Verh. Internat. Verein. Limnol.	XV	735—744	Stuttgart, Februar 1964

Determination of plankton biomass in Rongelap Atoll, Marshall Islands, by the use of a multi-net plankton pump[1]

OLE A. MATHISEN (SEATTLE, U.S.A.)

With 3 figures and 2 tables in the text

The Laboratory of Radiation Biology at the University of Washington has been engaged in recent years in a radioecological study of Rongelap Atoll, Marshall Islands, in the west equatorial Pacific Ocean. A part of this work involves estimation of the total standing crop of plankton inside the atoll, and this report describes the methods and results of such a determination.

This study is under the general direction of Dr. L. R. DONALDSON, Laboratory Director, and under the immediate supervision of Dr. E. E. HELD. Dr. G. J. PAULIK kindly reviewed and commented upon the statistical analysis of this report.

Description of the multi-net plankton pump

A preliminary sampling of plankton in Rongelap Atoll in 1958 with a standard ½ m net indicated that a more efficient gear was desired because the plankton density in the lagoon was low. Also, a single mesh-sized net may at times clog and it would not indicate any size composition of the plankton. To solve the above problems, a multi-net plankton pump sampler was developed.[2] ARON (1958, 1961) gives a complete bibliography of previous experience with plankton pumps.

The plankton pump consisted of two main parts: a collector and a water pump (Figs. 1 and 2). The pump used was a gasoline-driven 10.2 cm "Jaeger" centrifugal type, with a rated capacity of 1,924 l/min., but was usually operated at a rate of 1,500 l/min. Intake was through a 10.2 cm hose which could be lowered to a maximum depth of 22.9 m. Water volume entering the collector was determined by a "Sparling" water meter installed between the pump discharge and the collector.

The collector consisted of two stainless steel drums, one installed within the other. The outer drum was 57.2 cm in diameter and 90.2 cm deep with a 10.2 cm intake valve at one side near the bottom. The inner drum was 43.8 cm in diameter and 66 cm deep fastened to the bottom of the outer tank. A 15.2 cm discharge pipe led from the bottom of the tank through a 90° elbow to a 15.2 cm run-off

[1] Report No. UWFL-81 from Division of Biology and Medicine, United States Atomic Energy Commission.

[2] The pump was designed by E. E. HELD and built by JOHN STEVENS.

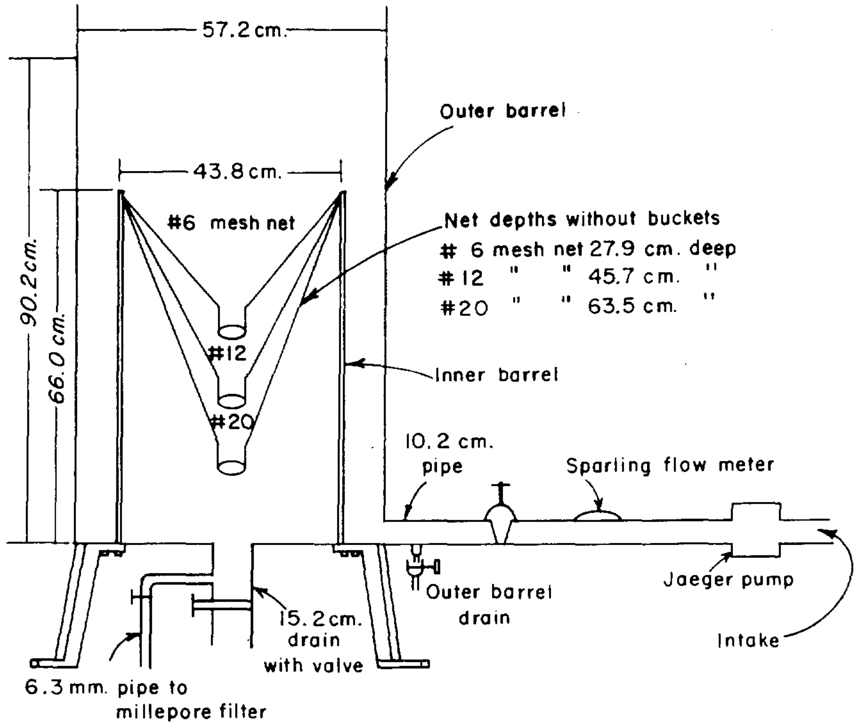


Fig. 1. Diagram of the plankton pump.

