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22 JAN. 1991

Nr. 5 - 1990



MANUAL FOR OPS
(Otolith Population Statistics)

ERLEND MOKSNESS

HAVFORSKNINGSINSTITUTTET
FORSKNINGSSTASJONEN FLØDEVIGEN
4817 HIS

ISSN 0800 - 7667

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MANUAL FOR OPS
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by

Erlend Moksness

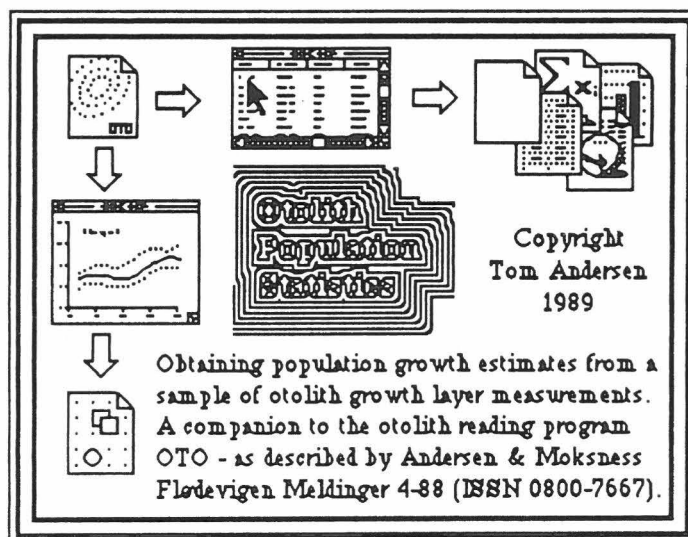


OPS

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Flødevigen Marine Research Station
4817 HIS, Norway.

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PREFACE

The initial programming, up to version 1.01, was done by Tom Andersen, Oslo. The final programming (version 2.01) has been done by Helge Nesteby (DigitalConsult, Oslo). Parts of the program are copyrighted by THINK Technologies, Inc. Macintosh is a trademark licensed to Apple Computers Inc., ImageWriter and LaserWriter are trademarks of Apple Computers Inc., SuperPaint is the trademark of Silicon Beach Software Inc., MacDraw is the trademark of Claris, and Microsoft Excel is a trademark of Microsoft Inc. Development of this program has partly been financed by the HELP-program (Institute of Marine Research, Bergen).

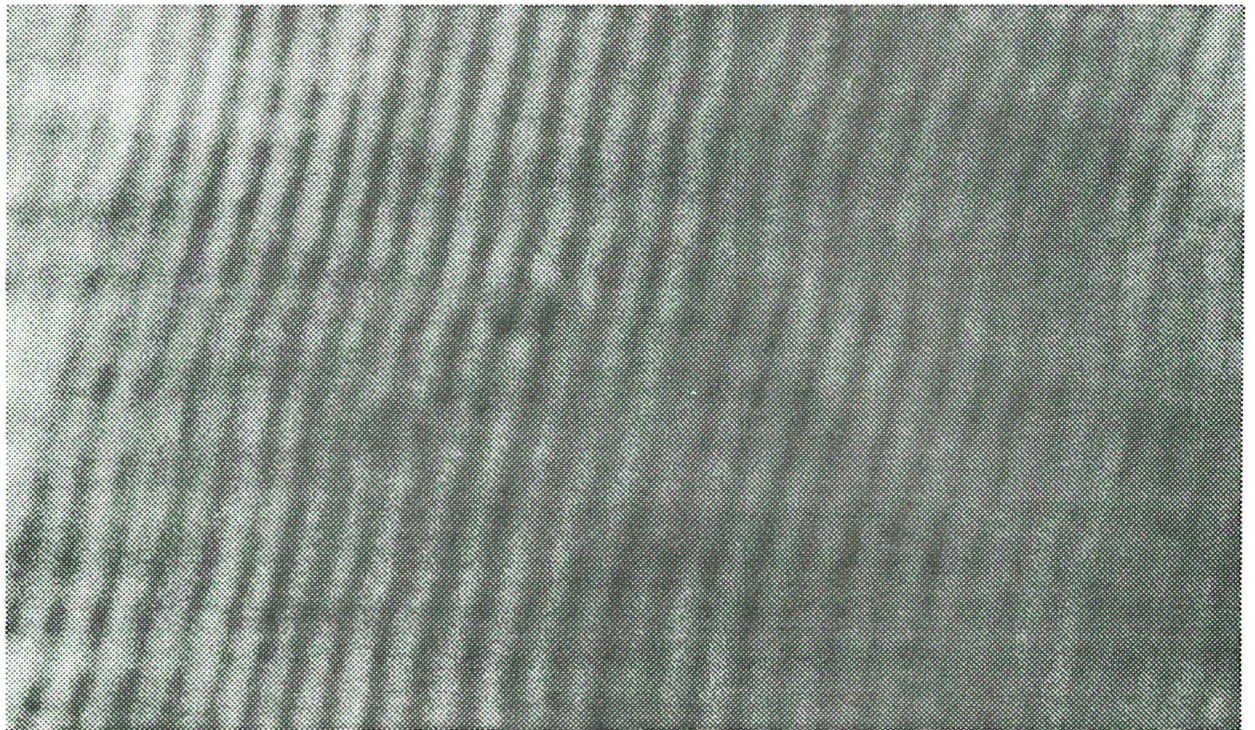


Figure 1. A herring otolith with daily increment, magnification 100 x.

