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A Preliminary Assessment of Age and growth
of Atlantic Skipjack Tuna, *Katsowonus pelamis*,
from Dorsal Fin Spines.

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ABSTRACT

Dorsal fin spine sections have been used in several studies for aging tunas. As far as skipjack is concerned the trials have been made in an isolated fashion. The present study is part of an international research program on Atlantic Skipjack coordinated by I.C.C.A.T. (International Commission for the Conservation of Atlantic Tunas). It discusses the methodology developed by a group of scientists as well as the comparison of readings on the same sample. The paper deals with the difficulties encountered in developing the method and the differences that exist between different readers. A first growth estimate is advanced by three scientists of the working group on three samples of different origins. Classical growth curves do not adjust well to the datas observed. Nevertheless, the data seems to indicate that growth is rather slow, approximately 5 cm per year. A certain doubt subsists regarding the periodicity of growth marks. This may be improved by tetracycline antibiotic marking. Indeed the first recoveries indicate that this substance is deposited on dorsal fin spines. A longer time at liberty (around one year) seems necessary in order to dispose of a reliable method of mark periodicity determination.

Different approaches have been taken for determining the age and growth of skipjack. A synthesis of past work may be found in JOSSE at al (1979). These have included : length frequencies distributions, modal progressions, marking and recapture, and readings of bony pieces : vertebrae, otoliths, dorsal fin spines. This last method is the one developed in the paper, following the observations by SHABOTINIETS (1968) and the work of BATTS (1972) and CAYRE (1979).

The rates of growth obtained by the different authors are very variable, these may go from simple to double or even triple. These differences may be partially explained by the diversity of methods and origins of samples. By choosing the method of dorsal fin spines we have wished to simplify the sampling and preparation as compared to other bony parts, such as vertebrae and otoliths. In addition, thanks to the sampling net installed for the International Skipjack Year Program (I S Y P), we dispose of samples from different locations in the Intertropical Atlantic.

MATERIALS AND METHODS.

Following the meeting of the ICCAT Skipjack Subcommittee in November 1980, the persons concerned by the study of skipjack hardparts decided to meet in order to calibrate the methods of preparation and reading of dorsal fin spines. Difficulties related to individual work and geographical distances did not allow all researchers to meet at one time. A first group met in Brest, France (4 persons) and then another meeting took place in February 1981 at Dakar, Sénégal (3 persons). Only one investigator was able to assist to both meetings. A method having been developed (see description in the next paragraph), it was decided to compile a series of photographs, which were sent to each person concerned, including those that had not been able to assist to either meeting. Each reader was to return, on transparent sheets of paper, his readings and interpretations (estimated age) to Brest. It was then possible to compare the eight different readings of the same sample.

The first dorsal fin spine has roughly the shape of an elongated cone the diameter of which is maximum just above the articulation. It is important to section the dorsal fin spine at the same relative position if one wishes to make measurements and to retrocalculate the length of the fish as a function of the dorsal fin spine diameter. One proceeds in the following manner : a series of sections (500 - 700 microns) are cut above the articulation (3-5 mm according to the length of the fish). The form of the sections will change as one moves away from the articulation ; figure 1 shows which is the section to be retained. Sections were performed with an Isomet low speed saw (Model 11-1180).

Dorsal fin spine sections present a succession of hyalin bands (clear in transmitted light and dark in incident light) and opaque bands (dark in transmitted light and clear in incident light). Microradiographs done on several sections demonstrated that the hyalin bands represented zones of higher Calcium Concentration, which are normally related to periods of inhibited growth (CASTANET, 1977 ; COMPEAN, 1980). Furthermore, the central area of sections undergoes an alteration of bony tissue which intensifies as the fish grows older.

The discrimination of hyalin and opaque bands is easened by a drop of liquid on the section (for example, alcohol 60° or 90°).

Sections are roughly cone-shaped. Thus, if one disposes of two sections care must be taken in order to arrange a section on its lower side and the other inversely ; this may facilitate readings when the quality of sections varies. When [illegible] interpret structures (hyalin or opaque) at the outermost edge of sections, care must be taken to arrange these on their lower side in order to avoid reading the upper side by transparency. Finally, if one disposes of second dorsal fin spine sections these may aid in interpreting readings.

The hyalin bands have been named "rings". Their aspect is variable from one section to another and whithin the same section. A code was defined to enable readers to understand the interpretations proposed. Considering the quasi-total absence of knowledge on the biology and behaviour of skipjack in the geographical areas (migration, reproduction), it seems rather difficult to classify rings as "accidental", "spawning", etc. The choice would be much too arbitrary. Thus, rings are notated and described according to a code.

Further on, it appears from our readings and in the work of others (CHI and YANG, 1973 ; COMPEAN, 1980 ; CAYRE and DIOUF, 1981) that rings are often present as doubles or as groups containing more than two rings. This has brought us to grouping rings in order to give an estimated age ; each group being supposed to represent an annual cycle.

7 out of 8 readers used the following code (3) :

A : ring

AR : ring present in altered central zone

AF : blurry ring ; not well marked ; limits sligthly marked

AE : narrow ring

AL : large ring

Ai : incomplete ring

Ad : ring partially dedoubled

A* : ring particularly well marked

The reader [illegible] by this code and then indicates which ones he retains or the groups that he defines, in order to age the sample.

Reading example :

$$\underbrace{AFR}_{1} + \underbrace{AE + AL}_{2} + \underbrace{A^{*} + AF}_{3} + \underbrace{A + A + AF}_{4} + \underbrace{A}_{+}$$

Number of rings = 9

Estimated age = 4 +

Two different methods were used for measuring sections : one consisted of a profile projector fit with a stage coupled with a micrometer, which measured the distance travelled ; the other consisted of a binocular lens fit with an ocular micrometer. Two measurement techniques were developed, a statistical test on a sample of 30 different section readings did not show a significant difference between the two methods ; these may then be regrouped for measurement purposes (Figures 2 and 3).

RESULTS AND DISCUSSION.

Comparison of readings.

This comparison was made on the readings of 8 investigators on a sample of 78 photographs of dorsal fin spine sections. The reader did not have the characteristics of the fish (length, origin), in order to avoid orienting his reading. The photographic magnification for all prints was the same. Readers 1, 2, 3, 5 and 7 participated in developing the reading code and applied it, readers 6 and 8 applied the code without having participated in its development, reader number 4 developed his own method. The 78 samples were deliberately chosen from fishes coming from different origins (Carribean, Central Atlantic, Gulf of Guinea). We will not try to interpret results from the point of view of skipjack growth.

From a total of 78 prints, 17 (21.8 %) were considered unreadable by at least one person ; only one print (1.3 %) gathered total agreement, plus two others when agreement was considerer at ± 0,5 year level (total 3.8 %). It is evident that there is a great difference in the different interpretations. It is noteworthy to mention that a similar comparison on cod otolith readings showed 39 % agreement between 10 readers (Lopez - Veiga, 1976).

Table I shows the agreement between different pairs of readers.

In all cases the percentage agreements are inferior to 40 % except for the pairs 2 - 4 (56 %) and 7 - 8 (73 %). The lowest values are observed for the pairs 4 - 6 (8 %) and 2 - 6 (9 %). The high level of agreement achieved by the pair 7 - 8 may be explained by the fact that they had the opportunity to work together and also because they did not try to estimate age with a ± .5 year precision. The pair 2 - 4 achieves a high level of agreement even though the methods used are different and that they never had the opportunity to work together.

