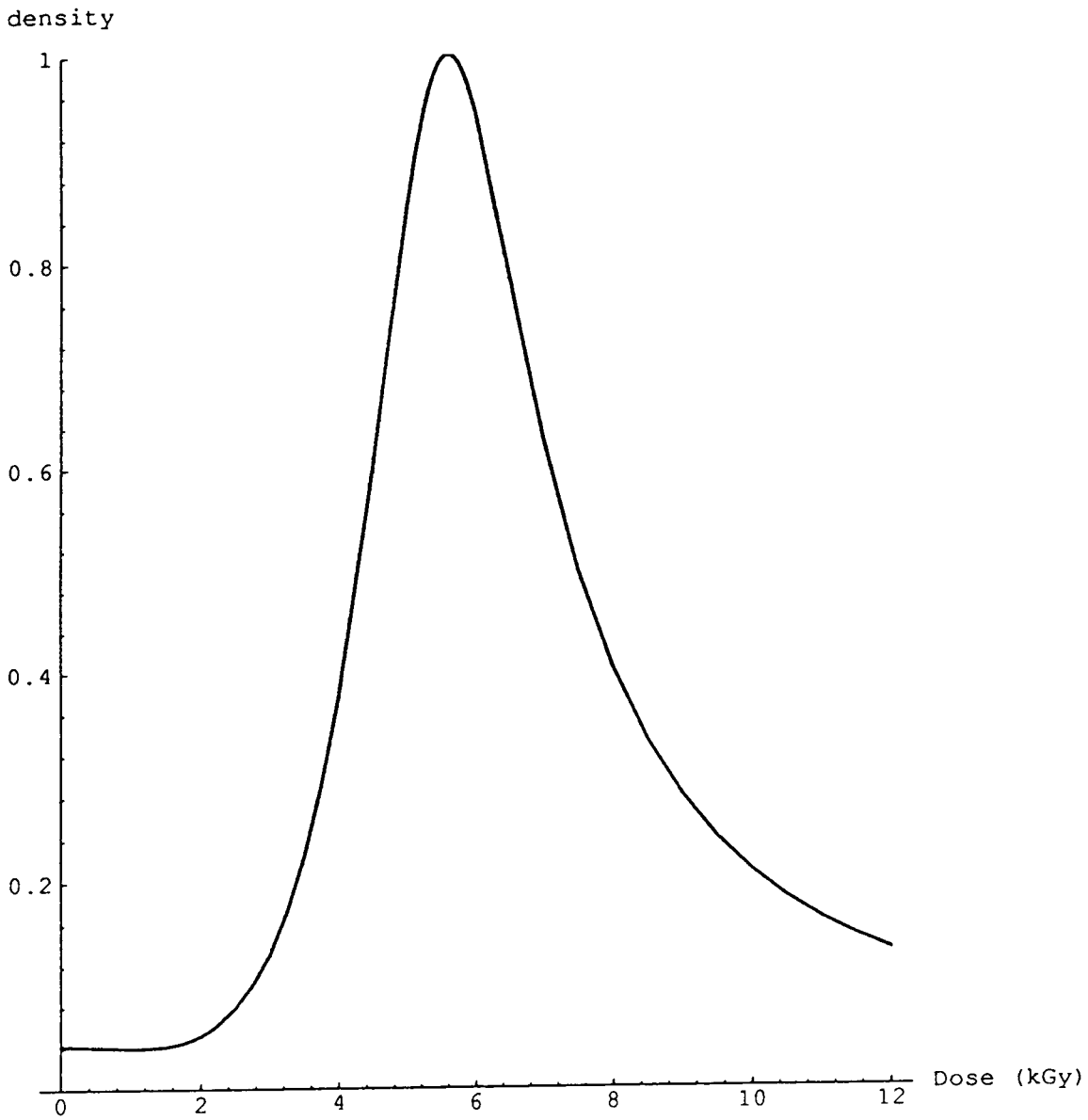


Figure 22 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the presence of air and a fixed bacteria population of 1 CFU/g

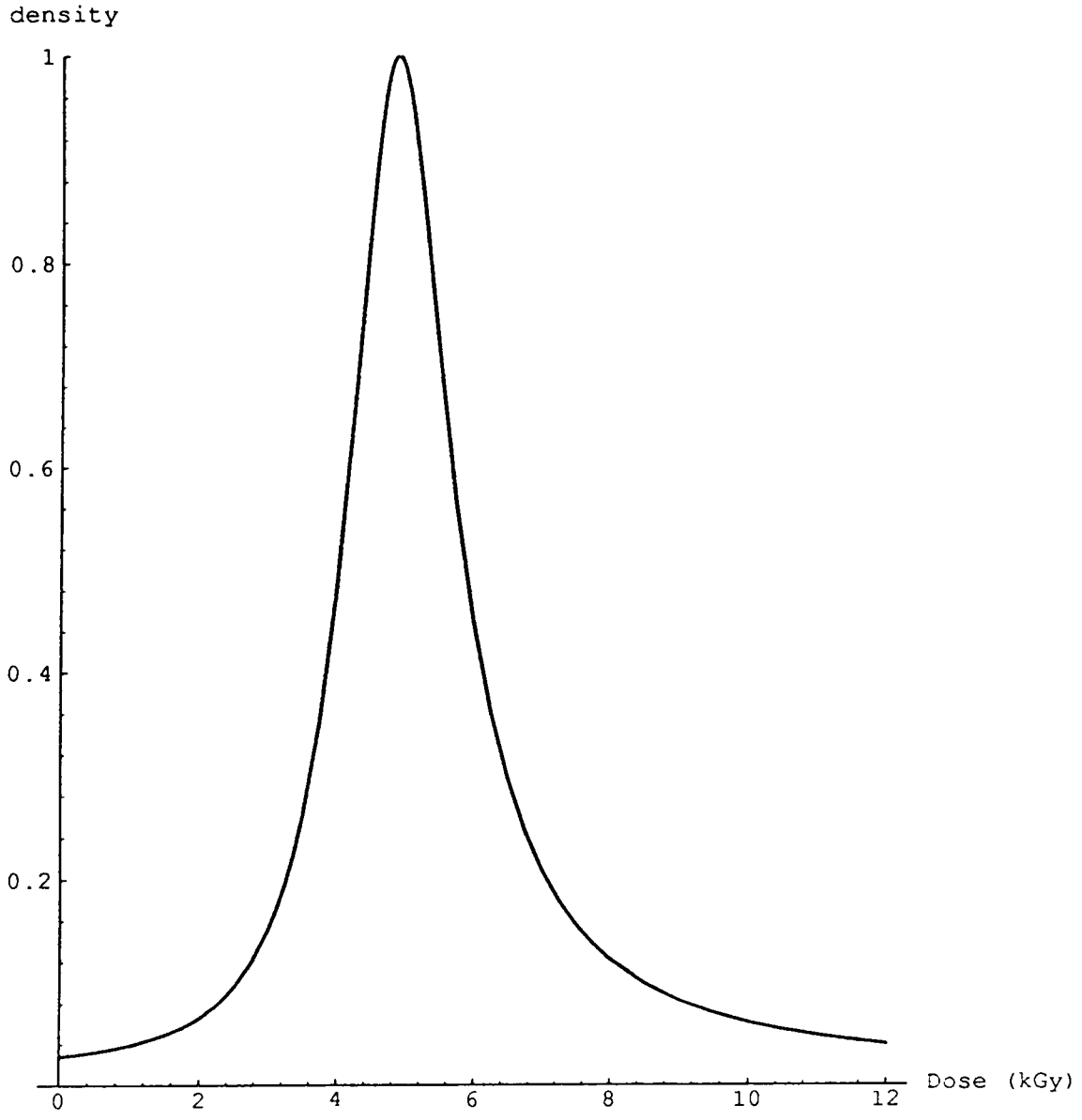


Figure 23 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed bacteria population of 100 CFU/g

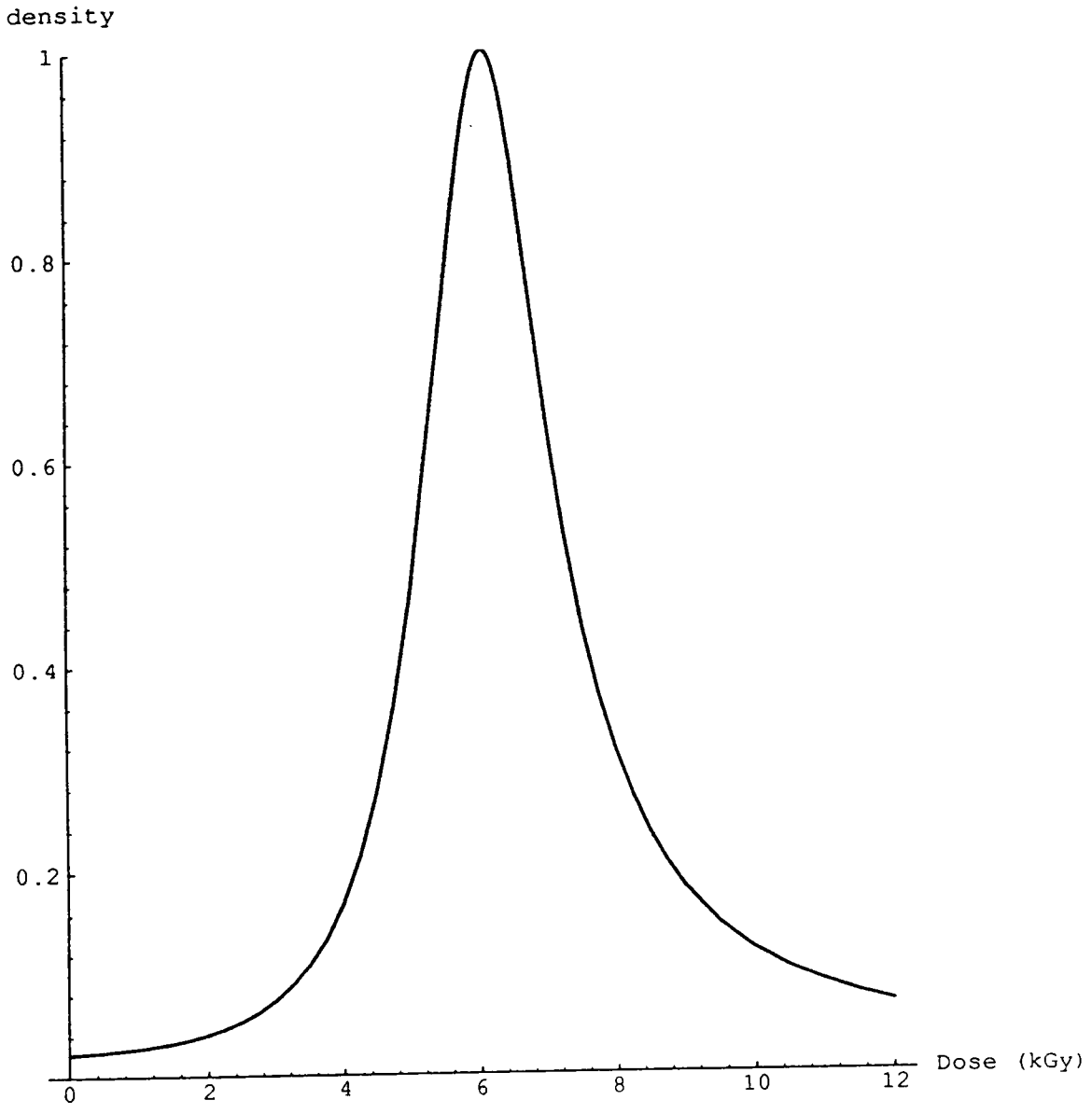
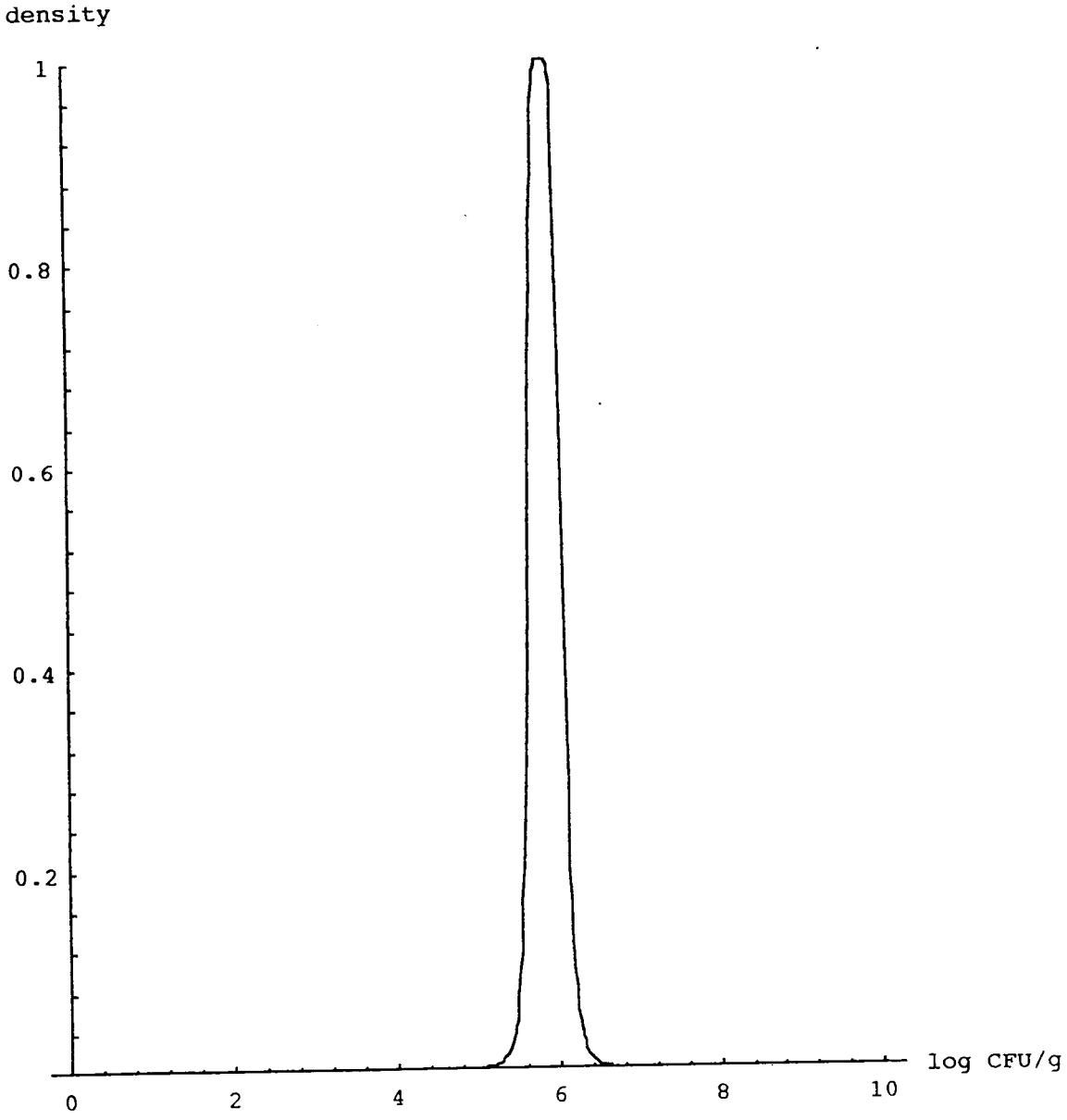


Figure 24 The calibrative density for mechanically deboned chicken meat irradiated at (-)20 °C, in the absence of air and a fixed bacteria population of 1 CFU/g

APPENDIX III

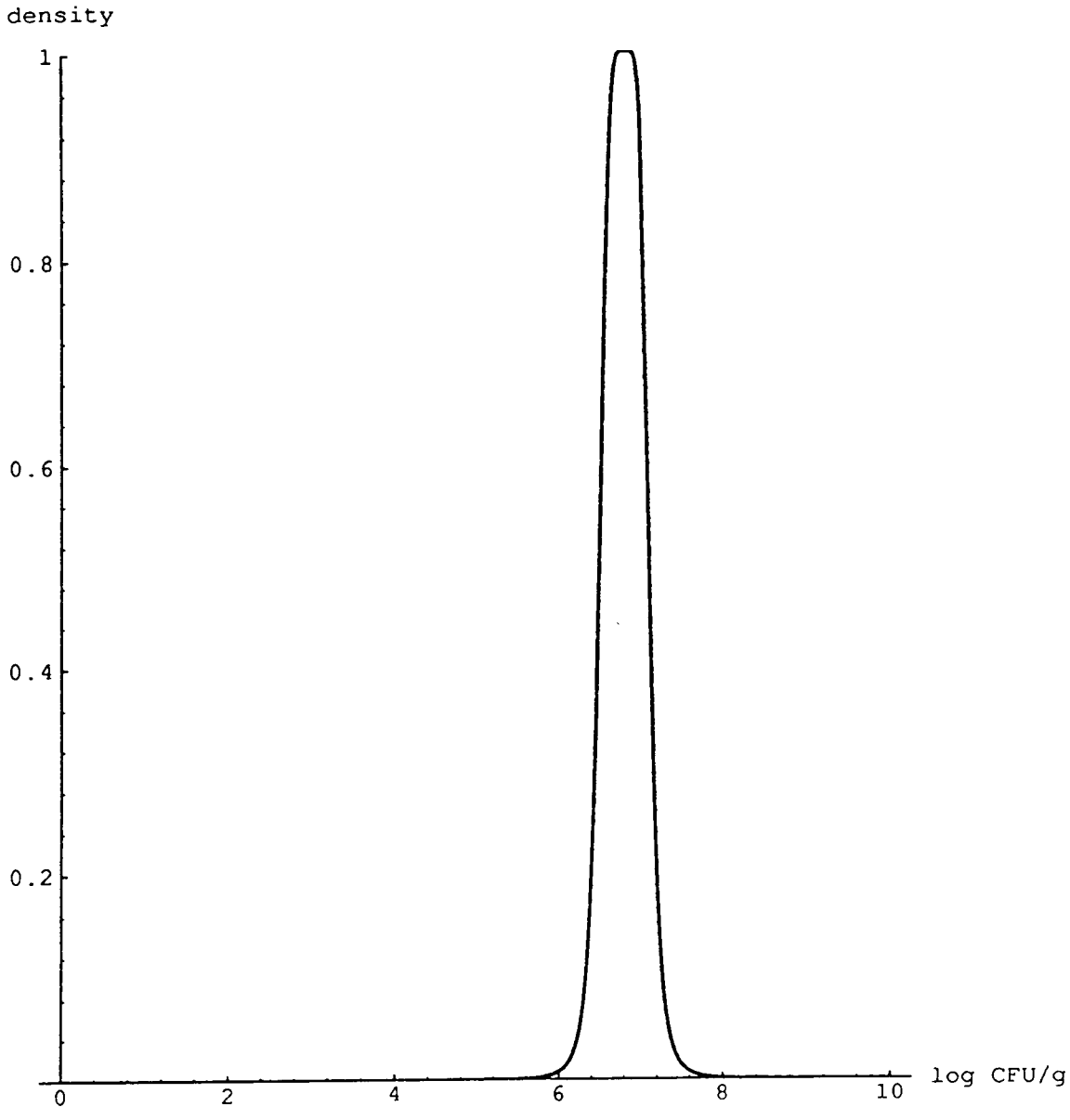
Mathematica programs

```
f[st_]:=  (1/((Beta[.5,2])*(Sqrt[4*0.01176])*(1+((u-5.859)^2
/(4*0.01176))^2.5))/3.45802)
Plot[f[st_],  {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log  CFU/g","density"}]
```