INTRODUCTION

There is an increasing interest in reading the daily increments in otoliths of marine fish larvae and juveniles, for ageing and estimation of daily growth rate (Anon. 1989). As part of a tool to read the daily increments, the software programme OTO (Andersen and Moksness 1988) was developed in 1988 for the Apple Macintosh computers. The basic functions in the OTO program are similar to the programme developed earlier for MS-DOS computers at SouthWest Fisheries Center, La Jolla, USA. The OTO programme can only be used to read manually the increments in otoliths by the use of an HEI Video digitizer and an Apple Macintosh computer, and to estimate the number of increments and the increment size in the otoliths from each individual fish. However, there are no features in the OTO-programme to calculate the average number of increments and the average increment size in the otoliths from more than one fish. To do such calculations, the data have to be transferred to spreadsheets like Microsoft Excel for processing. However, handling of data in such a way is time consuming. The programme OPS (Otolith population statistics) has been developed to reduce the handling time of the data to a minimum. The OPS programme has been developed to read the data files created by the programme OTO and process these files. The OPS programme can only be used on an Apple Macintosh computer with a mathematical coprocessor (Macintosh SE/30 or Macintosh II-serie).

Equipment needed

The following software and hardware are necessary for running the programme:

The **OTO** programme, version 1.06 or later

Data files from the **OTO** programme.

The **OPS** programme (address the 68881 coprocessor), version 2.1 or later, together with:

Apple Macintosh SE/30 or Macintosh II

It is recommended to use System 6.05, Finder 6.1 and Imagewriter 2.7 or later versions of system files for the Apple Macintosh computers. Use of MultiFinder (System 6.05, Finder 6.1) will make the work easier.

Also recommended are Imagewriter II and/or LaserWriter

What the OPS program does

The OPS programme reads the OTO data files and calculate the average number of increments and increment size from selected OTO data files. The results can be produced as tables and charts, which can be saved as either Text- or PICT-files.

1. STARTING THE PROGRAMME

The logos of the programme OPS and the data-files created by this programme is shown in Fig. 2. The **OPS** programme is to be used on a Macintosh SE/30 or Macintosh II-serie. The **OPS** (version 2.1 or later) address the 68881 coprocessor which increases the speed of the calculation in the programme. The programme is started either by double-clicking the programme logo or the data file logo. The logos of the OTO programme and the data files created by the OTO programme are shown in Fig. 3.

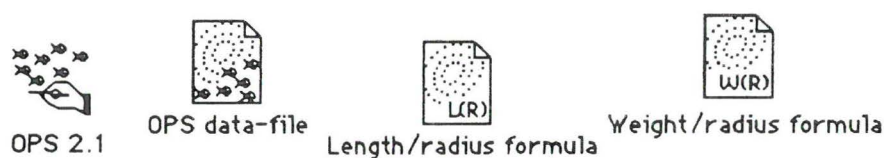


Fig. 2. The logos of the programme OPS and the data-files created by this programme. Length/radius formula and weight/radius formula are data files created by OPS and contain the length to otolith relationship and weight to otolith relationship respectively.

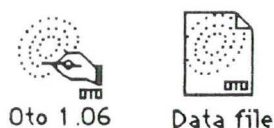


Fig. 3. The logos of the programme OTO and the data files created by this programme.

1.1. Creating a new "OPS" data file

1.1. Creating a new "OPS" data file

For opening a new data file, choose **New** from the **File** menu (Fig. 4). In the following window (Fig. 5) the user can either click on **Open all** and then click **Done** or **Open** for the selected OTO files that the user wants to have analyzed, and then click **Done**. While the programme processes the OTO data files, the computer leaves a window which indicates the processing of the OTO data files (Fig. 6). When all the OTO data files have been processed, the "**Processed files**" window automatically appears on the screen, showing all the OTO data files that have been processed.

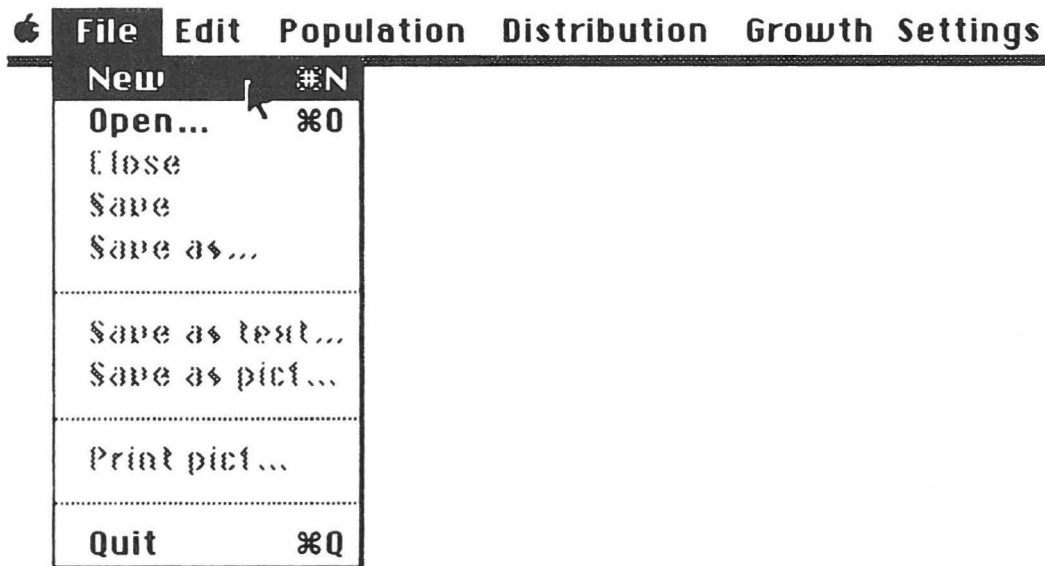


Fig. 4. Choosing **New** from the **File** menu.