Table II shows agreement between pairs of readers at ± .5 year level. The agreement is clearly much higher : 16 pairs out of 28 agree at more than 50 %, 13 at more than 60 %. The difference with the preceding table (table I) comes from the fact that readers may assign a different meaning to "+" when they use it and / or difficulties related to reading the structures (rings) close to the edge of the sections.

Tables 3 and 4 show the bias between pairs of readers. The bias being defined by the sum of overestimated and underestimated ages. The mean bias serves to indicate the tendency of a reader in relation to the whole set.

One can see that readers 2 and 4 clearly underestimate age in relation to the set of readers ; readers 7 and 8 have the inverse tendency. Readers 3, 5 and 6 overestimate age slightly ; reader I has the inverse tendency.

The examination of reading formulae furnished by each reader shows, except in several particularly easy cases (very well marked rings), that the number of rings seen or retained varied from one reader to another ; in general, those that distinguished the greater number of rings overestimate age. Nevertheless, differences may arise by the way in which rings are grouped.

Based on the interpretations of each reader, a mean age was established for the 61 sections that were read by the whole set of readers. It was not possible to ajust models of the Von Bertallanfy type. We have thus chosen to represent age as a function of length by a least squares linear model.

Table 5 shows the mean age obtained for 61 sections ; mean have been compared by a t test. Only 4 pairs out of 28 do not show a significant difference between them at the 5 % level, 7 pairs at the 1 % level.

An example of linear adjustement is given in figure 4. The equations of the lines (functional) are given in table 6.

The parameters a and b of the predictive regressions ($y = a + bx$) have been compared by means of their ellipses of joint confidence limits, the heterogeneity of the residual variances did not permit an analysis of variance.

Figure 5 shows that two groups may be clearly distinguished by non-overlapping of ellipses : readers 3,5,6,7 and 8 on the one hand and readers 2 and 4 on the other ; reader 1 occupies an intermediate position between both groups. Readers 3,5,6 may be mingled (quasi-concentrical ellipses).

The determination of age in skipjack by the use of dorsal fin spines remains difficult. Even when a common methodology has been developed, interpretations may show important divergences. Differences arise from the number of rings seen and retained and from the way in which these are grouped. The absence and/or the blurry nature of rings in the altered central zone most likely increases the bias in reading, especially when the fish are larger (more than 50 cm). Finally, the nature of the edge of the sections are not read in the same manner by the different readers.

Nevertheless, the development of a common methodology allows for comparisons and grouping of readers.

As far as possible, an effort must be made in order that a sample be read by several investigators before drawing conclusions on skipjack growth. Selection of easily read sections must also be avoided, in order to keep from introducing a bias related to the choice of individuals that may be different from the mean.

Growth

An application of the method described above was performed by readers 1,3 and 5. We have seen (tables 2 and 5) that there was a high level of agreement between these three readers. Three samples were studied : a sample from the landings at Cumana (Venezuela) another from Dakar (Senegal) and the third from Abidjan (Ivory Coast).

The relation between the diameter of the dorsal fin spine section and fork length was studied in order to obtain retrocalculated values of length at ring formation.

The relation was studied for the three samples by adjusting measurements to a linear model without data transformation. The equations of predictive regressions are shown in table 7. The comparison between the lines was performed on the predictive regressions : $y = a + bx$; residual variances not being homogenous (F test significant at the 5 % level) comparison was done by means of ellipses of joint confidence limits. Figure 6 shows that there does not seem to be a significant difference between the three samples.

The problem appeared of the possible dissappearance of rings within the central altered zone of sections, especially for lengths greater than 50 cm. The measurements at different rings have been grouped in the form of a histogram (figure 7). This figure suggests three individualized groups of measurements at 800,1000 and 1300 microns radius, which would correspond to the first three groups of rings. These three groups have been considered when estimating individual age within samples.

Two different methods have been used to study growth. In one case we have estimated length at different ring formation by back calculation ; in this case an individual is considered for several age-length estimates. In the other case we studied growth by the individual age-length relation.

The first method increases the number of observations but, bias may be introduced from back-calculation and from the non-independance of the different age-length estimates, the second lends it-self better for adjustement to a mathematical growth model.

For growth estimated by back-calculation, the predictive regressions obtained for each sample were used in calculations. The formula retained was :

$$LFi = a + (LF - a)\frac{Ai}{A}$$

Where, LF_i = Fork length at time i

a = Y intercept

LF = fork length of fish

A_i = distance to ring measured

A = radius of section

The results obtained per sample and for each group of rings (periods) are shown in table 8. Increments per period are in the order of 4 to 5 cm ; These are slightly stronger for the sample from Dakar. It does not seem justified to push further the comparison between the different samples, especially if one considers the fact that lengths at first and second groups of rings has been determined from figure 7, by attribution of rings when none were visible in the altered central zone.

For different pairs of age-length estimates the best adjustement was obtained by linear regression. The lines obtained are shown on figures 8a, 8b and 8c. Table 8 shows the lengths obtained for each period supposed annual. The values are very close to those obtained by back-calculation and are also slightly higher for the sample from Dakar. Comparisons will not be pushed further considering the heterogeneity of samples and variances.

We have mentioned above that the proposed hypothesis of double ring formation per year for several Tuna species including skipjack was retained for this work. A validation of this hypothesis was tried by the observation of the nature of the edge of skipjack dorsal spine sections. This study was carried out by one reader on skipjack landed at Dakar for which samples existed covering the year 1980. The proportion of hyalin edges was calculated per month. Figure 9 shows that from January to June there would be a long period of inhibited growth (hyalin edge). From July to September growth would resume (opaque edge), later in October a new hyalin edge appears ; and finally, growth would resume in November and December. This pattern seems to indicate the formation of two rings per year. Nevertheless, several reserves must be advanced.

a) Monthly samples are not numerous and do not take into account possible inter-school differences.

b) The interpretation of the edge of a section is difficult and it may vary from one reader to another.

c) The period of inhibited growth from January to July seems rather long, it is not excluded that several rings may have formed during this period.

On the basis of annual periodicity, the increaments obtained in this study (4 to 5 cm) are on the average two times less than other estimates for Atlantic skipjack (BATTS, 1972 ; CARLES-MARTIN, 1975 ; CAYRE, 1980), based on hard parts. It is thus evident that our results must be regarded as provisional. The different studies during the skipjack program should permit to consolidate certain hypothesis. In particular, Tetracycline marking may permit to clear doubts concerning ring formation periodicity. The first returns from Tetracycline injected skipjack show that the product is visible on dorsal spine sections under fluorescence microscopy. The present number of Tetracycline marked and recaptured fish and their time at liberty, is not sufficient to permit a study of growth by this method ; only fish with approximately one year at liberty should permit to verify ring periodicity and study annual growth.

At this point we would like to make the following remarks :

a) Inhibited growth marks are numerous and may be large, indicating frequent and/or long periods of inhibited growth.

b) Growth marks may be narrow, indicating short periods of rapid growth.

c) Marks are frequently different from one fish to another in one sample, which indicates a great variability of individual growth.

These remarks lead us to suppose a relatively opportunistic growth for skipjack, that is enhanced by local favorable conditions encountered ; this hypothesis has been already advanced for gonad maturation (CAYRE 1981).

This leads to the idea of studying skipjack at the individual school level.