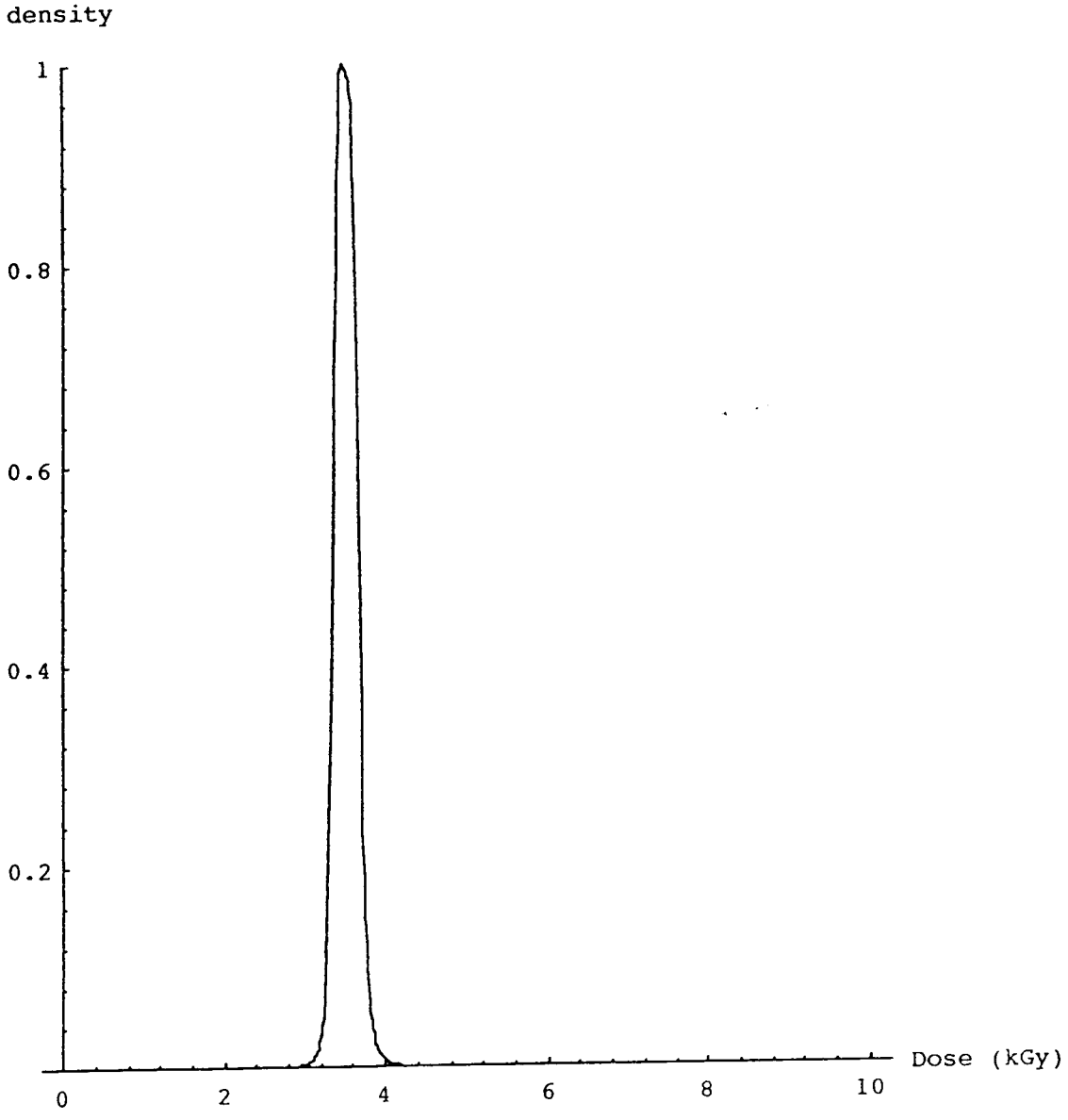
-Graphics-

```
f[st_]:= (1/((Beta[.5,2])*(Sqrt[4*0.0231])*(1+((u-6.80)^2
/(4*0.0231))^2.5))/2.46732)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



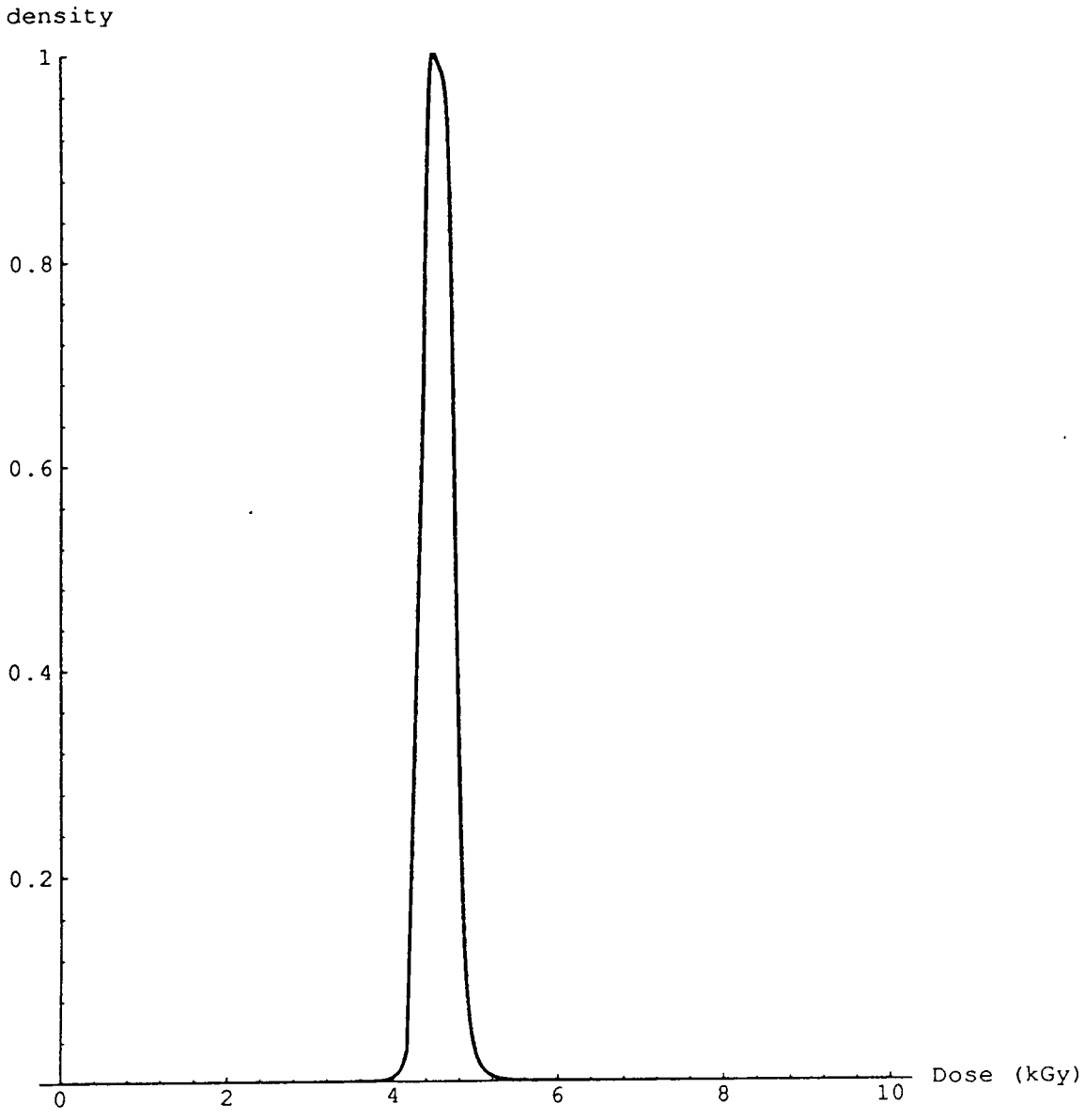
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```
f[st_]:=  (1/((Beta[.5,2])*(Sqrt[4*(.0116+(.00257*(u-1.25)^2))])
*(1+((2-((-1.921*u)+8.741))^2/(4*(.0116+(.00257*(u-1.25)^2))))
^2.5))/2.40678)
Plot[f[st_],  {u,0,10},AxesLabel->{"Dose  (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



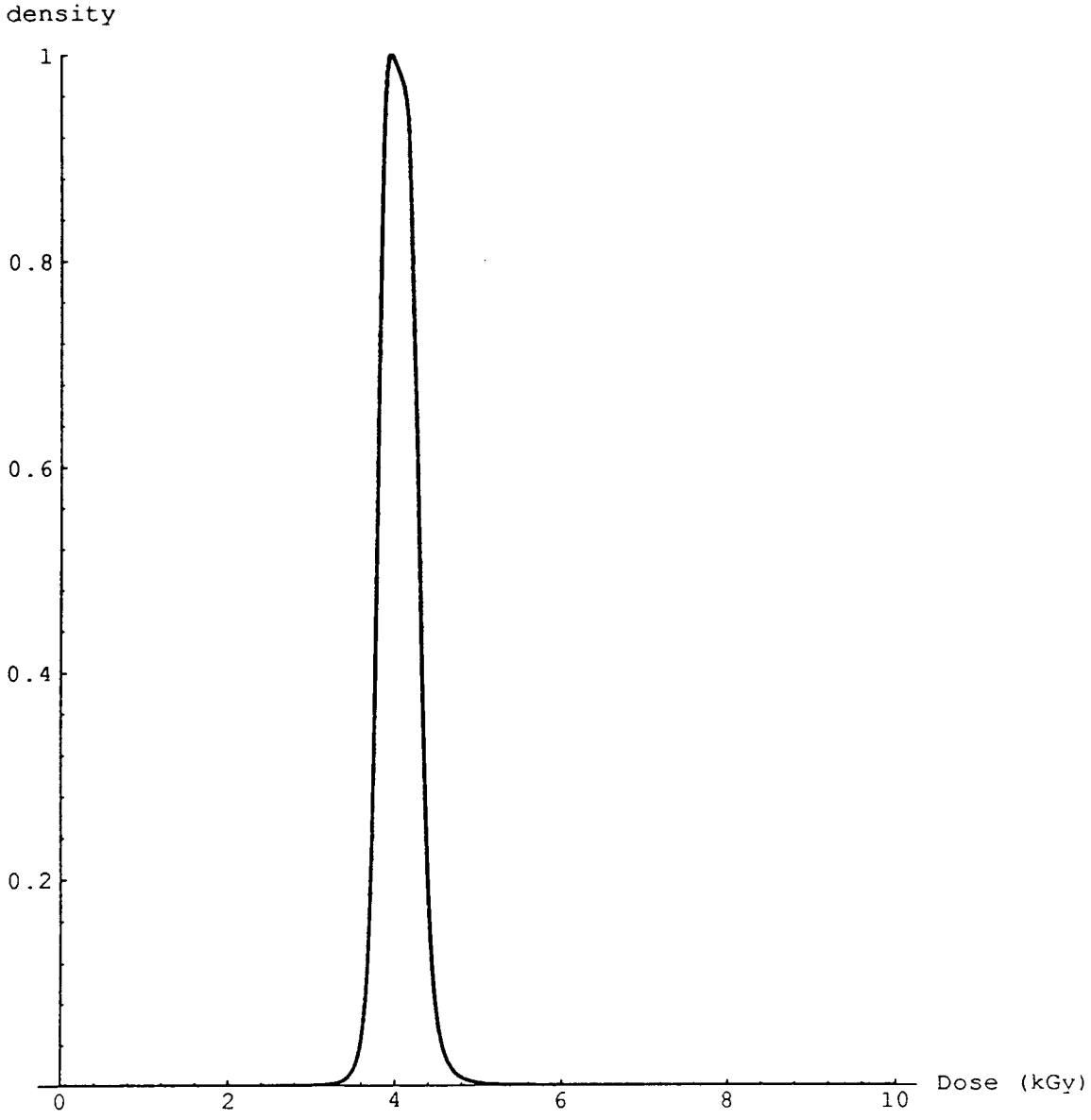
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```
f[st_]:= (1/((Beta[.5,2])*(Sqrt[4*(.0116+(.00257*(u-1.25)^2))])
*(1+((0-((-1.921*u)+8.741))^2/(4*(.0116+(.00257*(u-1.25)^2)))
)^2.5))/1.905)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



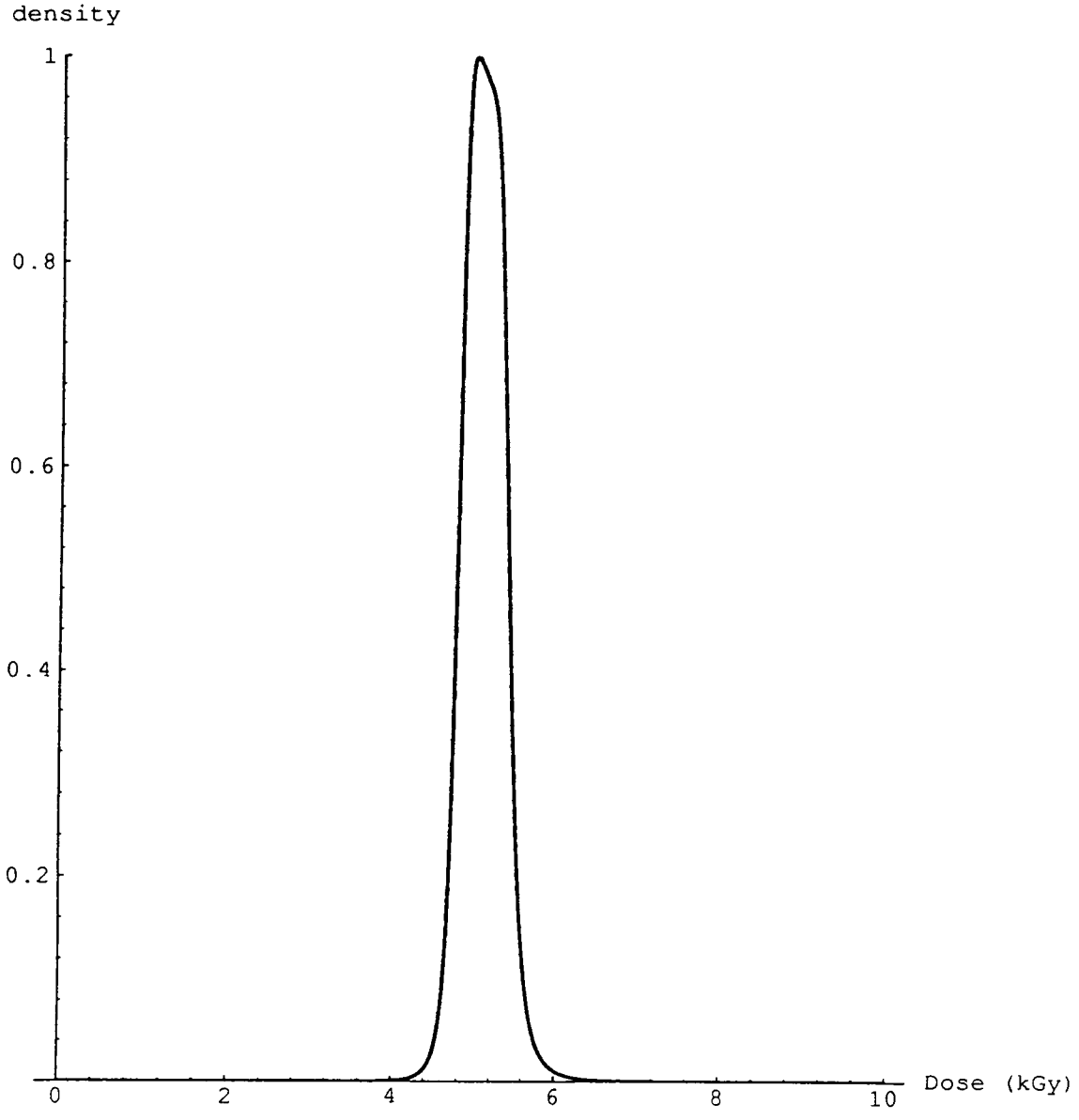
-Graphics-

```
f[st_] := (1/((Beta[.5,2])*(Sqrt[4*(.0228+(.00474*(u-1.25)^2))])
*(1+((2-((-1.895*u)+9.643))^2/(4*(.0228+(.00474*(u-1.25)^2)))
)^2.5))/1.55979)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



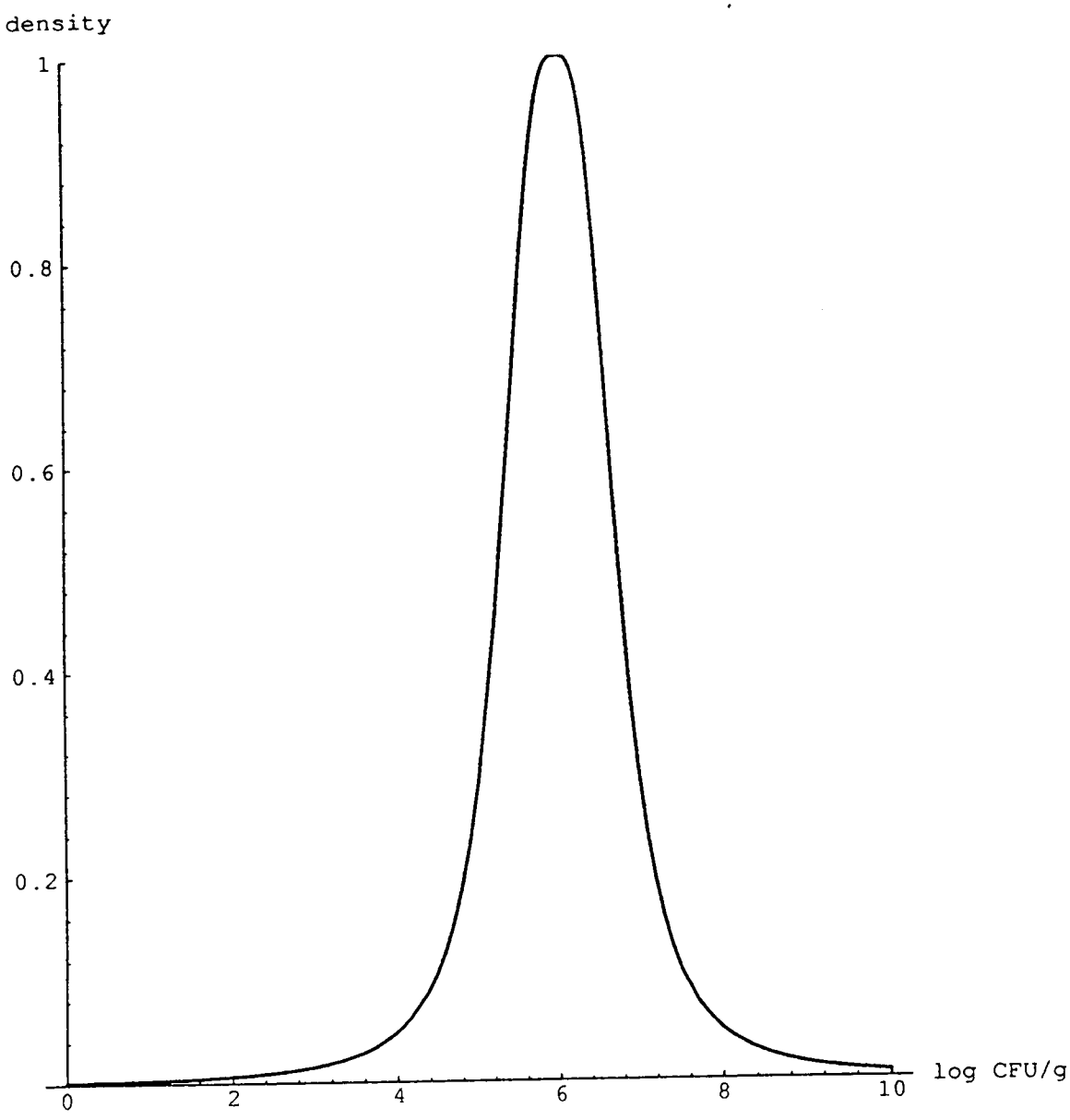
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```
f[st_]:= (1/((Beta[.5,2])*(Sqrt[4*(.0228+(.00474*(u-1.25)^2))])
*(1+((0-((-1.895*u)+9.643))^2/(4*(.0228+(.00474*(u-1.25)^2)))
)^2.5))/1.25262)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