File Edit Population Distribution Growth Settings

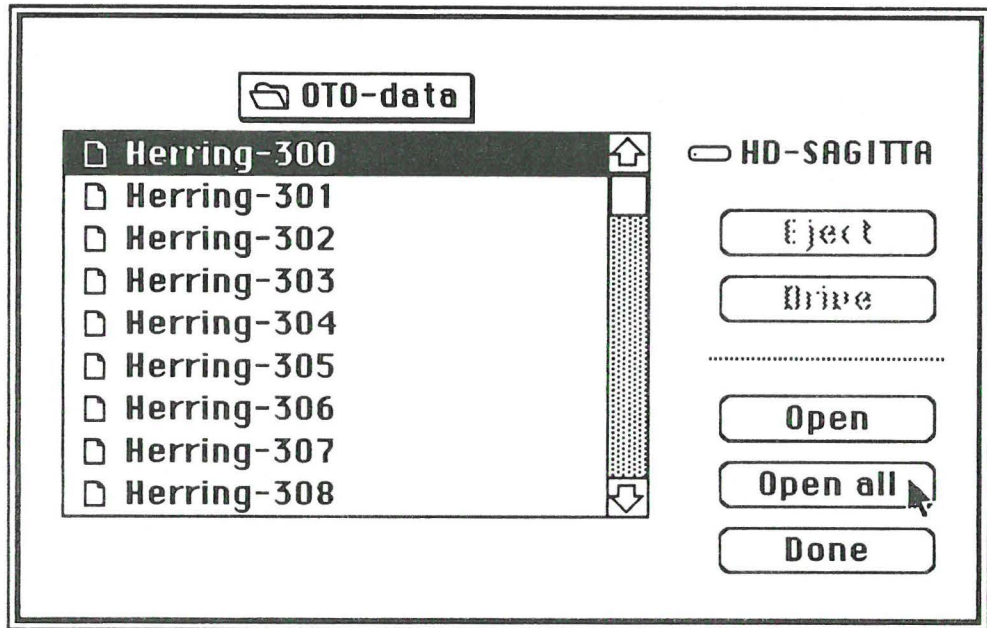


Fig. 5. The user can either click on **Open all** and the **Done** or click **Open** for the selected OTO files that the user wants to have analyzed, and the **Done**.

File Edit Population Distribution Growth Settings

Processed files				
Number	Experiment	Sample	Date	Species
300	Skagerrak-88	St.4	15/3/88	Herring
301	Skagerrak-88	St.4	15/3/88	Herring
302	Skagerrak-88	St.4	15/3/88	Herring



PROGRESS	
Processing file Herring-301	9 files
Combining transects	189.6 K

Fig. 6. The "Progress" Window which is shown while the OPS-programme processes the OTO-files.

Processed files					
Number	Experiment	Sample	Date	Species	
300	Skagerrak-88	St.4	15/3/88	Herring	↑
301	Skagerrak-88	St.4	15/3/88	Herring	□
302	Skagerrak-88	St.4	15/3/88	Herring	▒
303	Skagerrak-88	St.4	15/3/88	Sild	▒
304	Skagerrak-88	St.4	15/3/88	Herring	▒
305	Skagerrak-88	St.5	15/3/88	Herring	▒
306	Skagerrak-88	St.5	15/3/88	Herring	↓

Fig. 7. When the processing of OTO files is finished, the new window is usually left as shown in this figure.

1.2. Open an existing "OPS" data file

To open an existing data file, chose **OPEN** from the **FILE** menu (Fig. 8), and the desired data file is shown on the screen (Fig. 9).

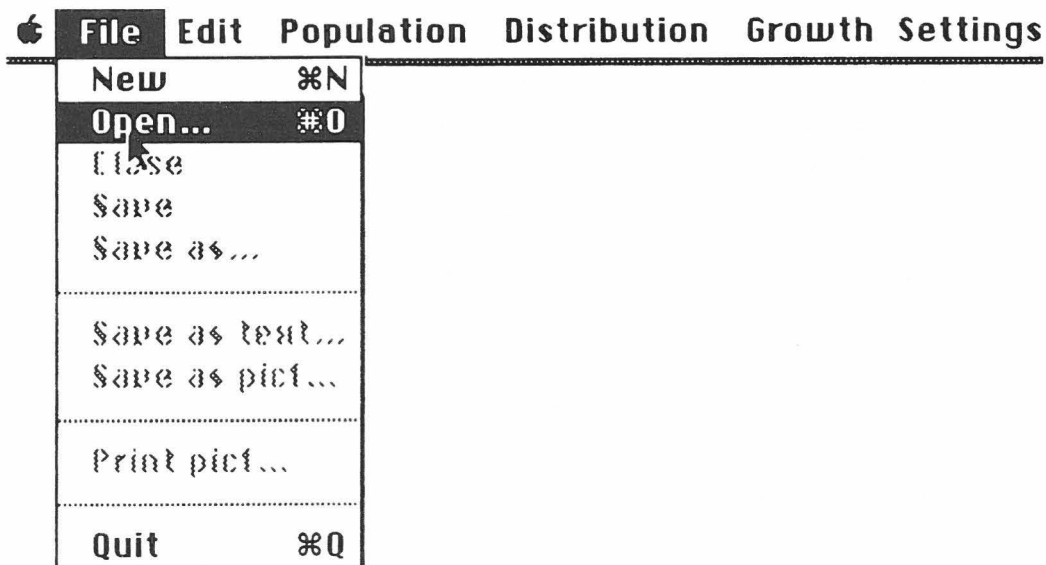


Fig.8. Choosing **Open** to fetch existing data file.

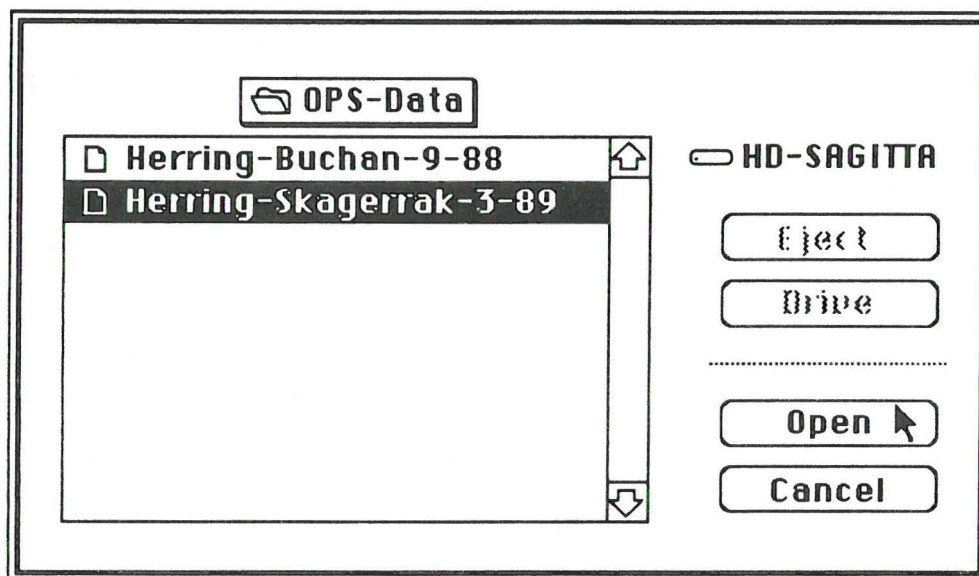


Fig. 9. The contents of a window after opening existing data file.