In conclusion, the authors estimate that reading skipjack age by dorsal fin spines, besides the difficulties encountered is an interesting method. It is the most simple to sample and to prepare.

The slow growth rate obtained in our study needs to be verified, particularly, by Tetracycline marking.

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TEXT FOOTNOTES

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3 - The code is derived from the french language which was adopted by the different readers.

TABLE LEGENDS

Table 1. Total agreement between pairs of readers.

Table 2. Agreement at 0.5 year level between pairs of readers.

Table 3. Bias between pairs of readers. Bias at ± 0.5 year level included. Bias = (% oversestimated age) - (% underestimated age).

Table 4. Bias between pairs of readers, for biais ± 1 year. Bias = (% oversestimated age) - (% underestimated age).

Table 5. Comparison of averaged estimated ages ; n = 61

Table 6a. Parameters of functional regressions : LF = a + b (age) for each reader.

Table 6b. Parameters of predictive regressions and correlation coefficients for age length estimates by each reader.

Table 7. Parameters of the regression : LF = a + bd where LF = fork length and d = section diameter.

Table 8. Periodical increments (supposed annual) obtained by back-calculation and by regression for 3 samples. L = fork length s = standard deviation.

Reader	1	2	3	4	5	6	7	8
1	x	31	38	31	30	23	13	14
2	31	x	25	56	31	9	13	10
3	38	25	x	20	39	24	38	30
4	31	56	20	x	24	8	16	13
5	30	31	39	24	x	14	23	26
6	23	9	24	8	14	x	21	21
7	13	13	38	16	23	21	x	73
8	14	10	30	13	26	21	73	x

Reader	1	2	3	4	5	6	7	8
1	x	67	66	62	61	50	40	46
2	67	x	48	72	41	54	25	22
3	66	48	x	46	63	65	67	61
4	62	72	46	x	35	50	26	24
5	61	41	63	35	x	60	45	48
6	50	54	65	50	60	x	66	61
7	40	25	67	26	45	66	x	73
8	46	22	61	24	48	61	73	x

Reader	1	2	3	4	5	6	7	8	mean bias
1	x	49	-26	47	-24	-23	-42	-64	-12
2	-49	x	-68	5	-61	-63	-64	-90	-56
3	26	68	x	70	- 2	2	-42	-46	11
4	-47	- 5	-70	x	-57	-41	-42	-87	-50
5	24	61	2	57	x	5	-29	-27	14
6	23	63	- 2	41	- 5	x	-45	-42	5
7	42	64	42	42	29	45	x	1	38
8	64	90	46	87	27	42	- 1	x	50

tab 4

Reader	1	2	3	4	5	6	7	8	mean bias
1	x	31	-20	36	-23	-16	-54	-44	-13
2	-31	x	-51	9	-57	-44	-76	-79	-47
3	20	51	x	54	0	-13	-27	-29	8
4	-36	- 9	-54	x	-58	-50	-74	-76	-51
5	23	57	0	58	x	- 3	-31	-29	11
6	16	44	13	50	3	x	-22	-28	11
7	54	76	27	74	31	22	x	1	41
8	44	79	29	76	29	28	- 1	x	41

Reader	1	2	3	4	5	6	7	8
averaged estimated age	3.25	2.78	3.55	2.69	3.58	3.56	4.07	3.95
sd ($\bar{x}$)	0.13	0.11	0.13	0.09	0.16	0.15	0.16	0.14

non significant difference at 5 % level	2 - 4 3 - 6 5 - 6 7 - 8
$1\ \% < \text{level} < 5\ \%$	1 - 3 1 - 5 1 - 6

[illegible] 6

6 a

Reader	a	b
1	27.57	7.77
2	25.72	9.78
3	24.06	8.12
4	21.98	1.47
5	29.23	6.47
6	26.34	7.39
7	26.14	6.54
8	22.19	7.60

6 b

reader	a	b	r	N
1	34.51	5.65	0.727	78
2	35.14	6.40	0.654	77
3	35.12	5.01	0.616	75
4	30.55	8.27	0.721	61
5	34.25	5.09	0.787	76
6	34.86	5.03	0.680	78
7	34.90	4.41	0.674	78
8	32.32	5.10	0.671	78

	a	b
CUMANA	19.6722	0.09275
ABIDJAN	19.8645	0.09133
DAKAR	19.4613	0.09216

age	Cumana back calculation	LF/age	Abidjan back calculation	LF/age	Dakar back calculation	LF/age
1	L = 34.1 δ = 1.81	L = 34.68 δ = 4.60	L = 34.5 δ = 2.21	L = 35.75 δ = 4.89	L = 34.2 δ = 12.00	L = 35.24 δ = 3.69
2	L = 39.0 δ = 2.70	L = 39.09 δ = 4.57	L = 38.8 δ = 2.72	L = 39.92 δ = 4.84	L = 39.5 δ = 2.55	L = 40.27 δ = 3.57
3	L = 44.1 δ = 2.90	L = 43.5 δ = 4.54	L = 43.2 δ = 3.08	L = 44.09 δ = 4.80	L = 45.1 δ = 2.87	L = 45.30 δ = 3.49
4	L = 47.9 δ = 2.95	L = 47.91 δ = 4.53	L = 47.5 δ = 3.52	L = 48.26 δ = 4.78	L = 49.8 δ = 2.08	L = 50.33 δ = 3.46
5	L = 51.6 δ = 3.69	L = 53.32 δ = 4.52	L = 52.4 δ = 4.68	L = 52.43 δ = 4.77	L = 54.0 δ = 3.16	L = 55.36 δ = 3.47
6	L = 53.6 δ = 5.13	L = 56.73 δ = 4.52	L = 55.6 δ = 5.54	L = 56.60 δ = 4.78	L = 57.7 δ = 3.69	L = 60.39 δ = 3.53
7	L = 62.8 δ = 6.18	L = 61.14 δ = 4.53	L = 58.7 δ = 3.78	L = 60.77 δ = 4.81		

FIGURE LEGENDS

Figure 1. a : sectioned too low ; b : sectioned correctly ; c : sectioned too high.

Figure 2. Profile projector measurements ; a1 a2 = symmetry axis ; d = section diameter ; r = radius : ring measurement.

Figure 3. Ocular micrometer measurements ; d : section diameter ; d1 : ring measurement ; $r' = d1 - d/2$

Figure 4. Example of length/age regression (reader n° 5)

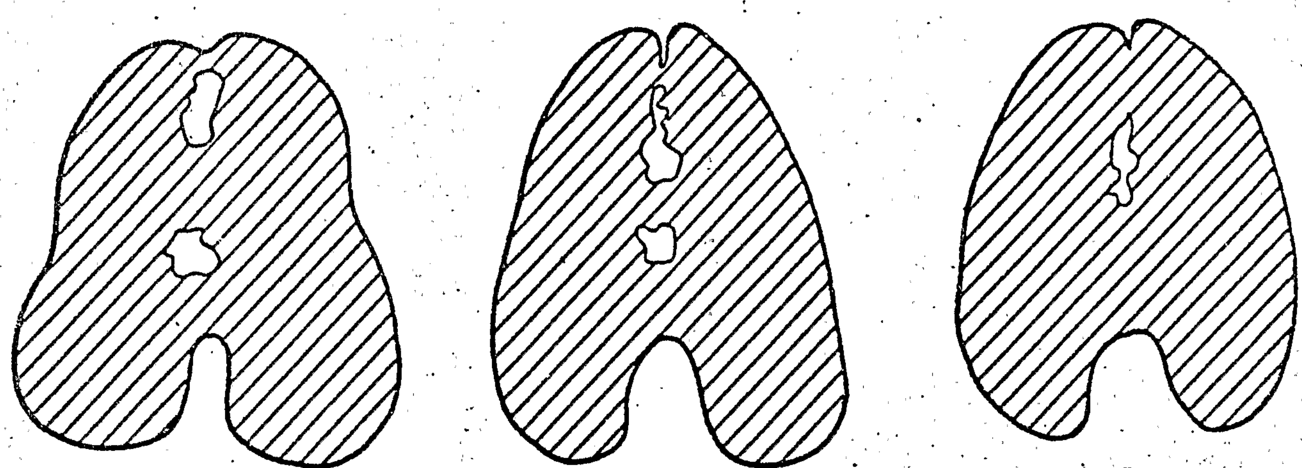
Figure 5. Ellipses of joint confidence limits for length/age regressions by different readers.

