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PROJECT CHARIOT--FINAL REPORT

ANALYSIS OF PLANT MATERIALS IN THE RUMEN
OF CARIBOU OF NORTHWESTERN ALASKA

MASTER

AEC CONTRACT NO. AT(04-3)-310

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ANALYSIS OF PLANT MATERIAL IN THE RUMEN
OF CARIBOU OF NORTHWESTERN ALASKA

Lloyd H. Swift and Richard H. Bishop

Over 60 caribou rumen samples were collected between January 1960 and June 1962 as a part of the University of Alaska's studies in northwestern Alaska under the U.S. Atomic Energy Commission's Project Chariot. Forty-three of these samples have been processed in varying degrees and are the basis of this report.

At the commencement of this study the techniques employed were largely those developed by Alan M. Courtright (Alaska Cooperative Wildlife Research Unit Quarterly Progress Report, 8(4):1-16, 1957.) and involved screening of caribou rumen samples and analysis of the coarser subsamples. At the conclusion of the study, emphasis was laid on procedures designed to test the hypothesis that the proportion of the lichen fraction is markedly and progressively greater in the subsamples of finer material than it is in the coarser subsamples.

The work covered in this report was done by a succession of individuals. The separation of subsamples was begun by Barbara Murray (See Project Chariot Caribou Progress Report, June 1961, p 11-14) and continued by Sally Crawford. Gail Mayo did most of the work of separating the subsamples into lichen, moss, and vascular fractions, though Ronald Fayer was also associated with this phase of the work. The junior author completed the work of subsample separation, continued the work of separating the coarser subsample fractions, and made analytical counts on randomly chosen portions of the subsamples of finer material. Carolyn Parker has assisted in this last phase of the work by randomizing material to be tested, by counting and weighing particles, and by making slides.

The following samples were screened:

	<u>Collection Number</u>	<u>Collection Date</u>	<u>Collection Data</u>
Collector: Peter C. Lent			
1.	PCL 1	14 Feb 60	male, 2 mi. E. of mouth of Ogotoruk Cr.
2.	PCL 13	30 May 60	female, yearling, Carbon Cr.
3.	PCL 16	4 June 60	female, Elusive Cr.
4.	PCL 31	18 July 60	male, Ukinyik Cr.
5.	PCL 32	3 Aug 60	male, Isiak Lake
6.	PCL 33	8 Aug 60	male, Cutler R.
7.	PCL 35	15 Aug 60	male, Tungnak R.
8.	PCL 36	25 Sept 60	male, Hunt R. near Kobuk R.
9.	PCL 37	25 Sept 60	#2 male, area as above
10.	PCL 41	24 Oct 60	male, Squirrel R.
11.	PCL 43	24 Oct 60	female, area as above
12.	PCL 45	8 Nov 60	female, Kelly R.
13.	PCL 46	14 Nov 60	male, Ogotoruk Cr.
14.	PCL 47	25 Nov 60	male, Noatak Flats, 10 mi. E. of Noatak
15.	PCL 48	25 Nov 60	#2 male, area as above
16.	PCL 62-3	22 June 62	female, Driftwood Air Strip
17.	PCL 62-6	22 June 62	#2 female, area as above
18.	PCL 97	6 Aug 61	male, in mts. 3 mi. W. of Anorat Cr., upper Noatak R.
Collector: William O. Pruitt, Jr.			
19.	WOP 1251	26 Apr 60	2 mi. NE. of mouth of Ogotoruk Cr.
20.	WOP 1253	26 Apr 60	4 mi. NE. of mouth of Ogotoruk Cr.
21.	WOP 1311	15 Oct 60	male, Wulik R.
22.		19 Apr 60	male, Ogotoruk Cr. region 2 mi. NE of mouth.
Collector: W. Weber			
23.	W. Weber-4	25 Apr 60	Ogotoruk Cr. region
Collector: Odd Lono			
24.	OL 6	9 July 61	male, Kugururok R.
25.	OL 31	18 Oct 61	male, Akillik R.
26.	OL 45	19 Nov 61	male, Rabbit R.
27.	OL 47	7 Dec 61	female, Rabbit R.
28.	OL 52	18 Jan 62	female, Squirrel R.
29.	OL 53	18 Jan 62	male, area as above
30.	OL 63	21 Feb 62	male, area as above
31.	OL 64	21 Feb 62	female, area as above

<u>Collection Number</u>	<u>Collection Date</u>	<u>Collection Data</u>
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Collector: Ernest S. Burch, Jr.