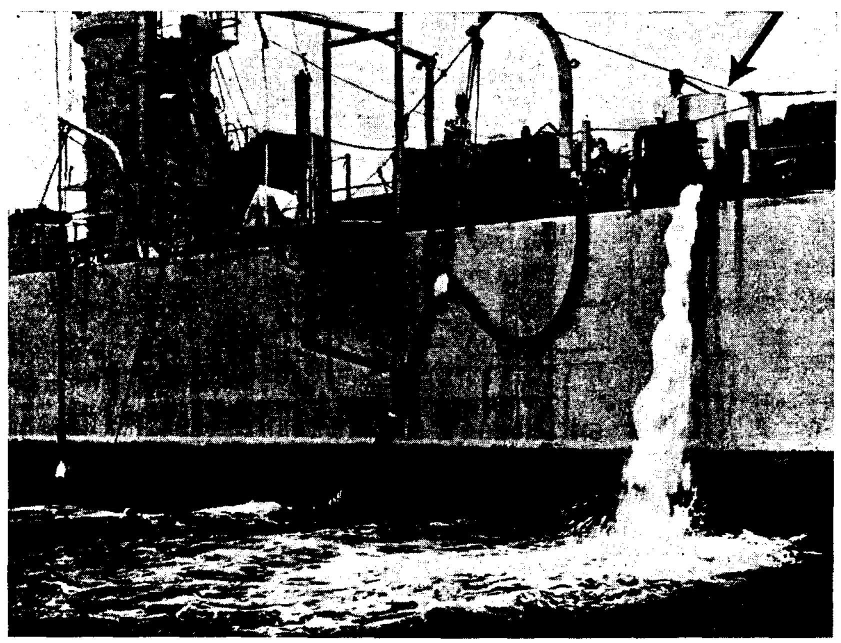
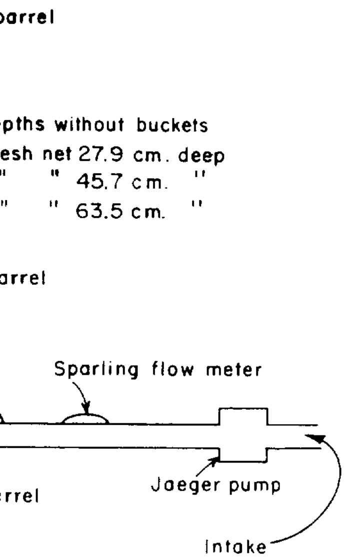


Fig. 2. Photograph of plankton pump in operation, showing intake hose (left) and filtering cylinder with discharge valve (right).



kton pump.

, showing intake hose (left) and
valve (right).

pipe. A 6.3 mm pipe and valve was placed between the discharge valve and the elbow to permit the continuous removal of a sample of the discharge water.

Inside the inner drum were installed three nylon nets with an inner diameter of 43.8 cm, of mesh sizes No. 6, No. 12, and No. 20, respectively. The nets with canvas facing were supported in the inner barrel by three stainless steel rings. The No. 6 mesh net was 27.9 cm deep; the No. 12 mesh net 45.7 cm deep, and the No. 20 mesh net was 63.5 cm deep.

Sea water was pumped into the outer drum, allowed to overflow the rim of the inner drum and thence flow by gravity through the three nets to a discharge valve.

Sampling procedure

Rongelap Atoll (Fig. 3) has a square-shaped surface area with an extension in the northeastern corner. Inside the surrounding reefs the depth increases rapidly to a maximum of about 61 m. The bottom is relatively even except for numerous coral heads which extend almost to the water surface.

New water is brought into the lagoon from the east side during high tide via overflow over the reefs. Outflow takes place mainly through the western and southern passes. Von Arx (1954) has estimated the water renewal of the lagoon to take about one month.

