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Computer Analysis of Cell Volume Distributions

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Computer Analysis of Cell Volume Distributions

by

Phillip N. Dean

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ABSTRACT

Two computer programs have been written to analyze cell volume distributions measured with a Coulter type of cell spectrometer. One of the programs, called AVØL, is used to calculate the mean cell volume of any type of distribution and to plot the data in various formats. The other program, called CELVØL, uses an iterative least-squares technique to fit either normal, log-normal, or skewed-normal distributions to the data. Either single- or double-peak distributions can be fitted. The program also converts the results of the fit to cell concentration in each peak in cells per cubic millimeter, standard deviation in cubic microns, and mean cell volume in cubic microns. For red blood cells the mean cell volume is also calculated from the hematocrit and included in the data output listing.

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CHAPTER 1

INTRODUCTION

The use of electronic particle counters to measure volume distributions of cells has brought forth a requirement for an accurate method of analyzing the volume spectra (1). To study the changes in volume distributions of various cells under different experimental conditions requires a precise knowledge of the parameters of the distribution. Some cell volume distributions can be fitted quite accurately with normal, log-normal, or skewed-normal functions. Such calculations require computer techniques. This report describes two computer programs which perform these calculations. The parameters calculated are the area (number of cells), standard deviation, mean, and, if applicable, coefficient of skewness. The CELVØL program will fit either single- or double-peak distributions. Two peaks with mean volumes of 50 and 60 cubic microns have been resolved by this program. The function chosen to fit the data is determined primarily by the type of cell being measured. Red and white blood cells appear to be fitted "best" by a skewed-normal function. Growing cell populations may have a distribution (determined by dynamics of growth) which cannot be fitted well with either function.

CHAPTER 2

AVØL COMPUTER PROGRAM

This program reads in the data, calculates the mean, standard deviation, and area for a selected band of channels, prints out the results, and then plots the data in various formats. There are options for plotting either linear or logarithmic coordinates in any combination. There are also options for excluding certain channels from the plots. All calculated data are also printed on the plot. The arithmetic mean of the distribution, μ , is calculated from the relation

$$\mu = \frac{\sum_{i=k}^l i f_i}{\sum_{i=k}^l f_i},$$

where i is the channel number, f_i is the number of cells in channel i , and k and l are the lower and upper channel numbers that define the band of data that are to be used in the calculation to exclude debris and pulser signals. This is shown in the example. The standard deviation, σ , is calculated from the relation

$$\sigma = \frac{\sum_{i=k}^1 f_i (i-\mu)^2}{\sum_{i=k}^1 f_i} ,$$

where the variables are as described above. The area is the summation $A = \sum_{i=k}^1 f_i$.

The format of the control card used with this program is shown in Table 1. If no lower and upper limits for the logarithmic plot are entered (columns 53 and 54), the limits are determined by the program. The program contains the option of having the entire spectrum shifted any number of channels (column 55). The plotting options can be used in any combination (columns 64 and 65). If zero, the plot will be linear only. If a logarithmic plot is called for, there will automatically be a linear plot as well. The ADD-1 value entered in columns 66-72 is the total number of pulses stored in the analyzer, obtained by counting the ADD-1 or stored pulses with an external scaler. The program will sum the counts in all channels and list this as well as the ADD-1, providing a check on accuracy of the data printout from the analyzer. The serial number is used to identify the data deck.

The control card is followed by the data cards. The data card format is shown in Table 2. For a 100-channel

Table 1. AVØL Control Card Format

Column	Information
1-52	Alpha-numeric information used to describe the data being analyzed
53	Lower limit for logarithmic plot of data; exponent of power of 10 (e.g., 2 for 100)
54	Upper limit for logarithmic plot of data; exponent of power of 10 (e.g., 4 for 10,000)
55	Number of channels spectrum is to be shifted
56	Set = 1, if only channels above 10 are to be plotted
57	Number of hundreds of channels to be read in (i.e., 1 for 100, 2 for 200, etc., up to 400)
58-59	First channel to be used in computation
60-62	Last channel to be used in computation
63	Set = 1, if printout of input and calculated data is desired
64	Set = 1 for plot of log X
65	Set = 1 for plot of log Y
66-72	ADD-1
73-80	Serial number

Table 2. AVØL Data Card Format

Column	Information
1-3	Sequence number of data card in the data deck, beginning with zero*
4-9	Channel 1
10-16	Channel 2
17-23	Channel 3
24-30	Channel 4
31-37	Channel 5
38-44	Channel 6
45-51	Channel 7
52-58	Channel 8
59-65	Channel 9
66-72	Channel 10
73-80	Serial number

*The second card would contain channels 11-20, the third channels 21-30, etc.

spectrum, there would be 10 cards. The serial number is repeated on each card.

The order of control card and data cards may be repeated as many times as desired. Figure 1 shows the card order of a problem processed by the AVØL program. Figure 2 shows the output listing. The selected area is the sum of the counts in channels 25-95. Data are printed out to channel 105, since the spectrum was shifted by 5 channels. Figure 3 is a log-log plot of the data, and Fig. 4 is a linear plot of the data. The plots also contain all computed values. A FORTRAN listing of the AVØL computer program, with the exception of the plotting subroutines, is contained in Appendix 1.

MEAN CELL VOLUME = 52.45 CH
 STANDARD DEVIATION = 13.99 CH
 SELECTED AREA = 19237. CELLS
 CHANNELS USED IN COMP. = 25 TO 95

ADD-1 TOTAL = 21146.
 SUM OF 105 CH = 21146.

	1	2	3	4	5	6	7	8	9	10
0	0	0	0	0	0	-0	-0	4	56	121
1	113	109	91	100	96	106	121	127	99	118
2	115	99	97	108	120	106	116	126	140	160
3	178	178	197	264	300	322	342	379	456	459
4	530	560	540	564	527	546	584	591	561	560
5	572	551	533	509	492	500	469	433	422	339
6	352	329	306	295	291	242	259	242	227	222
7	224	202	184	160	152	132	121	125	97	103
8	67	92	75	69	64	58	39	51	28	34
9	45	37	35	28	24	32	21	22	25	30
10	19	18	23	24	15	0	0	0	0	0

Fig. 2. AVØL output listing for the data shown in Fig. 1.

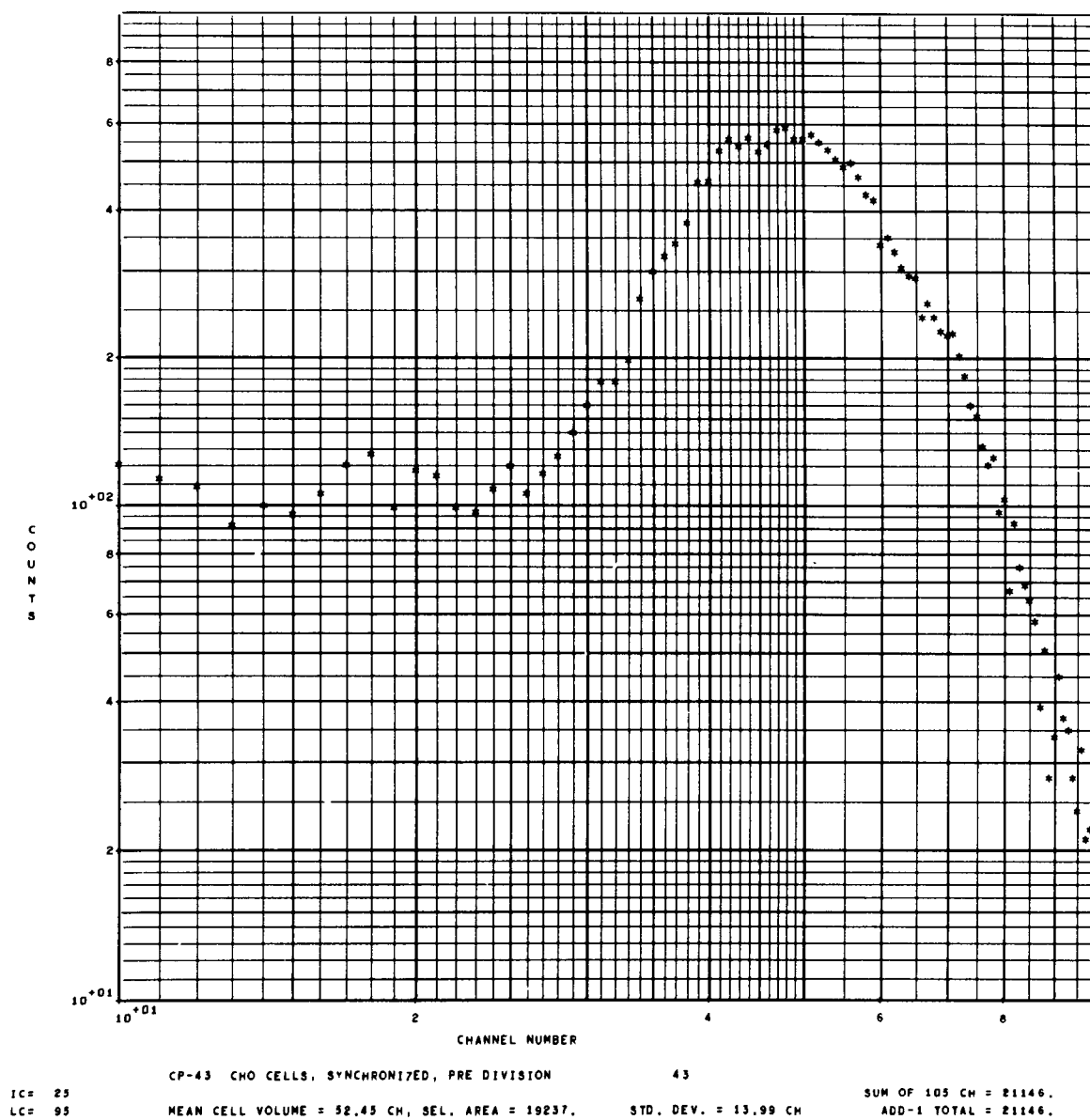


Fig. 3. A log-log plot of the count rate data of Fig. 2.

