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A NEW TWO-WAY MIXED ANOVA MODEL WITH AN UNBALANCED NESTED STRUCTURE IN GEOLOGY - ANOTHER DOWN-TO-EARTH DESIGN

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1. Introduction

It is often desired prior to implementing large scale regional geochemical surveys to determine with a small field experiment the effect of certain fixed treatments on the relative magnitudes of the data variability across the region, on some local scales, and in the laboratory. These fixed treatments might be the use of different field operators, soil horizons, stream sediment materials, or lake basins. Following these special, small scale, orientation-type studies, the routine regional surveys may then be better implemented, where the main interest lies in the investigation and analysis of the random effects due to the spatial and laboratory variability.

This paper describes a new two-way mixed analysis of variance (ANOVA) model, which employs a structured unbalanced nested design to meet the above objective as part of Canada's National Geochemical Reconnaissance Program. Implementation and interpretation of this model to stream and lake sediment surveys are highlighted along with the features of a tailor-made computer program. The outputs of this program consist of: (1) the usual ANOVA table entries - with focus on the (unbiased) estimates of the variance components; (2) a summary of the coefficients of the expected mean squares for all model terms; (3) a traditional set of hypothesis tests calculated under conservative conditions; and (4) a pooled ANOVA table - applicable when the interaction or nested component(s) of variance can be considered negligible. Further statistical areas of research for this model are briefly addressed in the concluding section.

Considerable research and insight into the properties and use of unbalanced "staggered" nested designs has been published in the statistical literature (e.g., see Bainbridge [2], Leone et al. [23], Searle [27], and Anderson [1]), including its use in a 2-way layout for completely random effects (Thitakomal [32]). However, the two-way mixed model application of this design is different from the work above. In the geological field, the usefulness of such a model was first hinted at by Flinn [6] for balanced data. Nevertheless, no further reference to this model application occurred until the mid-1970's, when the U.S. Geological Survey began using two-stage applications of unbalanced staggered designs as exemplified by McNeal [24]. In this case, a simple 2-way mixed model with pooled replication was analysed in parallel with an unbalanced staggered nested model involving only random effects. Although much information is obtained from this approach, the direct handling and analysis of the structured interaction-nested effects of the more appropriate 2-way mixed model is lost.

2. Statistical Model and Computational Method

As part of Canada's National Geochemical Reconnaissance Program, a new, 2-way mixed ANOVA mixed model that incorporates an unbalanced staggered nested design was developed. For illustration purposes the statistical model is expressed for a two-stage nested structure, as given by Figure 1; that is,

$$x_{ijkm} = \mu + A_i + b_j + c_{k(j)} + d_m(jk) + (Ab)_{ij} + (Ac)_{ik(j)} + (Ad)_{im(jk)}$$

where x_{ijkm} represents an individual analysis; μ is the true overall mean concentration of the element of interest; A_i represents the ith fixed (operator) effect; b_j, c_k, and d_m represent the random (location) effects where k is nested in j, and m within k; and, $(Ab)_{ij}$, $(Ac)_{ik}$, $(Ad)_{im}$ represent the various interaction effects between the fixed and random factors. It is assumed that the A_i's are constants, subject to the restriction $\sum_i A_i = 0$, $i = 1,2,\ldots,a$; and that b,c,d, Ab, Ac, Ad are all random variables, each normally and independently distributed with zero mean and constant variances: σ_b^2, σ_c^2, σ_d^2, σ_{Ab}^2, σ_{Ac}^2, σ_{Ad}^2 respectively, with $\sum_i (Ab)_{ij} = 0$ for all j. All covariances are assumed zero, except for $\sigma[(Ab)_{ij}, (Ab)_{i'j}] = [-1/(a-1)]\sigma_{Ab}^2$, for $i \neq i'$. The subscript representation and limits can be seen from the example layout of Figure 1. The above model description follows that of Model III of Hocking [20], and of Searle [27; pp. 401-403].

For this model formulation, our prime interest lies in the calculation of the components of variance and their relative contributions to the total variability. Secondly, it is of interest to test the usual hypotheses: $H_{01}: A_i = 0$, for all i; $H_{02}: \sigma_b^2 = 0$; $H_{03}: \sigma_c^2 = 0$; $H_{04}: \sigma_d^2 = 0$; $H_{05}: \sigma_{Ab}^2 = 0$; and $H_{06}: \sigma_{Ac}^2 = 0$. Also, appropriate and valid pooling of mean squares is desired.