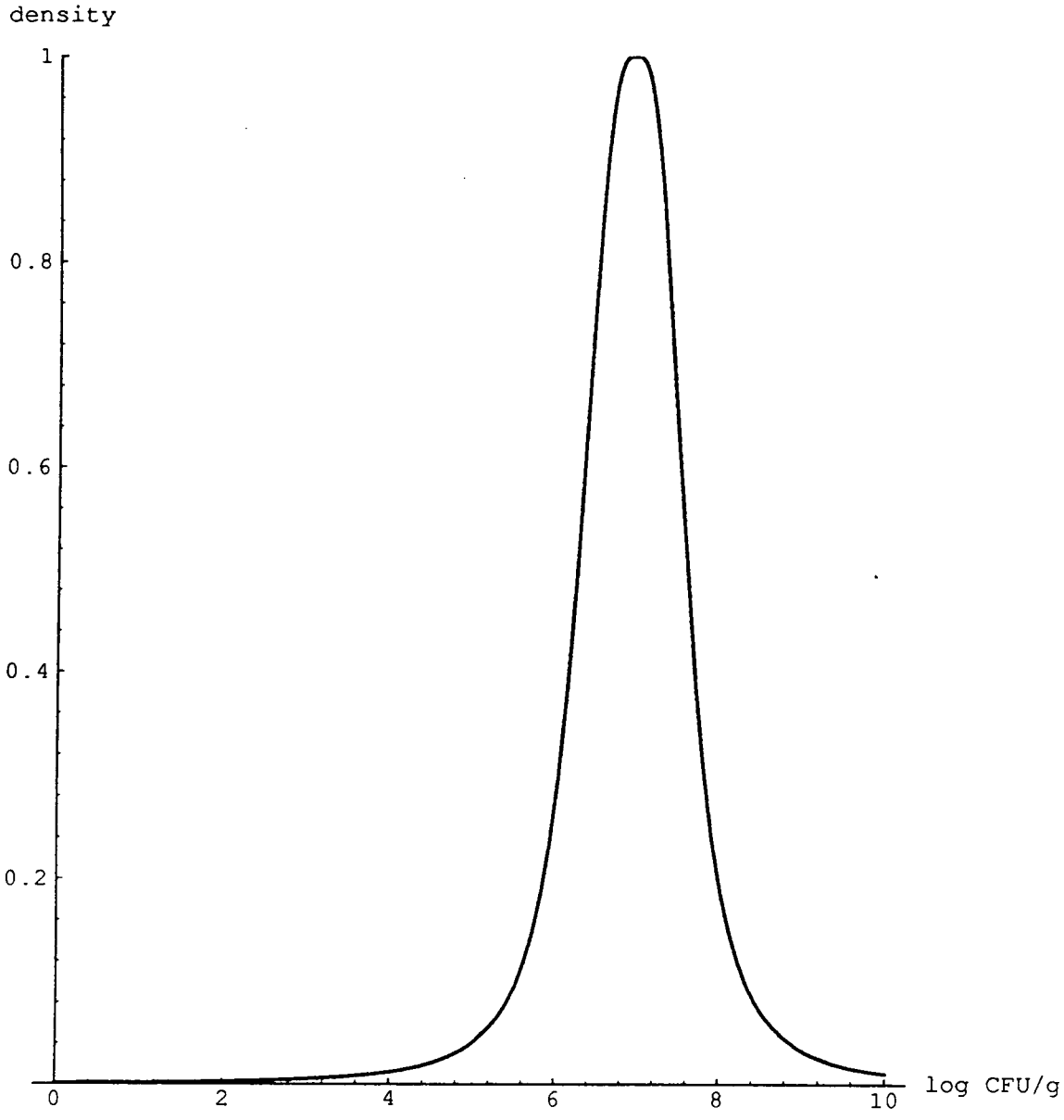
-Graphics-

```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*0.274])*(1+((u-6)^2
/(2*0.274))^1.5))/.675429)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



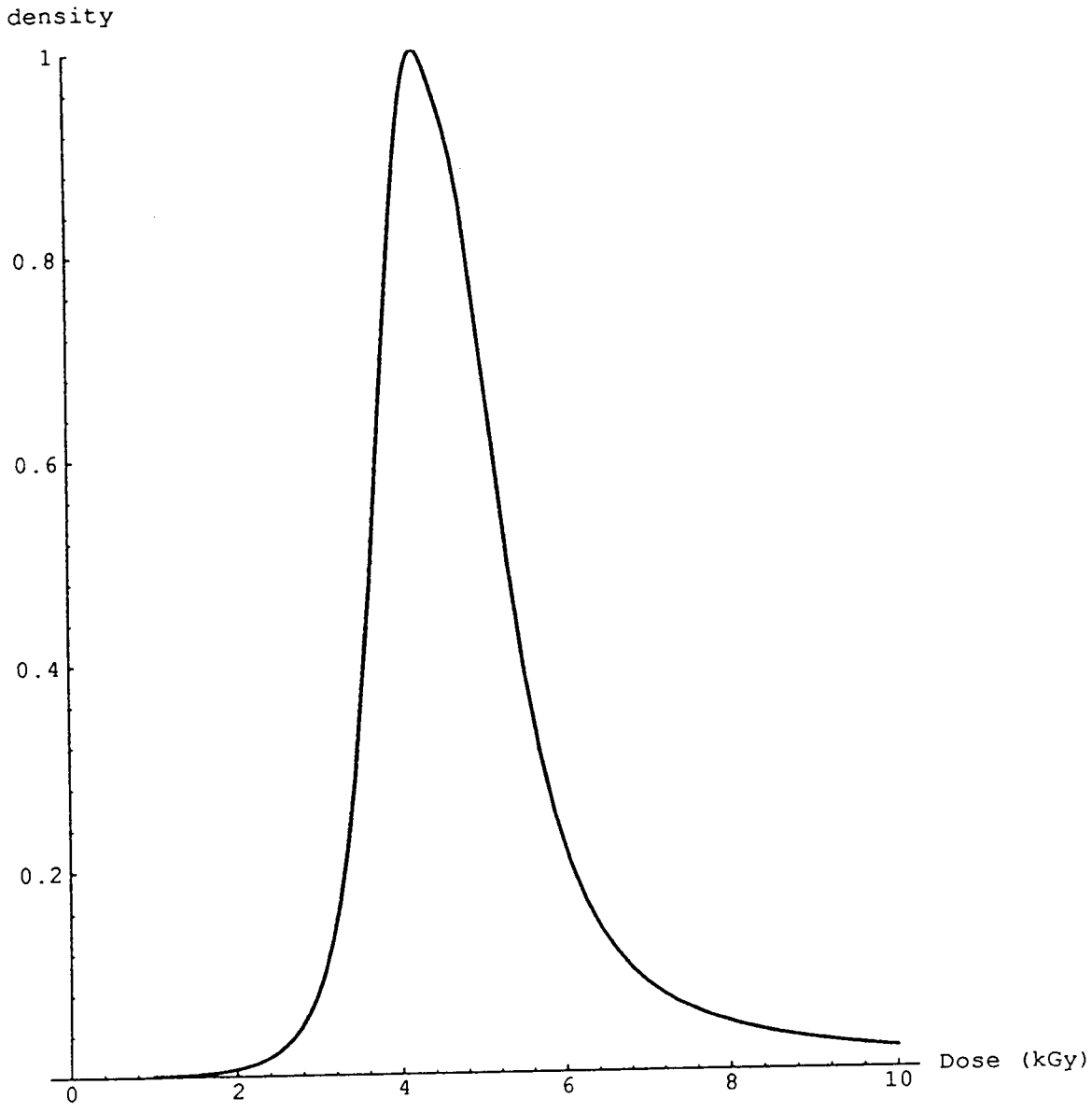
-Graphics-

```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*0.224])*(1+((u-6.93)^2
/(2*0.224))^1.5))/.747018)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



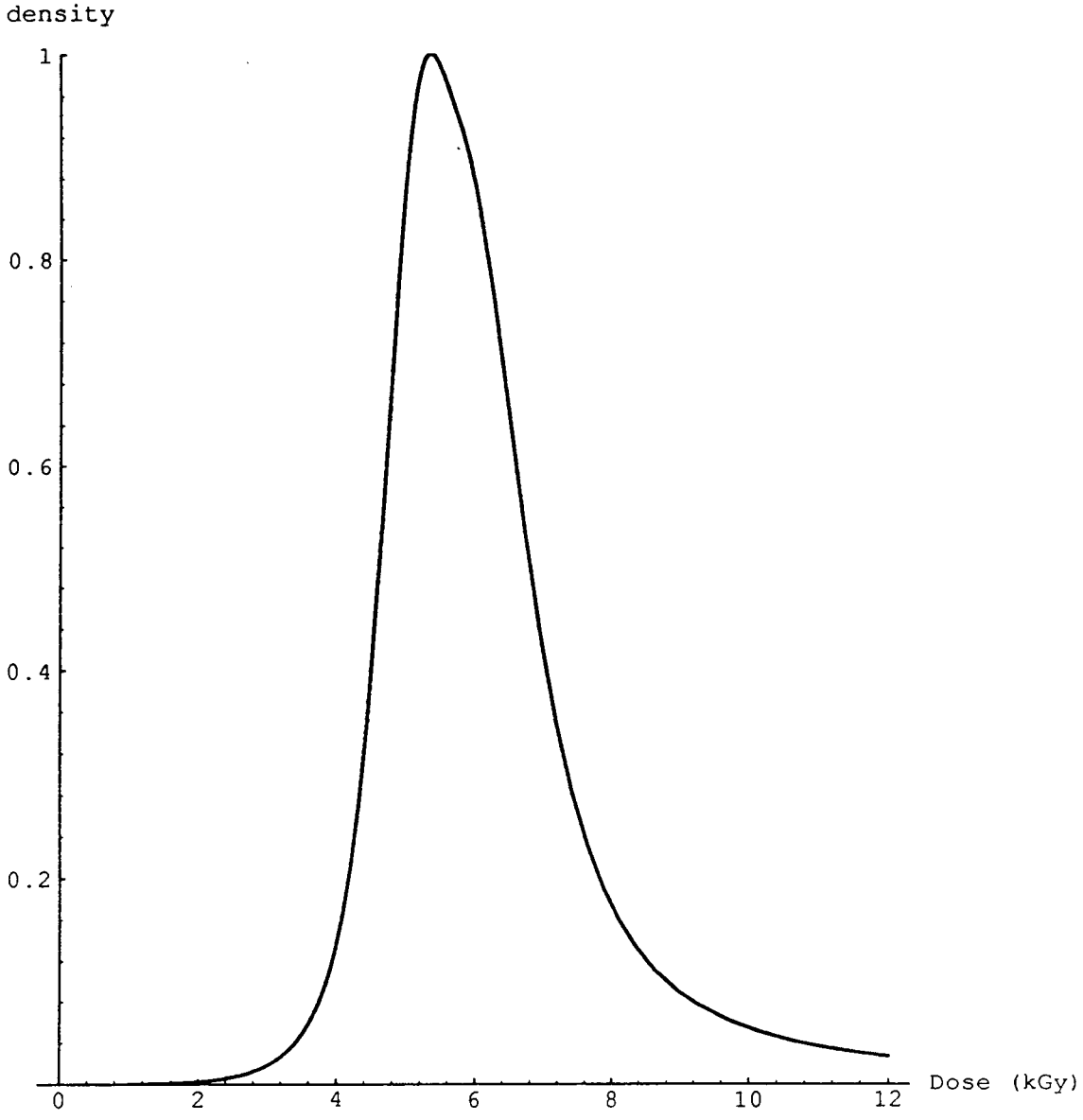
-Graphics-

```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*(.0486+(.0719*(u-.75)^2))])*(
1+((2-((-1.643*u)+9.265))^2/(2*(.0486+(.0719*(u-.75)^2))))^1.5))
/.364522)
Plot[f[st_], {u,1,10},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```

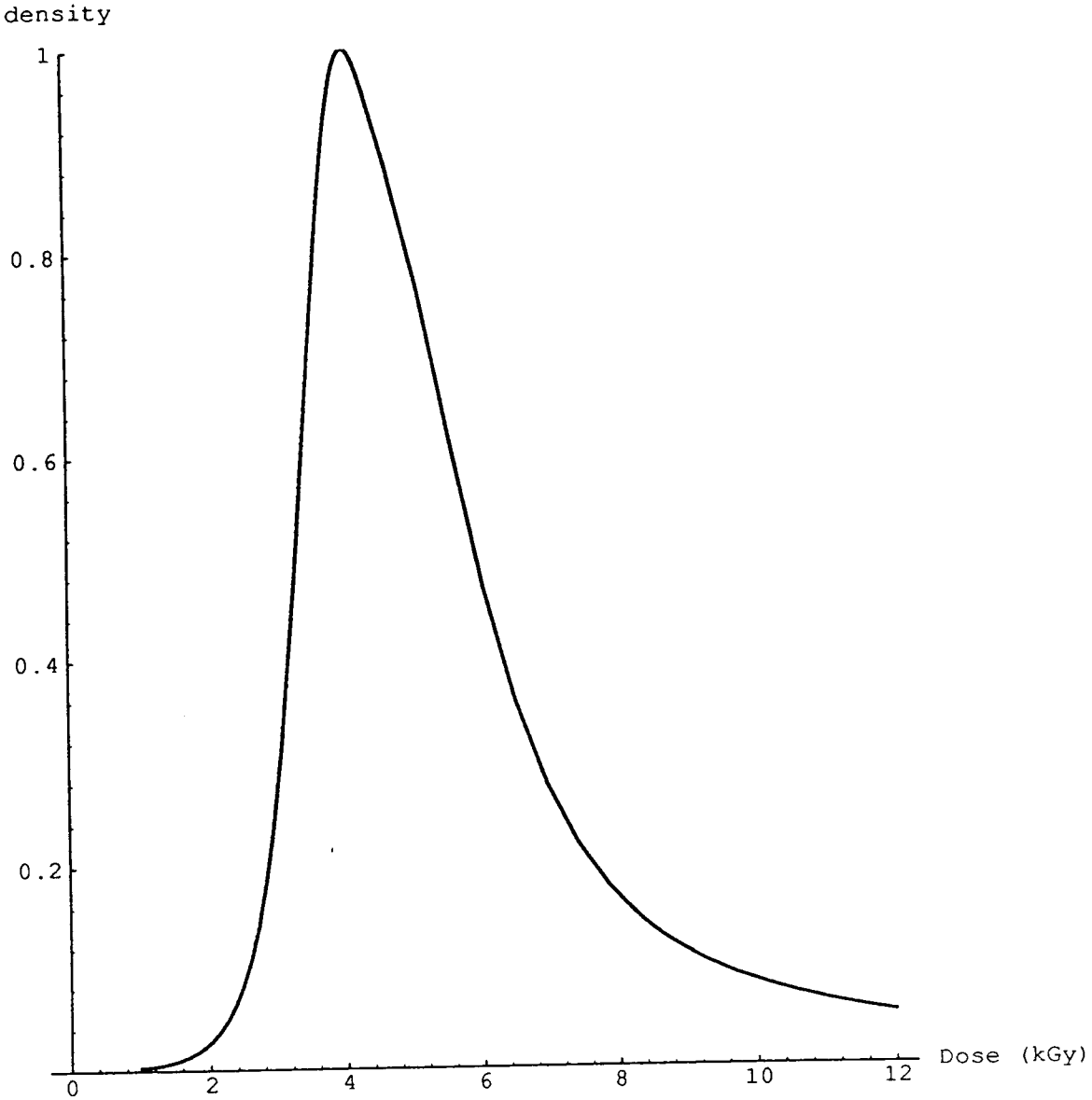


-Graphics-

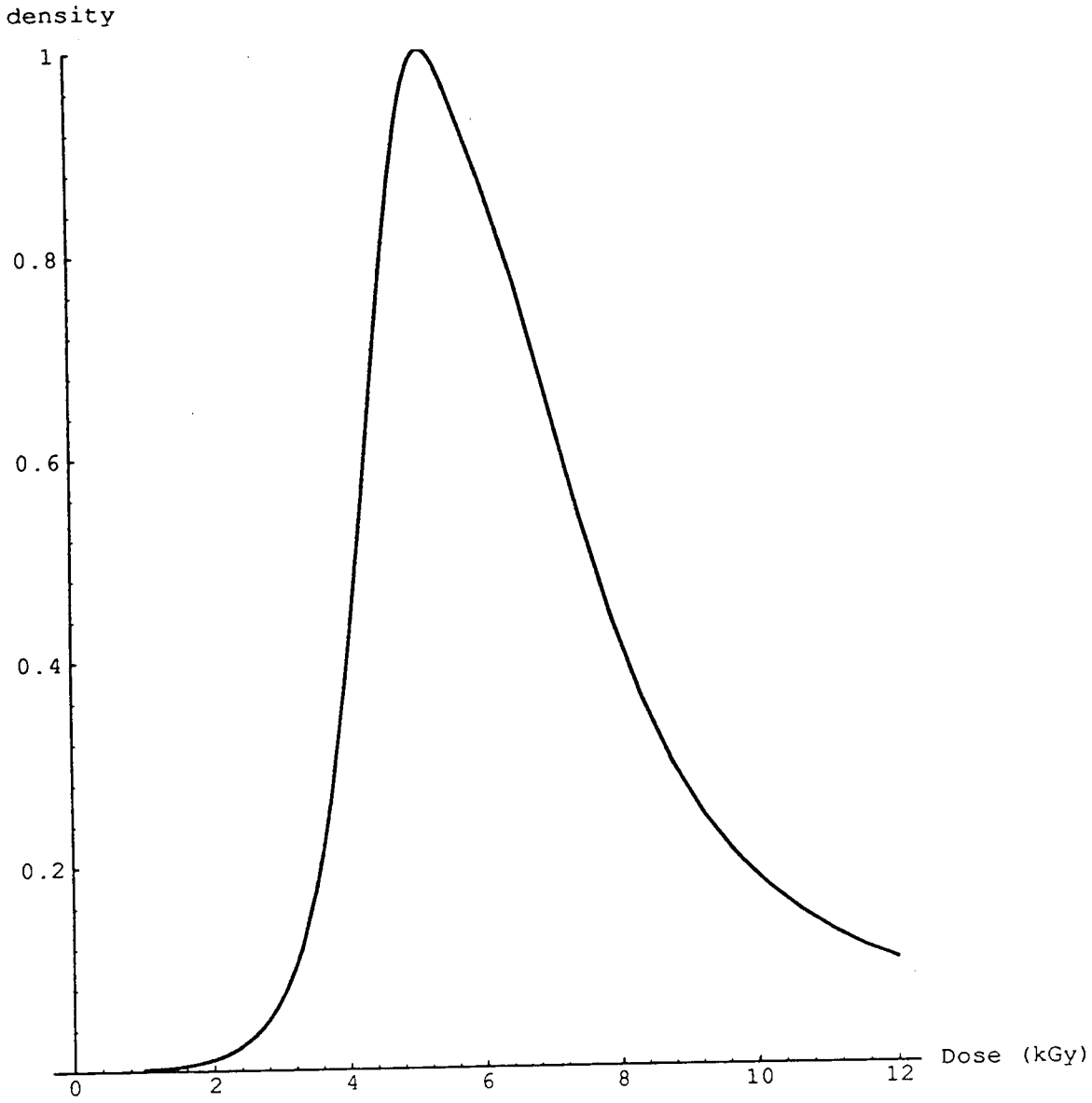
```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*(.0486+(.0719*(u-.75)^2))])*(1
+((0-((-1.643*u)+9.265))^2/(2*(.0486+(.0719*(u-.75)^2))))^1.5))
/.276835)
Plot[f[st_], {u,1,12},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```



```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*(.112+(.199*(u-.75)^2))])*(1
+((2-((-1.637*u)+9.390))^2/(2*(.112+(.199*(u-.75)^2))))^1.5))
/.224137)
Plot[f[st_], {u,1,12},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```

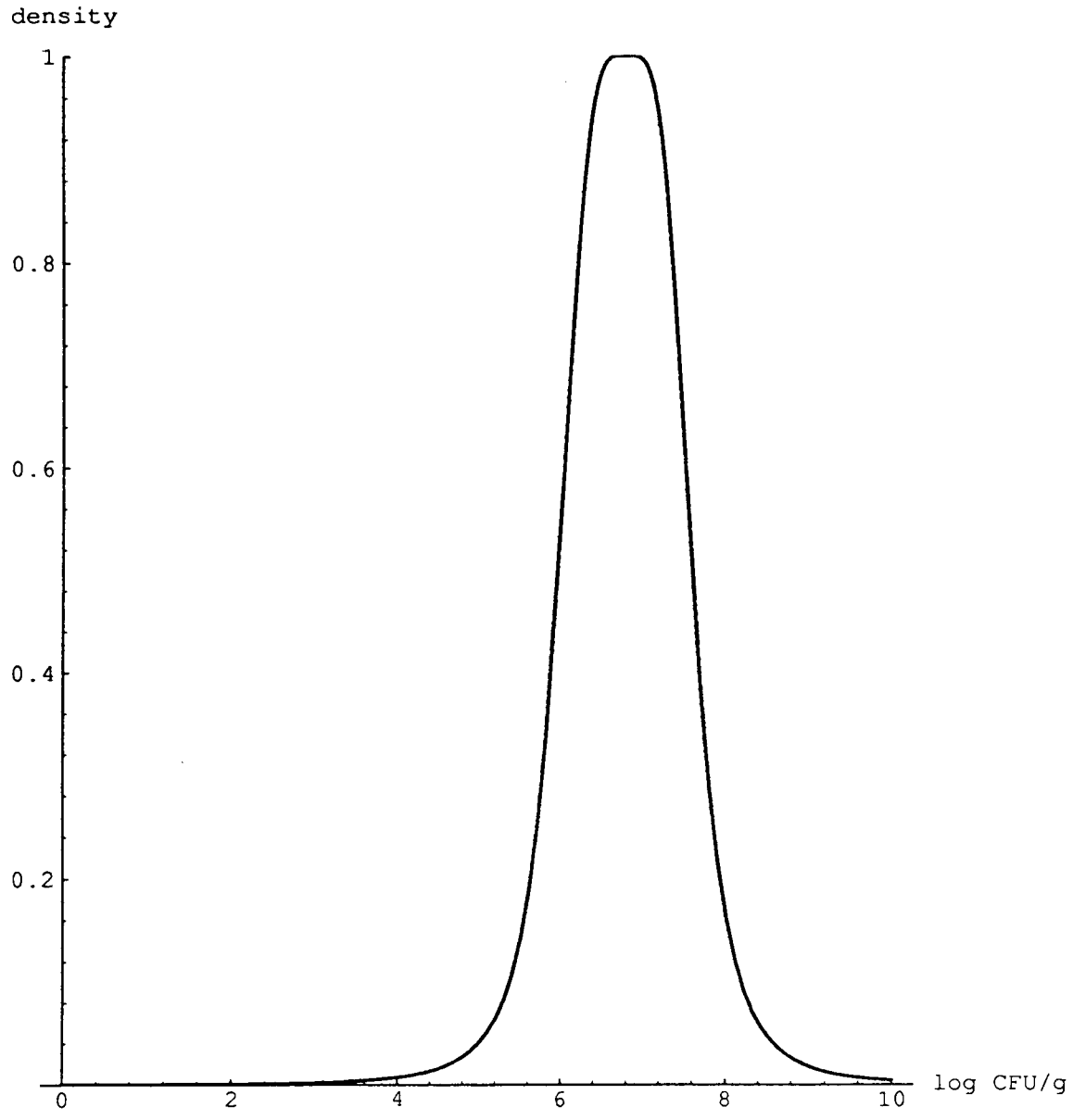