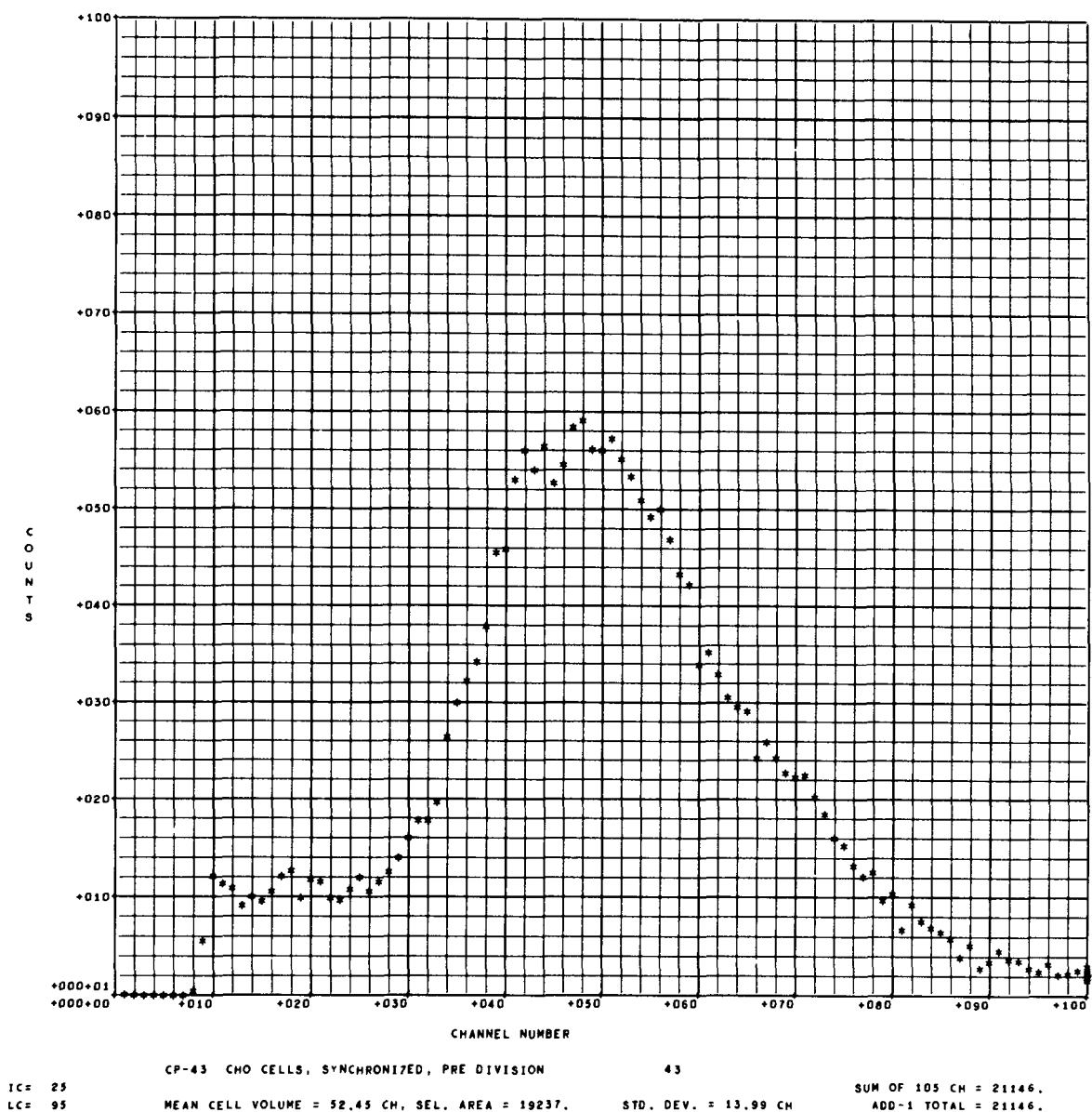


Fig. 4. A linear plot of the count rate data of Fig. 2.

CHAPTER 3

CELVØL COMPUTER PROGRAM

This program reads in the data, fits the peaks with normal, log-normal, or skewed-normal functions as specified by a control card, prints out the results, and plots the data points and computed curve. The function used in the normal fit is:

$$Y_1 = \frac{N_1}{\sqrt{2\pi} \sigma} \exp \left[-1/2 \left(\frac{X_1 - \mu}{\sigma} \right)^2 \right],$$

where Y_1 is the frequency (number of cells) in the i^{th} channel, N_1 is the area (total number of cells), σ is the standard deviation, and μ is the arithmetic mean of the distribution. A selected band of channels can be used in the fit to exclude debris and pulser signals from the computation. The function used in the log-normal fit is (2):

$$Y_1 = \frac{N_1}{\sqrt{2\pi} \sigma_{\log} X_1} \exp \left[-1/2 \left(\frac{\log X_1 - \mu_{\log}}{\sigma_{\log}} \right)^2 \right],$$

where $\sigma_{\log}$ is the standard deviation and $\mu_{\log}$ is the mean of the distribution of $\log X$'s. The interpretation of the results

of the fit is perhaps more convenient if the parameters are given for the distribution of X's rather than the log X's. The standard deviation and mean are transformed by the following relations (3):

$$\mu = \exp \left[\left(\mu_{\log} + \frac{\sigma_{\log}^2}{2M} \right) \frac{1}{M} \right]$$

$$\sigma^2 = \exp \left[\left(\mu_{\log} + \frac{\sigma_{\log}^2}{M} \right) \frac{2}{M} - \mu^2 \right],$$

where $M = \log_{10} e$.

The function used in the skewed-normal fit is (2):

$$Y_1 = \frac{N_i}{\sqrt{2\pi} \sigma} (1-A) \exp \left[-1/2 \left(\frac{X_1 - \mu}{\sigma} \right)^2 \right],$$

$$A = \frac{\alpha_3}{2} \left[\frac{X_1 - \mu}{\sigma} - 1/3 \left(\frac{X_1 - \mu}{\sigma} \right)^3 \right],$$

where α_3 is the coefficient of skewness and the other variables are as defined above. A coefficient of skewness of zero would result in a normal distribution.

The general least-squares procedure used to fit the data has been described in detail elsewhere (4). Briefly, the Gauss method of solving the nonlinear problem is utilized.

This method consists essentially of linearizing the desired function with respect to each of the parameters by means of a truncated Taylor's series. Initial "guesstimates" of the parameters are used to evaluate the coefficients of the expansion and to obtain new parameter estimates. Until some convergence criterion is satisfied, the iterative procedure is repeated. Our particular criterion is that every parameter estimate of the function must agree with its corresponding value in the previous iteration to within one part per million. The final "best" estimates of the parameters are then substituted in the classical least-squares equation to verify their validity.

If a single peak is being fitted, the program will compute the "guesstimates" by the method described in Chapter 2. If two peaks are being fitted, the "guesstimates" must be provided for the program.

The format of the control card used with this program is shown in Table 3. Polyethylene spheres are usually used to calibrate the gain of the counter. If a 1 is punched into column 69, the spectrum is treated as a calibration measurement. The program will calculate the mean channel number and then a ratio to the mean channel number of the original calibration (channel 50.3). The computed data are then corrected by this ratio. If spheres are measured both before and after a set of measurements, both mean channel numbers

Table 3. CELVØL Control Card Format

Column	Information
1-2	Experiment number
3-8	Serial number
9-24	Animal or subject identification (alpha-numeric)
26-32	Date, as 14SEP65
35	Number of peaks
38-39	Initial channel of data to be fitted
41-42	Last channel of data to be fitted
44-47	Time sample was counted, in minutes (decimal point between columns 46 and 47)
48-54	ADD-1 count (total number of pulses stored in analyzer)
55-59	Calibration factor to convert from channel number to cubic microns
60-63	Hematocrit
68	Set = 0 to fit data with skewed-normal function = 1 to fit data with normal function = 2 to fit data with log-normal function
69	Set = 1 if spectrum is of spheres (for calibration)
70	Set = 0 for main output sheet only = 1 for complete listing of input and calculated data = 2 for plot only
71	Parameter sign control = 1 for no control = 2 for sign fixed = 0 for sign fixed for 5 iterations
72	Set = 1 if "guesstimates" are to be read in (for 2 peaks)
73-80	Repeat of experiment number and serial number

are computed and then averaged. "Guesstimates" must be provided if the program is to fit two peaks to a set of data. The format of the "guesstimate" card is shown in Table 4. The data card format is the same as for the AVØL program and is shown in Table 2. The card order required by the program is shown in Table 5. There are several features built into the program to verify that all cards contain the correct information in the proper order. All cards of a data deck must contain the same serial number in columns 73-80. The 10 cards containing the spectrum are checked to verify that the sequence numbers in column 3 are in the correct order. The control card must contain the year in columns 31 and 32. It is used to identify the card as being the control card. If any errors are found, the program skips the spectrum, searches for the next sample, and continues computation.

Figure 5 shows the card order of two problems processed by the CELVØL program. Only one peak is fitted to the first spectrum and two to the second. Figures 6, 7, and 8 are the output listings for the first spectrum fitted with a skewed-normal function. Most of the information printed on the sheets is self-explanatory, although a few of the variables should be defined. The number of iterations required to reach the convergence criterion is printed. The maximum number of iterations that the program will attempt is 26. The

Table 4. CELVØL "Guesstimate" Card Formats

Column	Information
<u>For Normal and Log-Normal Fits</u>	
1-6	Area of first peak (number of cells)
7-14	Standard deviation of first peak (in channels)
15-22	Mean of first peak (in channels)
37-42	Area of second peak (number of cells)
43-50	Standard deviation of second peak (in channels)
51-58	Mean of second peak (in channels)
<u>For Skewed-Normal Fit</u>	
1-6	Area of first peak (number of cells)
7-14	Standard deviation of first peak (in channels)
15-22	Mean of first peak (in channels)
23-29	Coefficient of skewness of first peak
37-42	Area of second peak (number of cells)
43-50	Standard deviation of second peak (in channels)
51-58	Mean of second peak (in channels)
59-65	Coefficient of skewness of second peak

Table 5. CELVØL Card Order*

Card	Information
1	Control card
2	"Guesstimate" card, if required
3	First card of data deck, containing only the serial number in channels 73-80
4	Data deck (10 cards)

*This card order can be repeated as many times as desired.

704 LA S 080																			
PROGRAMMER										PROBLEM									
1	6	7	12	13	18	19	24	25	30	31	36	37	42	43	48	49	54	55	60
										DATE									
61	66	67	72	73	PAGE					OF									
1	6	7	12	13	18	19	24	25	30	31	36	37	42	43	48	49	54	55	60
11000085																			
0	3		5		3		8		8		10		17		17		30		37
1	33		46		58		66		64		93		90		123		159		192
2	240		335		520		749		1030		1501		2016		2560		3147		3623
3	3922		4050		4133		3883		3546		3119		2633		2187		1693		1399
4	1002		684		541		393		260		178		130		95		63		60
5	47		36		27		44		34		37		39		31		44		40
6	37		42		36		41		50		29		33		32		25		17
7	20		18		21		17		15		18		19		16		21		15
8	18		20		25		27		31		59		1533		1725		72		71
9	2		3		3		3		3		1		2		4		0		1
11000084																			
0			1		6		5		1		3		8		12		14		11
1	14		12		15		25		20		33		37		64		89		108
2	158		218		339		387		583		681		852		1007		1083		1049
3	1070		992		877		809		652		580		478		388		330		308
4	314		263		307		292		316		334		344		386		420		410
5	449		507		477		505		473		546		544		492		489		504
6	454		453		376		370		371		317		265		245		210		189
7	156		143		124		120		91		91		79		54		52		58
8	53		49		39		33		39		58		1342		2101		70		27
9	17		14		13		9		9		9		8		9		10		9

Fig. 5. Two examples of the card order required by the CELVØL computer program.