32.	C 12	23 Mar 61	female, Ukpiksugruk R.
33.	C 16	24 Mar 61	male, area as above
34.	C 17	25 Mar 61	male, area as above

Collector: Howard Kantner

35.	HK 1	13 Mar 61	female, Ogotoruk Cr.
36.	HK 8	22 Mar 61	female, area as above
37.	HK 15	3 Apr 61	male, area as above
38.	HK 19	21 Apr 61	female, area as above

Collector: Anonymous

39.	6.1	13 Oct 60	fawn, Kissimilouk Cr. near Sikrikpuk Mtn. Received from Ernie Carl
40.	6.2	Oct 60	male, Squirrel R.
41.	6.3	18 Sept 60	Kotzebue hunters approx. 10 mi W. of Ambler Village on Kobuk R.
42.	6.4	18 Sept 60	#2, data as above
43.	6.5	June 60	data lacking

As here used, the expression caribou rumen sample signifies a 250 ml sample of the contents of the rumen of a caribou taken as a sample of the rumen contents of a caribou population. The term subsample signifies that portion of a sample caught in a single screen of a set of screens. The term fraction refers to one of several constituent categories of material into which the samples and subsamples can be analysed.

The data of this report fall into four general categories: (1) sieve separation of subsamples; (2) analysis of the coarser subsamples into lichen, moss, and vascular fractions; (3) analysis of the subsamples of finer material made with one sieving only; and (4) analysis of the subsamples of finer material made with two sievings or with other modifications of technique.

The sieve series is based on a screen (200 U.S. Alternate Designation) which has a mesh opening of 74 microns. The next larger screen increases by the fourth root of 2. Thus 1 is to 1.189 as 74 microns is to the mesh opening of the next larger

screen--88 microns. Going up from the 200 screen each screen in a full sequence is larger in mesh size than the previous one by this factor. This gives a sequence of 26 screens in the range of mesh sizes dealt with in this paper. Using alternate screens, one would advance by the square root of 2, and in this case the areas of openings would double from screen to screen for 13 screens in the range of mesh sizes here dealt with. With such a series one can graph percentage of weight on the ordinate and the reciprocal of mesh size--conveniently approximated by number of mesh per unit length--logarithmically on the abscissa.

Sieve Separation of Subsamples

The 43 samples listed above were collected and stored in plastic bags with formalin as a preservative. In the sieving process, the material was washed through a column of testing sieves, and the amount of material in each subsample was determined by dry weight. The maximum amount of water used in washing one sample was 32.6 liters; the minimum was 5.65 liters; and the mean for 41 samples was 15.47 liters.

Unfortunately, two different sets of screens were used (See Table 1), and there is no record as to which set was used for a given sample; the interpretation of data assumes the Designation numbers given approximate the U.S. Alternate system.

Details of procedure have varied during the study, but the ultimate practice was to place a set of 11 screens in a stack over a 10 gallon milk can and seal the stack with sections of rubber tire tubing. The can had been fitted with a valve attached to a rubber hose connected with a vacuum line. Blinding of the screens--especially those with finer mesh--was a problem, the samples collected in the summer being especially difficult.

In 17 of the 43 samples, the sieve analysis includes data on the subsample passing the 200 screen. These data are based on the filtration of a quart of fluid that had passed the 200 screen.