```
f[st_]:= (1/((Beta[.5,1])*(Sqrt[2*(.112+(.199*(u-.75)^2))])*(1
+((0-((-1.637*u)+9.390))^2/(2*(.112+(.199*(u-.75)^2))))^1.5))
/.170807)
Plot[f[st_], {u,1,12},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```



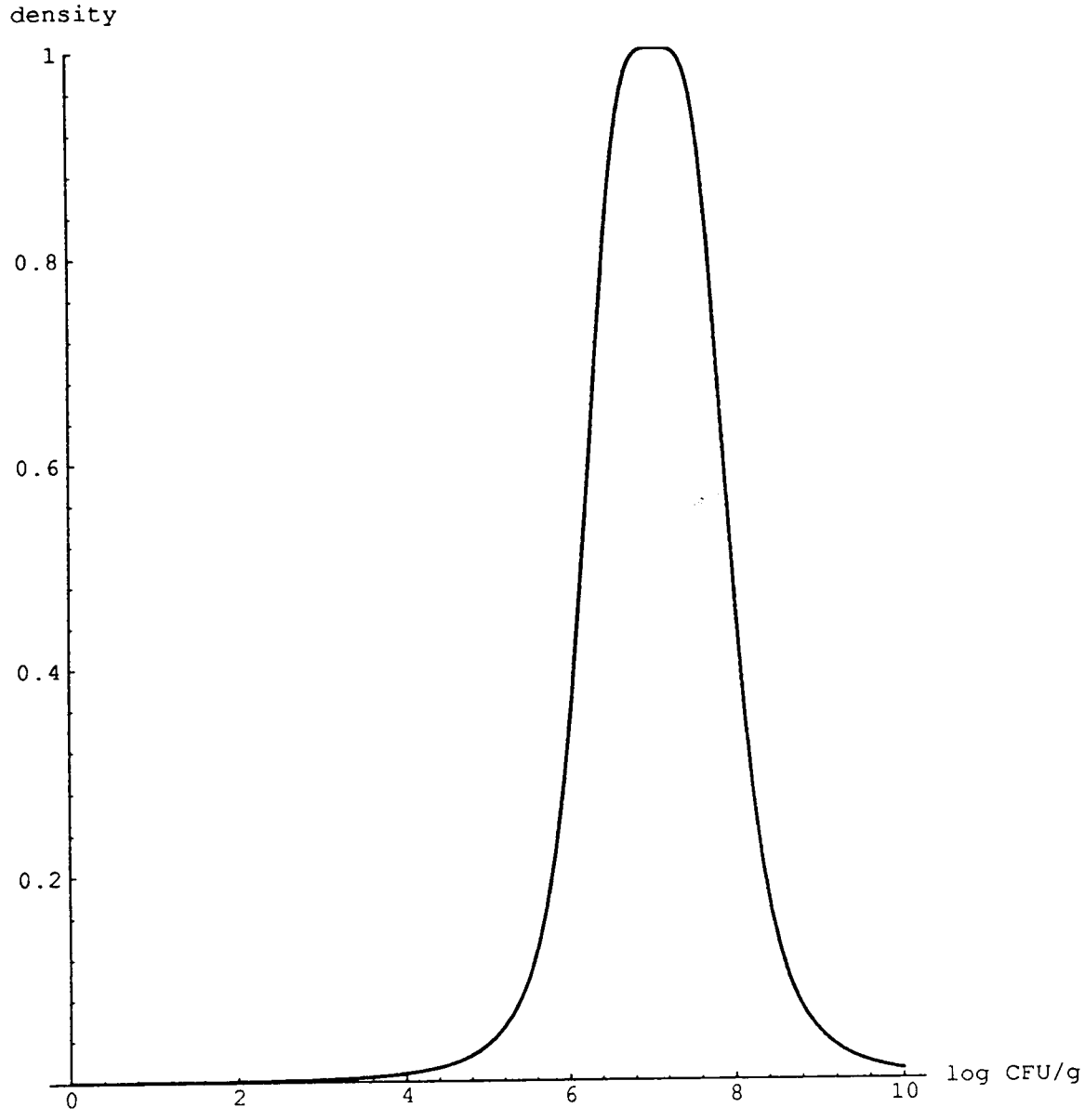
-Graphics-

```
f[st_]:= (1/((Beta[.5,1.5])*(Sqrt[3*0.2205])*(1+((u-6.792)^2
/(3*0.2205))^2))/.782736)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



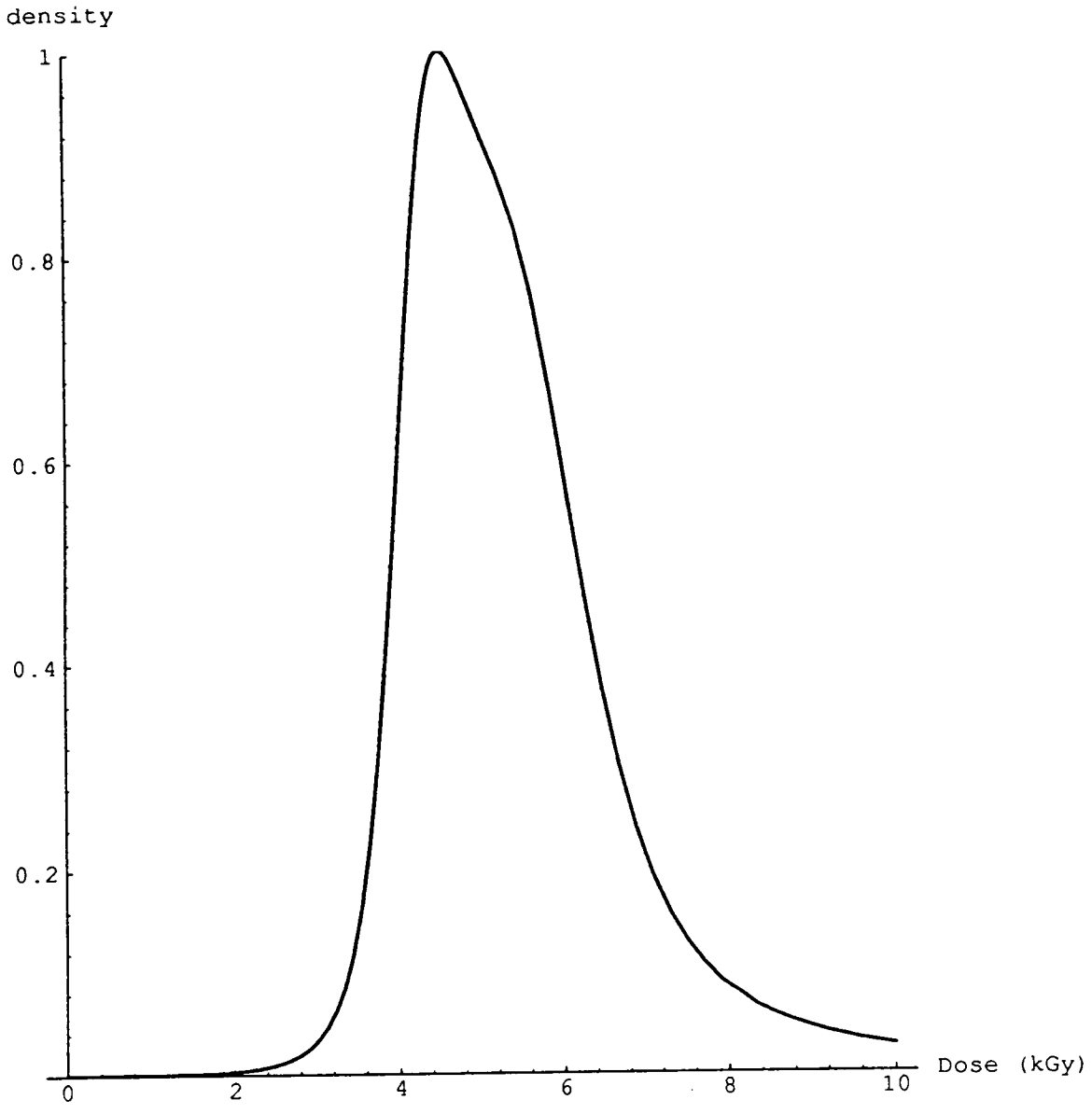
-Graphics-

```
f[st_]:= (1/((Beta[.5,1.5])*(Sqrt[3*0.273])*(1+((u-7.077)^2
/(3*0.273))^2))/.703458)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



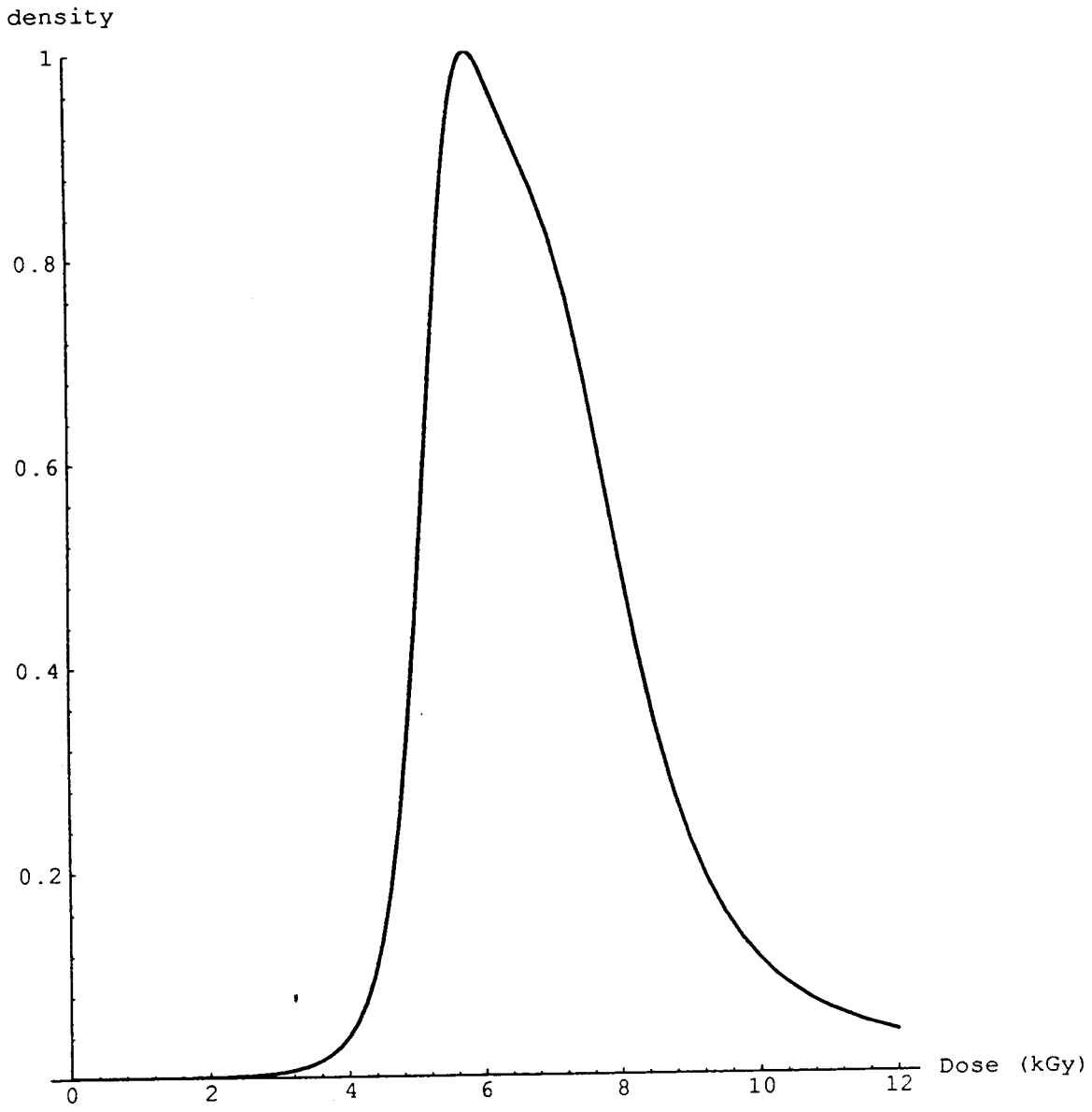
-Graphics-

```
f[st_]:= (1/((Beta[.5,3])*(Sqrt[3*(.220+(.0547*(u-1.38)^2))])*
(1+((2-(( 1.395*u)+8.885))^2/(3*(.220+(.0547*(u-1.38)^2))))^2))
/.607931)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



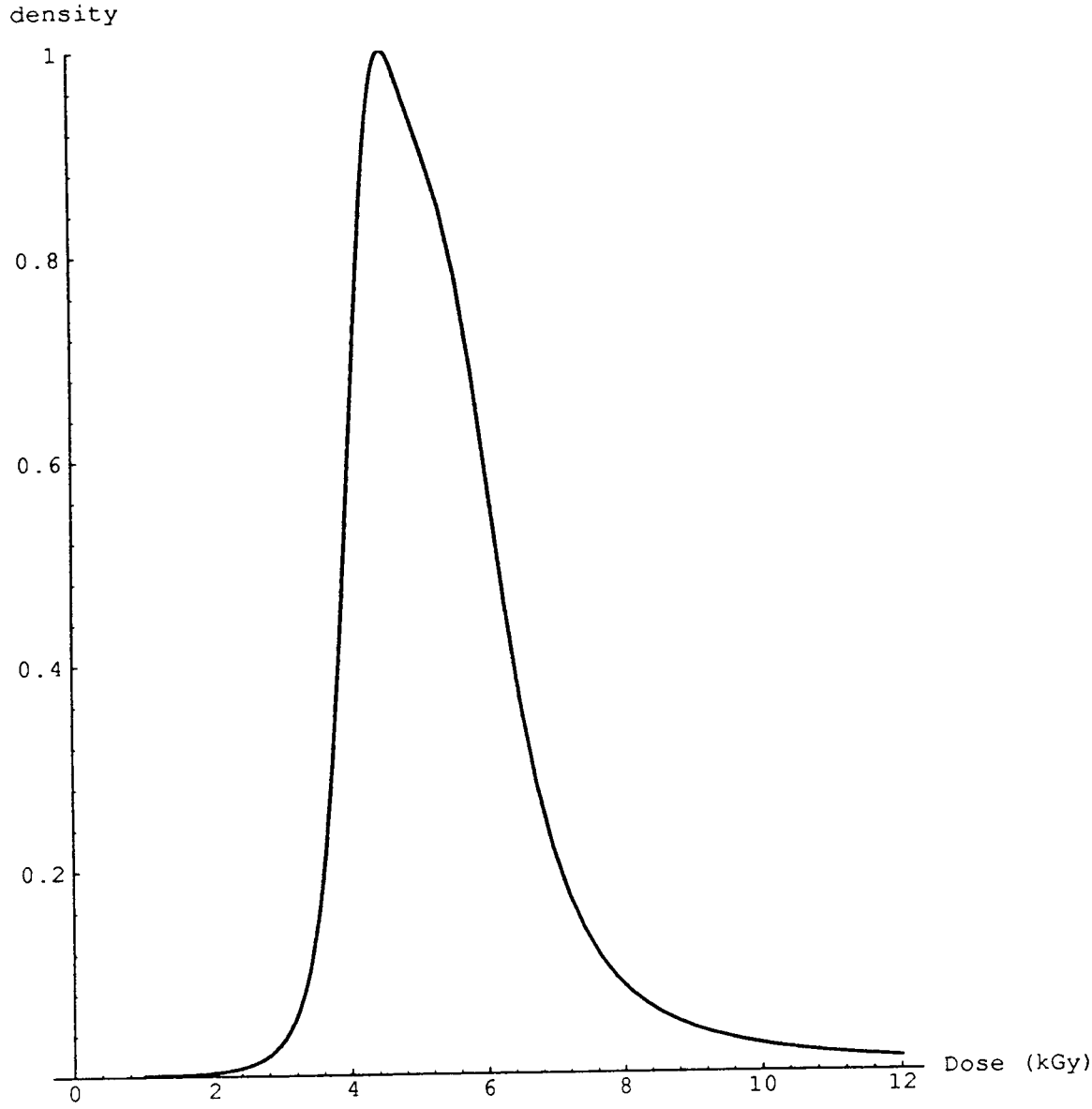
-Graphics-