EXPT. 11 MOUSE NO. 3C SERIAL 11000085 16OCT64

SKewed NORMAL DISTRIBUTION (1) FIT TO EXPERIMENTAL DATA.

INPUT DATA CONSISTS OF 100 DATA POINTS (OR CHANNELS) OF WHICH CHANNELS 20 TO 50 ARE USED IN FIT.
COUNTING TIME = T = 1.00 MINUTES LIVE.

SUM OF 100 CHANNELS IS	55197 COUNTS.	NUMBER ITERATIONS =	7
ADD-1 SCALER RECORDED	55197 COUNTS.	WEIGHTED VARIANCE =	1.79
DIFFERENCE IS	0 COUNTS.		
SUM CHANNELS FITTED IS	49884 COUNTS.	UNWGT SUM SQUARES	
		OF DEVIATIONS	= 1.30E 05

MEAN CHANNEL NUMBER OF SPHERE SPECTRUM (11000081) IS 52.42

MEAN CHANNEL NUMBER OF SPHERE SPECTRUM (11000087) IS 51.90

RBC = CELLS PER CUBIC MILLIMETER = (K/T)(SUM 100 CHANNELS) = 14.13 MILLION, WHERE K = 256.0
HCT = 59.0
MCV = 41.8 CUBIC MICRONS.

PARAMETER		GUESSTIMATE	BEST ESTIMATE	STD.DEV.	CALIBRATED BEST ESTIMATE (STD.DEV.)
PEAK 1	AREA	49884. CTS	49807.	299.	12.75 (0.077) MILLION *
	S.D.	4.99 CHS	4.99	0.024	8.27 (0.039) CU MICRONS **
	MEAN	32.93 CHS	32.92	0.031	54.60 (0.051) CU MICRONS **
	SKEW	0.200	0.189	0.020	0.189(0.020)

* FACTOR = K/T
** FACTOR = 1.72(0.964)

MEAN CHANNEL NUMBER AS CALCULATED FROM 100 EXPERIMENTAL DATA POINTS = 36.63

Fig. 6. First page of CELVØL output listing for an analysis of the first example of Fig. 5.

CHANNEL NUMBER	WEIGHT FACTOR	EXPERIMENTAL DATA	CALCULATED DATA	DIFFERENCE
1	0.	3.	-0.	0.0
2	0.	5.	-0.	0.0
3	0.	3.	-0.	0.0
4	0.	8.	-0.	0.0
5	0.	8.	-0.	0.0
6	0.	10.	-0.	0.0
7	0.	17.	-0.	0.0
8	0.	17.	-0.	0.0
9	0.	30.	-0.	0.1
10	0.	37.	-0.	0.2
11	0.	33.	-0.	0.3
12	0.	46.	-1.	0.6
13	0.	58.	-1.	0.9
14	0.	66.	-1.	1.1
15	0.	64.	-1.	0.8
16	0.	93.	1.	-1.1
17	0.	90.	7.	-6.8
18	0.	123.	20.	-19.9
19	0.	159.	47.	-46.9
20	0.00521	192.	97.	95.1
21	0.00417	240.	182.	57.7
22	0.00299	335.	318.	17.4
23	0.00192	520.	518.	1.9
24	0.00134	749.	796.	-47.0
25	0.00097	1030.	1156.	-126.4
26	0.00067	1501.	1593.	-92.4
27	0.00050	2016.	2087.	-71.3
28	0.00039	2560.	2604.	-44.1
29	0.00032	3147.	3099.	48.2
30	0.00028	3623.	3522.	101.1
31	0.00025	3922.	3828.	93.7
32	0.00025	4050.	3985.	64.9
33	0.00024	4133.	3978.	155.0
34	0.00026	3883.	3814.	69.4
35	0.00028	3546.	3517.	29.4
36	0.00032	3119.	3124.	-5.4
37	0.00038	2633.	2679.	-46.3
38	0.00046	2187.	2222.	-34.6
39	0.00059	1693.	1784.	-91.2
40	0.00071	1399.	1390.	9.2
41	0.00100	1002.	1051.	-49.3
42	0.00146	684.	773.	-88.8
43	0.00185	541.	552.	-11.0
44	0.00254	393.	383.	9.9
45	0.00385	260.	258.	1.9
46	0.00562	178.	169.	9.4
47	0.00769	130.	107.	23.3
48	0.01053	95.	65.	29.8
49	0.01587	63.	39.	24.5
50	0.01667	60.	22.	38.1
51	0.	47.	12.	-12.0
52	0.	36.	6.	-6.3
53	0.	27.	3.	-3.2
54	0.	44.	2.	-1.6
55	0.	34.	1.	-0.7
56	0.	37.	0.	-0.3
57	0.	39.	0.	-0.1
58	0.	31.	0.	-0.1
59	0.	44.	0.	-0.0
60	0.	40.	0.	-0.0

Fig. 7. Second page of CELVØL output listing for an analysis of the first example of Fig. 5.

61	0.	37.	0.	-0.0
62	0.	42.	0.	-0.0
63	0.	36.	0.	-0.0
64	0.	41.	0.	-0.0
65	0.	50.	0.	-0.0
66	0.	29.	0.	-0.0
67	0.	33.	0.	-0.0
68	0.	32.	0.	-0.0
69	0.	25.	0.	-0.0
70	0.	17.	0.	-0.0
71	0.	20.	0.	-0.0
72	0.	18.	0.	-0.0
73	0.	21.	0.	-0.0
74	0.	17.	0.	-0.0
75	0.	15.	0.	-0.0
76	0.	18.	0.	-0.0
77	0.	19.	0.	-0.0
78	0.	16.	0.	-0.0
79	0.	21.	0.	-0.0
80	0.	15.	0.	-0.0
81	0.	18.	0.	-0.0
82	0.	20.	0.	-0.0
83	0.	25.	0.	-0.0
84	0.	27.	0.	-0.0
85	0.	31.	0.	-0.0
86	0.	59.	0.	-0.0
87	0.	1533.	0.	-0.0
88	0.	1725.	0.	-0.0
89	0.	72.	0.	-0.0
90	0.	7.	0.	-0.0
91	0.	2.	0.	-0.0
92	0.	3.	0.	-0.0
93	0.	3.	0.	-0.0
94	0.	3.	0.	-0.0
95	0.	1.	0.	-0.0
96	0.	2.	0.	-0.0
97	0.	4.	0.	-0.0
98	0.	0.	0.	-0.0
99	0.	1.	0.	-0.0
100	0.	6.	-0.	0.

Fig. 8. Third page of CELVØL output listing for an analysis of the first example of Fig. 5.

"guesstimate," final value, and standard deviation (S.D.) for each parameter are printed. The final values are also multiplied by calibration factors to convert the area from counts to millions of cells and the standard deviation and mean from channels to cubic microns. The calibration factors are also listed, identified by asterisks. The number in parentheses is the ratio of 50.3 to the average mean channel number of the two sphere spectra. The last line of the printout has the mean channel number calculated by the method described in Chapter 2 for the AVØL program. The second page of the listing contains the weighting factor, the experimental and calculated spectra, and their difference channel by channel. For accurate least-squares analysis of counting data, it is important that the data be properly weighted. "Statistical" weighting is used (that is, the reciprocal of the variance of the count rate in each channel) so that the weighted variance should be 1. Figure 9 is a plot of the experimental data (open circles) and computed spectrum (solid line). The solid circles show the limits of the data used in the calculation. The peak in channel 88 is due to a pulser and is used to monitor the gain of the electronics.

Figures 10, 11, and 12 are the output listings for the two-peak problem. Two log-normal distributions were used. The results of fitting each peak are listed separately. The

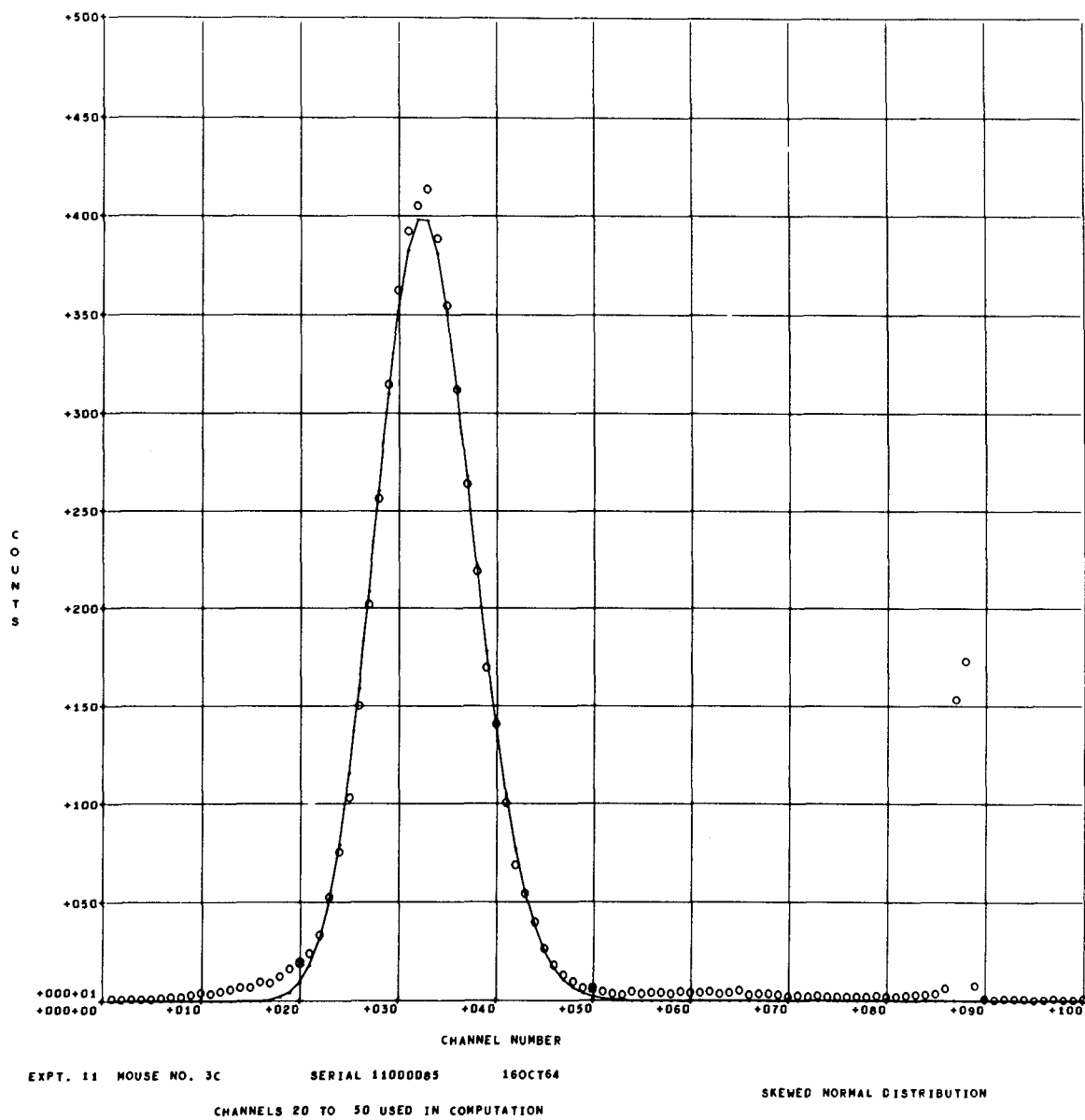


Fig. 9. A plot of the computed spectrum (solid line) and experimental data (open circles) of the example problem of Figs. 6, 7, and 8.