The surface current moves therefore in a westerly direction, but deeper layer waters have a counter current. Our sampling procedures were designed to study the variability in plankton abundance from east to west, from south to north, and from surface to deeper layers. Also considered were diurnal and seasonal variations.

While the ship was cruising normally at a speed of from 4 to 5 knots, samples could only be taken from surface and down to a depth of 3 m. At anchor, pumping could be done to a maximum depth of 22.9 m.

The basic sampling was done at the 61 cm depth with the ship cruising to-and-fro on an east-west direction with the course-lines 2½ minutes apart. A few replicate hauls were also made on the same day. The second principal sampling was done with the ship cruising in a north-south or south-north direction and with the pump intake at the 3 m depth, and with the course-lines 5 minutes apart. The pump was operated normally for one hour corresponding to a filtered volume of about 95,000 l of water before the nets were emptied and the collected plankton preserved. The pump was equipped with a double set of plankton nets so that change of nets could be accomplished in less than one minute without stopping the pump.

Finally, three fixed stations were established with continuous 24-hour sampling from the following depths: 61 cm, 3 m, 6.1 m, and 22.9 m, rotated after 68,000 l of water had been strained.

The plankton received in the three nets was filtered through a nylon filter of the same mesh as the net mounted in a millepore filter holder with the aid of an