Table 1. Size of openings in screens of various sizes

Mesh per Inch (Tyler Designation)	U.S. Alternate Designation	Mesh Size mm	First Screen Set Size mm	First Screen Set Designation Number
3 1/2	3 1/2	5.66	5.47	4
4	*4	4.76		
5	5	4.00		
6	6	3.36		
7	*7	2.83		
8	8	2.38	2.49	7
9	*10	2.00		
10	12	1.68	1.659	10
12	14	1.41		
14	16	1.19		
16	18	1.00		
20	*20	0.841	0.8371	20
24	25	0.707		
28	*30	0.595		
32	35	0.500	0.485 (Approx)	30
35	*40	0.420		
42	45	0.354	0.344 (Approx)	40
48	*50	0.297	0.306 (Approx)	50
60	*60	0.250	0.247	60
65	70	0.210		
80	*80	0.177	0.176	80
100	*100	0.149	0.148	100
115	120	0.125		
150	140	0.105		
170	170	0.088		
200	*200	0.074	0.074	200
250	230	0.063		
270	270	0.053		
325	325	0.044		
400	400	0.037		

*Indicates screens used in the second set; the data on the first set is based on the Courtright report. Data on Tyler and U.S. Alternate Designations and their mm mesh size are from the Tyler Company Handbook 53, 1963 edition.

Estimates of the weight percentage of dry material that passed the 200 screen varied from 1.3 per cent to 48.3 per cent, with a mean of 23.9 per cent.

Since the screen sizes used were apparently originally chosen arbitrarily by Courtright, analysis of results is difficult. However, the material caught in any given screen may be apportioned in theory to the missing intervening screens below the next higher screen. Thus, in the case of the 200 screen, the matter caught by it represents what would have been the cumulative catch by the 120, 140, 170, and 200 screens of a complete set. Therefore, for an estimate of the relationship of the normal catch of the 200 screen to that of other screens, the amount actually caught by the 200 screen must be divided by four. The operation of this principle may be illustrated by four samples extensively dealt with in this report. Table 2 lists data on the individual samples, totals, number of screen intervals used to divide the totals to obtain comparable numbers in arbitrary units to represent the relative theoretical catch of the various screens. These numbers are graphed as the Total line in Figure 2.

Volumetric analysis was made for 40 samples, 440 subsamples. This measurement was made by removing the damp subsample from its screen and placing it in a 100 ml graduate. To this 50 ml of water was added and the volume of damp plant material estimated by subtracting 50 ml from the total. Sample PCL 1 may be taken as an example. Figure 1 shows comparative per cent (uncorrected for unequal gradation in sieve size) for both volume and weight of this sample. If the total dry weight reported for this sample, 17.5 grams, is divided by the volumetric total, 22.9 ml, one obtains an estimate of 0.76 as the specific weight of the material. This figure varies considerably from sample to sample and from worker to worker. Thus, the value for one sample (6.4) has the relatively high value of 10.7 g/9.5 ml or 1.13, whereas another (6.5) has 13.0 g/96.0 ml or 0.136.

Table 2. Subsamples caught in screens--per cent

Screen Designation	PCL 16	PCL 32	OL 47	OL 53	Total
4	4.2	30.8	5.8	5.2	46.0
7	2.9	4.4	30.0	4.5	41.8
10	4.2	4.9	15.3	21.7	46.1
20	37.9	19.4	28.0	36.5	121.8
30	8.0	9.3	10.1	11.5	38.9
40	8.5	3.6	3.7	6.0	21.8
50	5.3	3.2	2.9	5.0	16.4
60	0.8	1.0	1.7	0.6	4.1
80	3.2	6.4	1.6	3.9	15.1
100	1.6	12.1	0.5	0.1	14.3
200	23.4	5.0	0.2	4.5	33.1

Comparable Figures for Different
Screens Obtained by Dividing Total
Figures by the Appropriate Division
Number

4	--	--
7	3	13.9
10	2	23.0
20	5	24.4
30	2	19.4
40	2	10.9
50	2	8.2
60	1	4.1
80	2	7.5
100	1	14.3
200	4	8.3

FIG. 1. Comparison of volumetric (solid lines) with weight (broken lines) analysis in terms of per cent (PCL 1).

FIG. 2. Comparative amounts of material caught in sieve series. Averaged results for four samples. Data adjusted to compensate for unequal gradations in mesh aperture.

Analysis of the Coarser Subsamples
into Lichen, Moss, and Vascular Fractions

An attempt was made to analyse the coarser subsamples (4, 7, and 10 screens) into their lichen, moss, and vascular plant components. The analysts felt a simple dissecting scope adequate for work at this level. Fourteen samples were analysed in toto, and comparable quantities were analysed from two others. The cumulative results are presented in Table 3.