EXPT. 11 MOUSE NO. 14 SERIAL 11000084 16OCT64

LOG NORMAL DISTRIBUTION (2) FIT TO EXPERIMENTAL DATA.

INPUT DATA CONSISTS OF 100 DATA POINTS (OR CHANNELS) OF WHICH CHANNELS 20 TO 80 ARE USED IN FIT.
COUNTING TIME = T = 1.00 MINUTES LIVE.

SUM OF 100 CHANNELS IS	29827 COUNTS.	NUMBER ITERATIONS =	9
ADD-1 SCALER RECORDED	29827 COUNTS.	WEIGHTED VARIANCE =	1.17
DIFFERENCE IS	0 COUNTS.		
SUM CHANNELS FITTED IS	25539 COUNTS.	UNWGT SUM SQUARES	
		OF DEVIATIONS	= 5.14E 04

MEAN CHANNEL NUMBER OF SPHERE SPECTRUM (11000081) IS 52.42

MEAN CHANNEL NUMBER OF SPHERE SPECTRUM (11000087) IS 51.90

RBC = CELLS PER CUBIC MILLIMETER = (K/T)(SUM 100 CHANNELS) = 7.64 MILLION, WHERE K = 256.0
HCT = 41.8
MCV = 54.7 CUBIC MICRONS.

PARAMETER	GUESSTIMATES	LOG NORMAL BEST ESTIMATE	STD.DEV.	TRANSFORMED BEST ESTIMATE	STD.DEV.	CALIBRATED BEST ESTIMATE (STD.DEV.)
PEAK 1 AREA	12110. CTS	13457.	145.	13457.	145.	3.44 (0.037) MILLION *
S.D.	0.072 CHS	0.074	0.001	5.39	0.062	8.93 (0.102) CU MICRONS **
MEAN	1.48 CHS	1.49	0.001	31.18	0.020	51.73 (0.034) CU MICRONS **
PEAK 2 AREA	13613. CTS	12253.	145.	12253.	145.	3.14 (0.037) MILLION *
S.D.	0.089 CHS	0.072	0.001	9.66	0.131	16.03 (0.218) CU MICRONS **
MEAN	1.74 CHS	1.76	0.001	57.76	0.033	95.81 (0.055) CU MICRONS **

* FACTOR = K/T
** FACTOR = 1.72(0.964)

MEAN CHANNEL NUMBER AS CALCULATED FROM 100 EXPERIMENTAL DATA POINTS = 48.94

Fig. 10. First page of CELVØL output listing for an analysis of the second example of Fig. 5.

CHANNEL NUMBER	WEIGHT FACTOR	EXPERIMENTAL DATA	CALCULATED DATA	DIFFERENCE
1	0.	-0.	0.	0.
2	0.	1.	0.	0.
3	0.	6.	0.	0.
4	0.	5.	0.	-0.0
5	0.	1.	0.	-0.0
6	0.	3.	0.	-0.0
7	0.	8.	0.	-0.0
8	0.	12.	0.	-0.0
9	0.	14.	0.	-0.0
10	0.	11.	0.	-0.0
11	0.	14.	0.	-0.0
12	0.	12.	0.	-0.0
13	0.	15.	0.	-0.0
14	0.	25.	0.	-0.1
15	0.	20.	0.	-0.3
16	0.	33.	1.	-1.4
17	0.	37.	5.	-4.7
18	0.	64.	13.	-13.4
19	0.	89.	32.	-32.3
20	0.00926	108.	68.	40.1
21	0.00633	158.	127.	31.2
22	0.00459	218.	213.	5.0
23	0.00295	339.	327.	12.4
24	0.00258	387.	462.	-74.6
25	0.00172	583.	607.	-24.2
26	0.00147	661.	749.	-68.0
27	0.00117	852.	872.	-20.5
28	0.00099	1007.	965.	41.6
29	0.00092	1063.	1020.	62.9
30	0.00095	1049.	1034.	14.9
31	0.00093	1070.	1010.	60.1
32	0.00101	992.	954.	38.2
33	0.00114	877.	874.	2.7
34	0.00124	809.	781.	28.3
35	0.00153	652.	682.	-30.1
36	0.00172	580.	586.	-5.8
37	0.00209	478.	498.	-19.9
38	0.00258	388.	422.	-34.2
39	0.00303	330.	361.	-31.3
40	0.00325	308.	316.	-8.2
41	0.00318	314.	287.	27.4
42	0.00380	263.	272.	-8.7
43	0.00326	307.	270.	37.2
44	0.00342	292.	279.	13.0
45	0.00316	316.	297.	18.9
46	0.00299	334.	322.	12.1
47	0.00291	344.	351.	-7.0
48	0.00259	386.	382.	3.7
49	0.00238	420.	414.	6.3
50	0.00244	410.	443.	-33.4
51	0.00223	449.	470.	-20.9
52	0.00197	507.	492.	15.0
53	0.00210	477.	509.	-31.9
54	0.00198	505.	510.	-15.0
55	0.00211	473.	525.	-52.0
56	0.00183	546.	524.	22.1
57	0.00184	544.	517.	26.9
58	0.00203	492.	505.	-13.0
59	0.00204	489.	488.	0.8
60	0.00198	504.	467.	36.5
61	0.00220	454.	443.	10.5

Fig. 11. Second page of CELVØL output listing for an analysis of the second example of Fig. 5.

62	0.00221	453.	417.	35.9
63	0.00266	376.	389.	-13.0
64	0.00270	370.	360.	10.1
65	0.00270	371.	330.	40.5
66	0.00315	317.	301.	15.7
67	0.00377	265.	273.	-7.8
68	0.00408	245.	245.	-0.4
69	0.00476	210.	219.	-9.4
70	0.00529	189.	195.	-5.9
71	0.00641	156.	172.	-16.2
72	0.00699	143.	151.	-8.4
73	0.00806	124.	132.	-8.4
74	0.00833	120.	115.	4.8
75	0.01099	91.	100.	-8.7
76	0.01099	91.	86.	5.0
77	0.01266	79.	74.	5.2
78	0.01852	54.	63.	-9.1
79	0.01923	52.	54.	-1.7
80	0.01724	58.	46.	12.4
81	0.	53.	39.	-38.5
82	0.	49.	32.	-32.5
83	0.	39.	27.	-27.2
84	0.	33.	23.	-22.8
85	0.	39.	19.	-19.0
86	0.	58.	16.	-15.8
87	0.	1342.	13.	-13.1
88	0.	2101.	11.	-10.9
89	0.	70.	9.	-9.0
90	0.	27.	7.	-7.4
91	0.	17.	6.	-6.1
92	0.	14.	5.	-5.0
93	0.	13.	4.	-4.1
94	0.	9.	3.	-3.3
95	0.	9.	3.	-2.7
96	0.	9.	2.	-2.2
97	0.	8.	2.	-1.8
98	0.	9.	1.	-1.5
99	0.	10.	1.	-1.2
100	0.	9.	1.	-0.9

Fig. 12. Third page of CELVØL output listing for an analysis of the second example of Fig. 5.

"guesstimates" are made and entered on the guesstimate card for the normal plane. The program then converts them to the log plane. The "best" fit values are then transformed back to the normal plane by the relations described earlier. The calibration values are as described for the first example. Figure 13 is a plot of the experimental data (open circles) and computed spectrum (solid line).

A FORTRAN listing of the CELVØL program, with the exception of the plotting and matrix inversion subroutines, is contained in Appendix 2.

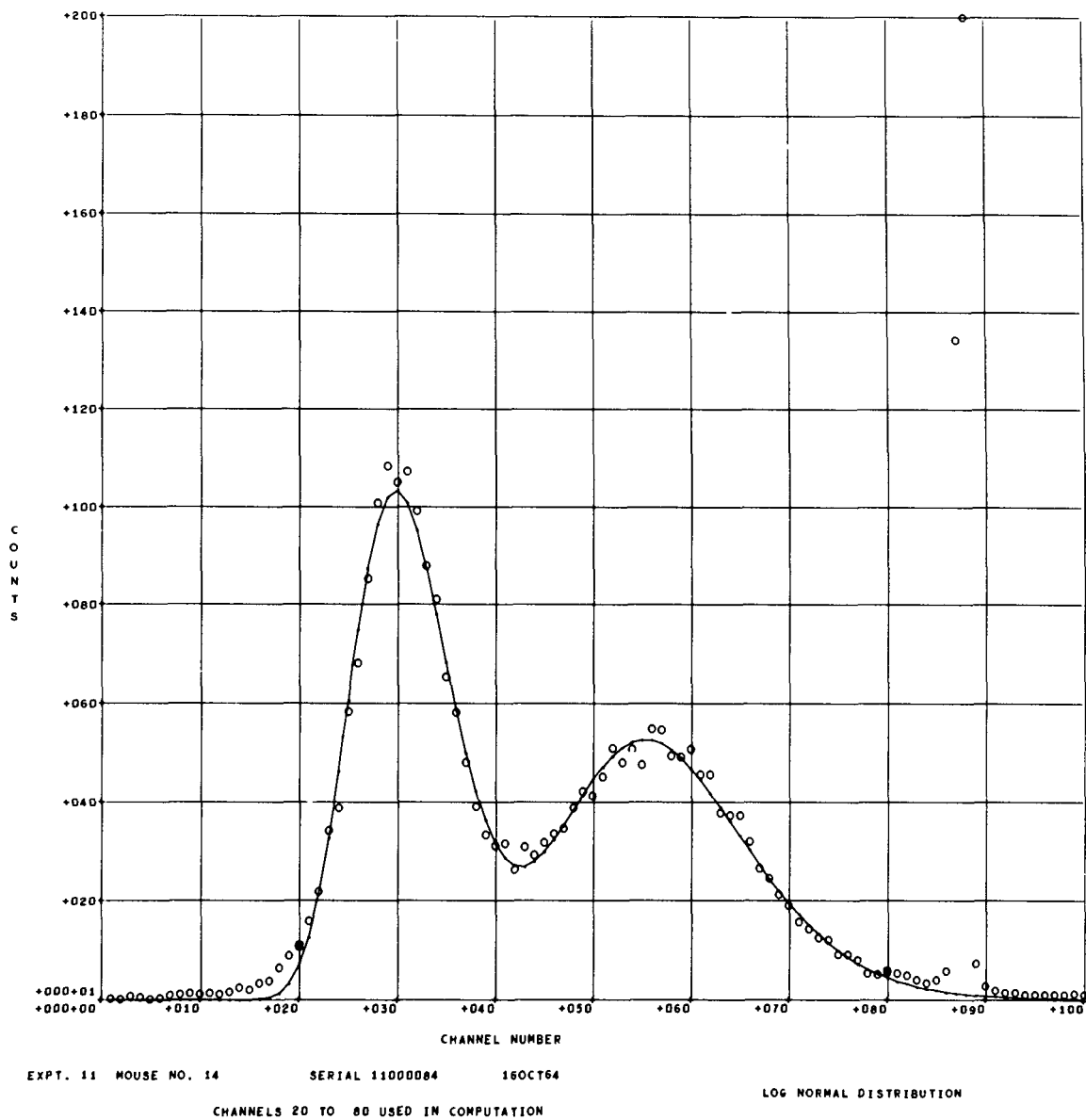


Fig. 13. A plot of the computed spectrum (solid line) and experimental data (open circles) of the example problem of Figs. 10, 11, and 12.