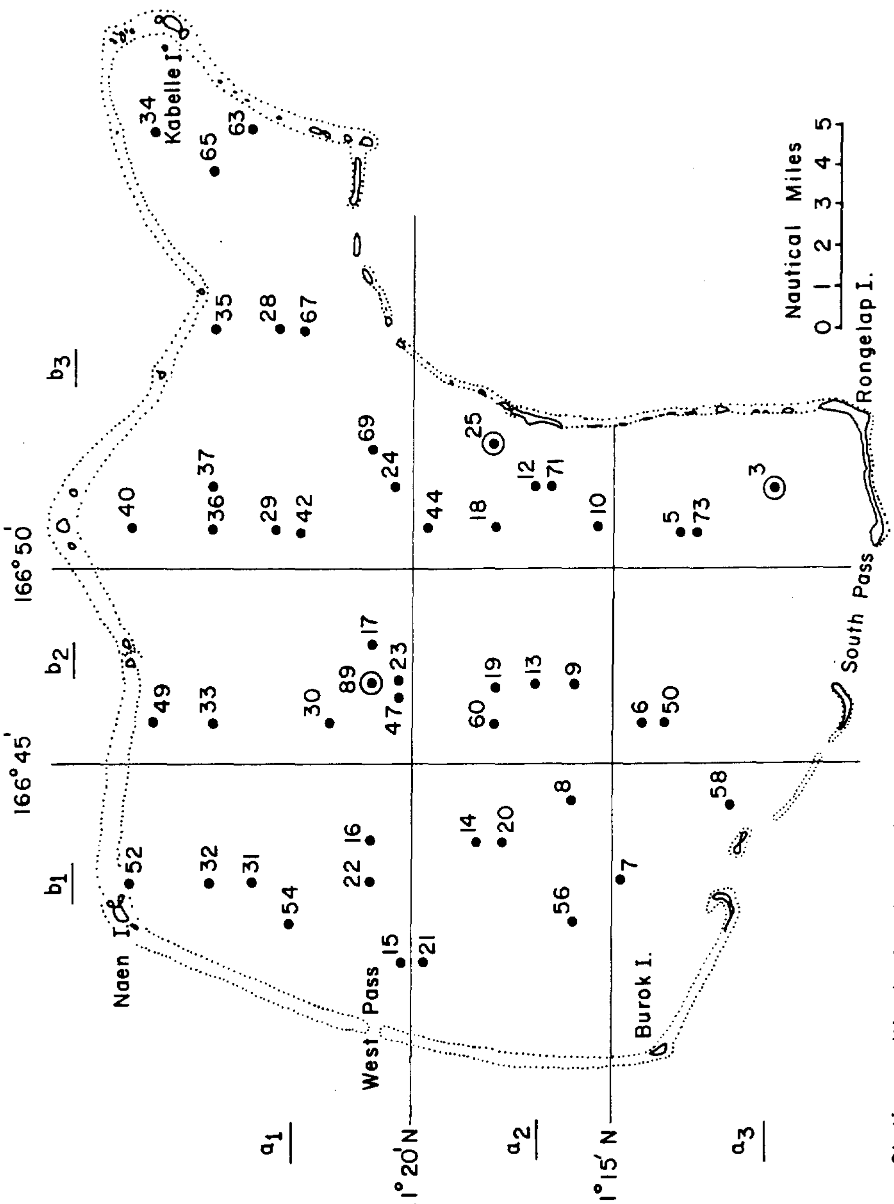


Fig. 3. Diagram of Rongelap Atoll giving station locations and employed grid system.

electric vacuum pump. One-fourth of the sample of each net from every station was preserved in formalin for species indentification and species composition. The remaining $^3/_4$ of the filter was dried in an electric oven and, after all moisture had evaporated, the samples were sealed in small plastic bags and shipped to the Seattle Laboratory, where the dry weight on each filter was ascertained. The work began September 4 and was concluded on September 21, 1959.

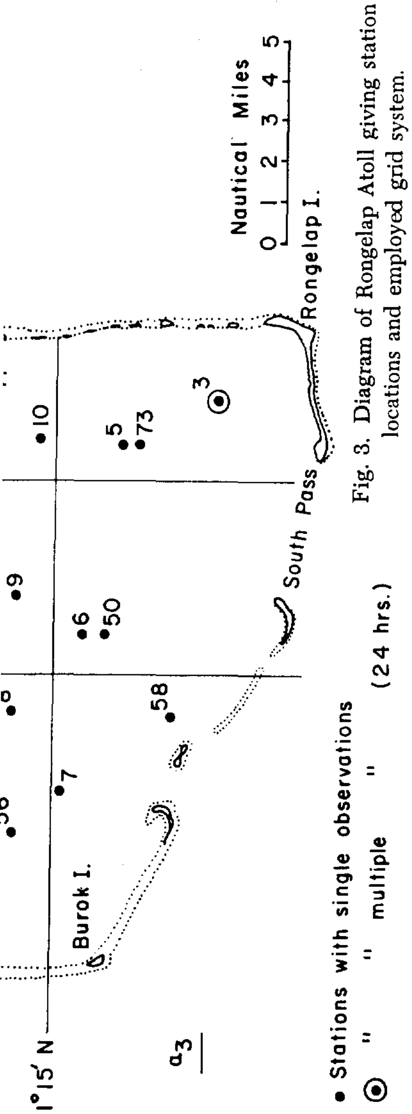


Fig. 3. Diagram of Rongelap Atoll giving station locations and employed grid system.

e of each net from every station ion and species composition. The oven and, after all moisture had plastic bags and shipped to the filter was ascertained. The work nber 21, 1959.

Analysis

For each sample the following information was entered on IBM cards: station and sample number, position at start and stop, date, time at start and stop, depth, volume of water strained, dry weight of sample from each of the three nets, and weather information.

The following definitions were introduced: Plankton was defined as all organisms retained by the three nets having mesh sizes No. 6, No. 12, and No. 20. The random variable, y, is the combined dry weight of all plankton obtained in the three nets in one sample calculated per cubic meter of water. Position of each sample means the mid-point between starting and stopping position. Likewise, time is taken as the mid-point between start and stop.

In order to study the effect of the position and depth of each sample, the surface of the lagoon was divided into 9 sections by latitudinal and longitudinal lines as indicated in Figure 3. For each of these factors there are three levels designated as a_i ($i = 1, 2, 3$) and b_j ($j = 1, 2, 3$), respectively.

Likewise, for depth there are three levels designated as c_k ($k = 1, 2, 3$) where the observations from the 61 cm and 3 m depths were considered to be representative of the water layers from the surface to a depth of 4.3 m, the observations from 6.1 m were considered to be representative of the water stratum from the 4.3 to 12.2 m depths, and finally, the observations from the 22.9 m depth were taken to be representative of the third and last depth stratum extending from 12.2 m to the bottom.