A number of different techniques exist in the statistical literature for estimating the variance components in mixed models from unbalanced data; for example, see Henderson [18], Hartley and Rao [15], Harvey [16], Searle [27], Rao [25], and Speed and Hocking [30]. See Harville [17] and Searle [28] for recent summaries of this area. Computational methods for computing the expected mean squares for such models are reviewed in Kennedy and Gentle [22]. Employing the popular method of equating mean squares to their expected values, Henderson's (1953) Method 3, and an approach suggested in Goodnight and Speed [11], Binder [3] derived unbiased estimators for the variance components of the mixed effects model for the unbalanced staggered nested design having any number of levels of nesting and 2-way factor interactions. This work extends the Bainbridge [2] and Leone et al. [23] analyses of the unbalanced staggered nested design to the 2-way mixed effects model.

Having accomplished the primary objective, the unbiased estimation of the variance components, attention was briefly given to the construction of F-tests for the desired hypotheses stated above. Snee [29] reviews the construction of synthesized F-tests for unbalanced completely nested designs. In balanced data situations, Graybill and Wang [13] discuss confidence interval estimation for proportions of variability in two-factor nested random models, and Cohen and Miller [4]discuss appropriate F-test statistics for the 2-way mixed model. See Hemmerle [19] for a recent treatment of standard hypothesis tests for the fixed-effects ANOVA model with unbalanced data involving missing cells. A simple, approximate approach to F-testing was chosen for our immediate purposes. The general F-test algorithm so employed is summarized below:

FACTOR (General Case)	MS Ratio	Testable Condition(s)
A (fixed effect)	A/Ab	None (always testable)
b (main random effect)	b/Ab	$\sigma^2_{Ab} = \sigma^2_c = \ldots = \sigma^2_t = 0$
c (1st nested random effect)	c/Ac	$\sigma^2_d = \ldots = \sigma^2_t = 0$
d (2nd nested random effect)	d/Ad	$\sigma^2_e = \ldots = \sigma^2_t = 0$
⋮	⋮	⋮
t (last nested random effect)	t/At	None (always testable)
Ab (main interaction effect)	Ab/At	$\sigma^2_{Ac} = \ldots = \sigma^2_{As} = 0$
Ac (1st nested interaction eff.)	Ac/At	$\sigma^2_{Ad} = \ldots = \sigma^2_{As} = 0$
Ad (2nd nested interaction eff.)	Ad/At	$\sigma^2_{Ae} = \ldots = \sigma^2_{As} = 0$
⋮	⋮	⋮
As (t-1 nested interaction eff.)	As/At	None (always testable)
At (last nested interaction eff.)	None	--

The MS pooling procedure follows the four rules listed next: (1) Beginning with the lowest interaction hypothesis test (on As), the error term (At) is appropriately pooled when a Non-Significant result occurs (following Storm [31], where pooling is only carried out when the computed F value is less than the critical value $2F_{.50}$ at the stated degrees of freedom);(2) Pooling continues in this fashion if Non-Significant results occur with the subsequent higher level interaction hypothesis tests;(3) When the first Significant result occurs, the pooling procedure terminates; (4) A similar pooling procedure is followed for the nested effects, nested effects being pooled from the last upwards when Non-Significant hypothesis tests occur. Once a Significant result occurs the pooling procedure terminates.

It is emphasized that this algorithm is only an approximate F-test procedure. A cautionary note on the use of these F-test results accompanies each ANOVA table printout. Further research is needed in exact F-test methods for this model application.

3. Illustration and Application of Computer Programs

A computer program incorporating the above model and computational methodology was developed. Tables 1 and 2 illustrate the outputs of this program for a particular stream sediment survey, which was conducted under the design configuration as shown in Figure 1. Note that the "expected mean square coefficients" displayed in Tables 1 and 2 have their own particular factor ordering, that is: A, b, Ab, c, Ac, d, Ad when starting from the upper left-hand corner. Also, note that the "percent of total" column excludes the fixed effect, and treats negative variance components as zero contributions. The remaining information is largely self-explanatory.