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- (1) Los Alamos Scientific Laboratory Report LA-3132-MS
(1964), pp. 142-232.
- (2) F. E. Croxton and D. J. Cowden, eds., Applied General Statistics, Prentice-Hall, Englewood Cliffs, New Jersey
(1958), pp. 617-619.
- (3) J. F. Kenney and E. S. Keeping, Mathematics of Statistics, 2nd edition, Van Nostrand, New York (1951),
p. 122.
- (4) R. H. Moore and R. K. Zeigler, Los Alamos Scientific
Laboratory Report LA-2367 (1960).

APPENDIX 1

AV0L

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C   THIS PROGRAM COMPUTES MEAN CELL VOLUME, TOTAL NUMBER OF CELLS
C   AND STANDARD DEVIATION.
C   THE SPECTRUM IS PLOTTED WITH OPTIONS OF LINEAR OR LOG SCALES.
C   DATA CARD FORMAT IS COULTER COUNTER FORMAT.
C   DIMENSION ALAB(11),CH(450),S(15),BND(4),AB(450),Y(450),TEMP(450),
      BLAB(1),CLAB(3),NM(10),ICH(450),YCH(450)
      1 READ INPUT TAPE 10,100,(ALAB(I),I=1,9),IZ3,IZ4,IZ1,IZ2,NC,
      11C,LC,IP,IX,IY,ADD1,ALAB(10),ALAB(11)
100  FORMAT(8A6,A4,5I1,I2,I3,3I1,F7.0,A6,A2)
      DO 200 I=1,450
      CH(I)=0.
      ICH(I)=0.
      YCH(I)=0.
      AB(I)=0.
200  Y(I)=0.
      IF(NC-1) 2,2,3
      2  NC=1
      INC=1
      3  N=NC*100+IZ1
      M=N
      IW=IZ1+1
      READ INPUT TAPE 10,101,(CH(I),I=IW,N)
      DO 201 I=1,N
201  AB(I)=I
      IX3=0
101  FORMAT(3X,F6.0,9F7.0)
      BCH=0.
      DO 7 I=1,N
      7  BCH=BCH+CH(I)
C   COMPUTE MEAN CHANNEL NUMBER
      SMCN=0.
      CHS=0.
      IC=IC+IZ1
      LC=LC+IZ1
      DO 6 I=IC,LC
      A=I
      SMCN=SMCN+A*CH(I)
      6  CHS=CHS+CH(I)
      BMCN=SMCN/CHS
C   BMCN=MEAN CHANNEL NUMBER
C   CALCULATE SIGMA
      RHM=0.
      DO 8 I=IC,LC
      A=I
      8  RHM=RHM+CH(I)*(BMCN-A)**2
      SIGMA=SQRTF(RHM/CHS)
      AREA=CHS
C   PLOT ROUTINE
58  CONTINUE
      BND(1)=0.
      BND(3)=0.
      IF(IY) 9,9,13
      9  S(1)=5.0
      S(2)=10.0
      S(3)=20.0

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      DO 90 I=1,N
90  Y(I)=CH(I)
      DO 10 I=4,15,3
      S(I)=S(I-3)*10.
      S(I+1)=S(I-2)*10.
10  S(I+2)=S(I-1)*10.
      ISY1=10
      ISY2=6
      ILY=6
      GO TO 60
13  DO 15 I=1,6
15  S(I)=10.**I
      DO 30 I=1,N
30  Y(I)=LOG10F(CH(I))
60  CONTINUE
      CALL MAX3(CH,-1,N,IPX,CHMAX)
      DO 12 I=1,15
      IF(CHMAX-S(I)) 11,11,12
11  BND(4)=S(I)
      J=1
      GO TO 16
12  CONTINUE
16  IF(IY) 18,18,17
17  BND(4)=J
      JY=0
      DO 170 I=IW,N
      JY=JY+1
170  YCH(JY)=CH(I)
      L=NC*100
      CALL MIN3(YCH,-1,L,IPY,CHMIN)
      CI=1.
      JI=0
      IF(CHMIN-10.) 65,64,64
64  CI=CI*10.
      JI=JI+1
      IF(CHMIN-CI) 62,62,64
62  BND(3)=JI-1
65  CONTINUE
      ISY1=BND(4)-BND(3)+10.
      ISY2=0
      ILY=ISY1
18  IF(IX) 19,19,21
19  BND(2)=NC*100
      ISX1=10
      ISX2=6
      ILX=6
      DO 190 I=1,N
190  AB(I)=1
      GO TO 25
21  IF(NC-1) 23,23,22
22  INC=2
23  BND(2)=INC+1
      ISX1=11+INC
      ISX2=0
      ILX=11+INC
54  IF(IZ2) 52,52,51

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51 BND(1)=1.
   ISX1=10+INC
   ILX=10+INC
   JY=0
   DO 55 I=10,N
   JY=JY+1
   AB(JY)=LOG10F(AB(I))
55 Y(JY)=Y(I)
   GO TO 25
52 DO 24 I=1,N
   A=I
24 AB(I)=LOG10F(A)
25 CONTINUE
   IF(IZ3) 251,251,250
250 ISY1=IZ4-IZ3+10
   ILY=ISY1
   BND(3)=IZ3
   BND(4)=IZ4
251 CONTINUE
   CALL ADV FLM(0,1)
   CALL SEL GRD(ISX1,ISY1)
   CALL SEL GRD(ISX2,ISY2)
   CALL LBL GRD(ILX,ILY,BND)
   CALL PLFILM(M,TEMP,AB,-1,Y,-1,BND,44,0)
   BLAB(1)=6HCOUNTS
   CLAB(1)=6HCHANNE
   CLAB(2)=6H NUMB
   CLAB(3)=2HER
   CALL WFILM(1,6,BLAB,0,30.0,6.0)
   CALL WFILM(0,14,CLAB,0,59.0,55.0)
   CALL WFILM(0,66,ALAB,0,61.0,20.0)
   CALL LINCNT(62)
105 FORMAT(5H IC=I4,91X,6HSUM OF I4,5H CH =F7.C)
   WRITE OUTPUT TAPE 12,105,IC,N,BCH
   CALL LINCNT(63)
   WRITE OUTPUT TAPE 12,106,LC,BMCN,AREA,SIGMA,ADD1
106 FORMAT(5H LC=I4,11X,18HMEAN CELL VOLUME =F6.2,
14H CH,12H SEL. AREA =F7.0,6X,11HSTD. DEV. =,
2F6.2,3H CH9X,13HADD-1 TOTAL =F7.0)
107 FORMAT(6X18HMEAN CELL VOLUME =F6.2,3H CH,
12X13HADD-1 TOTAL =F8.0/
26X20HSTANDARD DEVIATION =F6.2,3H CH,
31X6HSUM OF I4,5H CH =F8.0/
46X15HSELECTED AREA =F8.0,6H CELLS/
56X24HCHANNELS USED IN COMP. =I3,3H T014)
108 FORMAT(1H ,5X10I7)
109 FORMAT(1H0I6,10I7)
110 FORMAT(1H5,10A6,A2)
   IF(IX3) 57,57,59
57 IF(IX) 40,40,41
40 IF(IY) 59,59,41
41 IX=0
   IY=0
   IZ2=0
   IX3=1
   IZ3=0