The dividing points at the 4.3 and 12.2 m depths correspond approximately to the geometric means between the 3 and 6.1, and 6.1 and 22.9 m depths. The random variable can thus be described as y_{rijk} where r or replications within each cell ranged from 2 to 13 and where the three main factors, latitude, longitude, and depth, each have three levels.

However, observations from the levels of c_2 and c_3 exist only in cells $a_1 b_2 c_2$; $a_1 b_2 c_3$; $a_2 b_3 c_2$; $a_2 b_3 c_3$; $a_3 b_3 c_2$; and $a_3 b_3 c_2$, and there are 12 cells with missing observations. There thus exists a strongly unbalanced incomplete $3 \times 3 \times 3$ factorial design with the three main factors considered fixed.

The basic model used was one with fixed variables

$$y_{rijk} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{kj} + \varepsilon_{ijk}$$

where α stands for latitudinal position effect, β for longitudinal position effect and γ for depth effect. The second order interactions were considered non-significant and included in the experimental error, ε_{ijk}. The somewhat complicated analysis of the given data can speedily be done with a BIMD # 14 (1961) program which analyzes the statistical significance of independent variables that can be formulated in terms of general linear hypothesis model. Since the variables may be either classification or analysis of variance variables or regression variables of covariates, it is possible to introduce as covariates x_{ijk} or time of day of a sample and z_{ijk} or day of sampling. The expectation within each cell would then be:

$$E(y_{rijk}) = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{kj} + \upsilon_1 x_{ijk} + \upsilon_2 z_{ijk}$$

with the following linear restraints:

$$\sum_i \alpha_i = \sum_j \beta_j = \sum_k \gamma_k = \sum_{i,j} (\alpha\beta)_{ij} = \sum_{i,k} (\alpha\gamma) = \sum_{j,k} (\beta\gamma) = 0$$

A $\log_{10}$ y transformation of the random variable was introduced as recommended by WINSOR and CLARKE (1940). The results of this analysis are given in the following analysis of variance table:

H:	$\alpha_1 = \alpha_2 = \alpha_3 = 0$	$\beta_1 = \beta_2 = \beta_3 = 0$	$\gamma_1 = \gamma_2 = \gamma_3 = 0$	$(\alpha\beta)_{ik} = 0$
d.f.	2 and 105	2 and 105	2 and 105	5 and 105
F	.00	1.74	.00	3.29**

$(\alpha\gamma)_{ik} = 0$	$(\beta\gamma)_{jk} = 0$
4 and 105	2 and 105
.37	.00

No significant difference could be established in regard to the main factors and the first order interaction between latitude or longitude and depth. However, the interaction between latitude and longitude or position within the lagoon proved to be highly significant $P < .01$. This result is not surprising considering the current regimen inside the lagoon. In looking at the cell means, pronounced differences were evident with the lowest values of the cell means found near the west and south passes with outward current. Consequently, the best total estimate of the plankton biomass within Rongelap Atoll will be derived by stratifying the lagoon and by using total volume within each stratum, N_i as weighting factors.

Estimate of total plankton biomass

The term "plankton" is again defined as the organisms retained by the employed nets, and weight refers to dry weight. Repeated observations exist for each of the 9 cells in the water layer from surface to 4.3 m. For the two deeper layers extending from 4.3 to 12.2 m and from 12.2 m to the bottom, respectively, all three cells within each row or latitudinal direction were combined in both cases giving a total of 15 strata. The necessary data are listed in Table 1. The weighted over-all mean or population mean of dry weight of plankton per cubic meter in Rongelap Atoll is then (COCHRAN 1953),

$$\bar{y}_{st} = \frac{\sum_{i=1}^{15} N_i \bar{y}_i}{N} = .008{,}246 \text{ g/m}^3$$

where $\bar{y}_{st}$ = weighted over-all mean, $\bar{y}_i$ = mean weight of plankton per cubic meter and N_i = volume in m^3 within each stratum. Since the sampled volume is extremely small in relation to the total volume of each stratum the finite popula-

Table 1. Basic data from plankton sampling at Rongelap Atoll, 1959.