```
f[st_]:= (1/((Beta[.5,3])*(Sqrt[3*(.220+(.0547*(u-1.38)^2))])*
(1+((0-((-1.395*u)+8.885))^2/(3*(.220+(.0547*(u-1.38)^2))))^2))
/.464495)
Plot[f[st_], {u,0,12},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



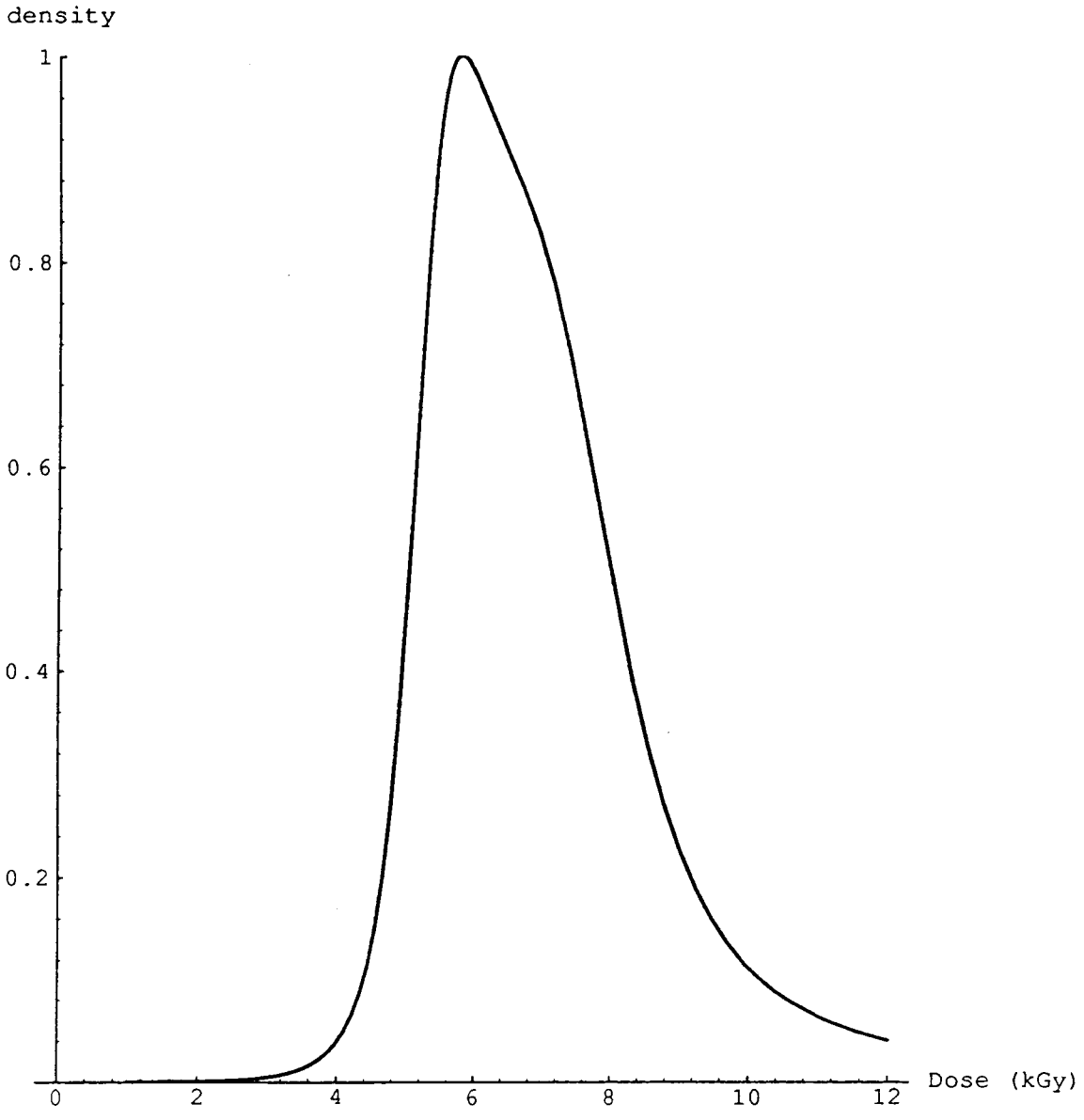
-Graphics-

```
f[st_]:= (1/((Beta[.5,1.5])*(Sqrt[3*(.220+(.0547*(u-1.38)^2))])
*(1+((2-((-1.395*u)+8.885))^2/(3*(.220+(.0547*(u-1.38)^2))))^2))
/.412822)
Plot[f[st_], {u,1,12},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```



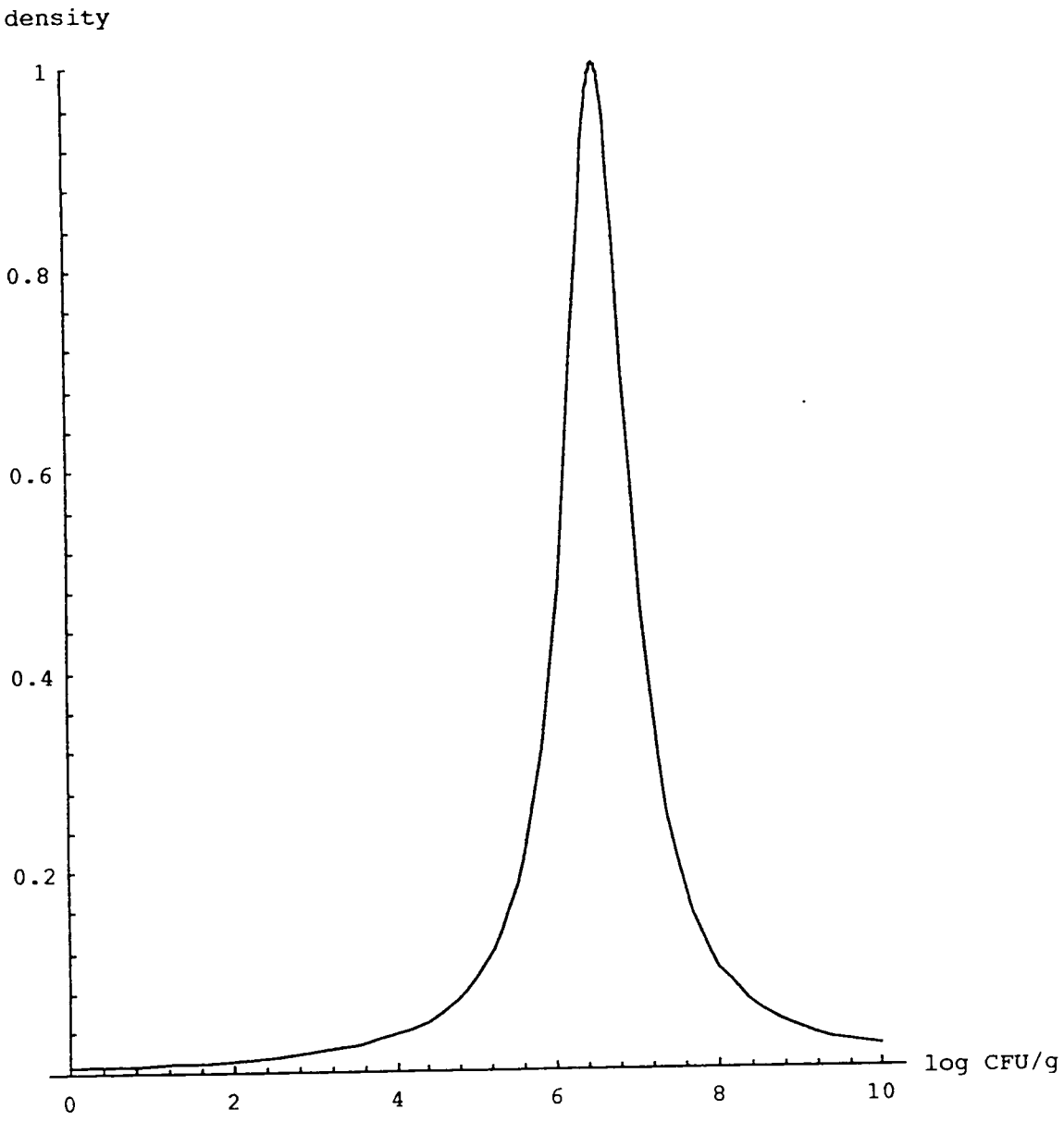
-Graphics-

```
f[st_]:= (1/((Beta[.5,1.5])*(Sqrt[3*(.220+(.0547*(u-1.38)^2))])
*(1+((0-((-1.395*u)+8.885))^2/(3*(.220+(.0547*(u-1.38)^2))))^2))
/.315395)
Plot[f[st_], {u,1,12},PlotRange->{0,1},AxesLabel
->{"Dose (kGy)","density"},AspectRatio->1.2]
```



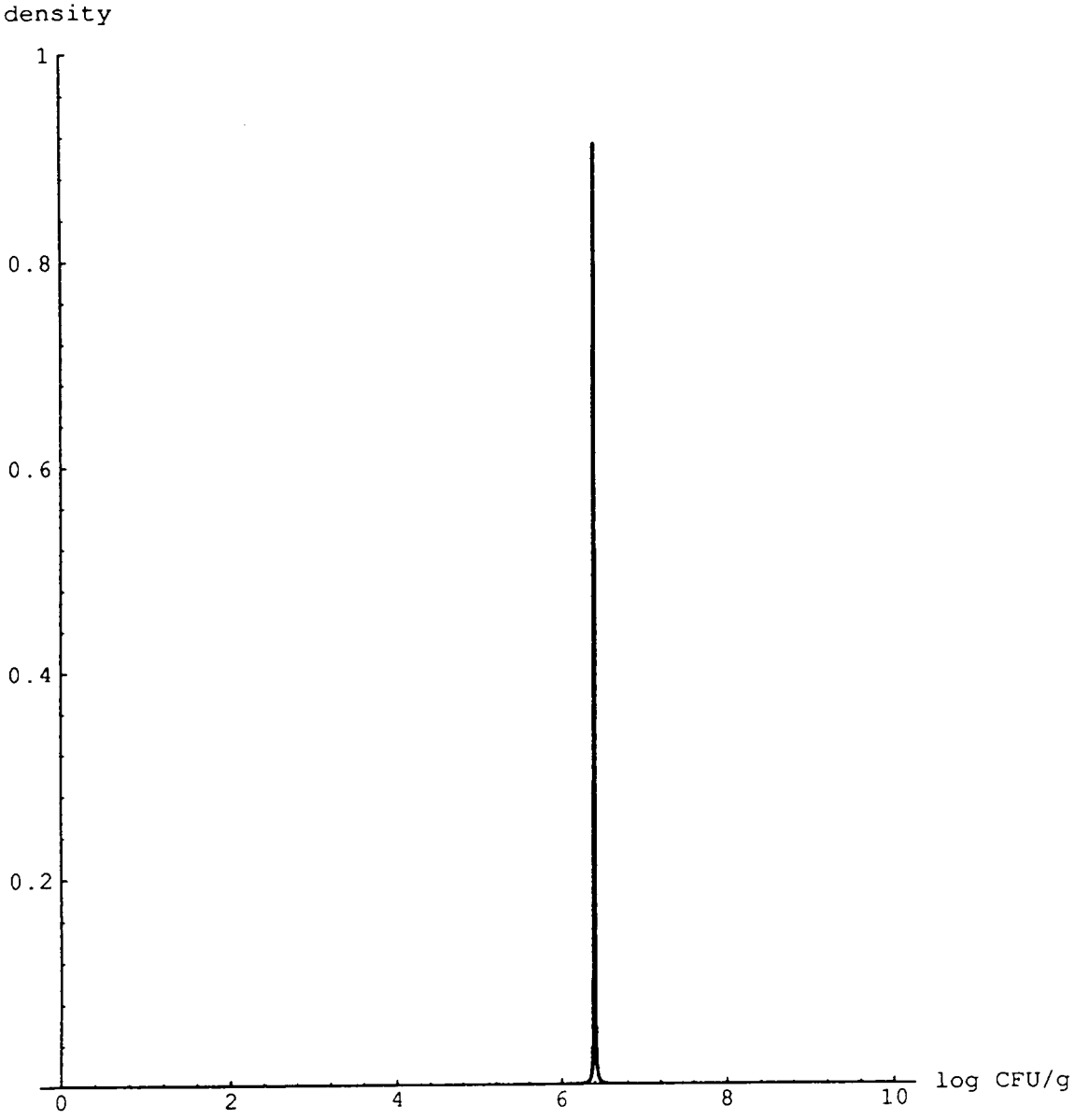
-Graphics-