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GO TO 58
59 CONTINUE
  IF(IP) 1,1,26
26 WRITE OUTPUT TAPE 9,110,{ALAB(I),I=1,11}
  WRITE OUTPUT TAPE 9,111
  WRITE OUTPUT TAPE 9,107,BMCN,ADD1,SIGMA,N,8CH,AREA,IC,LC
  WRITE OUTPUT TAPE 9,111
111 FORMAT(1H-)
DO 27 I=1,10
27 NM(I)=I
  WRITE OUTPUT TAPE 9,108,{NM(I),I=1,10}
  M=NC*10+1
  IJ=1
  IK=10
DO 270 I=1,N
270 ICH(I)=CH(I)
DO 28 I=1,M
  J=I-1
  WRITE OUTPUT TAPE 9,109,J,{ICH(K),K=IJ,IK}
  IJ=IJ+10
28 IK=IK+10
GO TO 1
END(1,0,0,0,0,0,1,0,0,1,0,C,0,0,0,0)
```

APPENDIX 2

CELVOL

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C   THIS PROGRAM IS WRITTEN TO FIT ONE OR MORE SKEWED GAUSSIANS TO
C   CELL VOLUME SPECTRA TAKEN WITH THE COULTER COUNTER
C   PROGRAM COMPUTES ESTIMATES FOR A SINGLE PEAK.
C   FOR TWO PEAKS THE ESTIMATES MUST BE GIVEN TO THE PROGRAM.
C   THE PROGRAM CONTAINS THE OPTION OF FITTING THE DATA WITH NORMAL,
C   SKEWED NORMAL, OR LOG NORMAL FUNCTIONS
C   WRITTEN IN JANUARY 1965
C   DIMENSION ANID(3),CH(100),AH(100),AMCN(2),IEXS(2),IS1S(2),IS2S(2),
1BB(2),TEMP(100),AB(100),S(15),BND(4)
C   DIMENSION Y(100),W(100),YC(100),DY(100),PG(40),P(40),SP(40),
1BM(40,41),X(1,100),B(100),ALAB(1),BLAB(3),AN(40),PART(40),
2Z(40),AM(40,40),DP(40),PC(40)
C   DIMENSION PCC(6),AR(2),PERS(2),PERM(2),PERA(2),SPC(6)
COMMON Y,X,W,Z,PG,P,SP,YC,DY,BM,B,AN,WVAR,SSQ,PART,TEST,NIT,IFG
1,IFF,NPK
C   FORMATS FOR DATA INPUT
1   FORMAT(I2,I3,2A6,A4,A6,I2,I3,I4,I3,F5.1,F7.0,F5.1,F4.1,4X,5I1,
12I4)
2   FORMAT(2X,I1,F6.0,9F7.0,2I4)
100  READ INPUT TAPE 10,1,IEX,KS1,KS2,ANID(1),ANID(2),ANID(3),DATE,IYR,
1NPK,ICH,LCH,CT,ADD1,FK,HCT,IFF,IS,IP,IFG,IG,IS1,IS2
IF(IYR) 50,50,101
50  WRITE OUTPUT TAPE 9,51,IS1,IS2
51  FORMAT(21H1 SPECTRUM NUMBER2I4,44H IS IN DOUBT. THERE ARE TOO
1MANY DATA CARDS.)
GO TO 100
C   READ IN DATA CARDS (SHOULD BE 11 OF THEM)
101 IF(IG) 102,102,103
103 IF(1FF-1) 107,105,108
105 READ INPUT TAPE 10,106,(PG(I),I=1,6)
106 FORMAT(F6.0,2F8.2,14X,F6.0,2F8.2)
GO TO 102
107 READ INPUT TAPE 10,104,(PG(I),I=1,8)
104 FORMAT(F6.0,2F8.2,F7.2,7X,F6.0,2F8.2,F7.2)
GO TO 102
C   READ IN ESTIMATES FOR LOG NORMAL FIT
108 READ INPUT TAPE 10,106,(PG(I),I=1,6)
C   CONVERT ESTIMATES
D=PG(2)
A=SQRTF(PG(2)**2+PG(3)**2)
B=LOG10F(A/PG(3))
PG(2)=SQRTF(B)
PG(3)=LOG10F(PG(3))
D=PG(5)
A=SQRTF(PG(5)**2+PG(6)**2)
B=LOG10F(A/PG(6))
PG(5)=SQRTF(B)
PG(6)=LOG10F(PG(6))
102 K=-1
READ INPUT TAPE 10,2,IEC,(AH(I),I=1,10),JS1,JS2
IF(JS1+JS2-IS1-IS2) 37,3,37
C   IF NOT ZERO SERIAL NUMBERS DO NOT CHECK, SKIP SAMPLE
3   I1=1
I2=10
DO 6 M=1,10

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```

      K=K+1
      READINPUT TAPE 10,2,IEC,(CH(I),I=11,12),JS1,JS2
      IF(IEC-K) 39,4,39
C     IF NOT ZERO CARDS ARE OUT OF SEQUENCE
      4 IF(JS1+JS2-IS1-IS2) 37,5,37
      5 I1=11+10
      6 I2=12+10
C     IF IS =1, THIS IS SPHERE SPECTRUM, COMPUTE MCN AND STORE
      7 IF(IS) 12,12,8
      8 JS0=1
      IF(ISONE) 10,10,9
      9 JS0=2
     10 SMCN=0.
      CHS=0.
      AMCN(2)=0.
      DO 11 I=30,70
      A=I
      SMCN=SMCN+A*CH(I)
     11 CHS=CHS+CH(I)
      AMCN(JS0)=SMCN/CHS
      IEXS(JS0)=IEX
      IS1S(JS0)=KS1+1000
      IS2S(JS0)=KS2+1000
      ISONE=1
      GO TO 100
     12 ISONE =0
C     COMPUTE MEAN CHANNEL NUMBER
      SMCN=0.
      CHS=0.
      DO 13 I=ICH,LCH
      A=I
      SMCN=SMCN+A*CH(I)
     13 CHS=CHS+CH(I)
      BMCN=SMCN/CHS
      SMCN=0.
      CHS=0.
      DO 130 I=1,100
      A=I
      SMCN=SMCN+A*CH(I)
     130 CHS=CHS+CH(I)
      GMCN=SMCN/CHS
C     GMCN= MEAN CHANNEL NUMBER CALCULATED FROM ALL DATA POINTS
      NCHF=LCH-ICH+1
      N=100
      IF(IFF-1) 133,131,131
     133 IK=NPK*4
      GO TO 132
     131 IK=NPK*3
     132 CONTINUE
      M=1
      J=ICH-1
      DO 14 I=1,J
      W(I)=0.
     14 Y(I)=0.
      J=LCH+1
      DO 15 I=J,100

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```

      W(I)=0.
15  Y(I)=0.
      TCH=0.
      DO 16 I=ICH,LCH
      TCH=TCH+CH(I)
      W(I)=1./CH(I)
16  Y(I)=CH(I)
      SCH=0.
      DO 17 I=1,100
      X(I,I)=1
17  SCH=SCH+CH(I)
170 IF(IG) 171,171,19
171 IF(IP-1) 172,172,2010
2010 DO 2011 I=1,100
      YC(I)=0.
2011 AB(I)=I
      GO TO 2012
C    CALCULATE SIGMA
172 RHM=0.
      DO 18 I=ICH,LCH
18  RHM=RHM+Y(I)*(BMCN-X(I,I))**2
      SIGMA=SQRTF(RHM/TCH)
      AREA =TCH
      IF(IFF-1) 463,463,462
C    COMPUTE ESTIMATES FOR LOG NORMAL FIT
462 D=SIGMA
      A=SQRTF(D**2+BMCN**2)
      B=LOG10F(A/BMCN)
      SIGMA=SQRTF(B)
      BMCN=LOG10F(BMCN)
463 CONTINUE
C    ASSIGN ESTIMATES FOR COMPUTATION
C    MEAN CHANNEL NUMBER ESTIMATE, BASED ON FITTED CHANNELS
      PG(1)=AREA
      PG(2)=SIGMA
      PG(3)=BMCN
      PG(4)=0.2
19 TEST=0.000001
      CALL CELLP(M,N,IK)
C    WRITE ROUTINE
20 FORMAT(11H1      EXPT.13,2A6,A4,8X,6HSERIAL15,I4 ,6X,A6,I2//)
21 FORMAT(34H0      SKEWED NORMAL DISTRIBUTION (11,27H) FIT TO EXPERIM
      ENTAL DATA.////////)
      IS2=IS2+10000
      WRITE OUTPUT TAPE 9,20,1EX,ANID(1),ANID(2),ANID(3),IS1,IS2,
      1DATE,1YR
450 IF(IFF-1) 451,452,454
451 WRITE OUTPUT TAPE 9,21,NPK
      GO TO 457
452 WRITE OUTPUT TAPE 9,453,NPK
453 FORMAT(27H0      NORMAL DISTRIBUTION (11,27H) FIT TO EXPERIMENTAL D
      IATA.////////)
      GO TO 456
454 WRITE OUTPUT TAPE 9,455,NPK
455 FORMAT(31H0      LOG NORMAL DISTRIBUTION (11,27H) FIT TO EXPERIMENT
      IAL DATA.////////)

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456 INK=NPK*3+1
    P(INK)=0.
    PG(INK)=0.
    SP(INK)=0.
457 CONTINUE
22 FORMAT(76H-      INPUT DATA CONSISTS OF 100 DATA POINTS (OR CHANNEL
15) OF WHICH CHANNELS 13,3H TO 14,17H ARE USED IN FIT./
225H      COUNTING TIME = T =F5.2,14H MINUTES LIVE.)
    WRITE OUTPUT TAPE 9,22,ICH,LCH,CT
23 FORMAT(34H-      SUM OF 100 CHANNELS IS 13,14,8H COUNTS.,17X,
119HNUMBER ITERATIONS =13/34H      ADC-1 SCALER RECORDED 13,
214,8H COUNTS./34H      DIFFERENCE IS 17,8H COUNTS.,
317X,19HWEIGHTED VARIANCE =F6.2//34H      SUM CHANNELS FITTED
4 IS 13,14,8H COUNTS.17X 17HUNWGT SUM SQUARES/1H ,65X,13HOF DEVIATIO
5NS5X1H=1PE9.2)
    ID=TCH*0.0001
    D=ID*10000
    JD=TCH-D+10000.
    IE=SCH*0.0001
    D=IE*10000
    JE=SCH-D+10000.
    IF=ADD1*0.0001
    D=IF*10000
    JF=ADD1-D+10000.
    JG=SCH-ADD1
    WRITE OUTPUT TAPE 9,23,IE,JE,NIT,IF,JF,JG,4HVAR,ID,JD,SSQ
    IF(JS0) 29,29,24
24 DO 25 I=1,JS0
25 WRITE OUTPUT TAPE 9,26,IEXS(I),IS1S(I),IS2S(I),AMCN(I)
26 FORMAT(46H-      MEAN CHANNEL NUMBER OF SPHERE SPECTRUM (12,213,4H)
1 ISF6.2)
27 FORMAT(11H0)
    K=(14-JS0*2)/2
    DO 28 I=1,K
28 WRITE OUTPUT TAPE 9,27
    GO TO 310
29 DO 30 I=1,7
30 WRITE OUTPUT TAPE 9,27
31 FORMAT(68H      RBC = CELLS PER CUBIC MILLIMETER = (K/T)(SUM 100
1 CHANNELS) =F6.2,19H MILLION, WHERE K =F6.1/11H      HCT =F5.1/
211H      MCV =F5.1,15H CUBIC MICRONS.//)
310 RBC=FK/CT*SCH*0.000001
    VCM=10.*HCT/RBC
    WRITE OUTPUT TAPE 9,31,RBC,FK,HCT,VCM
34 FORMAT(11H 71X14H* FACTOR = K/T/
11H 70X16H** FACTOR = 1.721H(F6.3,1H))
35 FORMAT(75H-      MEAN CHANNEL NUMBER AS CALCULATED FROM 100 EXPERIM