1.3. Add OTO-files to an existing "OPS" data file

If the user wants to add one or more OTO files to the one that is already processed by the OPS programme, he/she can use **Select population** from the **Population** menu (Fig. 10). A new window will appear on the screen (Fig. 11) from which the user can select the OTO files he/she would like to add to the already processed OTO files. The proceeding windows are the same as in Fig. 6 and 7.

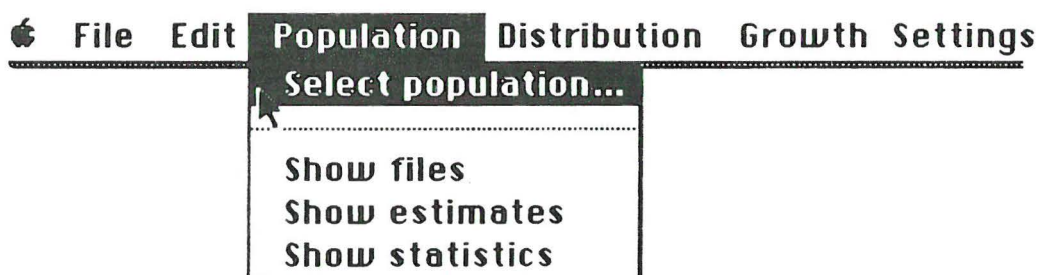


Fig. 10. The **Select population** option in the **Population** menu when more OTO files are to be added to already processed OTO files.

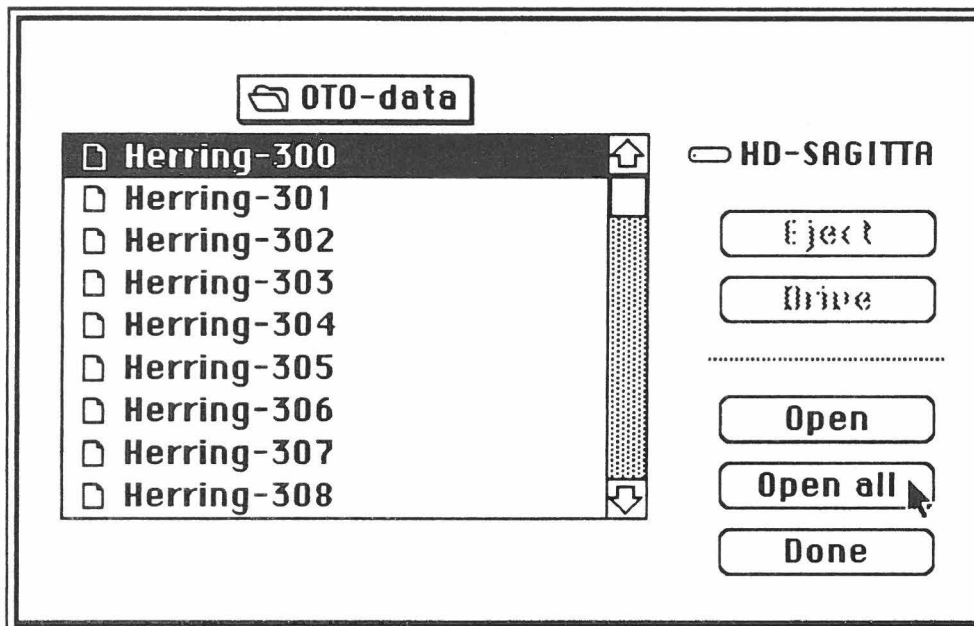


Fig. 11. Choosing the **OTO-datafiles**.

1.4. Saving data

When you have processed OTO data files (data files created by the OTO programme), choose **Save as** from the **File menu** (Fig. 12) to save the processed OTO data files as a OPS data file. A window appears as shown in Fig. 13 with a default name given. In the example given in Fig. 13 the default name has been changed.

After minor changes have been made to the data, choose **Save** from the **File menu**, since the file has already been established. If you leave the programme by **Close** or **Quit** (Fig. 14 and 15) and data are not saved, a window will appear (Fig. 16) asking if the changes made are to be saved. If you have made changes and want to save them, click **Yes**.

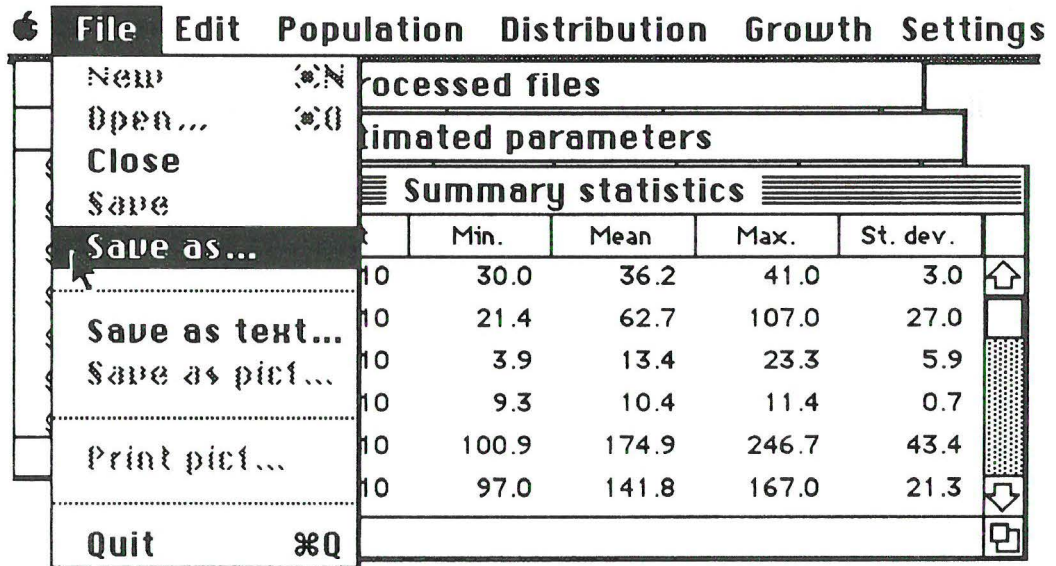


Fig. 12. Choosing **Save as** for saving data the first time.