i = stratum number	Stratum location	N_i = 10^6 stratum volume in m^3	n_i	$\bar{y}_i$ = 10^{-6} mean weight gm/m^3	S_i = 10^{-6} Standard deviation of y_i	S_i^2 = 10^{-6} variance of y_i	$S^2(\bar{y}_i) \times 10^{-6}$
1	$a_1 b_1 c_1$	575.438	7	2,960	1,120	1.254,400	.179,200
2	$a_1 b_2 c_1$	501.728	18	20,466	24,133	582.401,689	32.355,649
3	$\sum_{j=1}^{3} a_1 b_j c_2$	3,799.840	6	7,573	2,893	8.369,449	1.394,908
4	$\sum_{j=1}^{3} a_1 b_j c_3$	19,073.773	7	7,146	1,653	2.732,409	.390,344
5	$a_1 b_3 c_1$	1,007.964	13	3,627	1,200	1.440,000	.110,769
6	$a_2 b_1 c_1$	569.213	5	3,680	2,560	6.553,600	1.310,720
7	$a_2 b_2 c_1$	357.099	4	4,400	3,387	11.471,769	2.867,942
8	$a_2 b_3 c_1$	294.410	13	5,813	3,493	12.201,049	.938,542
9	$\sum_{j=1}^{3} a_2 b_j c_2$	2,233.277	7	7,933	2,120	4.494,400	.642,057
10	$\sum_{j=1}^{3} a_2 b_j c_3$	11,724.756	7	7,733	2,493	6.215,049	.887,864
11	$a_3 b_1 c_1$	285.679	2	3,800	1,853	3.433,609	1.716,805
12	$a_3 b_2 c_1$	405.978	2	3,387	1,133	1.283,689	.641,845
13	$a_3 b_3 c_1$	348.655	12	17,400	19,626	385.179,876	32.098,323
14	$\sum_{j=1}^{3} a_3 b_j c_2$	1,883.261	10	16,826	16,533	273.340,089	27.334,009
15	$\sum_{j=1}^{3} a_3 b_j c_3$	8,300.482	11	10,706	5,893	34.727,449	3.157,041
	Totals	$51,361.552 \times 10^6$	124				

tion correction, fpc, has been omitted here and in subsequent developments. The variance of the weighted mean is:

$$V(\bar{y}_{st}) = \sum_{i=1}^{15} W_i^2 \frac{s_i^2}{n_i}$$

where $W_i = \frac{N_i}{N}$ and s_i^2 = estimated sample variance of the y values in the i^{th} stratum, and n_i = number of replication with each stratum. By substitution of the values given in Table 1 into the general formula for $V(\bar{y}_{st})$ the following estimate of variance is obtained omitting the fpc correction:

$$s^2(\bar{y}_{st}) = \sum_{i=1}^{15} W_i^2 \cdot s^2(\bar{y}_i) = 233.2 \cdot 10^{-3} \text{ and}$$

$$s(\bar{y}_{st}) = .483 \cdot 10^{-3}$$

The best estimate of the total biomass, B, in dry weight inside Rongelap Atoll is then $B = N\bar{y}_{st} = 423.5$ metric tons, and with 5‰ confidence limits ($t = 1.98$ with 123 d.f.) of

$$B = N\bar{y}_{st} + tNs(\bar{y}_{st}) = 472.6 \text{ metric tons and}$$
$$B = N\bar{y}_{st} - tNs(\bar{y}_{st}) = 374.4 \text{ metric tons.}$$

Optimum sample allocation

The obtained estimates of the standard deviation of the random variable, weight of plankton per cubic meter, in each stratum can be used to estimate the allocation of samples given a fixed sample size, n, which will produce minimum variance of $\bar{y}_{st}$ by random sampling in each stratum.

By selecting a Lagrange multiplier λ the problem is to minimize

$$V(\bar{y}_{st}) + \lambda(n_1 + n_2 + \ldots\ldots + n_{15} - n)$$

Again, by omitting the fpc correction this leads to the following expression for n_i (Cochran 1953)

$$n_i = n \cdot \frac{N_i s_i}{\sum_{i=1}^{15} N_i s_i}$$

and the obtained values are given in Table 2. It is quite evident that the employed sample design failed to give proper attention to the deeper and larger strata, which would have reduced the confidence limits of the estimated biomass of plankton with the same sampling effort.