It appears from Table 3 that the lichen fraction in the 7 and 10 screens is markedly greater than that in the 4 screen. The three screens, however, account for only 10.4 per cent of the 13 samples (out of the 16 in this group) for which a value has been assigned to the subsample passing the 200 screen. It has, therefore, seemed questionable to generalize as to the caribou diet on the basis of this 10 per cent, for the proportion of lichen in the 7 and 10 screens is significantly higher than that in the 4 screen.

Table 4 analyses the same 16 samples as to their coarsest subsamples and shows a marked seasonal variation in the lichen fractions, the spring samples having the lowest percentage of lichen.

Analysis of the Subsamples of Finer
Material Made with One Sieving Only

The 20 and 30 subsamples in the above mentioned 13 samples contain 17.5 per cent and 14.9 per cent of the total, respectively. Thus, together they retain three times the total amount caught by the 4, 7, and 10 screens. An attempt was therefore made to analyse the 20 and 30 subsamples, but because of the smaller particle size, only a fraction of the material was identified--not less than 1/512th of the total in the subsample. Four 20 subsamples were analysed. The 4, 7, and 10 subsamples of the corresponding samples had in the previous analysis of coarse material been found to have average lichen percentages of 7.8, 29.8, and 28.4, respectively. The weight

Table 3. Analysis of lichen, moss, and vascular fractions
of coarser subsamples

Collection Number	4 Screen			7 Screen			10 Screen		
	% L	% M	% V	% L	% M	% V	% L	% M	% V
PCL 1	0.2	3.3	96.4	4.6	8.7	86.6	7.7	8.1	84.2
PCL 13	6.5	--	93.5	6.0	2.0	91.9	4.9	4.7	90.4
PCL 16	0.3	--	99.6	1.7	4.1	94.2	4.6	7.7	87.7
PCL 31	12.0	4.3	83.7	20.4	6.0	73.6	21.1	8.0	70.9
PCL 32	12.6	2.0	85.4	22.7	3.0	74.3	17.1	3.9	79.0
PCL 36	29.6	1.2	69.1	44.7	3.0	52.2	47.3	0.9	51.8
PCL 37	7.6	9.4	83.0	19.1	4.1	76.7	39.1	2.3	58.2
PCL 47	--	0.5	99.5	15.7	3.7	80.5	21.9	3.0	75.1
PCL 48	1.0	0.8	98.2	4.6	0.9	94.5	10.2	2.1	87.7
WOP 1251	0.4	1.8	97.8	0.8	7.5	91.7	3.9	8.9	87.1
WOP 1311	15.9	2.8	81.3	31.0	4.8	64.2	32.8	4.0	63.1
OL 47	10.0	--	90.0	54.6	0.8	44.6	62.6	0.3	37.1
OL 53	8.2	0.8	91.0	40.2	5.0	54.8	33.2	3.9	63.0
C 12	7.3	1.7	91.0	12.9	3.7	83.3	22.7	5.4	71.8
C 16	5.9	1.5	92.6	14.3	1.9	83.8	19.1	3.3	77.0
Kotzeb 6.3	17.2	14.1	68.7	52.4	1.0	46.6	55.2	1.4	43.3
Average	8.4	2.8	88.8	21.6	3.6	74.6	25.2	4.3	70.4

Table 4. Analysis of lichen percentages by subsamples of coarser material and by season

Season	Number of Samples Analysed	Average % of Lichen in 4 Screen	Average % of Lichen in 7 Screen	Average % of Lichen in 10 Screen	Average % for Seasons for 3 Screens
Jan. & Feb.	2	4.2	22.4	20.5	15.7
Mar. & Apr.	3	4.5	8.1	15.2	9.3
May & June	2	3.4	3.9	4.8	4.0
Jul. & Aug.	2	12.3	21.5	19.1	17.6
Sept. & Oct.	4	23.4	36.8	43.6	34.6
Nov. & Dec.	3	3.7	25.0	31.6	20.1
Average of % for Screens for all Seasons		8.5	19.5	22.2	

estimates gave 33.7 as the average lichen percentage for the four 20 subsamples, and this suggested a progressive increase in the lichen fractions going from coarser to finer material.