1ENTAL DATA POINTS =F6.2)
    IF(1FF-1) 338,337,336
336 WRITE OUTPUT TAPE 9,330
    FORMATS FOR PRINTING LOG NORMAL FITS
C 330 FORMAT(11H041X10HLOG NORMAL16X11HTRANSFORMED20X10HCALIBRATED/
11H 11X9HPARAMETER4X12HGUESSTIMATES3X13HBEST ESTIMATE,
23X8HSTD.DEV.3X13HBEST ESTIMATE3X8HSTD.DEV.2X,
313HBEST ESTIMATE11H (STD.DEV.))
    GO TO 339

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337 WRITE OUTPUT TAPE 9,32
GO TO 339
338 WRITE OUTPUT TAPE 9,32
32 FORMAT(1H0,77X,10HCALIBRATED/1H 12X9HPARAMETER4X,11HGUESSTIMATE,
15X,26HBEST ESTIMATE STD.DEV.4X24HBEST ESTIMATE (STD.DEV.))
339 CONTINUE
I=1
NPKL=NPK
DO 36 K=1,NPKL
IF(IFF-1) 424,424,420
C CONVERT LOG NORMAL PARAMETERS TO LINEAR PLANE
C STD. DEV. CONVERTED ON PERCENT BASIS
420 CONTINUE
PCC(I)=P(I)
SPC(I)=SP(I)
PCC(I+1)=P(I+1)
SPC(I+1)=SP(I+1)
PCC(I+2)=P(I+2)
SPC(I+2)=SP(I+2)
PERA(K)=SP(I)/P(I)
PERS(K)=SP(I+1)/P(I+1)
PERM(K)=SP(I+2)/P(I+2)
P(I+2)=EXP((P(I+2)+0.5/0.434294*P(I+1)**2)/0.434294)
SP(I+2)=P(I+2)*PERM(K)
AY=2.0*PCC(I+2)+2.0/0.434294*P(I+1)**2
P(I+1)=EXP(AY/0.434294)-P(I+2)**2
P(I+1)=SQRT(P(I+1))
SP(I+1)=P(I+1)*PERS(K)
424 CONTINUE
BEA=P(I)*FK/CI*0.000001
BEAE=SP(I)/P(I)*BEA
IF(JS0) 352,352,351
351 AS0=JS0
SAVG=(AMCN(1)+AMCN(2))/AS0
C0F=50.3/SAVG
GO TO 353
352 C0F=1.
353 BESD=P(I+1)*1.72*C0F
BEM=P(I+2)*1.72*C0F
BESDE=SP(I+1)/P(I+1)*BESD
BEME=SP(I+2)/P(I+2)*BEM
BEC=P(I+3)
BECE=SP(I+3)
IF(IFF-1) 333,334,335
333 CONTINUE
WRITE OUTPUT TAPE 9,33,K,PG(I),P(I),SP(I),BEA,BEAE,PG(I+1),P(I+1),
1SP(I+1),BESD,BESDE,PG(I+2),P(I+2),SP(I+2),BEM,BEME,PG(I+3),P(I+3),
2SP(I+3),BEC,BECE
33 FORMAT(10H0 PEAK12,7H AREA6XF7.0,4X3HCTSSXF7.0,9XF5.0,7XF5.2
1,2H (F5.3,14H) MILLION */1H 14X4HS.D.10XF5.2,5H CHS9XF5.2,10X,
2F5.3,4XF5.2,2H (F5.3,15H) CU MICRONS **/
31H 14X4HMEAN10XF5.2,5H CHS9XF5.2,10XF5.3,3XF6.2,2H (F5.3,15H) CU
4MICRONS **/
51H 14X4HKEW11XF5.3,14XF5.3,9XF5.3,5XF5.3,1H(F5.3,1H)//)
GO TO 340
334 WRITE OUTPUT TAPE 9,332,K,PG(I),P(I),SP(I),BEA,BEAE,PG(I+1),P(I+1),

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      1,SP(I+1),BESD,BESDE,PG(I+2),P(I+2),SP(I+2),BEM,BEME
332 FORMAT(10H0   PEAK12,7H   AREA6XF7.0,4X3HCTS5XF7.0,9XF5.0,7XF5.2
      1,2H (F5.3,14H) MILLION   */1H 14X4HS.D.10XF5.2,5H  CHS9XF5.2,10X,
      2F5.3,4XF5.2,2H (F5.3,15H) CU MICRONS **/
      31H 14X4HMEAN10XF5.2,5H  CHS9XF5.2,10XF5.3,3XF6.2,2H (F5.3,15H) CU
      4MICRONS **//)
      GO TO 340
335 WRITE OUTPUT TAPE 9,331,K,PG(I),PCC(I),SPC(I),P(I),SP(I),
      1BEA,BEAE,PG(I+1),PCC(I+1),SPC(I+1),P(I+1),SP(I+1),BESD,BESDE,
      2PG(I+2),PCC(I+2),SPC(I+2),P(I+2),SP(I+2),BEM,BEME
331 FORMAT(10H0   PEAK12,6H   AREA6XF7.0,4X3HCTS3XF7.0,
      18XF5.0,7XF7.0,8XF5.0,5XF5.2,2H (F5.3,14H) MILLION   */
      21H 13X4HS.D.10XF6.3,4H  CHS7XF6.3,8XF5.3,8XF5.2,
      39XF5.3,F7.2,2H (F5.3,15H) CU MICRONS **/
      41H 13X4HMEAN10XF5.2,5H  CHS7XF5.2,9XF5.3,8XF5.2,
      59XF5.3,F7.2,2H (F5.3,15H) CU MICRONS **//)
340 IF(1FF-1) 360,361,361
360 I=I+4
      GO TO 36
361 I=I+3
36 CONTINUE
      WRITE OUTPUT TAPE 9,34,C0F
      WRITE OUTPUT TAPE 9,35,GMCN
      GO TO 200
37 WRITE OUTPUT TAPE 9,38,IS1,IS2
38 FORMAT(        66H1SERIAL NUMBER ON FIRST CARD DOES NOT AGREE WITH I.
      1D. CARD, SERIAL15,14,43H, SPECTRUM SKIPPED AND COMPUTATION RESUMED
      2.)
      GO TO 41
39 WRITE OUTPUT TAPE 9,40,IS1,IS2,IEC
40 FORMAT(19H1   SERIAL NUMBER 214,28H , CARD OUT OF ORDER, NUMBER1
      12,42H,SPECTRUM SKIPPED AND COMPUTATION RESUMED.)
41 READ INPUT TAPE 10,1,1EX,KS1,KS2,ANID(1),ANID(2),ANID(3),DATE,IYR,
      1NPK,ICH,LCH,CT,ADD1,FK,HCT,1FF,IS,IP,IFG,IG,IS1,IS2
      IF(IYR) 41,41,101
C  PL0T ROUTINE
200 DO 201 I=1,N
      Z(I)=X(I,I)
      IF(1FF-1) 403,404,405
403 CALL YP1(YT,IK)
      GO TO 407
404 CALL YP2(YT,IK)
      GO TO 407
405 DO 406 ICI=1,NPK
      P(3*ICI-2)=PCC(3*ICI-2)
      P(3*ICI-1)=PCC(3*ICI-1)
406 P(3*ICI)=PCC(3*ICI)
      CALL YP3(YT,IK)
407 CONTINUE
      YC(I)=YT
201 AB(I)=I
2012 S(1)=5.0
      S(2)=10.0
      S(3)=20.0
      DO 202 I=4,13,3
      S(I)=S(I-3)*10.

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      S(I+1)=S(I-2)*10.
202  S(I+2)=S(I-1)*10.
      BND(1)=0.
      BND(2)=100.
      BND(3)=0.
      CALL MAX3(Y,-1,100,IPX,CHMAX)
      DO 205 I=1,15
      IF(CHMAX-S(I)) 204,204,205
204  BND(4)=S(I)
      GO TO 206
205  CONTINUE
206  CALL ADV FLM(0,1)
      CALL SEL GRD (6,6)
      CALL LBL GRD (6,6,BND)
      CALL PLFILM(100,TEMP,AB,-1,CH,-1,BND,38,0)
      CALL PLFILM(100,TEMP,AB,-1,YC,-1,BND,42,1)
      AB(1)=ICH
      AB(2)=LCH
      BB(1)=CH(ICH)
      BB(2)=CH(LCH)
      CALL PLFILM(2,TEMP,AB,-1,BB,-1,BND,44,0)
      CALL PLFILM(2,TEMP,AB,-1,BB,-1,BND,63,0)
      ALAB(1)=6HCOUNTS
      BLAB(1)=6HCHANNE
      BLAB(2)=6HL NUMB
      BLAB(3)=2HER
      CALL WFILM(1,6,ALAB,0,30.0,6.0)
      CALL WFILM(0,14,BLAB,0,59.0,55.0)
      CALL LINCNT(61)
      WRITE OUTPUT TAPE 12,212,1EX,ANID(1),ANID(2),ANID(3),IS1,IS2,
1DATE,IYR
      CALL LIN CNT (62)
      IF(1FF-1) 341,342,343
341  WRITE OUTPUT TAPE 12,344
344  FORMAT(90X,26H SKEWED NORMAL DISTRIBUTION)
      GO TO 347
342  WRITE OUTPUT TAPE 12,345
345  FORMAT(90X,19H NORMAL DISTRIBUTION)
      GO TO 347
343  WRITE OUTPUT TAPE 12,346
346  FORMAT(90X,23H LOG NORMAL DISTRIBUTION)
347  CONTINUE
      CALL LINCNT(63)
      WRITE OUTPUT TAPE 12,207,ICH,LCH
      CALL ADV FLM(0,1)
207  FORMAT(27X,8H CHANNELS 13,3H TO 14,20H USED IN COMPUTATION)
208  CONTINUE
C    PRINT ROUTINE
      IF(IP-1) 100,209,100
209  WRITE OUTPUT TAPE 9,210
210  FORMAT(61H1      CHANNEL WEIGHT EXPERIMENTAL CALCULATED DIFFE
1RENCE/45H      NUMBER FACTOR DATA DATA//)
      WRITE OUTPUT TAPE 9,211,((1,W(I),CH(I),YC(I),DY(I)),I=1,100)
211  FORMAT(110,F11.5,F12.0,F13.0,F13.1)
212  FORMAT(11H      EXPT.13,2A6,A4,8X,6H SERIAL 15,14,6X,A6,I2//)
      GO TO 100

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END(1,0,0,0,0,0,1,0,0,1,0,0,0,0,0)

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SUBROUTINE CELLP(M,N,IK)
  DIMENSION Y(100),W(100),YC(100),DY(100),PG(40),P(40),SP(40),
  IBM(40,41),X(1,100),B(100),ALAB(1),BLAB(3),AN(40),PART(40),
  ZZ(40),AM(40,40),DP(40),PC(40)
  COMMON Y,X,W,Z,PG,P,SP,YC,DY,BM,B,AN,WVAR,SSQ,PART,TEST,NIT,IFG
  1,IFF,NPK
  IM=0
  ITS=0
  IHSP=0
  M2SC=0
1125 CONTINUE
  SENSELIGHT 0
  VAR = 0.0
  SS = 0.0
  LIES=0
1100 DO 1101 I=1,IK
  DP(I)=0.0
  PC(I)=PG(I)
  BM(I,1) = 0.0
  SP(I)=0.0
1101 P(I)=PG(I)
  LIT = 0
  LIE = 0
  51 LICK = 0
  K=IK-IM
1103 IT=0
  KP=K+1
  IF(K) 1102, 11011,11032
11011 LIES = 1
  GO TO 11087
11032 DO1108I=1,K
  DO1108J=1,KP
  IF(J-KP)1104,1105,1105
1104 AM(I,J)=0.0
1105 IF(I+1-J)1106,1107,1106
1106 BM(I,J)=0.0
  GO TO 1108
1107 BM(I,J)=1.0
1108 CONTINUE
  H = 1.0
  IT=IT+1
11087 DO21122L=1,N
  DO 11084 J = 1, M
11084 Z(J) = X(J,L)
  66 IF(1FF-1) 407,408,409
  407 CALL YP1(YT,IK)
  GO TO 410
  408 CALL YP2(YT,IK)
  GO TO 410
  409 CALL YP3(YT,IK)
  410 CONTINUE
11085 IF(LIES)11086,11086,1115
11086 JACK=0
  DO9003JUK=1,IK
9002 JAKE=JUK-JACK