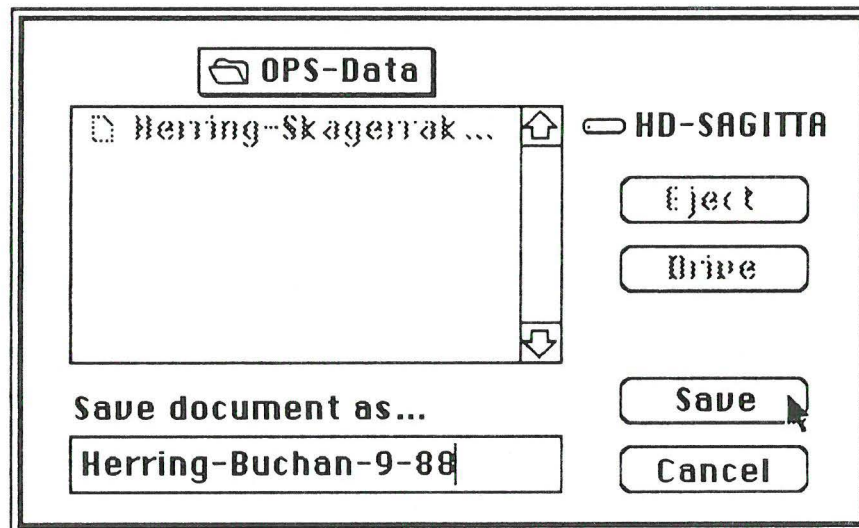


Fig. 13. Window after choosing **Save as**. The user can choose any name for the data file he/she wishes.

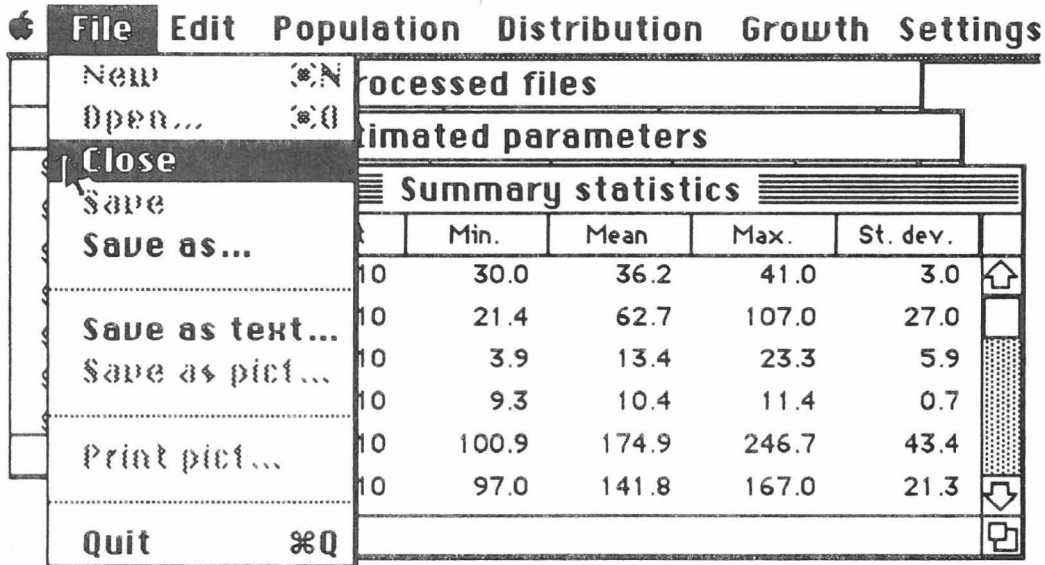


Fig. 14. Choosing **C**lose from the **F**ile menu.

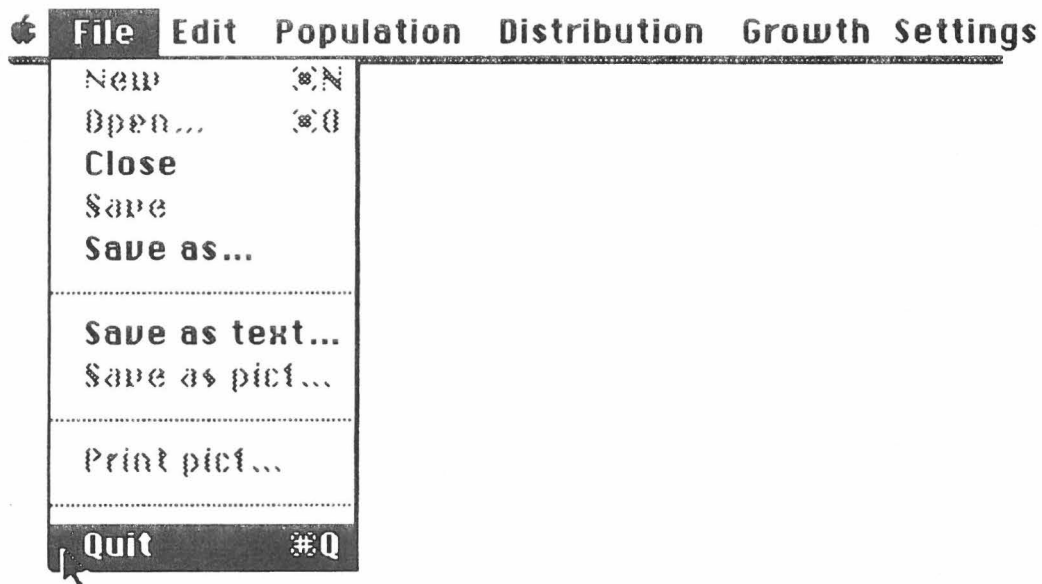


Fig. 15. Chosing **Q**uit from the **F**ile menu.