```
f[st_]:=  (1/((Beta[.5,.5])*(Sqrt[1*.242])*(1+((u-6.55)^2
/(1*.242))^1))/.647057)
Plot[f[st_],  {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log  CFU/g","density"}]
```



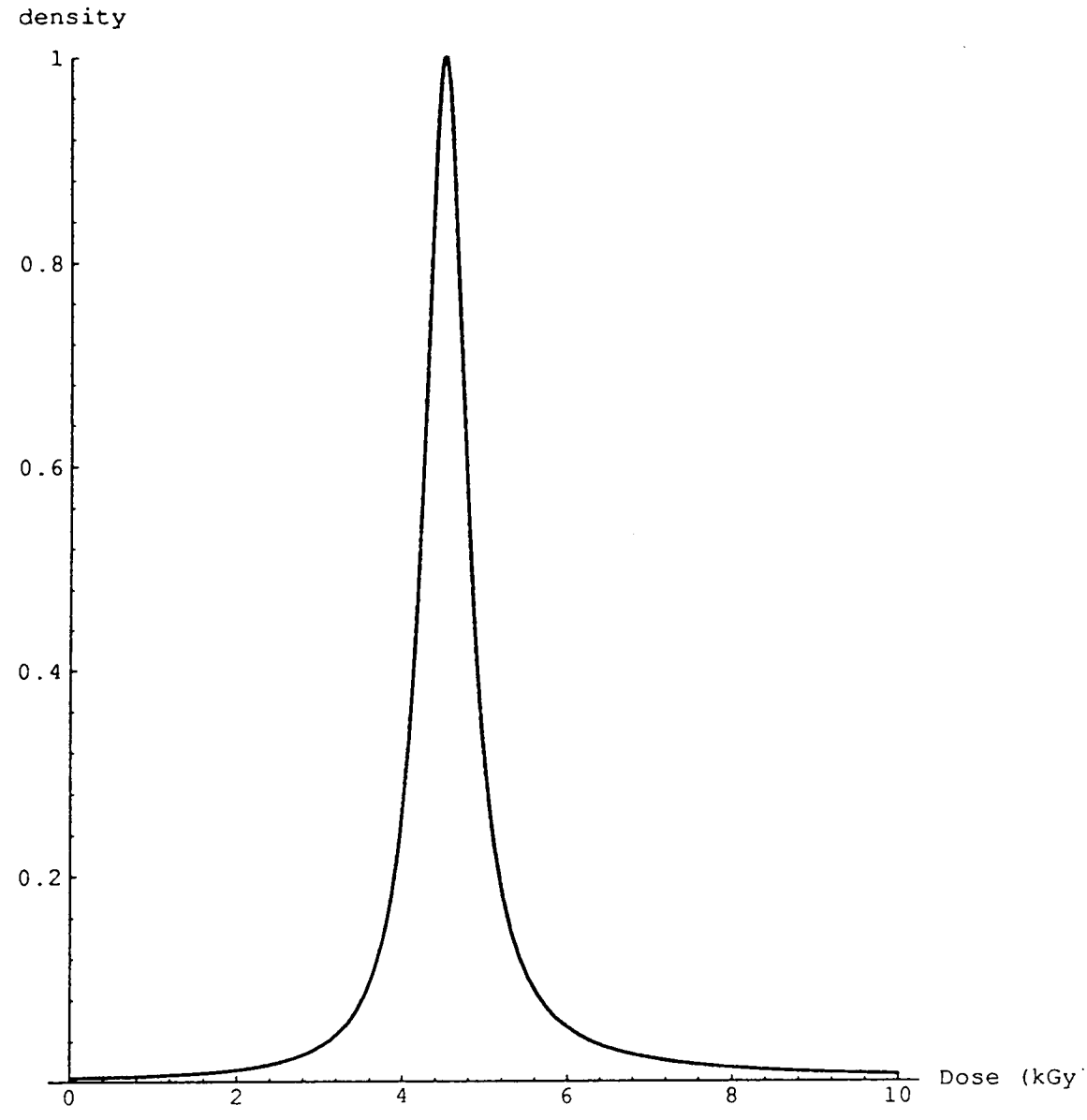
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*0.0000068])*(1+((u-6.403)^2
/(1*0.0000068))^1))/52.5349)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



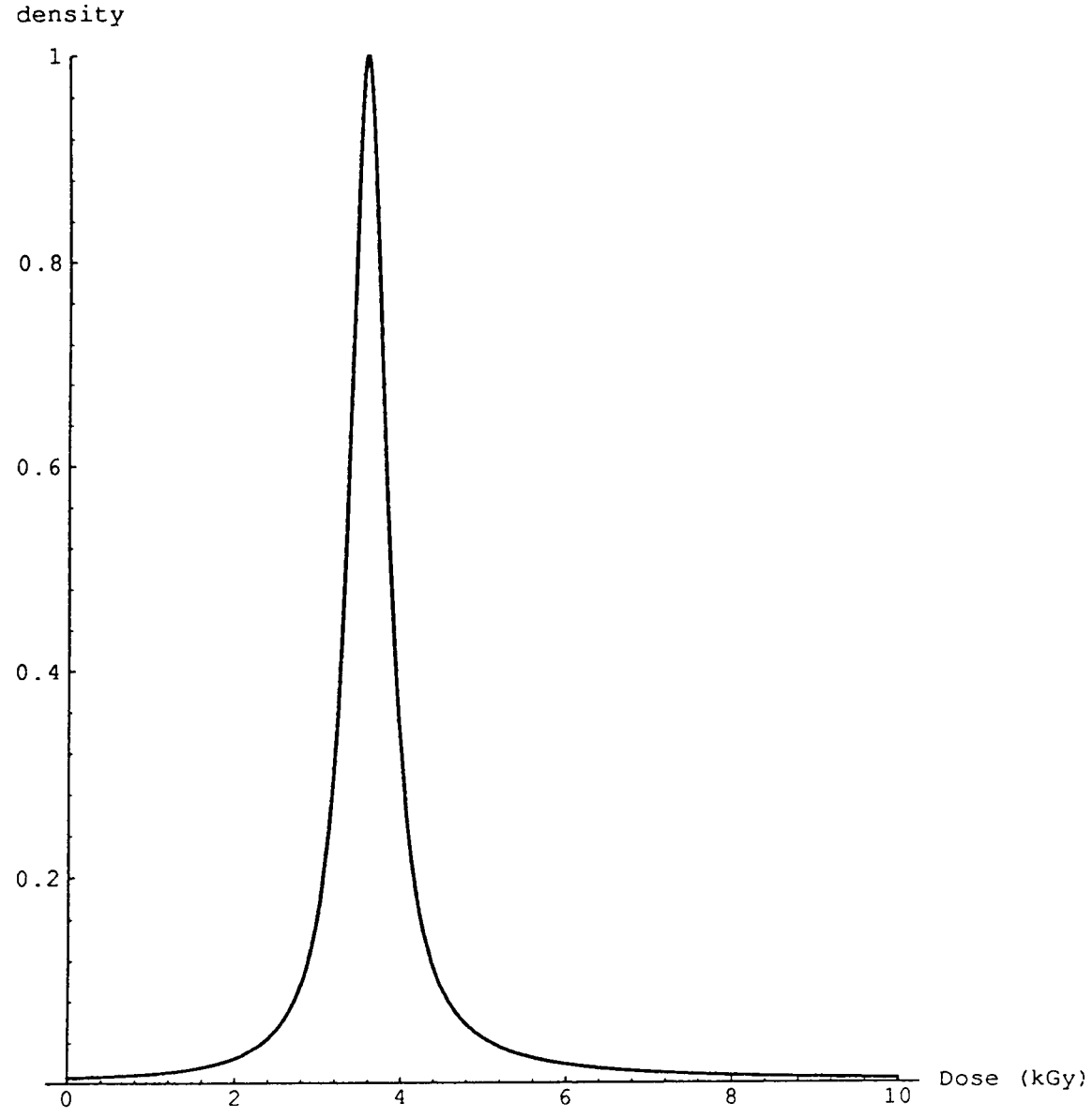
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.242+(.0259*(u-1.5)^2))])
*(1+((0-((-2.18*u)+9.82))^2/(1*(.242+(.0259*(u-1.5)^2))))^1))
/.461707)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



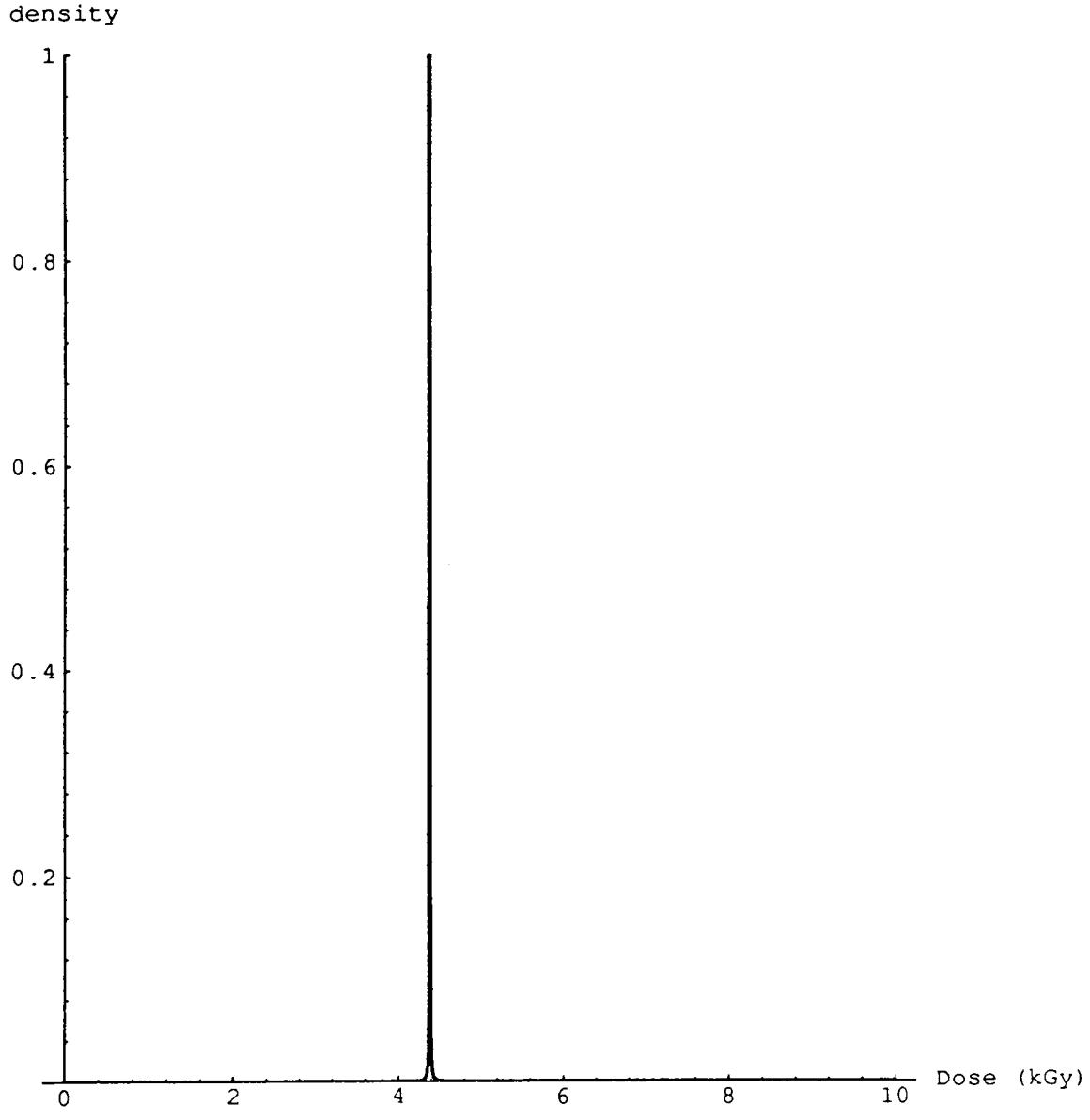
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.242+(.0259*(u-1.5)^2))])
*(1+((2-((-2.18*u)+9.82))^2/(1*(.242+(.0259*(u-1.5)^2))))^1))
/.534585)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



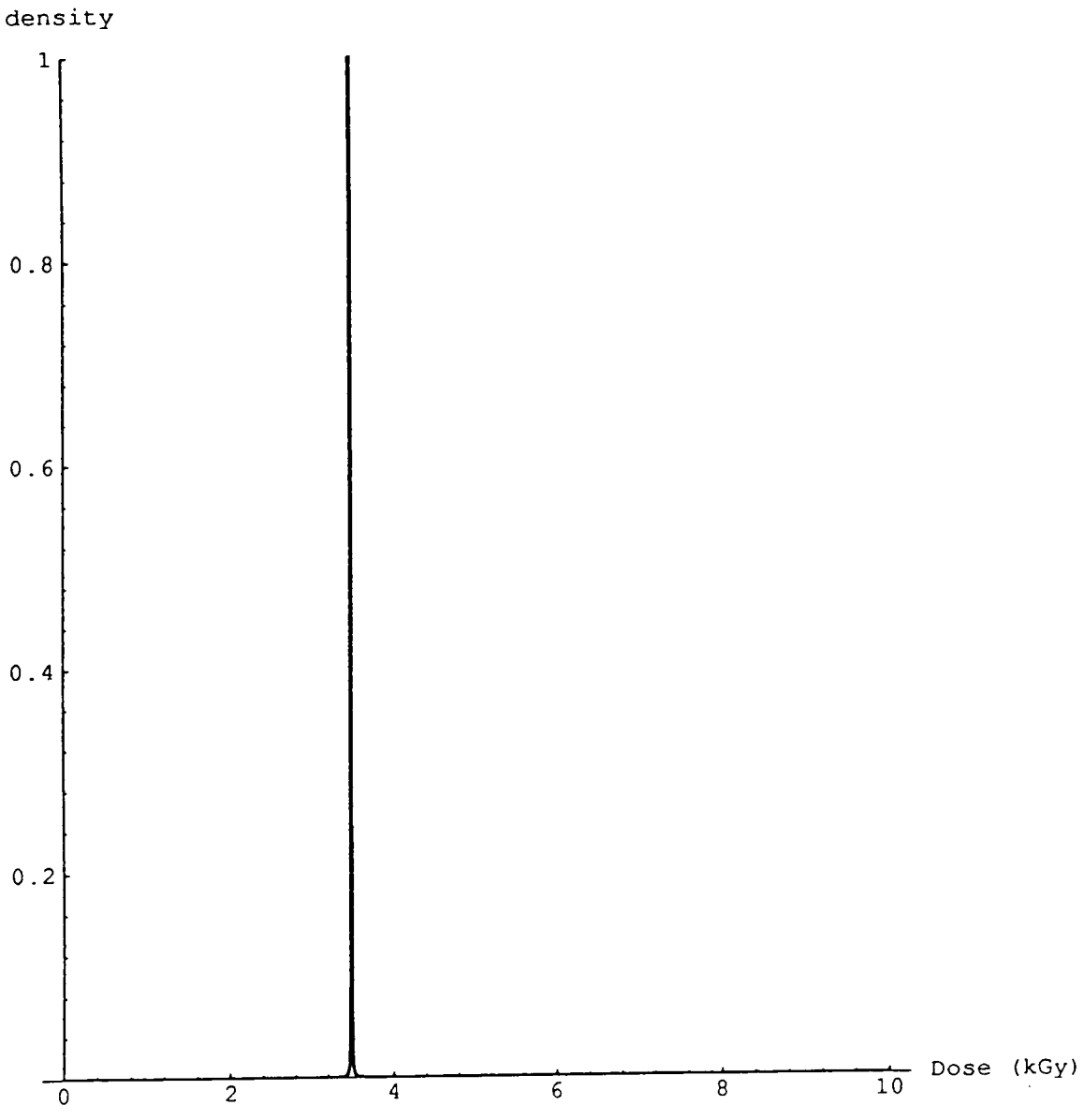
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.00000682+(.00000073
*(u-1.8)^2))])*(1+((0-((-2.221*u)+9.735))^2/(1*(.00000682+
(.00000073*(u-1.8)^2))))^1))/17.8449)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2,PlotPoints->50]
```