Figure 6. Ellipses of joint confidence limits for dorsal fin spine radius/fork length regressions.

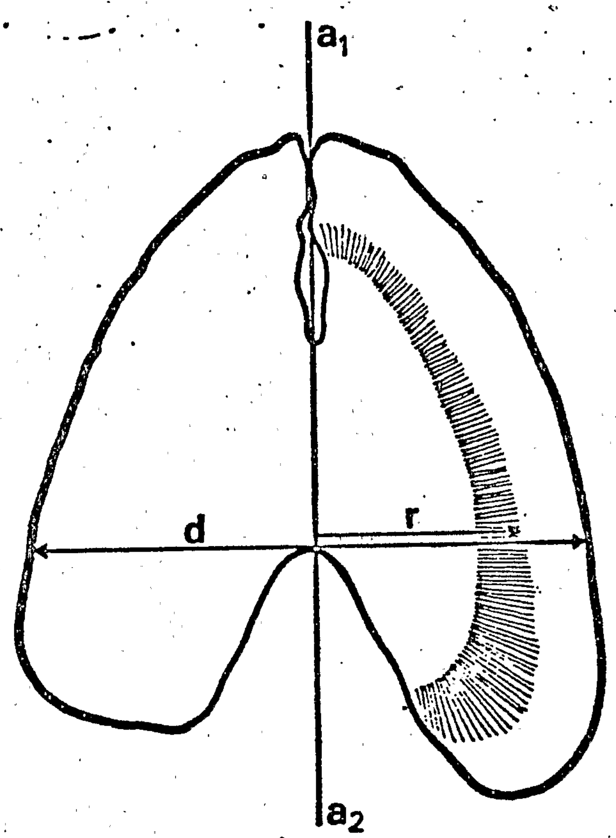
Figure 7. Ring radius frequencies, all samples combined.

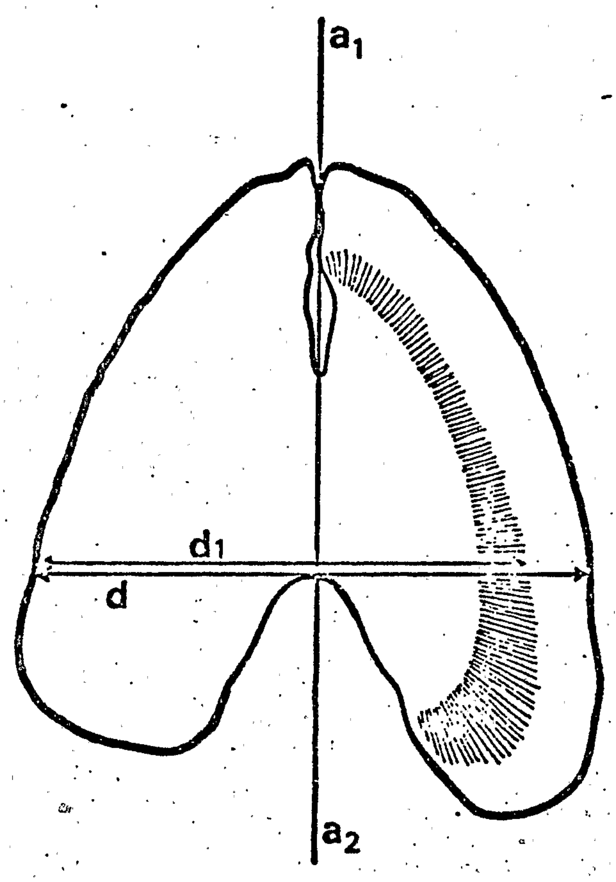
Figure 8. a) length/age regression, sample from Cumana.
b) Length/age regression, sample from Abidjan.
c) Length/age regression, sample from Dakar.

Figure 9. Hyaline edge frequencies through the year.