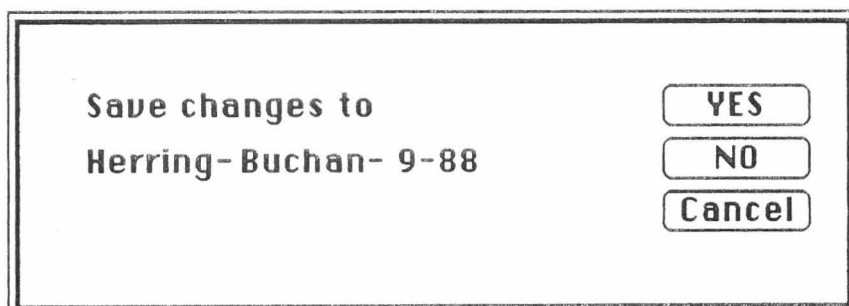


Fig. 16. Window after choosing **Q**uit or **C**lose.

2. STATISTICS AND DISTRIBUTIONS

The OPS programme will automatically show the OTO files that have been processed in the "**Processed files**" window. This window can also be called up by choosing "**Show files**" from the "**Population**" menu. In addition, the length, wet weight, dry weight, the measured distance from the centre of the otolith to the first increment observed (**Focus**), the measured distance from the nucleus to the margin of the otolith (**Radius**) and the number of increment estimated (**# Inc**), will all be shown in the "**Estimated parameters**" window. This window will appear after choosing "**Show estimates**" from the "**Population**" menu. A "**Summary statistics**" window gives you the basic statistics of length, wet weight, dry weight, Focus, Radius and # Inc and the window are chosen from the "**Population**" menu as "**Show statistics**". The three windows are shown in Fig. 17. All three windows can be saved as text files by choosing "**Save as Text...**" from the File menu. None of the windows can be copied to the scrapbook or to another programme.

File Edit Population Distribution Growth Settings							
Processed files							
Number	Experiment	Sample	Date	Species			
300	Skagerrak-88	St.4	15/3/88	Herring			
301	Skagerrak-88	St.4	15/3/88	Herring			
302	Skagerrak-88	St.4	15/3/88	Herring			
303	Estimated parameters						
304	Number	Length	Wet wt.	Dry wt.	Focus	Radius	# Inc.
305	300	41.0	107.0	23.30	10.2	246.7	167
306	301	37.0	61.5	13.32	10.5	170.9	153
	302	36.5	77.6	16.39	11.4	192.6	143
303	Summary statistics						
304	Param.	Count	Min.	Mean	Max.	St. dev.	
305	Length	10	30.0	36.2	41.0	3.0	
306	Wet wt.	10	21.4	62.7	107.0	27.0	
	Dry wt.	10	3.9	13.4	23.3	5.9	
	Focus	10	9.3	10.4	11.4	0.7	
	Radius	10	100.9	174.9	246.7	43.4	
	# Inc.	10	97.0	141.8	167.0	21.3	

Fig. 17. The three windows shown by the **Population** menu.

All the parameters in the "**Summary statistics**" window can be produced as graphs. This is done by choosing either of them from the "**Distribution**" menu (Fig. 18 and 19). The scale of the x- and y-axis can be changed by double clicking on one of the two axis (Fig. 23).

A special case is when creating a graph of the hatch distribution (Fig. 20). If the date-format used is not equal to the default date-format in the OPS programme, a window will appear to get the user to define the used date-format (Fig. 21). If, somehow, something is wrong, the programme will inform the user that the chart could not be made (Fig. 22).

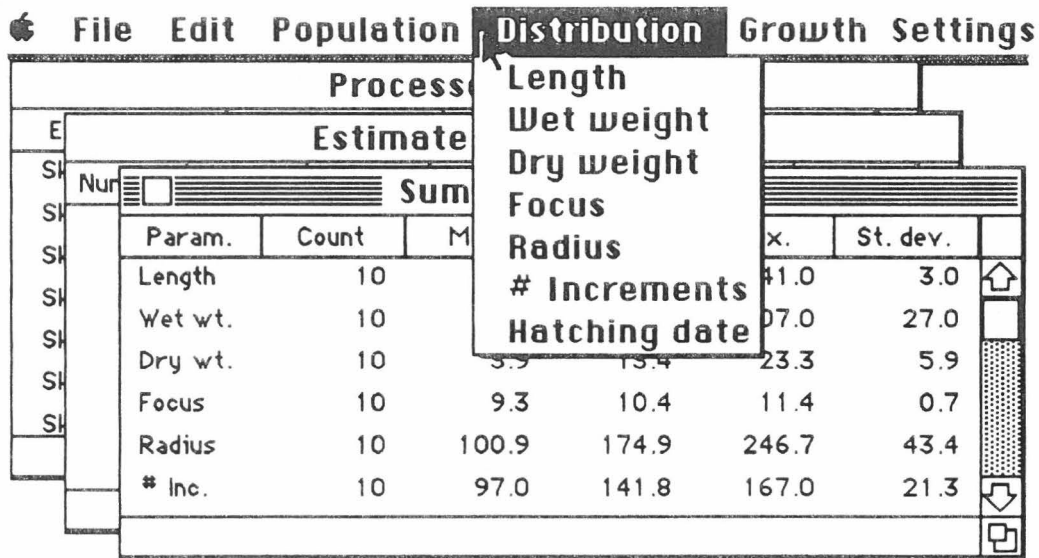


Fig. 18. Choosing charts from the **Distribution** menu.