To motivate and describe the application of this model and the interpretations underlying the resulting ANOVA calculations, two small detailed studies carried out in Canada during 1978 and 1979 will be highlighted. Both studies sought to investigate those aspects of stream sediment geochemical variability in the Yukon and lake sediment geochemical variability in northern Saskatchewan deemed important to subsequent large scale, regional geochemical exploration surveys. Each study involved unbalanced staggered nested designs in a two-way mixed effects model framework.

i) Example 1: Stream Sediment Survey

In stream sediment geochemical reconnaissance surveys fine silt is collected in the field for subsequent laboratory analysis. Often it is difficult to collect the ideal fine active silt sample material from the flowing part of the stream, and it has been proposed that the silt trapped in moss at the water's edge may be a suitable alternative sample material (see Goodfellow et al.[10]). It is recognized that local and analytical variability exists in stream sediment surveys, and the differences due to operators collecting the two sample types should be viewed in terms of the local variability. For logisitic reasons, 10m and 100m were the scales of local variability studied (see Garrett [7]); the resulting nested structure is shown in Figure 1. Prior to data analysis an investigation was made of data normality and homogeneity of variance under different transforms; the conclusion of this work was that a logarithmic transform best helped fulfill the requirements of the ANOVA method. The two-way mixed ANOVA model was used to determine if the fixed operator/sample material effects observed at randomly selected sampling locations were significant. The ANOVA results are summarized in Table 3; of the 10 elements studied, only two showed significant fixed operator/sample effects. The geochemical explanation is that the loosely held Zn or Fe in the stream sediments extractable with EDTA solution is also extractable by the weak acids present in the moss. Mosses are known to be Zn accumulators in the natural environment. The results for 5 elements are displayed in a means plot in Figure 2. In addition to the significant operator/sample fixed effect for Zn-EDTA a significant interaction effect for U-EDTA occurred. This interaction is not at the location level but at the highest nested level of 100m. The significant interaction reflects an inhomogeneity in the data at the 100m spatial level for U-EDTA. Clearly, sampling locations 3 and 4 behave differently from locations 1 and 2, a tendency to this situation is also revealed from Zn-EDTA. No geochemical explanation for this can be offered at this time, but it outlines an area of study which could be pursued in the future.

ii) Example 2: Lake Sediment Survey

In geochemical reconnaissance lake sediment surveys, lakes with a single central deep basin are usually sampled, as this is the preferred lake type (see Coker et al. [5]). However, sometimes larger multi-basin lakes are all that are available in the local environment. Prime interest was to study the variability between two basin types within single lakes, the basins which would have been routinely sampled and the basins which would have been left unsampled, over several sampling (lake) locations, in terms of analytical, sample (20m) and site (300m) variability (see Hornbrook and Lund [21]). To determine if the sampler could inadvertently introduce bias into a survey by selecting one particular basin from a multi-basin lake, 18 randomly chosen, multi-basin lakes were sampled using a three-level unbalanced staggered nested design. Within each lake basin, two sites were visited; at one of these sites, two samples were collected with one of these samples analyzed in duplicate. The design is illustrated by Figure 3; two levels of nesting were used. In an initial investigation of the data, 3 sampling locations were rejected as anomalous in nature due to the presence of atypical lake environments, which included major differences in geology and the presence of mineralization. The data for the 15 acceptable sampling locations were reviewed with consideration to normality and homogeneity of variance, and appropriate transforms were employed in the subsequent ANOVA.

The ANOVA results are summarized in Table 4, where it may be seen that for all 7 elements studied, the fixed basin effect is non-significant; that is, in the context of geochemical reconnaissance surveys it is immaterial which basin is sampled. It is worthwhile to note in Table 4 that approximately 45-65% of the variability for each element lies between lakes, and in general, far less than 8% of the systematic nested variability lies within single lake basins. This fact is gratifying as it demonstrates the extreme homogeneity of the centre-lake basin sample material and emphasizes that most of the variability in these lake systems lies between the lakes which are the basic sampling units in reconnaissance surveys. The interaction terms are of interest; in particular, note that the highest portion of the interaction variability is at the between sites in lake basins level, with U being the only exception. This indicates that site level variability is inconsistent as to whether site 1 or site 2 in a basin has a higher metal content from basin to basin in single lakes. This is both the expected and desirable location for the variability. If geochemically meaningful amounts of variability occurred at the site, sample or analytical nested levels, then it would indicate significant differences where none should be expected due to the randomization procedures introduced in the experimental design. In that sense the variability described by the interaction effects is akin to the lower levels of variability determined in a one-way nested design study. For further details see Hornbrook and Lund [21].