Minimum sample size needed to meet a specified degree of accuracy of total biomass estimate

Finally, it may be asked what is the minimum sample size needed to give the estimate of biomass with a prescribed degree of accuracy.

The objective chosen was to obtain a coefficient of variation of 5%. Since B = 423.5 tons, the desired standard variation is (.05) (423.5) = 21.177 tons and the desired variance $V = 21.177^2 = 474.25$ again following COCHRAN (1953) and omitting the very small fpc correction

$$n_{min} = \frac{(\Sigma N_i s_i)^2}{V} = 69.9 \simeq 70$$

by substitution of the values developed above.

Finally, the optimum sample distribution within the strata can be calculated for the minimum sample size n_{min} by multiplication of the sample sizes in Table 2 by the factor $K = \frac{70}{124} = .5645$.

Table 2. Distribution of sample size.

Stratum	Actual sample size	Optimum sample distribution	Optimum sample dist. by minimum sample size
1	7	.4	.2
2	18	8.2	4.6
3	6	7.5	4.2
4	7	21.5	12.1
5	13	.8	.5
6	5	1.0	.6
7	4	.8	.5
8	13	.7	.4
9	7	3.2	1.8
10	7	19.9	11.2
11	2	.4	.2
12	2	.3	.2
13	12	4.7	2.7
14	10	21.2	12.0
15	11	33.3	18.8
Totals	124	123.9	70.0

Summary

1. Construction of a multi-net plankton pump sampler has been described. The sampler was used to assess the biomass of plankton in Rongelap Atoll in 1959. Nets having mesh sizes No. 6, No. 12, and No. 20 were used.

2. The volume of water in the lagoon was calculated to be $51{,}361 \cdot 10^6$ m³.

3. The mean dry weight of plankton weighted by the volume of each stratum was estimated to be .008,246 gm/m³.

4. The total biomass of plankton was estimated to be 423.5 tons with 5% confidence limits of 472.6 tons — 374.4 metric tons.

5. The variances of the mean weight in each stratum were utilized to calculate an optimum sample allocation.

6. The minimum sample size needed to give a 5% coefficient of variation was calculated to be 70 observations, taken randomly but with optimum distribution within each stratum.

References

Aron, W. 1958. The use of a large capacity portable pump for plankton sampling, with notes on plankton patchiness. — *J. Mar. Res.*, **16**, 158—173.

— In press. Some aspects of sampling the macroplankton. — *Rapp. Cons. Explor. Mer*, Symposium on Zooplankton Production, Copenhague, Oct. 1961.

BIMD Computer Programs Manual. 1961. Division of Biostatistics, Dept. of Preventive Medicine and Public Health, School of Medicine, Univ. of Calif., Los Angeles. [Processed.]

Cochran, W. G. 1953. *Sampling techniques.* New York: Wiley. 330 pp.

Von Arx, W. S. 1954. Circulation systems of Bikini and Rongelap Lagoons. — *Prof. Pap. U. S. Geol. Surv.* **260**, 265—273.

Winsor, C. P., and Clarke, G. L. 1940. A statistical study of variation in the catch of plankton nets. — *J. Mar. Res.* **3**, 1—34.

Discussion

J. Clark: Was phytoplankton included in biomass estimate?

Mathisen: Yes, but only those forms retained by the nets used in this particular pump.

MacCoy: What is the nature of the pump and what is its effect on the plankton?

Mathisen: The plankton was, as a rule, better preserved than plankton obtained with a standard ½ m net. One-fourth of all our samples was preserved in formalin for species identification.

Langford: What was the extent of variation from one sampling period to another?

Mathisen: Variability between replicate hauls was small, while variability between cell means could be substantial as indicated by the significant first order interaction for position effect.

Allanson: Do you think that there is an effective size limit, especially for smaller boats?

Mathisen: No, pumps of this type have been used with a capacity of 300 l per minute and with an inside diameter of the intake hose of about 5 cm.

Tonolli: Can you give a crude estimation of the renewal of water inside the atoll because of tides?

Mathisen: Von Arx has estimated water renewal inside Rongelap Atoll to take about one month.