Attempts at weight analysis for the 30 screen proved impracticable because of the smallness of the particles. It was thought, however, that the number of particles in a given fraction might give a useful indication as to the weight of that fraction relative to other weights. For such counts a dissecting microscope was used for particles larger than those of the 40 subsample, and a compound microscope was used for particles of the 40 and finer subsamples.

The question arises as to whether particles of lichen, moss, and vascular plants have substantially the same average weight. For this reason, portions of the 4, 7, and 10 screen subsamples were both counted and weighed. There was considerable fluctuation in average particle weights, but no pronounced general weight difference between lichen, moss, and vascular material. Particle counts were therefore made in the subsamples of finer material as a means of estimating the relative proportions of the lichen, moss, and vascular fractions in subsamples 20 to 200--the values for the 4, 7, and 10 screens being already at hand. The values arrived at are presented in Table 5, where they are corrected by the method explained in connection with Table 2. The comparable figures are graphed in Figure 2. The cumulative per cent averages are graphed in Figure 3. Progressive change of ratio between the three fractions distinguished, is shown for one sample (PCL 16) in Figure 4.

Analysis of the Subsamples of Finer Material Made

With Two Sievings or With Other Modifications of Technique

One would anticipate that the ratios of the average particle weights of one subsample to the average particle weights of other subsamples would parallel the ratios of the cubes of the mesh apertures of the screens from which the subsamples were taken. Average particle weights may be estimated by dividing the weight of a subsample by the estimated number of particles in that subsample. The number of

Table 5. Lichen, moss, and vascular percentages in four samples--
averages, total of per cent, and division numbers used to
calculate comparable figures

Screen Size	Lichen				Average Lichen	Moss				Average Moss	Vasc.				Average Vasc.	Total
	PCL 16	PCL 32	OL 47	OL 53	%	PCL 16	PCL 32	OL 47	OL 53	%	PCL 16	PCL 32	OL 47	OL 53	%	Average %
4	0.0	3.9	0.6	0.4	1.2	0.0	0.6	0.0	0.0	0.1	4.2	26.3	5.2	4.8	10.1	11.5
7	0.1	1.0	16.4	1.8	4.8	0.1	0.1	0.2	0.2	0.1	2.7	3.3	13.4	2.5	5.5	10.4
10	0.8	9.6	7.2	4.4	0.3	0.2	0.0	0.8	0.3	3.7	3.7	3.9	5.7	13.7	6.7	11.5
20	2.2	3.7	14.1	12.5	8.1	3.7	1.1	3.2	2.6	2.8	32.0	14.6	10.7	21.4	19.7	30.4
30	1.1	2.1	7.1	3.8	3.5	1.3	5.9	1.7	0.6	2.4	5.6	1.3	1.3	7.1	3.8	9.7
40	1.2	1.6	2.6	2.8	2.0	1.7	0.5	0.4	0.9	0.9	5.6	1.5	0.7	2.3	2.5	5.4
50	0.8	1.4	2.1	2.6	1.7	1.1	0.3	0.3	0.8	0.6	3.4	1.5	0.5	1.6	1.7	4.1
60	0.2	0.7	1.2	0.5	0.6	0.1	0.0	0.2	0.0	0.1	0.5	0.3	0.3	0.1	0.3	1.0
80	1.6	4.2	1.3	2.4	2.4	0.7	0.5	0.1	0.6	0.5	0.9	1.7	0.2	0.9	0.9	3.8
100	0.7	9.4	0.5	0.1	2.7	0.4	1.4	0.0	0.0	0.4	0.5	1.3	0.0	0.0	0.4	3.6
200	19.4	4.1	0.2	3.9	6.9	1.5	0.4	0.0	0.5	0.6	2.5	0.5	0.0	0.1	0.8	8.3
TOTAL					38.3					8.8					52.4	