4. Conclusions

The two-way mixed ANOVA model is extremely relevant to detailed studies in geochemistry, and other natural sciences. The natural sciences provide abundant examples of random effects models, often nested, where one wishes to extrapolate the results of an analysis to a wider universe. However, fixed effects are not uncommon in preliminary orientation and detailed surveys. As with all experimental design the worker is often brought to his knees by the costs of employing balanced designs. In previous geochemical studies the two-way nested problem has been approached in steps (e.g., McNeal [24], and Hornbrook and Lund [21]), using separate two-way replicated and one-way unbalanced nested analyses. The two-way mixed ANOVA model employing a staggered nesting offers both an elegant, and useful, practical tool. Importantly, the structuring of the replication yields extremely useful additional information on the location of variability in the design, and under some circumstances allows mean squares to be pooled to provide more powerful tests of significance for the variance components.

Much theoretical research can still be done with this model, particularly with respect to more accurate F-testing over higher levels of nesting. In the simpler, completely nested unbalanced model a number of specific areas of study, including F-testing and confidence interval procedures, have been outlined in Goss and Garrett [12]; when those areas of concern are combined with the approach and references given in this paper, it is clear that much work lies ahead. Finally, much insight and understanding of the model's properties would be gained if computational comparisons and/or efficiency studies were carried out among competing variance estimation methods (e.g., ANOVA, maximum likelihood, MINQUE, etc.).

The computer program is still under development. It is planned to generalize the code to handle up to 9 levels of unbalanced, staggered nesting in a framework of up to 9 fixed treatments and 99 random effects for the 2-way mixed ANOVA model. Appropriate hypothesis testing and pooling will be carried out automatically. The code and examples will likely be published in Computers and Geosciences as done with previous work by Garrett and Goss [8].

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TABLE 1: ANOVA TABLE FOR ZN-EDTA STREAM SEDIMENT SURVEY

BASIN VARIABILITY STUDY, NTS 105B, YUKON, SUMMER 1978 NESTED DESIGN OPERATOR COMPARISON 15/03/80

RESPONSE IS ZN - EDTA

COLUMNS (TREATMENTS) ARE FIXED EFFECTS FOR OPERATORS
ROWS (BLOCKS) ARE RANDOM EFFECTS FOR LOCATIONS

DATA TRANSFORM - LOG10

NUMBER OF COLUMNS = 2
NUMBER OF ROWS = 4
NUMBER OF LEVELS = 2

SOURCE OF VARIATION	SUM OF SQUARES	DOF	MEAN SQUARE	VARIANCE COMPONENT	PERCENT OF TOTAL	F-RATIO	PROB. VALUE	SIGNIFICANCE (1)
OPERATORS	.42107	1	.42107	.32169E-01	.00	12.02	.9596	*
LOCATIONS	.49980	3	.16663	.16329E-01	26.15	N/C (2)		
100M SITES	.18990	4	.47475E-01	.21517E-01	30.76	13.92	.9672	-
10M SITES	.42685E-01	4	.10671E-01	-.66597E-02	0.00	.44	.2260	NS
OPERATORS X LOCATIONS	.10511	3	.35037E-01	.61273E-02	8.76	1.46	.6455	NS
OPERATORS X 100M SITES	.13666E-01	4	.34166E-02	-.15431E-01	0.00	.14	.0427	NS
OPERATORS X 10M SITES	.95963E-01	4	.23991E-01	.23991E-01	34.29			
TOTALS	1.3663	23		.69960E-01				

EXPECTED MEAN SQUARE COEFFICIENTS:

```
1.00  0.00  1.67  0.00  3.00  0.00 12.00
1.00  2.00  1.67  3.33  0.00  6.00
1.00  2.00  1.33  2.67
1.00  2.00
1.00  0.00  1.67  0.00  3.00
1.00  0.00  1.33
1.00
```