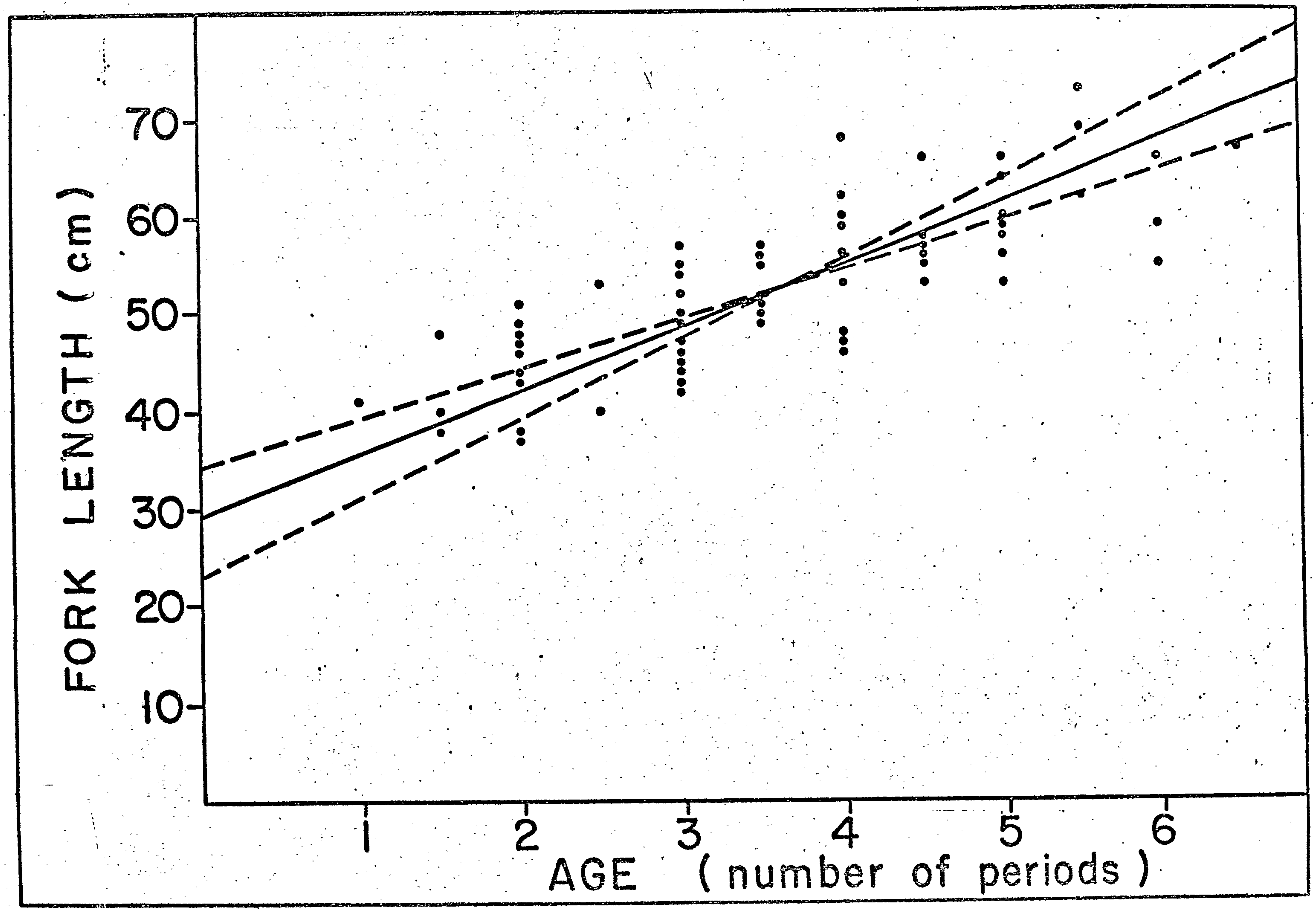


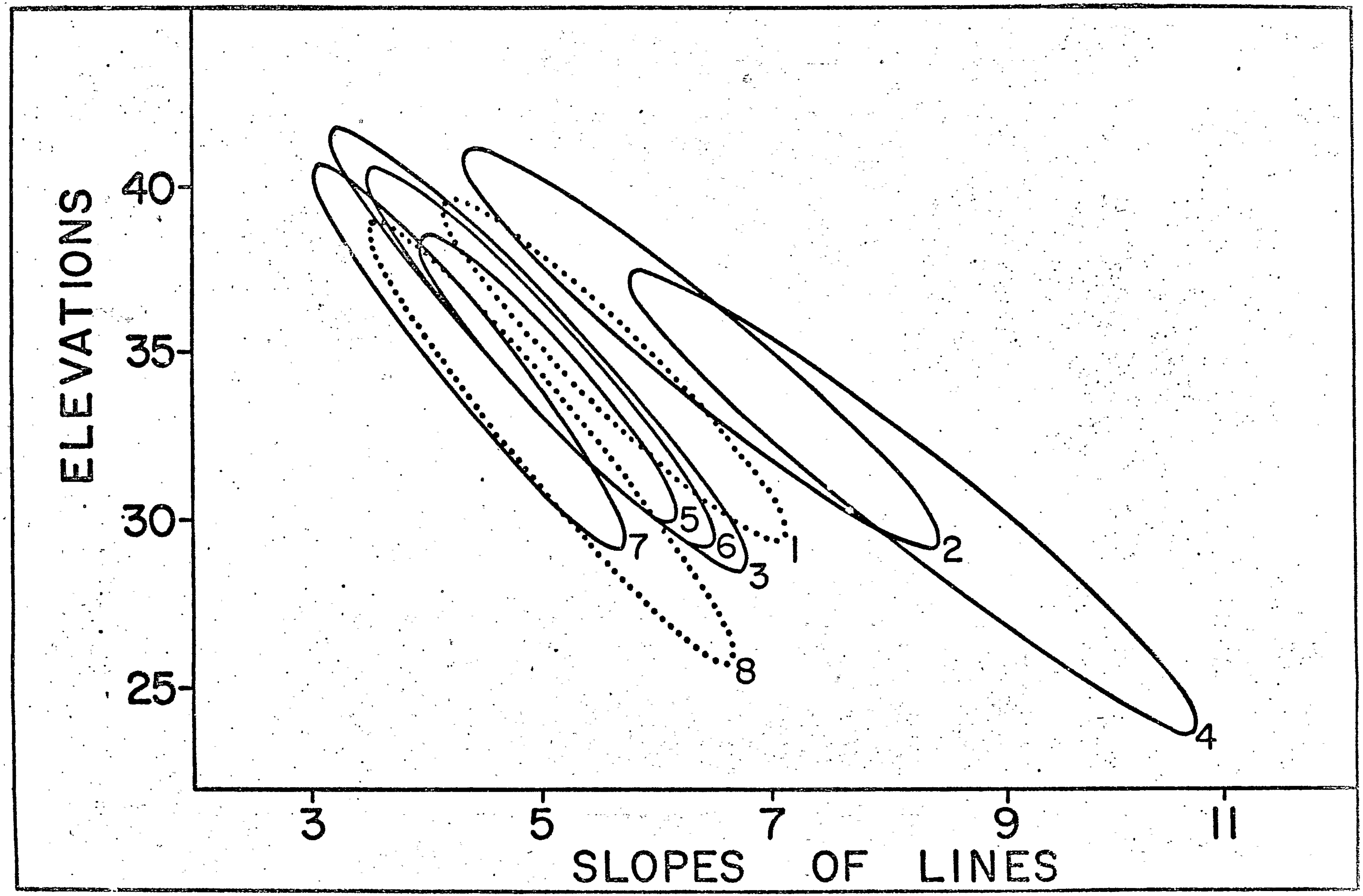
a b c

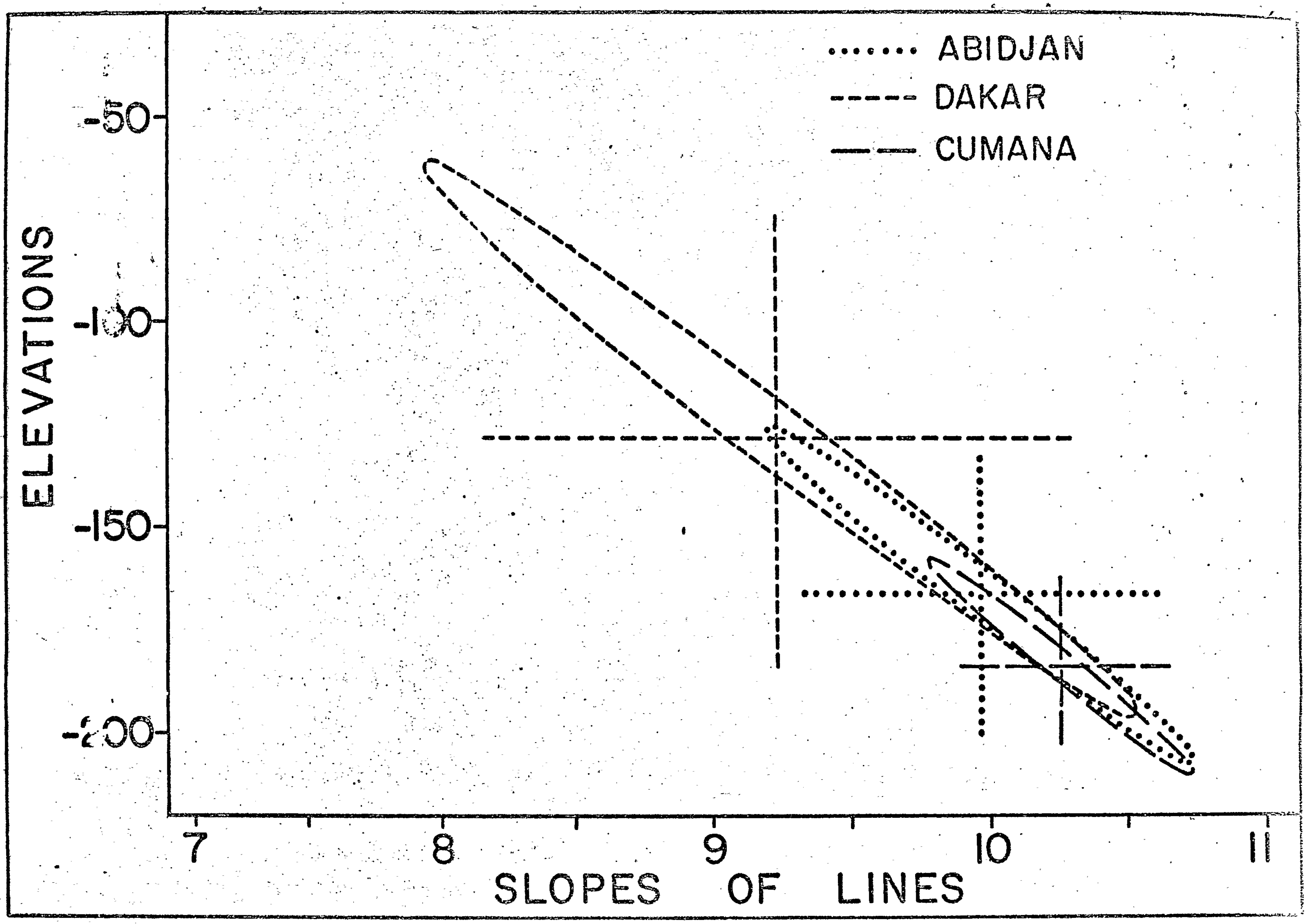