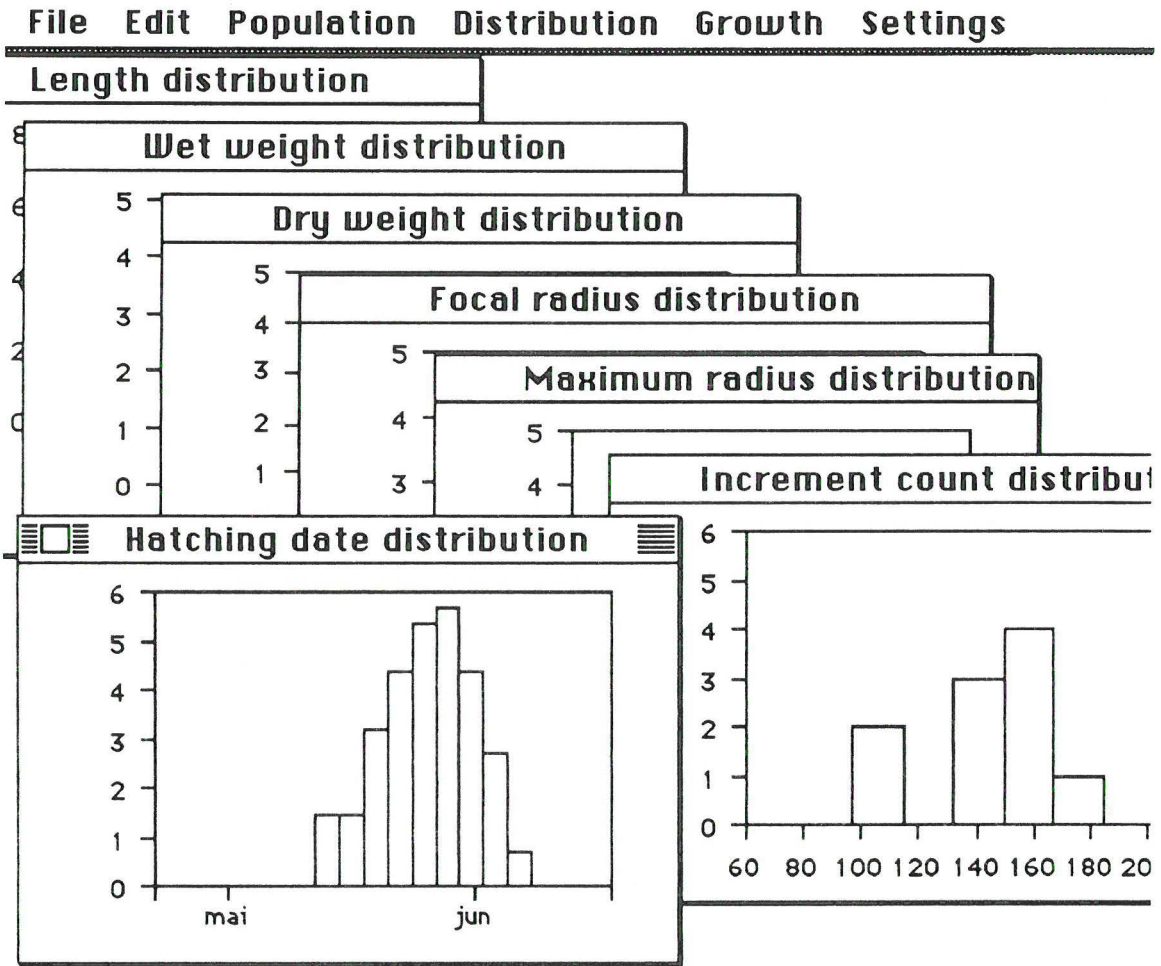


Fig. 19. The seven charts created by the **Distribution** window.

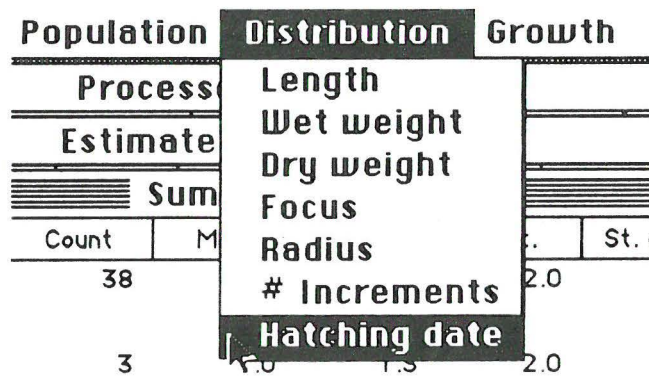


Fig. 20. Choosing **Hatching date** from the **Distribution** menu.

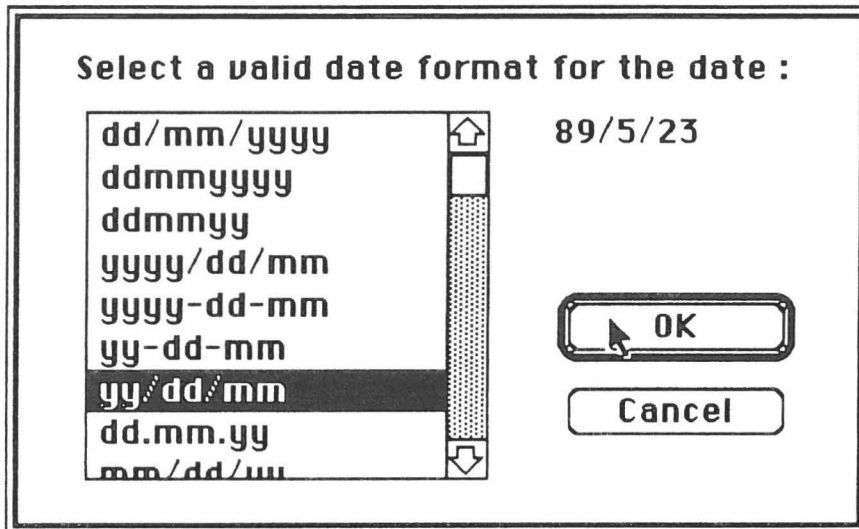


Figure 21. Selecting date format.