(1) NS = NOT SIGNIFICANT, F .LE. 0.95
* = F .GT. 0.95 BUT .LE. 0.99
** = F .GT. 0.99 BUT .LE. 0.999
*** = F .GT. 0.999

(2) N/C = HYPOTHESIS TEST NOT CALCULABLE DUE TO SIGNIFICANT NESTED TERM LOWER IN THE DESIGN

TABLE 2: POOLED ANOVA TABLE FOR ZN-EDTA STREAM SEDIMENT SURVEY

BASIN VARIABILITY STUDY, NTS 105B, YUKON, SUMMER 1978 NESTED DESIGN OPERATOR COMPARISON 15/03/80

RESPONSE IS ZN - EDTA

COLUMNS (TREATMENTS) ARE FIXED EFFECTS FOR OPERATORS
ROWS (BLOCKS) ARE RANDOM EFFECTS FOR LOCATIONS

DATA TRANSFORM - LOG10

NUMBER OF COLUMNS = 2
NUMBER OF ROWS = 4
NUMBER OF LEVELS = 2

--- POOLED ANOVA TABLE ---

SOURCE OF VARIATION	SUM OF SQUARES	DOF	MEAN SQUARE	F-RATIO	PROB. VALUE	SIGNIFICANCE (1)
OPERATORS	.42107	1	.42107	12.02	.9596	*
LOCATIONS	.49980	3	.16663	4.76	.6637	NS
100M SITES	.23250	8	.29073E-01	2.12	.8461	NS
OPERATORS X LOCATIONS	.10511	3	.35037E-01	2.56	.8717	NS
OPERATORS X 100M SITES	.10963	8	.13704E-01			
TOTALS	1.3663	23				

(1) NS = NOT SIGNIFICANT, F .LE. 0.95
* = F .GT. 0.95 BUT .LE. 0.99
** = F .GT. 0.99 BUT .LE. 0.999
*** = F .GT. 0.999

TABLE 3: SUMMARY OF ANOVA FINDINGS FOR STREAM SEDIMENT SURVEY

ANALYSES OF VARIANCE

	Operator Effect	Percentage Variance: Nested Effects: Locations	Nested Effects: 100m Sites	Nested Effects: 10m Sites	Interaction Effects: Locations	Interaction Effects: 100m Sites	Interaction Effects: 10m Sites
Zn-AR	NS	87.4***	0.0	0.0	0.0	0.0	12.6
Zn-EDTA	*	26.2	30.8	0.0	8.8	0.0	34.2
U-DNC	NS	81.9**	0.0	1.1	0.0	5.6	11.3
U-EDTA	NS	49.0°	0.0	0.0	0.0	45.0*	5.9
U-Fusion	NS	3.7	16.3	0.0	0.0	0.0	80.0
Fe-AR	NS	84.0*	2.9	0.0	3.5	0.0	9.6
Fe-EDTA	+	41.3+	0.0	17.7	0.0	29.6	11.4
Mn-AR	NS	78.8°	9.7	0.0	5.3*	0.0	6.2
Mn-EDTA	NS	48.9*	34.2	0.0	4.4	2.5	10.0
L.O.I.	NS	50.1***	0.0	7.1	0.0	0.0	42.8

Note: Significance < 0.001 ***
< 0.01 **
< 0.05 *
< 0.1 +

NS Operator Effect not significant
o Variance Component not testable

TABLE 4: SUMMARY OF ANOVA FINDINGS FOR LAKE SEDIMENT SURVEY

ANALYSES OF VARIANCE

	Basin Effect	Percentage Variance: Nested Effects: Lakes	Nested Effects: Sites	Nested Effects: Samples	Nested Effects: Analyses	Interaction Effects: Lakes	Interaction Effects: Sites	Interaction Effects: Samples	Interaction Effects: Analyses
U	NS	61.6°	6.1	0.0	1.7	12.3°	8.6°	5.8*	5.9
Zn	NS	56.6°	0.0	0.0	0.0	6.6°	31.6°	8.4***	0.8
Cu	NS	59.7°	0.0	1.2	0.0	6.5°	22.9°	5.0*	4.7
Ni	NS	66.4°	0.0	0.0	0.0	7.0°	15.3**	3.7	7.6
Co	NS	60.6°	0.0	0.0	0.1	2.1°	19.0**	7.4	10.8
Mn	NS	48.7°	0.0°	1.6*	0.0	12.5°	35.2°	1.7***	0.2
Fe	NS	45.3°	0.0	0.3	0.0	4.8°	48.0°	1.4***	0.1