Screen Size	Division Number	Total Lichen	Comparable Lichen Figures	Total Moss	Comparable Moss Figures	Total Vasc.	Comparable Vasc. Figures	Total	Comparable Total Figures
4	-	4.9	--	0.6	--	40.5	--	46.0	--
7	3	19.3	6.4	0.6	0.2	21.9	7.3	41.8	13.9
10	2	17.8	8.9	1.3	0.6	27.0	13.5	46.1	23.0
20	5	32.5	6.5	10.6	2.1	78.7	15.7	121.8	24.4
30	2	14.1	7.0	9.5	4.7	15.3	7.6	38.9	19.4
40	2	8.2	4.18	3.5	1.7	10.1	5.0	21.8	10.9
50	2	6.9	3.4	2.5	1.2	7.0	3.5	16.4	8.2
60	1	2.6	2.6	0.3	0.3	1.2	1.2	4.1	4.1
80	2	9.5	4.7	1.9	0.9	3.7	1.8	15.1	7.5
100	1	10.7	10.7	1.8	1.8	1.8	1.8	14.3	14.3
200	4	27.6	6.9	2.4	0.6	3.1	0.8	33.1	8.3

FIG. 3. Cumulative per cent of material retained in the various screens for the four samples of Table 5.

FIG. 4. Progressive changes in ratios between lichen, moss, and vascular fractions of subsamples of PCL 16.

particles in the subsample may be estimated by counting the number of particles in a definite portion of that subsample and then multiplying the counted number by the reciprocal of the ratio of the portion of the subsample to the total subsample.

The number of particles in a subsample may be estimated from subsample weights and mesh apertures by the following method. Somewhat more precise more complicated methods are possible, but this one seems adequate for the present discussion. If the number of particles in the 4 subsample is estimated by the method described above, this number multiplied by two other numbers will give an estimate for another subsample. The first number is the ratio of the reciprocal of the cube of the mesh aperture of the screen of the subsample being estimated to that of the 4 screen taken as 1. The second number is the ratio of the weight of the material in the subsample being estimated to that of the 4 screen taken as 1. Thus two estimates for the number of particles in a given sample may be obtained--the first based on counts on each subsample, and the second based on a count of the 4 subsample only. The procedure may be illustrated with sample PCL 16 as shown in Table 6. The results are graphed in Figure 5.

The discrepancy between estimates may be attributed to a variety of factors. In the first place, distribution of subsamples among the theoretical screens of a full set, so that those intervening between screens actually used might be evaluated, and a basing of cube size on a mean between the mesh apertures for each sequential theoretical pair of screens would substantially reduce the estimate based on mesh apertures, since most of this estimate comes from the 200 screen. On the other hand, the estimate based on particle counts seems clearly too low. One is forced to consider the possibility that many of the particles counted are, in fact, aggregates of the particles that entered a given subsample. The main problem here is to determine to what extent, and for which subsamples, aggregation may have biased the relative counts for the lichen, moss, and vascular fractions. The problem is clearly

Table 6. Estimated particle numbers in sample PCL 16

	Particle Numbers in Subsamples Estimated by Counts	Cumulative Particle Numbers Estimated By Counts	Mesh Apertures in Inches	Cubes of Apertures in Inches	Reciprocal of Cube of Apertures
4	537	537	.1850	.006331625	157.9280
7	468	1005	.1100	.001331	751.3148
10	974	1979	.0780	.000474552	2107.038
20	121125	123104	.0328	.000035287552	28336.64
30	57100	180204	.0232	.000012487168	80073.29
40	186730	366934	.0164	.000004410944	226706.0
50	450020	816954	.0116	.0000013456	742942.1
60	15675	832629	.0097	.00000088529281	1129561.0
80	208650	1041279	.0069	.000000328509	3044140.0
100	19963	1061242	.0058	.000000195112	5125577.0
200	1441670	2502912	.0029	.000000024389	41000410.0

	Ratios of Reciprocals of Cubes of Apertures, 4 Screen Taken as 1	Subsample Weights	Ratio of Weights of Sub- samples	Product of Ratios x 537	Cumulative Product of Ratios x 537
4	1	0.584	1.00	537	537
7	4.78	0.397	0.68	1745	2282
10	13.4	0.583	1.00	7200	9482
20	181.5	5.291	9.06	878000	887482
30	510.1	1.109	1.90	520600	1408082
40	1444	1.193	1.92	1488000	2896082
50	4770	0.739	1.27	3258000	6154082
60	7195	0.110	0.19	734000	6888082
80	19400	0.458	0.78	10420000	17308082
100	32700	0.227	0.39	6850000	24158082
200	261500	3.261	5.59	78430000	102588082

Number of
Particles

19

100,000,000

10,000,000

1,000,000

100,000

1. The first part of the report is a summary of the work done during the past year. It is a brief review of the progress made in the various fields of research.