a1
d
r
a2

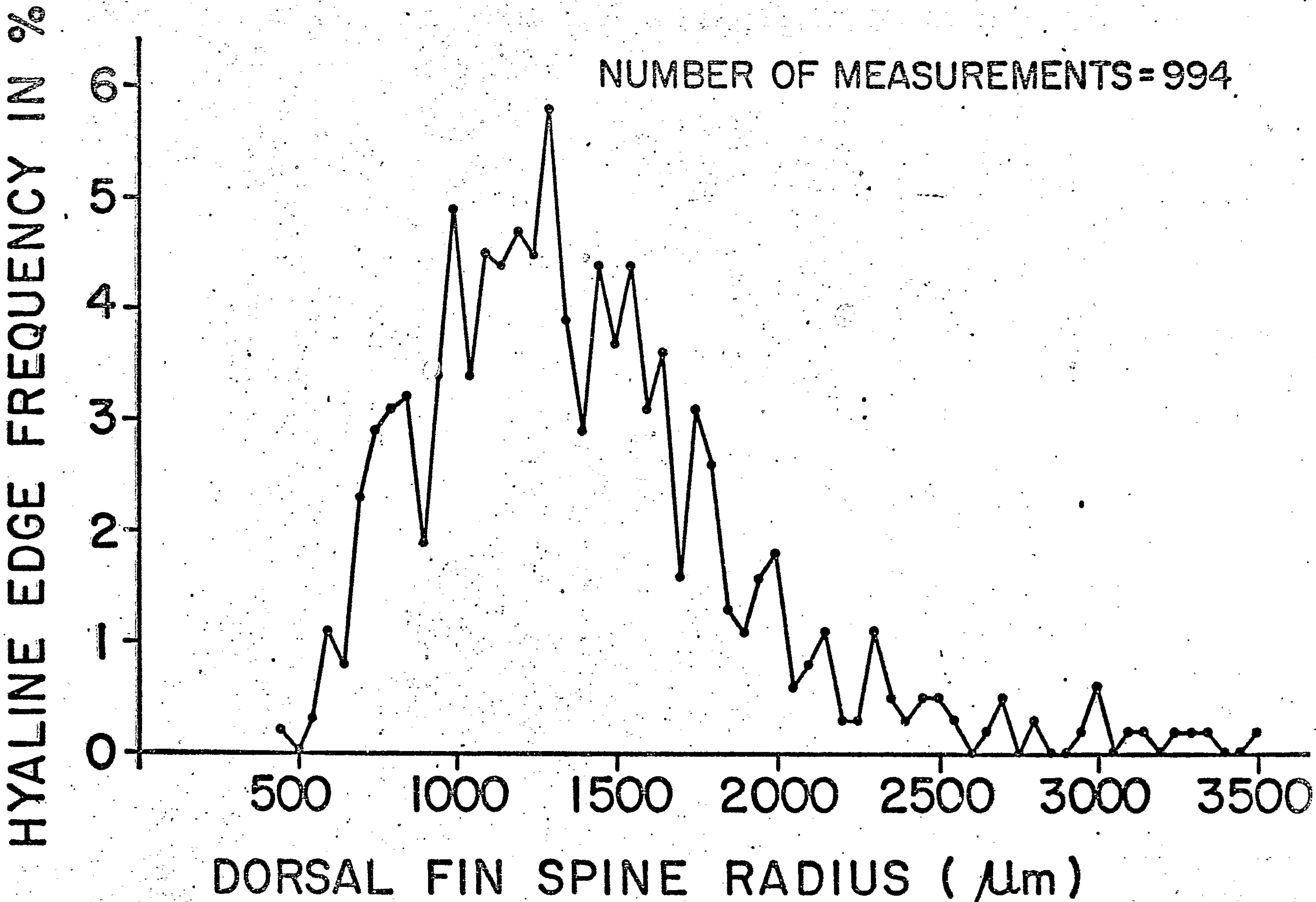


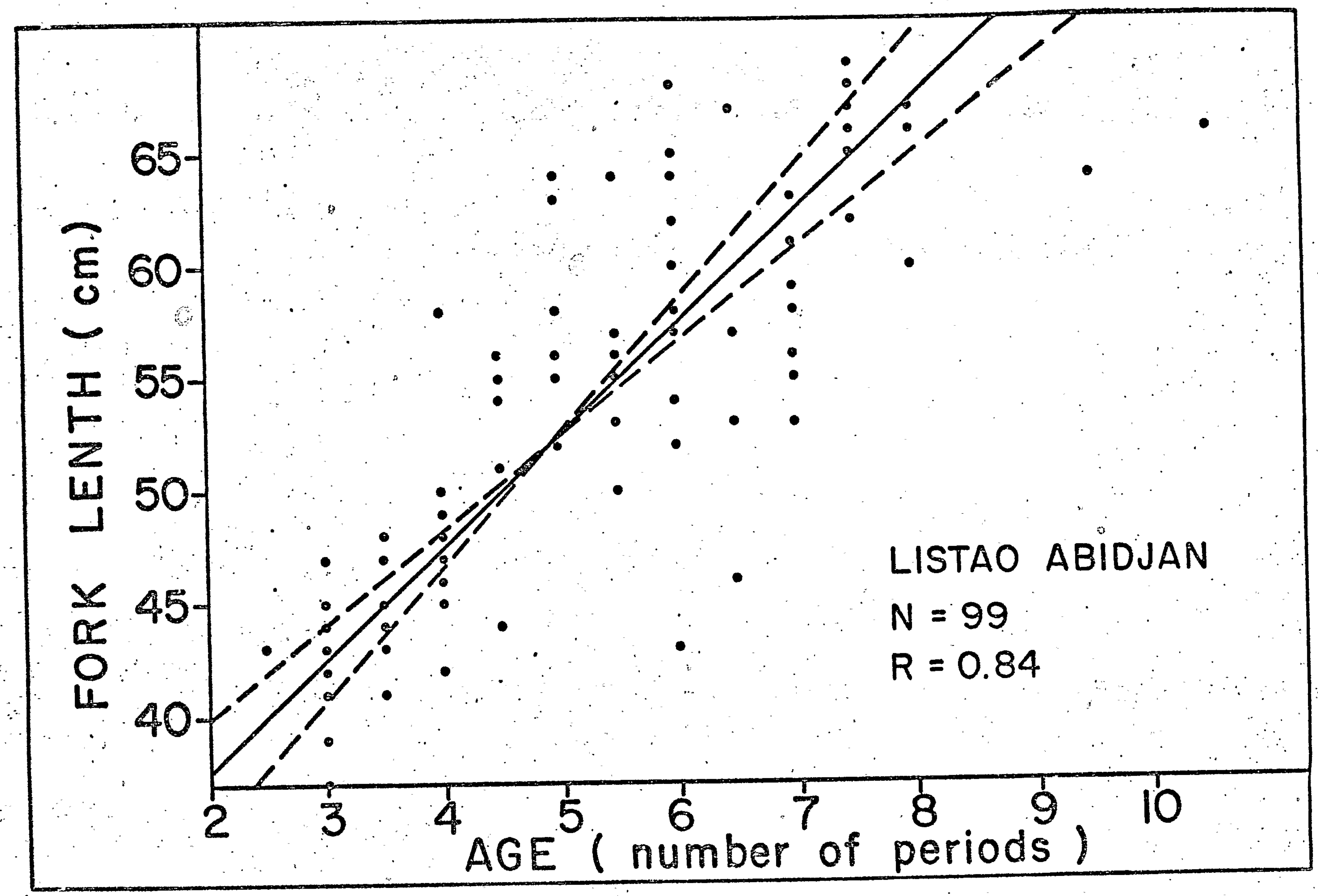
a1
d1
d