Note: Significance <0.001 ***
<0.01 **
<0.05 *

NS Basin Effect not significant
o Variance Component not testable

FIGURE 1: TWO-STAGE STAGGERED NESTED STRUCTURE FOR STREAM SEDIMENT SURVEY

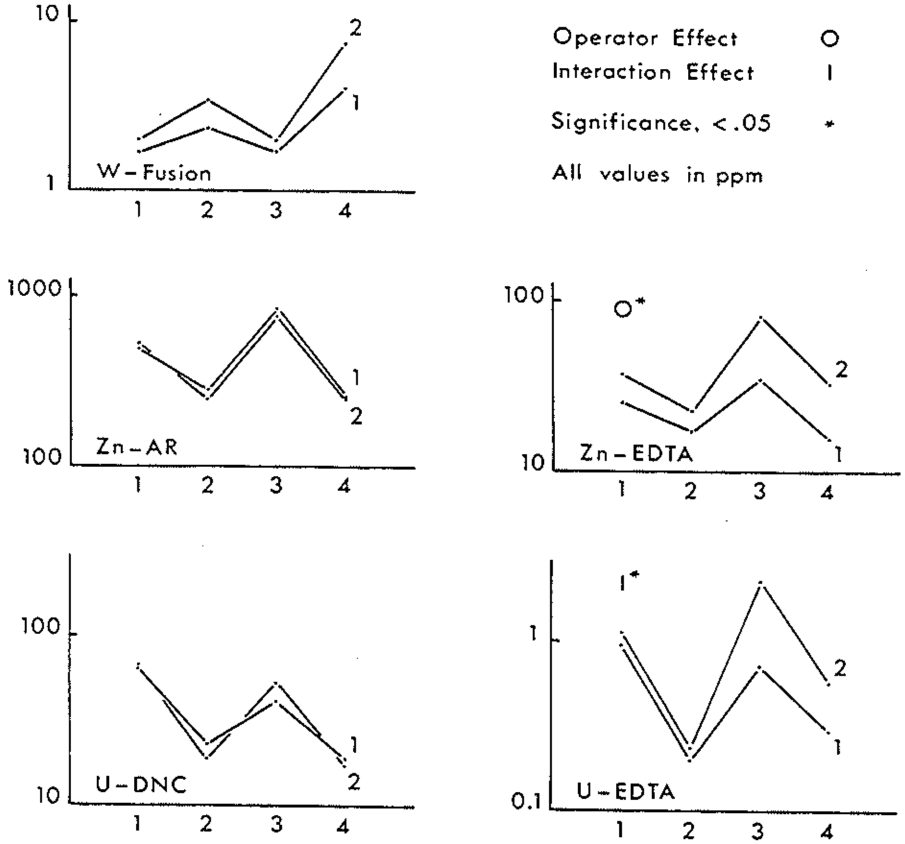

FIGURE 2: INTERACTION PLOTS FOR STREAM SEDIMENT EXAMPLE

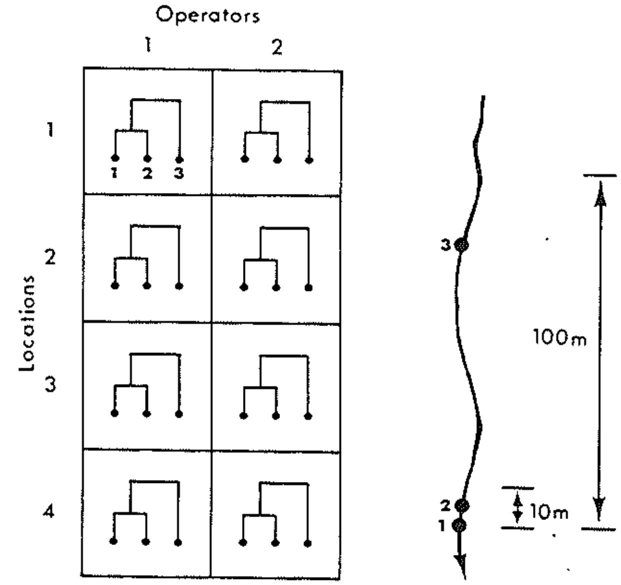

FIGURE 3: THREE-STAGE STAGGERED NESTED STRUCTURE FOR LAKE SEDIMENT SURVEY