 

greatest in connection with the subsamples of finer material, for here absolute average difference in size is practically lost among the counted particles. Table 7 shows relative average sizes of cubes of mesh apertures with the value for the 40 subsample taken as unity. Similarly, the relative average weights for subsamples of sample PCL 32 are given with the 40 subsample again taken as unity.

It is apparent from Figure 6 that adhesion seems by no means confined to the subsamples of finer materials, but occurs to some extent even in the coarser ones. To obtain analysis of material of unquestionably proper size, small portions of subsamples of fine material in a measured amount of water were repassed through the appropriate screen for the given subsamples, the appropriate screen for the 200 subsample being the 100. The results obtained are presented in Table 8. The comparative total particle estimates for the subsamples are graphed in Figure 7. Ocular measurements were made of representative once-screened particles and compared with measurements of those that had been twice screened. The former were indeed larger, but not as markedly so as to account for discrepancies in count estimates. Similarly, aggregation was greater in the once-screened particles, but the aggregation factor appeared less important than had been anticipated.

Some of the twice-screened material from the finer subsamples was analysed for lichen, moss, and vascular content. The results are presented in Table 9. The amount of the analysed twice-screened material is small, but it does not appear that the second screening has seriously affected the previous estimates as to the proportion of lichen in the subsamples of finer material.

Table 7. Comparison of relative average weights of subsample particles of sample PCL 32 with cubes of the relative mesh apertures of corresponding sieves

	Total Particles Estimated by Count	Average Weight of Particles in Subsamples (Grams)	Relative Average Weights of Particles	Logarithms of Relative Average Weights
4	11140	0.000874	90.9	9.958
7	3889	0.0003627	37.71	1.576
10	7069	0.0002218	23.05	1.362
20	148982	0.00004133	4.3	0.633
30	297100	0.00000984	1.024	0.010
40	117296	0.00000961	1.00	0.000
50	59880	0.00001739	1.804	0.265
60	23046	0.00001392	1.449	0.161
80	1164100	0.000001753	0.1823	9.261-10
100	512800	0.00000746	0.776	9.890-10
200	848400	0.000001774	0.1846	9.266-10

	Total Weights of Subsamples	Cubes of Mesh Apertures (mm)	Relative Sizes of Cubes of Mesh Apertures	Logarithms of Relative Sizes of Cubes
4	9.735	164.026	4014.0	3.604
7	1.407	15.419	377.5	2.577
10	1.567	4.566	111.7	2.048
20	6.154	0.5866	14.36	1.157
30	2.925	0.1140	2.79	0.446
40	1.127	0.04089	1.00	0.000
50	1.041	0.02871	0.7025	9.847-10
60	0.323	0.01518	0.3715	9.570-10
80	2.040	0.005461	0.1336	9.126-10
100	3.828	0.003242	0.0793	8.899-10
200	1.505	0.0004052	0.00992	7.997-10

Table 8. Summary of results on analysis of material from subsamples of finer material from sample PCL 16 rescreened to break down particles larger than the mesh aperture of the appropriate screens.

	Drops	Drops x Volume of Drop 0.0526 (ml)	Total Plant Particles in Drops	Total Fluid Used in Screening (ml)	Estimated Total Particles in Screened Portion of Subsample
20	7	0.368	8	96	2088
30	8	0.422	7	220	3649
40	7	0.368	26	215	15190
50	6	0.316	8	195	4940
60	7	0.368	5	193	2622
80	6	0.316	7	157	3477
100	6	0.316	31	128	12550
200	4	0.211	137	353	229000

	Total Dry Weight of Particles in Screened Portion (gm)	Total Weight of Subsample (gm)	Total Particles Estimated by Count After First Screening	Total Particles Estimated by Count After Second Screening	Total Particles Estimated by Mesh Apertures
20	0.005	5.29	121,125	2,210,000	878,000
30	0.005	1.11	57,100	809,500	520,600
40	0.005	1.19	186,730	3,614,000	1,488,000
50	0.002	0.74	450,020	1,827,000	3,258,000
60	0.002	0.11	15,675	272,000	734,000
80	0.002	0.46	208,650	722,000	10,420,000
100	0.002	0.23	19,963	2,700,000	6,850,000
200	0.087	3.26	1,441,670	14,540,000	78,430,000