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PPAK026
PPAK027
PPAK028

PPAK033
PPAK034
PPAK035
PPAK036
PPAK037
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PPAK039
PPAK040
PPAK041
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PPAK043
PPAK044
PPAK057
PPAK058
PPAK059
PPAK060
PPAK061
PPAK062
PPAK063
PPAK064
PPAK065
PPAK066
PPAK067
PPAK068
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PPAK070
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PPAK075
PPAK076
PPAK077

PPAK079
PPAK080
PPAK081
PPAK086

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AN(JAKE) = PART(JUK)
9003 CONTINUE
1115 YC(L) = YT
      DY(L) = Y(L) - YC(L)
      IF(LIES)21116,21116,31117
31117 VAR=VAR+W(L)*DY(L)**2
      SS=SS+DY(L)**2
      GO TO 21122
21116 IF(K)1102,21122,1117
      DO1122I=1,K
      DO 1122 J=I,KP
      IF(J-KP)1118,1119,1119
      1118 AM(I,J)=AM(I,J)+AN(I)*AN(J)*W(L)
      GO TO 1122
      1119 BM(I,1)=BM(I,1)+AN(I)*DY(L)*W(L)
      1122 CONTINUE
21122 CONTINUE
      V = 0.0
      DO 60 L = 1,N
      60 V = V + W(L) *DY(L) **2
      IF (LIES) 21128, 21128, 1153
21128 IF (K - 1) 21129, 21129, 31123
31123 DO31124 J=2,K
      JIG = J - 1
      DO 31124 I=1,JIG
31124 AM(J,I) = AM(I,J)
21129 IF (K) 1102, 1123, 11221
11221 IF(SENSELIGHT1)1123,11251
      1123 SENSELIGHT1
      IF (K) 1102, 1152, 21123
21123 CONTINUE
11251 IF(LIE) 1102, 6000,7000
      6000 KT = 1
      GO TO 8000
      7000 KT=KP
      8000 IF (K - 1) 1152, 3000, 4000
      3000 DET = AM(1,1)
      BM(1,1) = BM(1,1)/AM(1,1)
      BM(1,2) = 1.0/AM(1,1)
      GOT01131
      4000 CALLLEQD(AM,BM,K,KT,40,40,DET)
C      CHANGES FOR 7090...
      1131 IF(SENSELIGHT1)1132,1133
      1132 CONTINUE
      SENSELIGHT1
      1133 JUK = 0
      DO 11351 I=1,IK
14332 CONTINUE
11343 JAKE = I - JUK
      IF(IFG-1)27002,22353,6661
27002 IF(IT-5)6661,6661,22353
      6661 DP(I) = BM(JAKE, 1)
16666 PC(I) = P(I) + H* DP(I)
C      CHANGES FOR 7090...
      IF(SENSELIGHT1)7777,26666
26666 IF(IFG-1)27000,11351,6667

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PPAK087
PPAK090
PPAK091
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PPAK147
PPAK148
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PPAK150
PPAK151
PPAK152
PPAK153

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27000 IF(IT-5)6667,6667,11351
7777 SENSELIGHT1
GOTO11351
6667 IF(P(I)*PC(I))6668,11351, 11351
6668 H = H/2.0
IF (H - 0.0000000001) 12352, 16666, 16666
11351 CONTINUE
GO TO 1139
22353 IF(IT-25)6661,6661,13000
13000 ITS=IT
11341 IF(ABSF(DP(I)) - ABSF(BM(JAKE,1)))11342, 11342, 6661
11342 H = H/2.0
IF(H-.0000000001)12352,6661,6661
12352 IHSP=1
GOTO1102
1139 IF(SENSELIGHT1)1147,1140
1140 GOTO1142
1142 NIT=IT
1145 JERK = 0
DO 1146 I = 1,IK
IF (P(I)) 11451, 11452, 11451
11451 IF(ABSF((PC(I)-P(I))/P(I))- TEST ) 1146, 1146, 1148
11452 JERK = JERK + 1
1146 CONTINUE
IF (JERK - IK) 1147, 1148, 1148
1147 SENSELIGHT1
LIE = 1
M25C=1
1148 DO1149I=1,IK
1149 P(I)=PC(I)
IF (LICK) 1102, 1150, 1152
1150 IF(SENSELIGHT1)11501,1151
11501 SENSELIGHT1
LICK = 1
1151 IF(M25C)29768,29768,11032
29768 IF(IT-25)11032,11512,11032
11512 SENSELIGHT 1
IT=0
ITS=26
GO TO 1147
1152 VAR=0.0
SS=0.0
LIES = 1
GO TO 11087
1153 DF=N-K
IDF=N-K
IF(K) 1102, 21155, 21154
21154 CONTINUE
21998 IF(ITS)13002,13004,13002
13002 IT=ITS
13004 NIT=IT
21155 WVAR = VAR/DF
SSQ = SS
JACK = 0
DO 21160 I = 1,IK
1158 JAKE = I - JACK

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PPAK154
PPAK155
PPAK156
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PPAK158
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PPAK206
PPAK213
PPAK214
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PPAK217
PPAK218
PPAK219
PPAK224

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      J = JAKE + 1
1159 SP(1) = SQRTF(BM(JAKE,J)*WVAR)
21160 CONTINUE
1102 SENSELIGHT 0
      IF(IHSP122100,22108,22100
22100 WRITEOUTPUTTAPE9,22102
22102 FORMAT(75H1THE PROGRAM QUIT ITERATING SINCE THE PARAMETER(S) INSIS
      XT ON CHANGING SIGNS)
22108 RETURN
      END(1,0,0,0,0,0,1,0,0,1,0,0,0,0,0)
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PPAK225
PPAK226
PPAK229
PPAK230
PPAK231
PPAK232
PPAK234
PPAK235

YP1

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SUBROUTINE YP1(YT,IK)
SUBROUTINE YP1 IS FOR SKEWED NORMAL CURVE
DIMENSION Y(100),W(100),YC(100),DY(100),PG(40),P(40),SP(40),
1BM(40,41),X(1,100),B(100),ALAB(1),BLAB(3),AN(40),PART(40),
2Z(40),AM(40,40),DP(40),PC(40)
COMMON Y,X,W,Z,PG,P,SP,YC,DY,BM,B,AN,WVAR,SSQ,PART,TEST,NIT,IFG
1,IFF,NPK
3 YT=0.
C P(41-3)= AREA, P(41-2) = STANDARD DEVIATION,
C P(41-1) = MEAN, P(41) = COEFF OF SKEWNESS
XX=1./SQRTF(2.*3.1415926)
6 IND=(K/4
10 DO16 J1=1,IND
T1=P(4*J1-3)*XX/P(4*J1-2)*EXP(-0.5*
X(Z-P(4*J1-1))**2/(P(4*J1-2)**2))
T2=(Z-P(4*J1-1))/P(4*J1-2)
T3=(Z-P(4*J1-1))**3/(P(4*J1-2)**3)
T4=T2**2
YT=YT+T1*(1.-P(4*J1)*0.5*T2*(1.-(T4/3.)))
PART(4*J1-3)=T1/P(4*J1-3)*(1.-P(4*J1)*0.5*
X(T2-T3/3.))
PART(4*J1-2)=T1*(-1./P(4*J1-2))*(1.-P(4*J1)*0.5
X*T2+P(4*J1)*T3/6.)+T2**2/P(4*J1-2)*(1.-P(4*J1)
X*.5*T2+T3*P(4*J1)/6.)+P(4*J1)*0.5*T2/
XP(4*J1-2)-P(4*J1)*0.5*T3/P(4*J1-2))
PART(4*J1-1)=T1*(T2/P(4*J1-2)*(1.-0.5*P(4*J1)
X*(T2-T3/3.))+0.5*P(4*J1)/P(4*J1-2)-0.5*
XP(4*J1)*(Z-P(4*J1-1))**2/P(4*J1-2)**3)
16 PART(4*J1)=T1*(-0.5*(T2-T3/3.))
4 RETURN
END(1,0,0,0,0,0,1,0,0,1,0,0,0,0,0)

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YPG017

YP2

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SUBROUTINE YP2(YT,IK)
C  SUBROUTINE YP2 IS FOR NORMAL CURVE
   DIMENSION Y(100),W(100),YC(100),DY(100),PG(40),P(40),SP(40),
   BM(40,41),X(1,100),B(100),ALAB(1),BLAB(3),AN(40),PART(40),
   ZZ(40),AM(40,40),DP(40),PC(40)
   DIMENSION G(4)
   COMMON Y,X,W,Z,PG,P,SP,YC,DY,BM,B,AN,WVAR,SSQ,PART,TEST,NIT,IFG
   I,IFF,NPK
   YT=0.
   DO 4 I=1,NPK
   G(I)=(0.3989423*P(3*I-2)/P(3*I-1))*EXP(-0.5*((Z(I)-P(3*I))
   1/P(3*I-1))**2)
   YT=YT+G(I)
   PART(3*I-2)=G(I)/P(3*I-2)
   PART(3*I-1)=(((Z(I)-P(3*I))**2/(P(3*I-1))**3)
   1-(1.0/P(3*I-1)))*G(I)
4  PART(3*I)=(((Z(I)-P(3*I))/(P(3*I-1))**2))*G(I)
   RETURN
   END(1,0,0,0,0,0,1,0,0,1,0,0,0,0,0)

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YP3

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SUBROUTINE YP3(YT,IK)
C  SUBROUTINE YP3 IS FOR LOG NORMAL CURVE
    DIMENSION Y(100),W(100),YC(100),DY(100),PG(40),P(40),SP(40),
    IBM(40,41),X(1,100),B(100),ALAB(1),BLAB(3),AN(40),PART(40),
    Z(40),AM(40,40),DP(40),PC(40)
    DIMENSION G(4)
    COMMON Y,X,W,Z,PG,P,SP,YC,DY,BM,B,AN,WVAR,SSQ,PART,TEST,NIT,IFG
    1,IFF,NPK
    YT=0.
    DO 4 I=1,NPK
        ZLOG=LOG10F(Z(1))
        G(I)= 0.3989423*0.4342945*P(3*I-2)/P(3*I-1)/Z(1)*
        1EXP(-0.5*((ZLOG-P(3*I))/P(3*I-1))**2)
        YT=YT+G(I)
        PART(3*I-2)=G(I)/P(3*I-2)
        PART(3*I-1)=(((ZLOG-P(3*I))**2/(P(3*I-1))**3)
        1-(1.0/P(3*I-1)))*G(I)
    4 PART(3*I)=(((ZLOG-P(3*I))/(P(3*I-1))**2)*G(I)
    RETURN
    END(1,0,0,0,0,0,1,0,0,1,0,0,0,0,0)

```