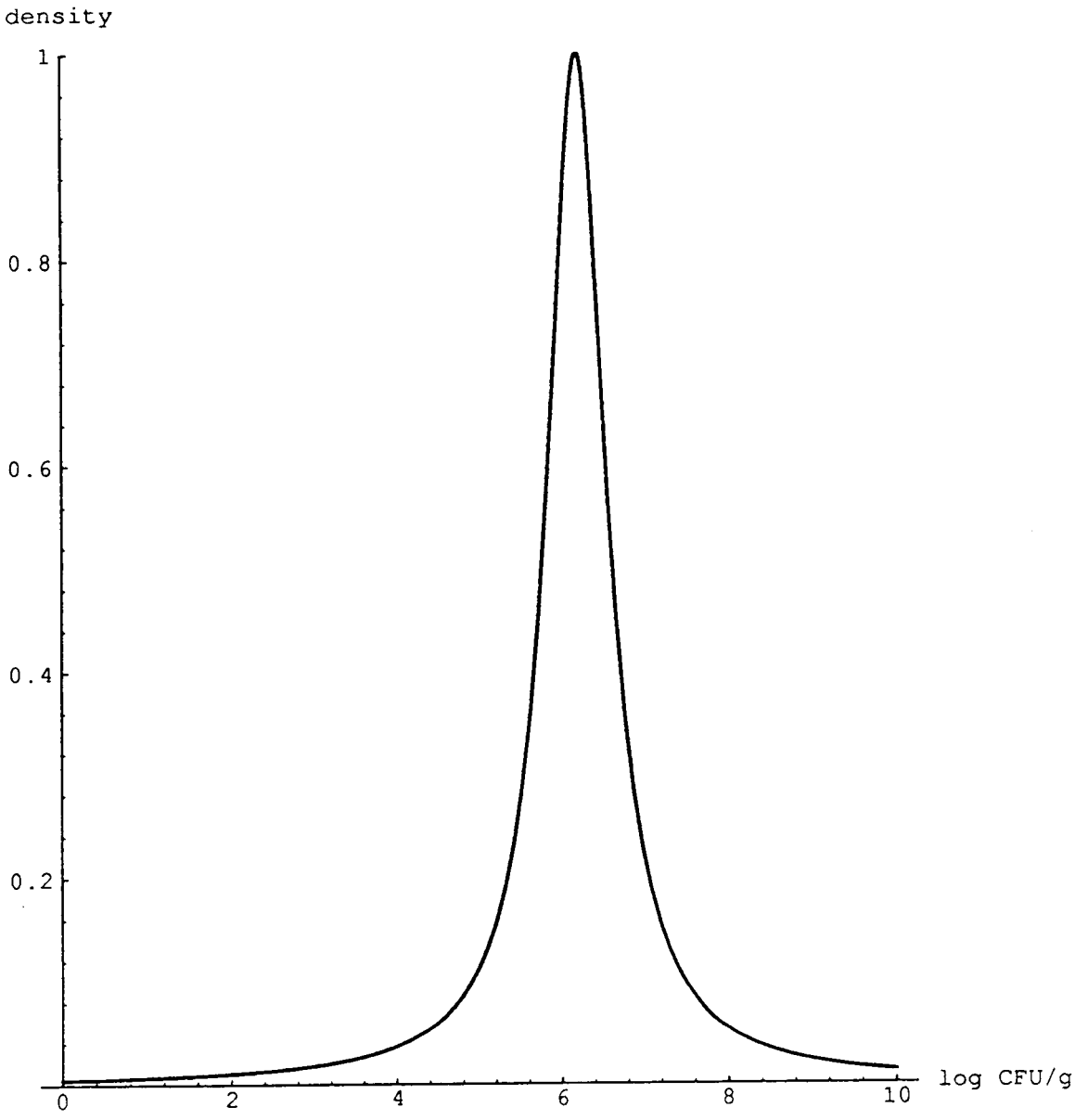
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.00000682+(.00000073
*(u-1.8)^2))])*(1+((2-((-2.221*u)+9.735))^2/(1*(.00000682+
(.00000073*(u-1.8)^2))))^1))/21.5942)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2,PlotPoints->50]
```



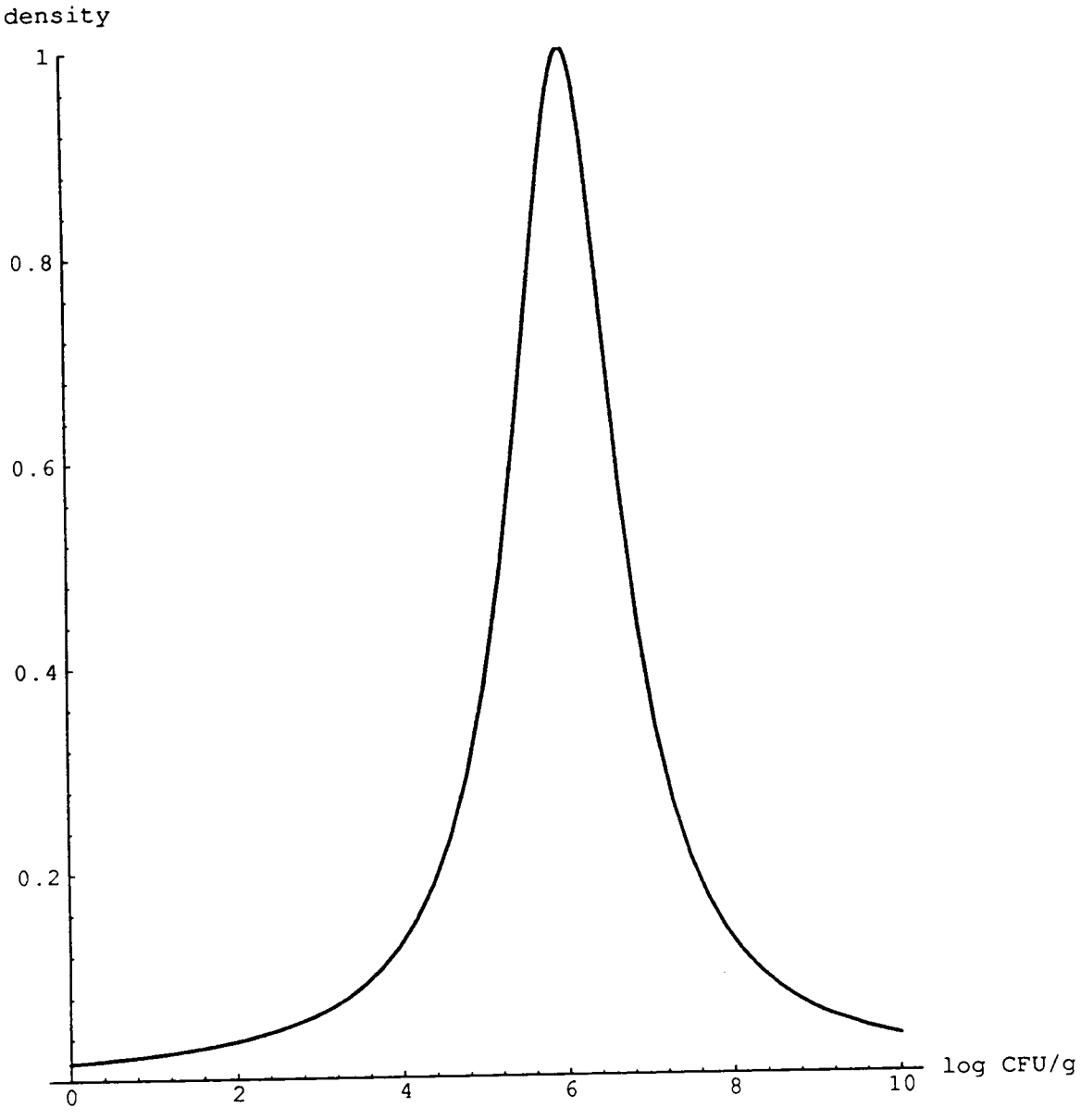
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*0.1829])*(1+((u-6.195)^2
/(1*0.1829))^1))/.74419)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



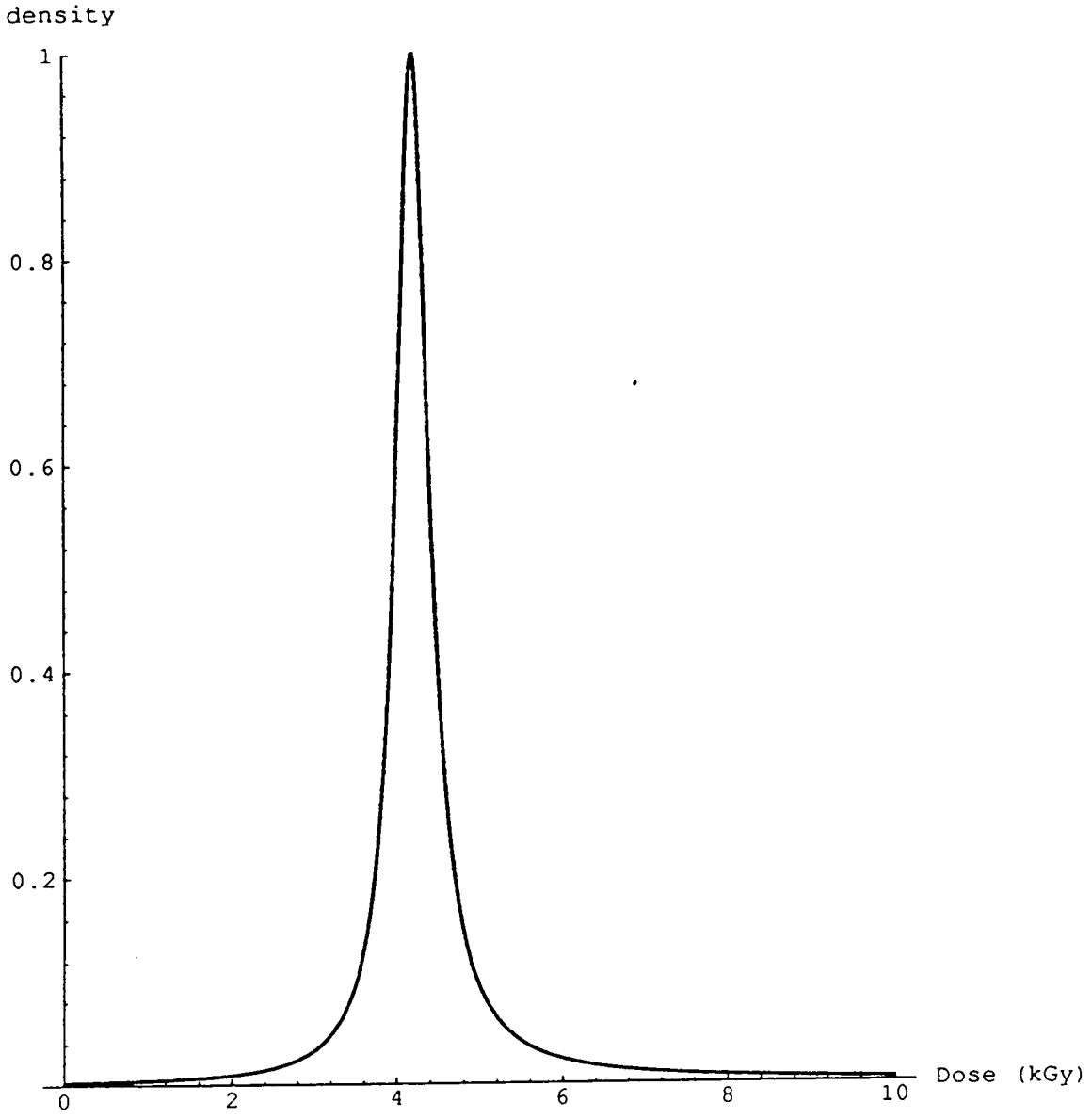
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*0.6036])*(1+((u-5.997)^2
/(1*0.6036))^1))/.409703)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



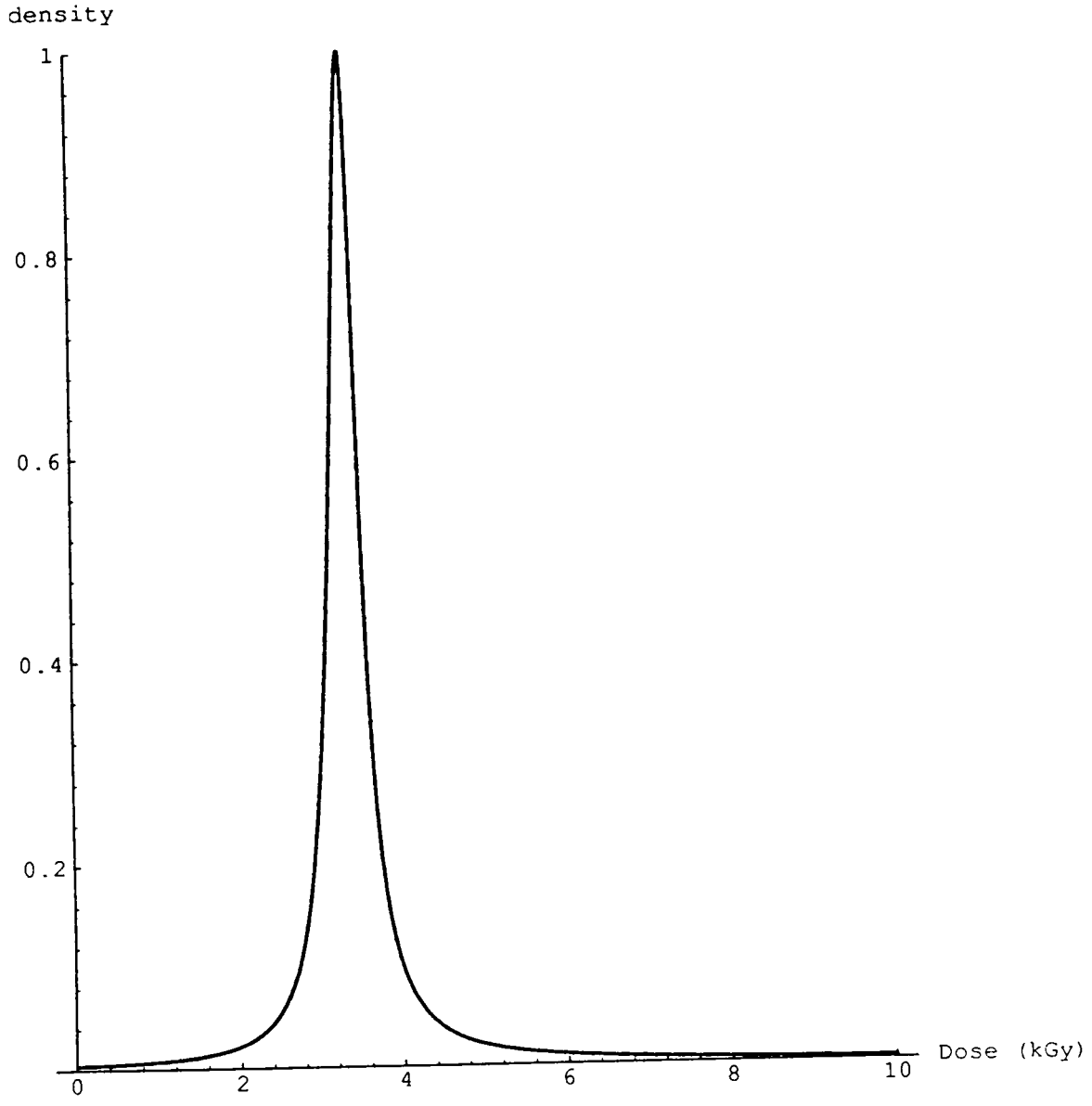
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.181+(.021*(u-1.8)^2))])
*(1+((0-((-2.29*u)+9.63))^2/(1*(.181+(.021*(u-1.8)^2))))^1))
/.578986)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



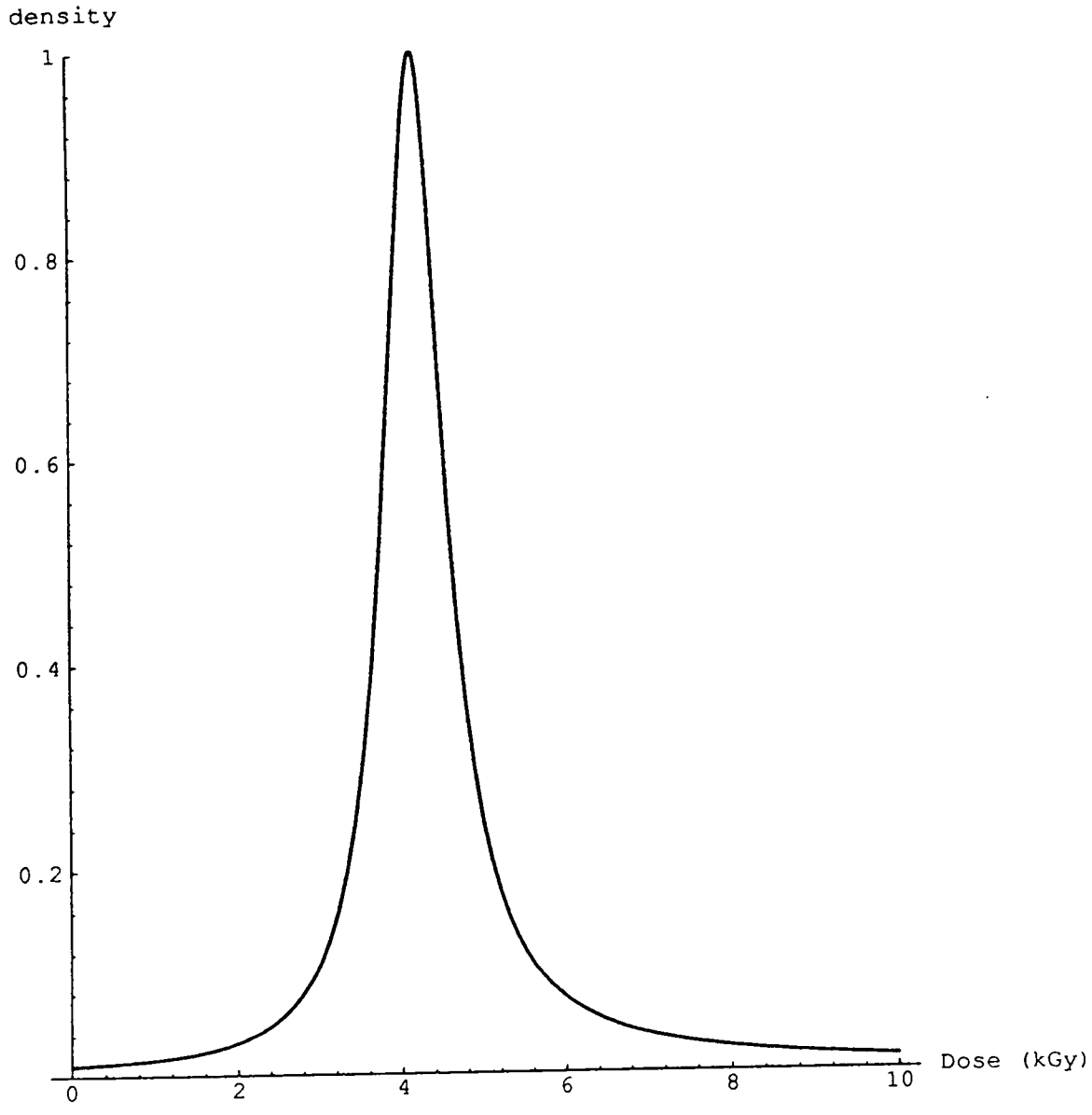
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.181+(.021*(u-1.8)^2))])
*(1+((2-((-2.29*u)+9.63))^2/(1*(.181+(.021*(u-1.8)^2))))^1))
/.66344)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



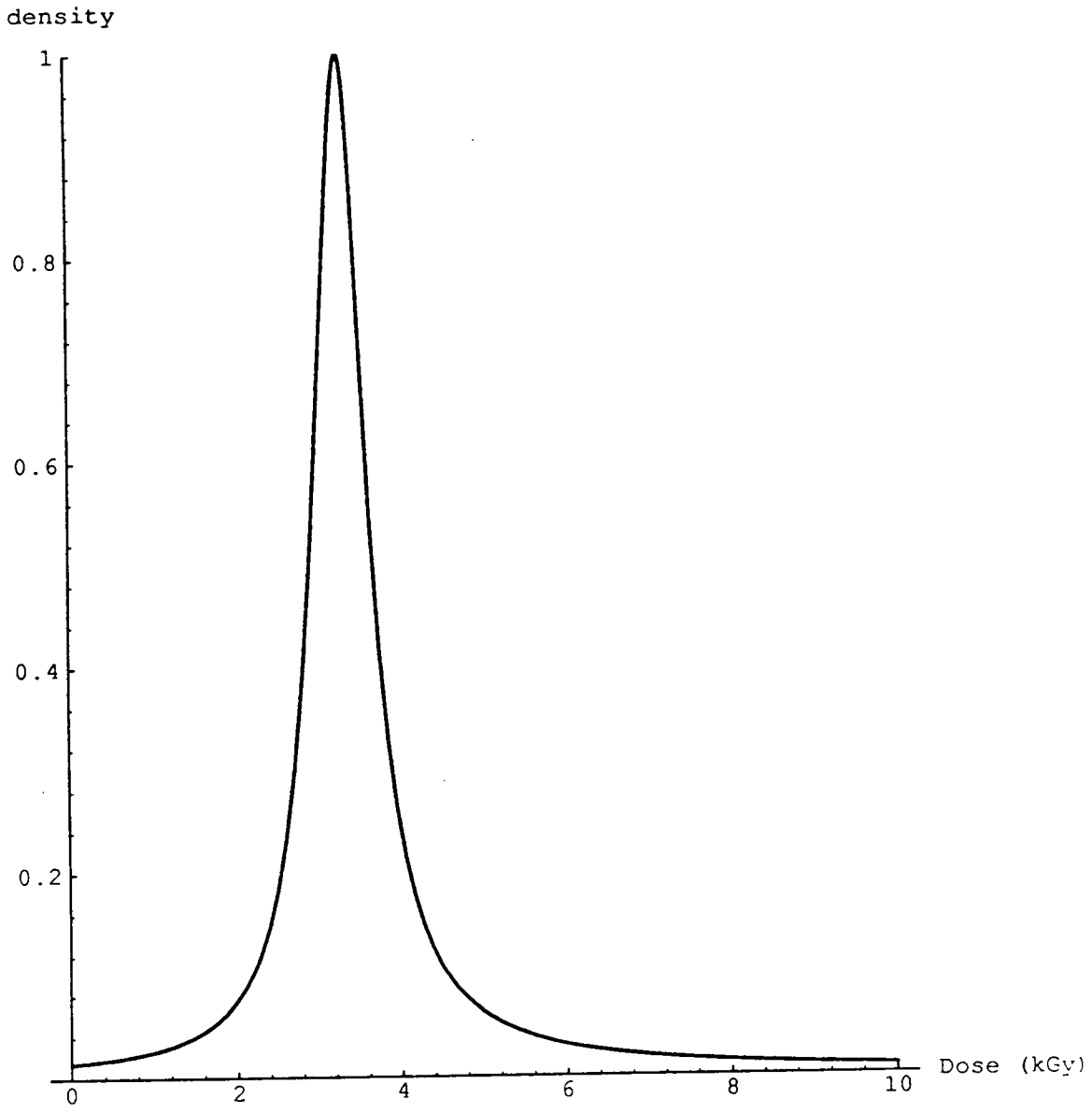
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.597+(.0693*(u-1.8)^2))])
*(1+((0-((-2.233*u)+9.347))^2/(1*(.597+(.0693*(u-1.8)^2))))^1))
/.320114)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



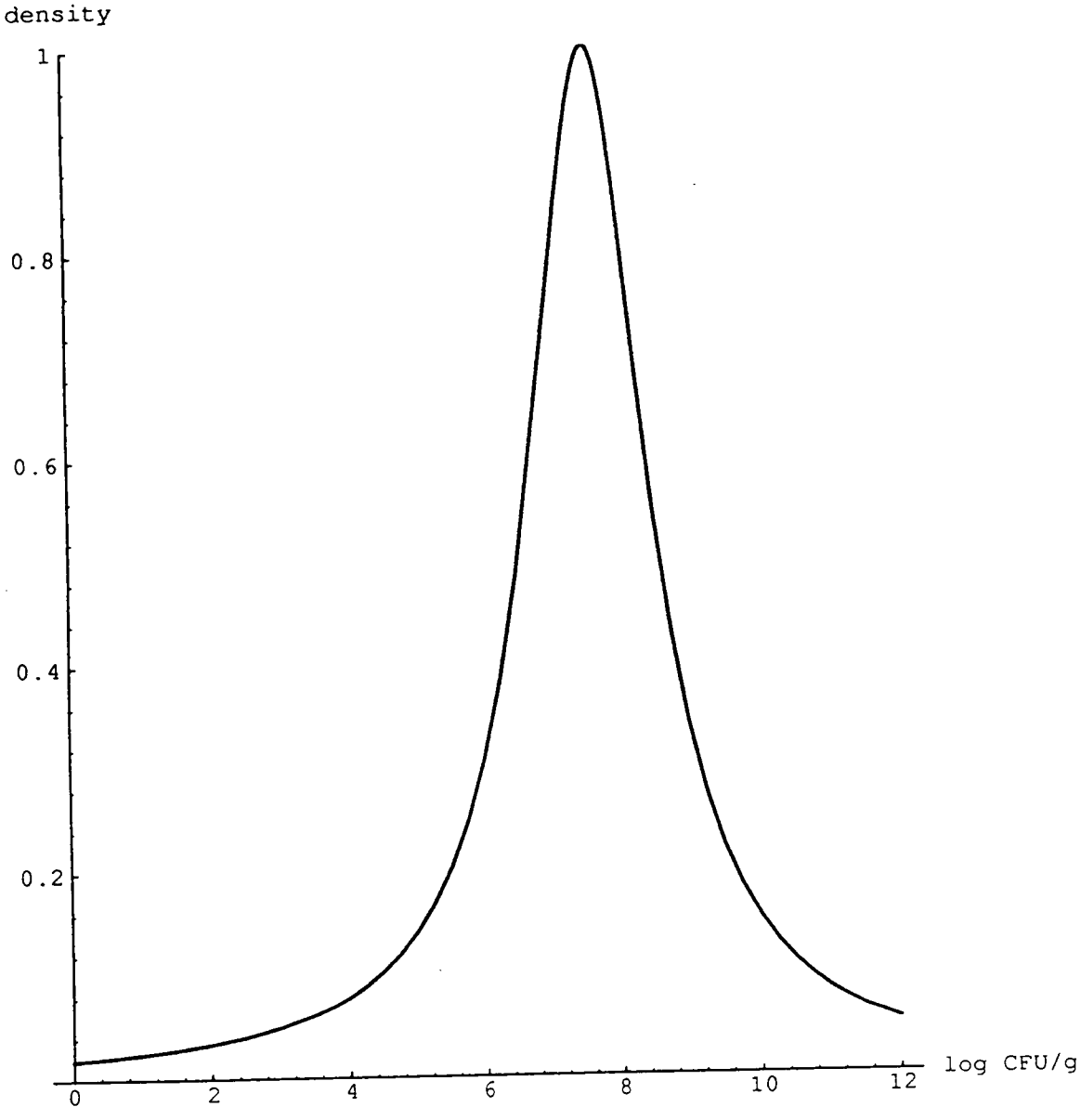
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.597+(.0693*(u-1.8)^2))])
*(1+((2-((-2.233*u)+9.347))^2/(1*(.597+(.0693*(u-1.8)^2))))^1))
/.367594)
Plot[f[st_], {u,0,10},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



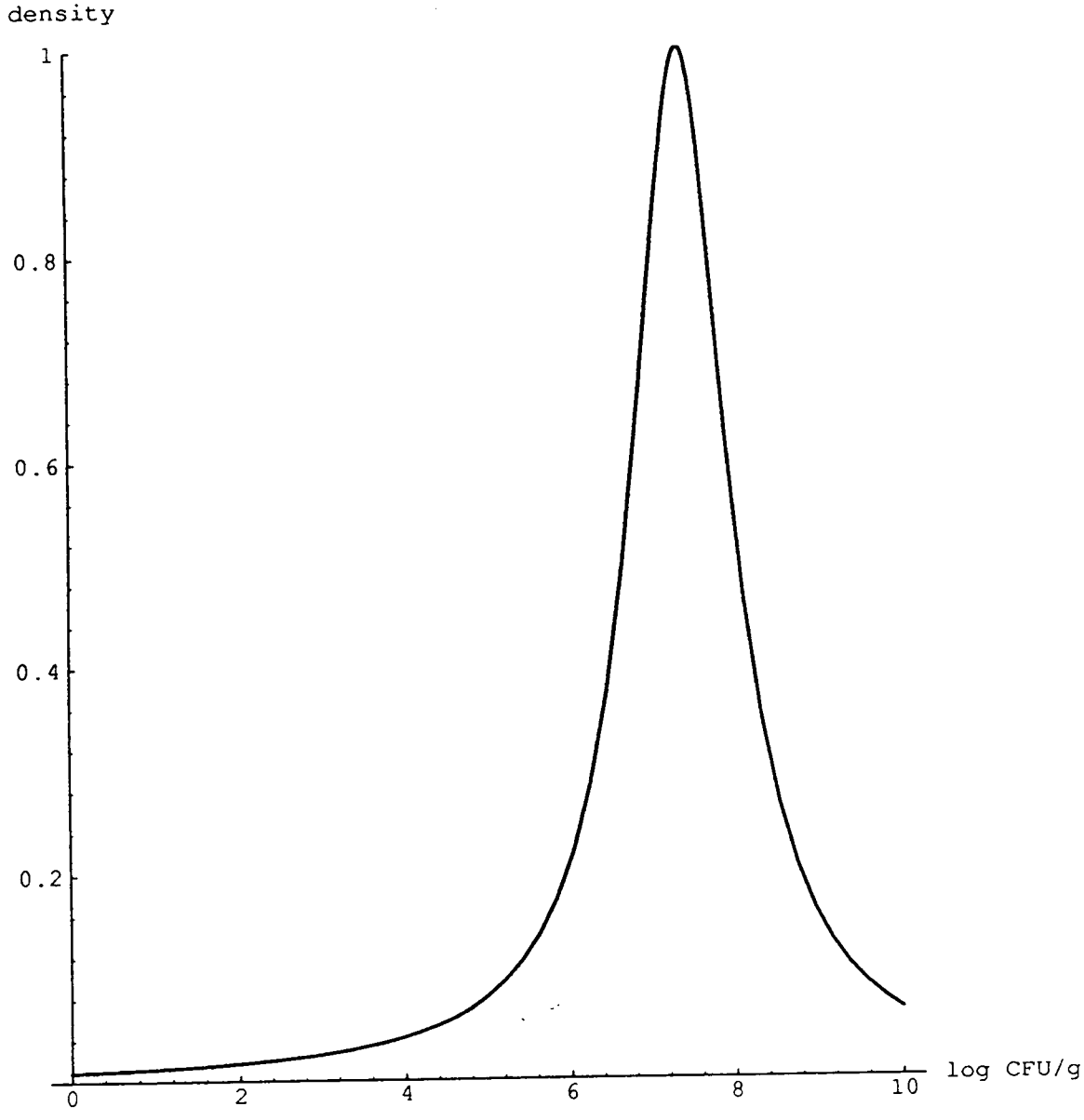
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*1.08])*(1+((u-7.56)^2
/(1*1.08))^1))/.306294)
Plot[f[st_], {u,0,12},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



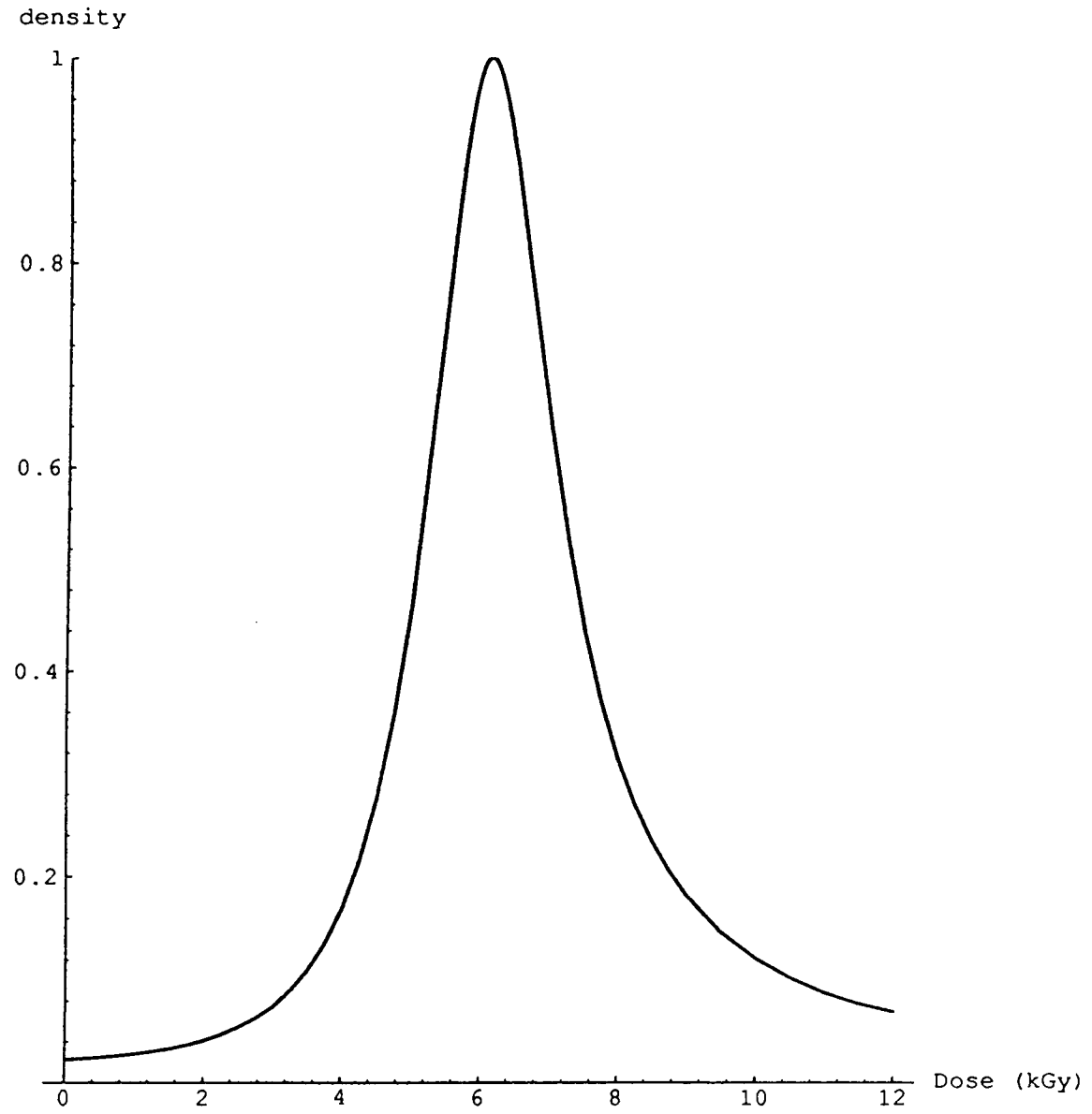
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*.497])*(1+((u-7.369)^2
/(1*.497))^1))/.451514)
Plot[f[st_], {u,0,10},PlotRange->{0,1},AspectRatio->1.2,
AxesLabel->{"log CFU/g","density"}]
```