a2

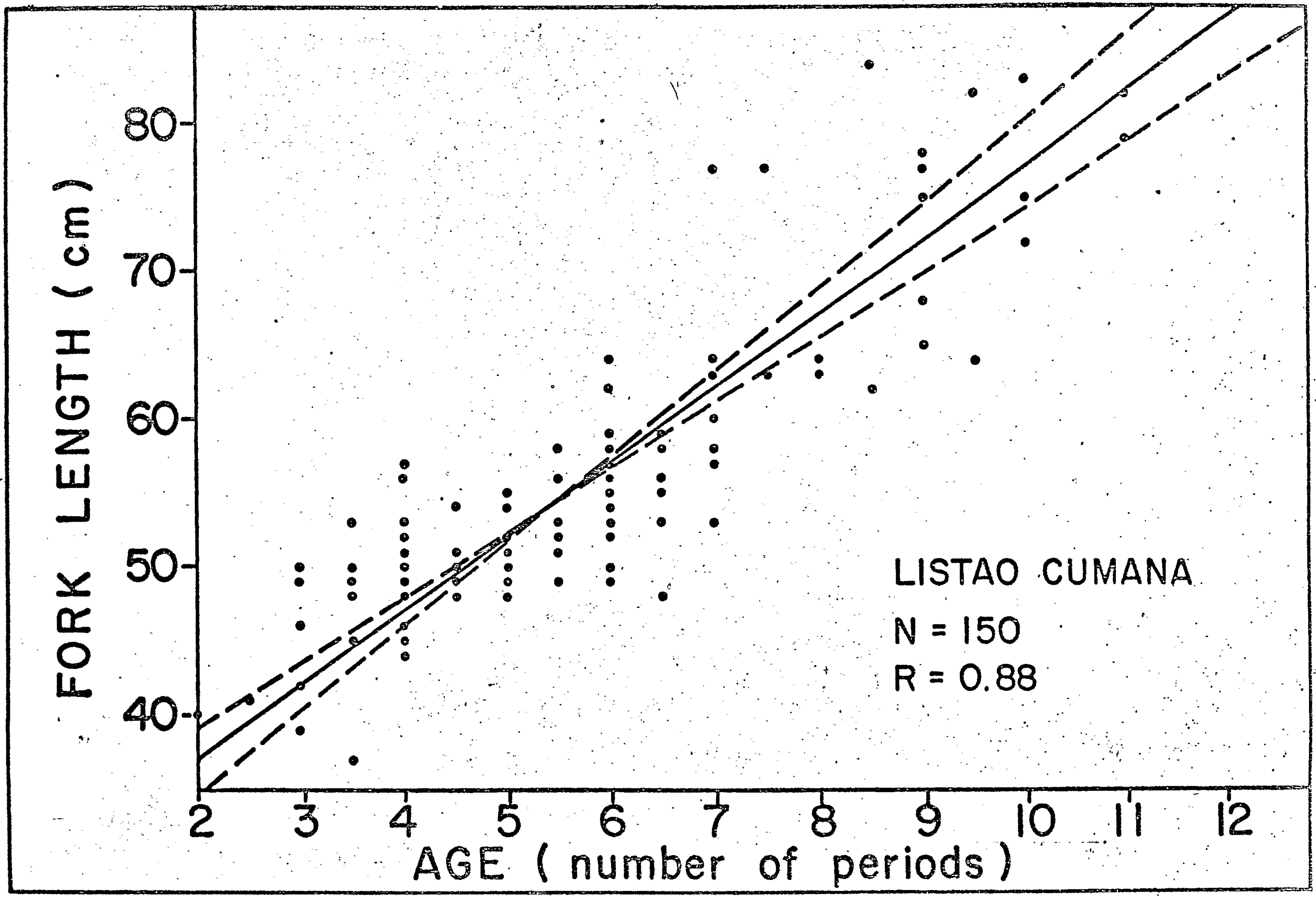

FORK LENGTH (cm)
70
60
50
40
30
20
10
1
2
3
4
5
6
AGE (number of periods)