Table 9. Analysis of twice-screened material from the subsamples of finer particles for lichen, moss, and vascular content (PCL 16)

Twice-Screened Material						
	Particle Numbers			Per Cent		
	Lichen	Moss	Vascular	Lichen	Moss	Vascular
50	1	0	7	12.5	0	77.5
60	2	0	3	40	0	60
80	6	0	1	85	0	14
100	18	7	6	58	23	19
200	128	6	3	93	4	2

Once-Screened Material			
	Per Cent		
	Lichen	Moss	Vascular
50	42	16	42
60	61	9	30
80	63	13	24
100	75	12.5	12.5
200	84	7	9

Ratio of Percentage of Twice-Screened Lichen to Percentage of Once-Screened Lichen

50	.298
60	.656
80	1.365
100	.774
200	1.108

FIG. 6. Logarithms of relative average particle weights for different subsamples of sample PCL 32 and logarithms of cubes of mesh apertures, the 40 screen taken as zero in both cases.

FIG. 7. Total subsample particles estimated by mesh apertures (solid line), by count after first screening (dotted line), by count after second screening (broken line) PCL 16.

Conclusions

The data from the count analysis thus corroborate the hypothesis of progressive increase in the lichen fraction from coarser to finer subsamples, as indicated in Table 10 and Figure 8, but they also indicate that the importance of this increase may be less than one might suppose from the examination of Figure 8, for the total lichen fraction of the four samples analysed, as represented in all subsamples except that passing the 200 screen, comes to about 38.3 per cent of the total material (total of Average Lichen % column, Table 5). This compares with 31.2 per cent lichen if the total lichen be estimated from the coarser (4, 7, and 10) subsamples only (10.4 lichen, 22.8 non-lichen, gives the 31.2 per cent lichen estimate from the 4, 7, and 10 subsamples, Table 5). This emphasizes the importance of the subsamples of intermediate texture, especially the 20 and 30 screen. Note that the major peak of the total line of Figure 2 comes at the 20 screen and represents the culmination of the vascular fraction, whereas the second peak represents chiefly lichen. As for the slight fall of the line at the 20 screen in Figure 8, it may be observed that this marks the point of change from the weight to the count technique, and that if one substitutes the weight value cited on page 13 (33.7) the irregularity is removed.

The course of this work has ranged from the macroscopic to the highly microscopic, and difficulties have been encountered in making the transition. In order to obtain precision in analysis of the finer material, additional techniques must be utilized. There is an extensive literature on such that should first be investigated. Papers on the effect of viscosity on the composition of subsamples of fine material should be located. The literature on lichen morphology should be examined for its own sake and for hints as to microtechnique. The effect of protozoons on aggregation should be assessed. Histochemical methods of analysis should be brought into play--especially stain technology. The process of fixation

Table 10. Per cent of lichen in the various subsamples of four samples

	Screen Designation Numbers										
	4	7	10	20	30	40	50	60	80	100	200
PCL 16	0.4	0.2	4.6	5.9	13.4	14.5	15.4	22.7	50.2	44.9	83.0
PCL 32	12.6	22.7	17.1	18.9	22.2	43.7	43.6	70.0	65.6	77.8	82.9
OL 47	10.0	54.6	62.6	50.0	70.0	71.6	71.8	71.6	84.0	89.0	86.0
OL 53	8.2	40.0	33.2	34.2	33.0	46.3	69.8	61.7	71.7	61.6	85.0
Avg	7.8	29.4	29.4	27.2	34.6	44.0	50.1	56.5	67.9	70.8	84.2

FIG. 8. Average per cent of lichen in the subsamples of four samples.

should be reexamined with lichen requirements given primary consideration. The portion of a subsample dried to obtain weight data should not be the portion analysed into lichen, moss, and vascular fractions. Physical methods such as differential centrifugation and the techniques of pharmacognosy should be explored, and perhaps radioautography.