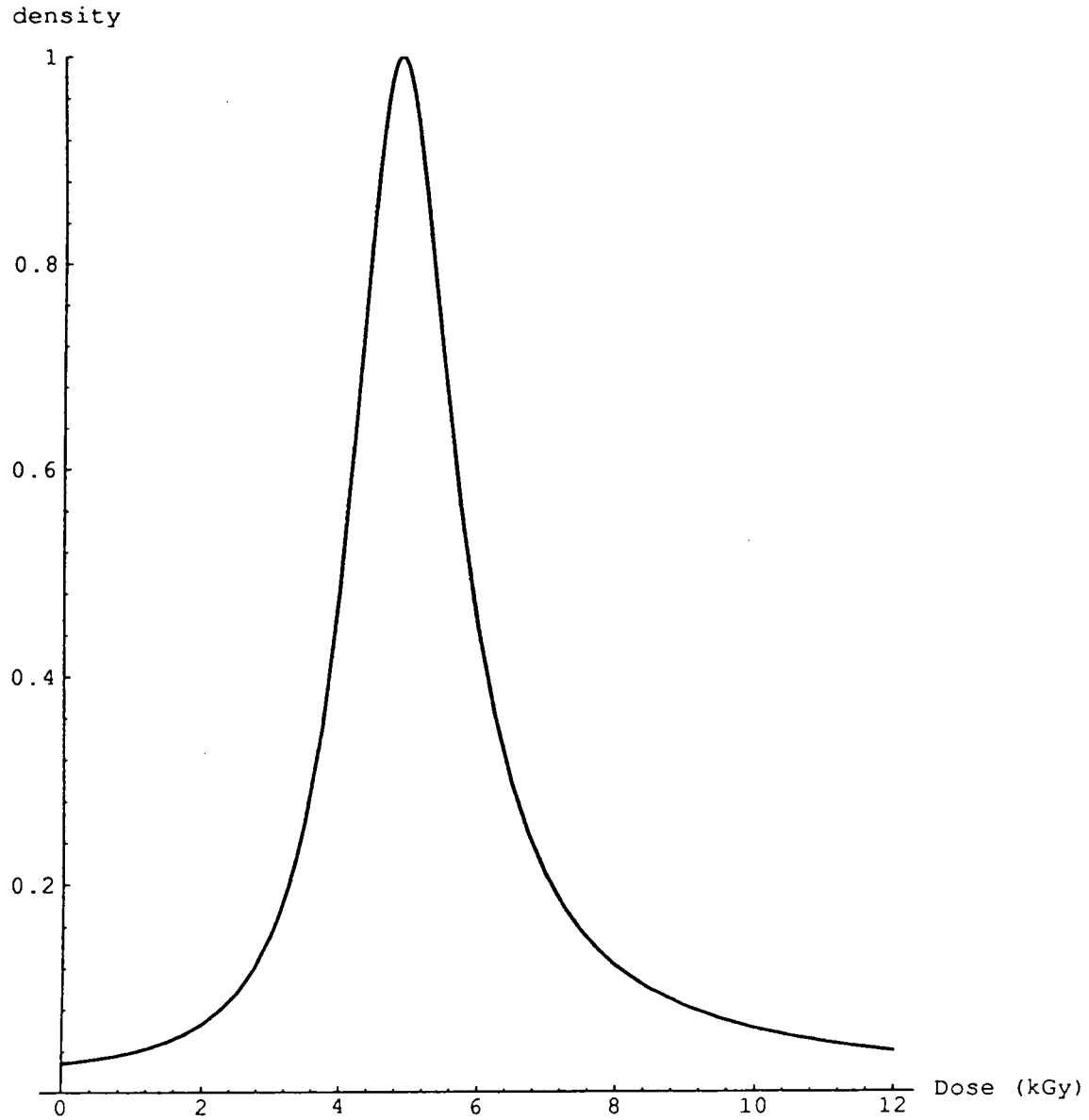
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(1.07+(.125*(u-1.8)^2))])
*(1+((0-((-1.61*u)+9.98))^2/(1*(1.07+(.125*(u-1.8)^2))))^1))
/.171821)
Plot[f[st_], {u,0,12},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



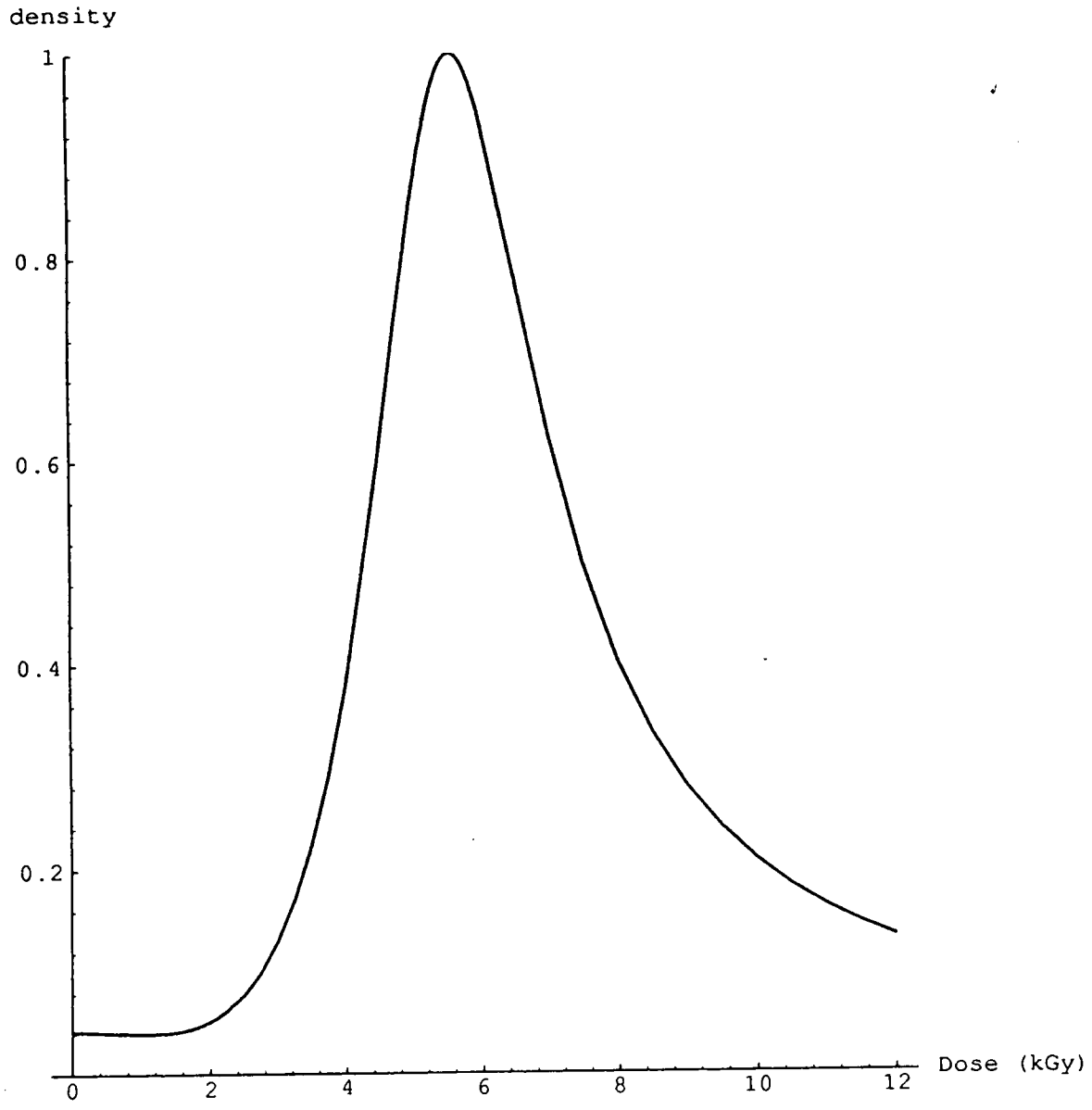
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(1.07+(.125*(u-1.8)^2))])
*(1+((2-((-1.61*u)+9.98))^2/(1*(1.07+(.125*(u-1.8)^2))))^1))
/.210517)
Plot[f[st_], {u,0,12},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



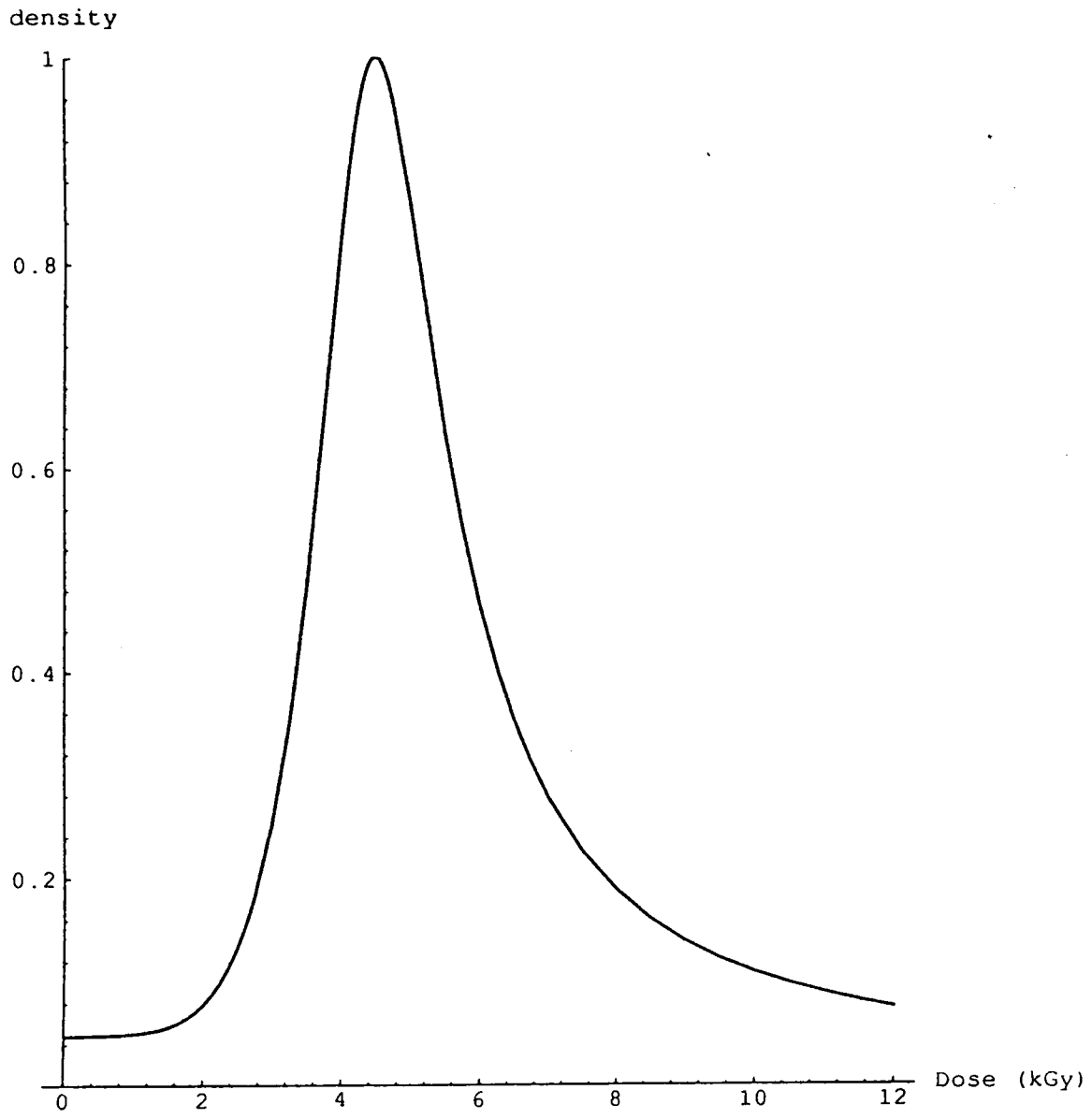
-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.597+(.492*(u-1.8)^2))])
*(1+((0-((-1.667*u)+9.870))^2/(1*(.597+(.492*(u-1.8)^2))))^1))
/.110593)
Plot[f[st_], {u,0,12},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



-Graphics-

```
f[st_]:= (1/((Beta[.5,.5])*(Sqrt[1*(.597+(.492*(u-1.8)^2))])
*(1+((2-((-1.667*u)+9.870))^2/(1*(.597+(.492*(u-1.8)^2))))^1))
/.150738)
Plot[f[st_], {u,0,12},AxesLabel->{"Dose (kGy)",
"density"},PlotRange->{0,1},AspectRatio->1.2]
```



-Graphics-

VITA

Michael Edward Howard was born in Silver Spring, Maryland on November 19, 1969. His family moved to Clearwater, Florida in 1975. In June of 1988 he graduated from Pinellas Park High School in Largo. He attended the University of South Florida before transferring to the University of Tennessee, Knoxville in May 1990. In December of 1992 he received his Bachelors degree in Nuclear Engineering. He then enrolled in the Masters program at the University of Tennessee, Knoxville and received his degree in May 1995.