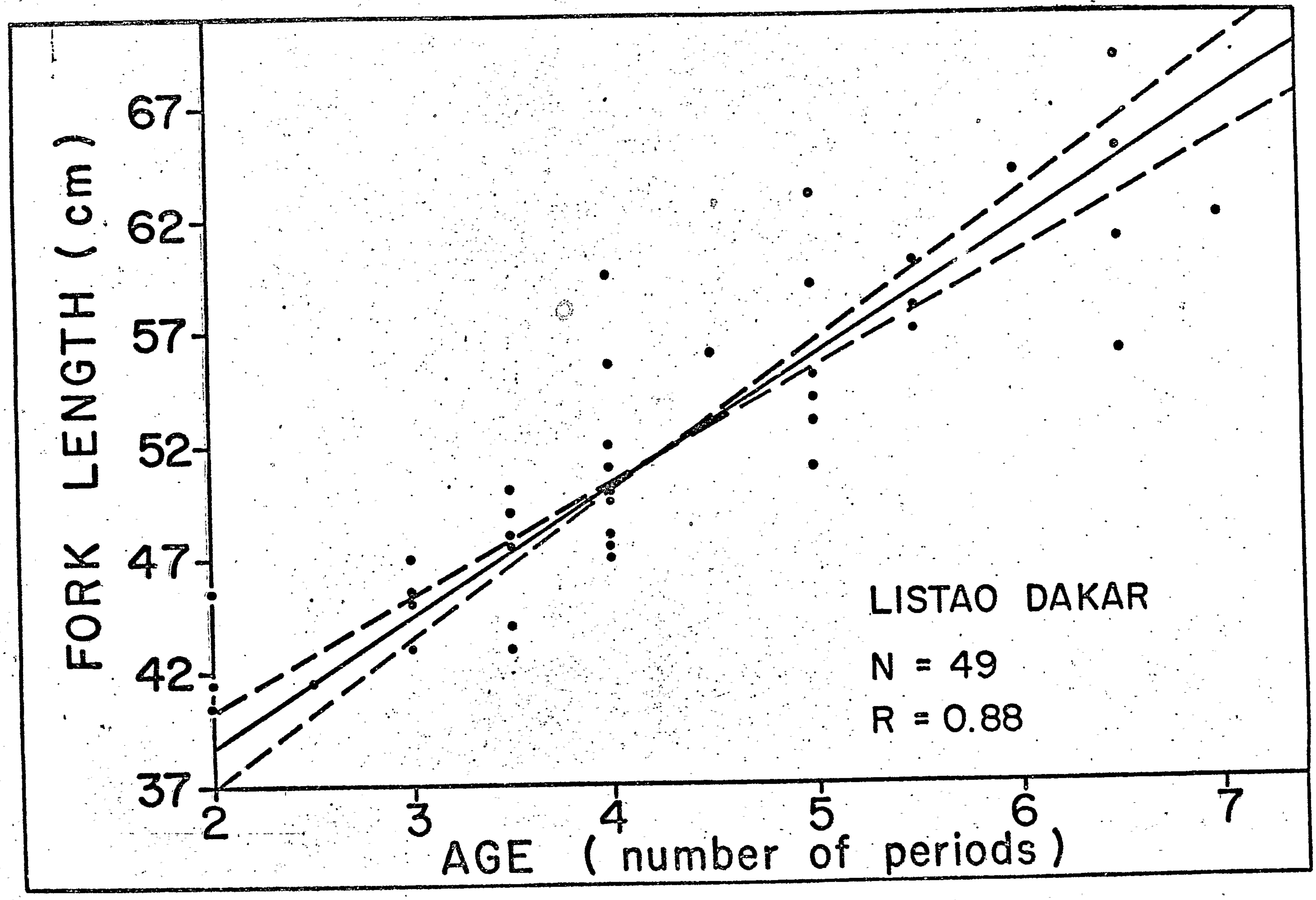

ELEVATIONS
40
35
30
25
3
5
7
9
11
SLOPES OF LINES
1
2
3
4
5
6
7
8


ABIDJAN
DAKAR
CUMANA
ELEVATIONS
-50
-100
-150
-200
7
8
9
10
11
SLOPES OF LINES

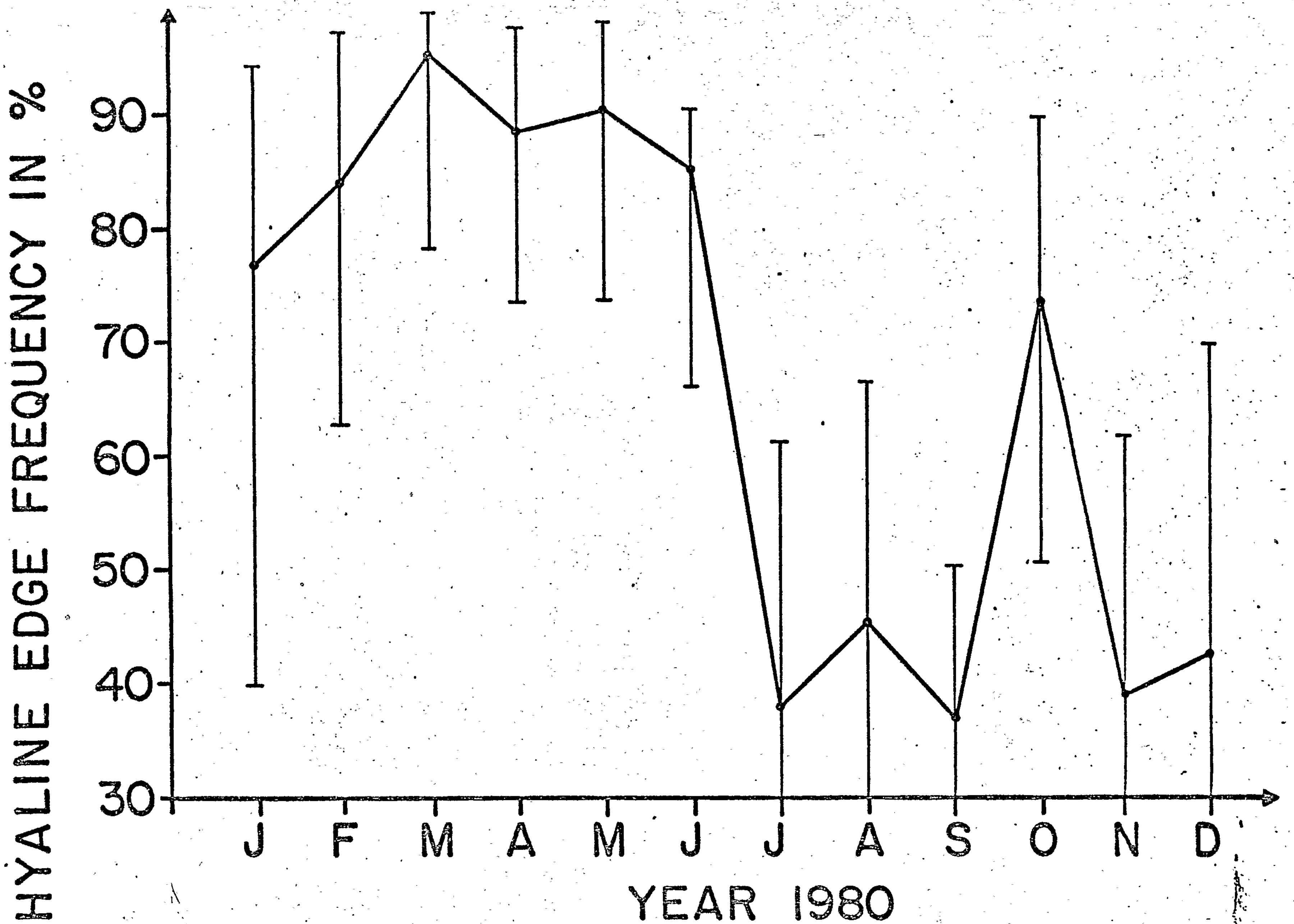

NUMBER OF MEASUREMENTS = 994
HYALINE EDGE FREQUENCY IN %
0
1
2
3
4
5
6
500
1000
1500
2000
2500
3000
3500
DORSAL FIN SPINE RADIUS (µm)


FORK LENTH (cm)
65
60
55
50
45
40
2
3
4
5
6
7
8
9
10
AGE (number of periods)
LISTAO ABIDJAN
N = 99
R = 0.84


FORK LENGTH (cm)
80
70
60
50
40
2
3
4
5
6
7
8
9
10
11
12
AGE (number of periods)
LISTAO CUMANA
N = 150
R = 0.88



FORK LENGTH (cm)
67
62
57
52
47
42
37
2
3
4
5
6
7
AGE (number of periods)
LISTAO DAKAR
N = 49
R = 0.88


HYALINE EDGE FREQUENCY IN %
90
80
70
60
50
40
30
J
F
M
A
M
J
J
A
S
O
N
D
YEAR 1980