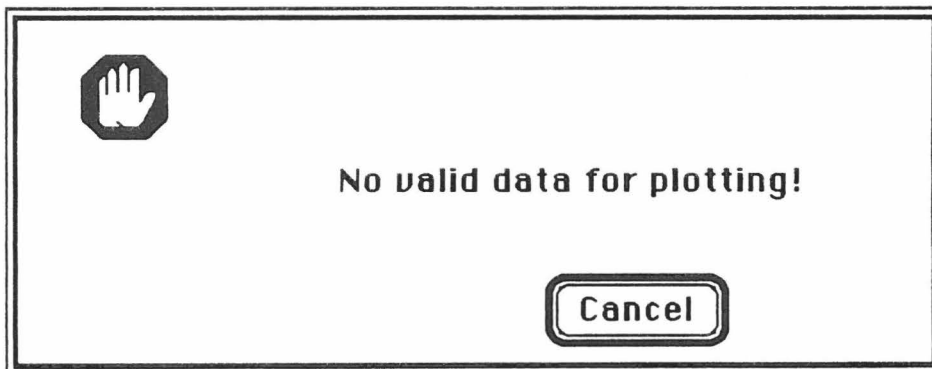


Fig. 22. Appearing window if the hatching chart can not be made.

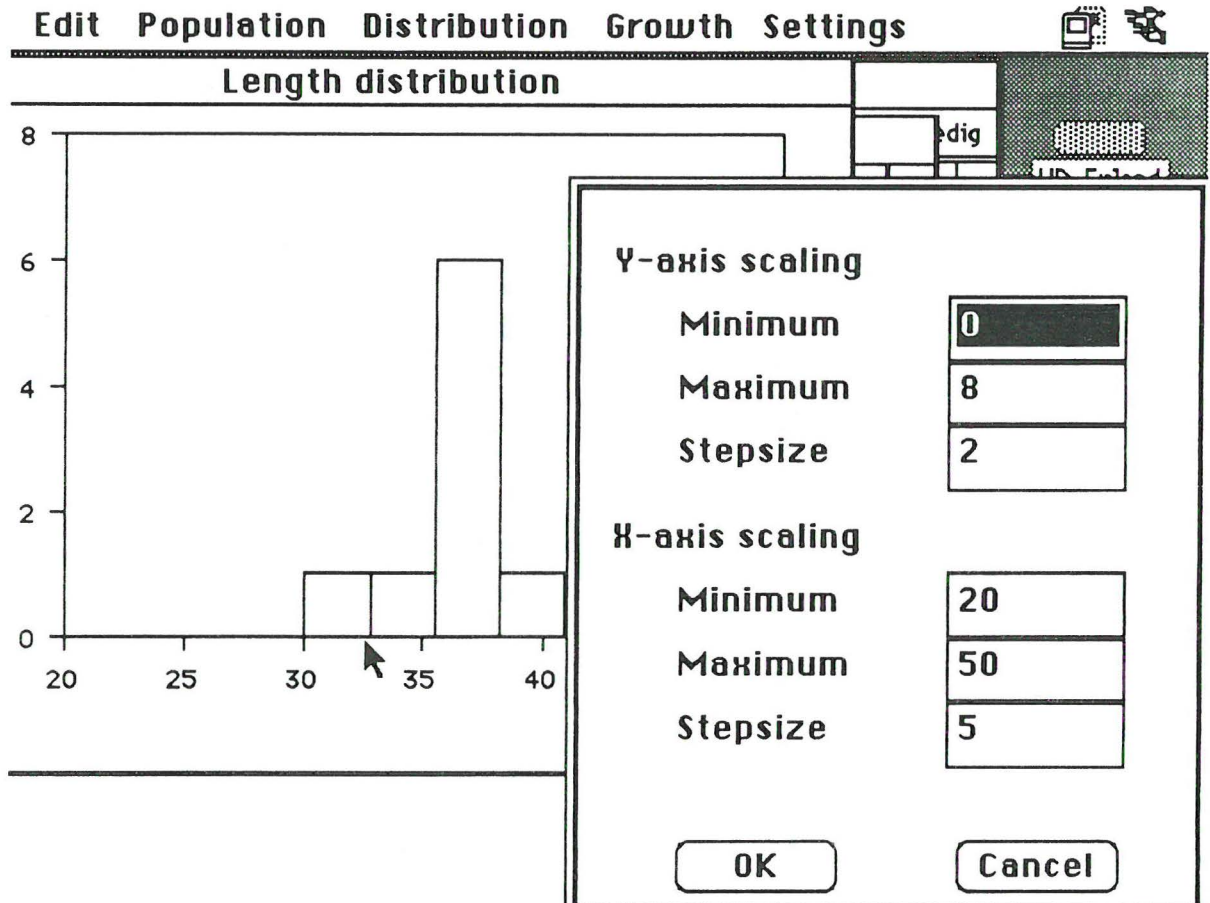


Fig. 23. To change the scale of the figure, double-click on one of the axes and a new window will appear in which the user can change the axes.

2.1 Saving and printing windows from Population and Distribution menu

The windows created by the **Population** menu can all be saved as **Text** files by choosing "**Save as text...**" from the file menu (Fig. 24). The OPS-programme will give the user a default name for the **Text** file, but this can be changed by the user (Fig. 25).

The graphs produced by the **Distribution** menu can either be saved as **Text**-files (Fig. 26 and 27) or **PICT** files (Fig. 29) by choosing "**Save as text...**" or "**Save as pict...**" from the File-menu. In addition the graphs can be printed on paper by choosing the "**Print pict...**" option in the **File** menu.

The Text files can be opened by a spreadsheet as MS Excel. In addition the Text file can either be saved with a comma or a period. When using the US format configuration (selected from the Control Panel in the Apple menu) the Text file will be saved with a period and when using the Norwegian format configuration, the Text file is saved with a comma.

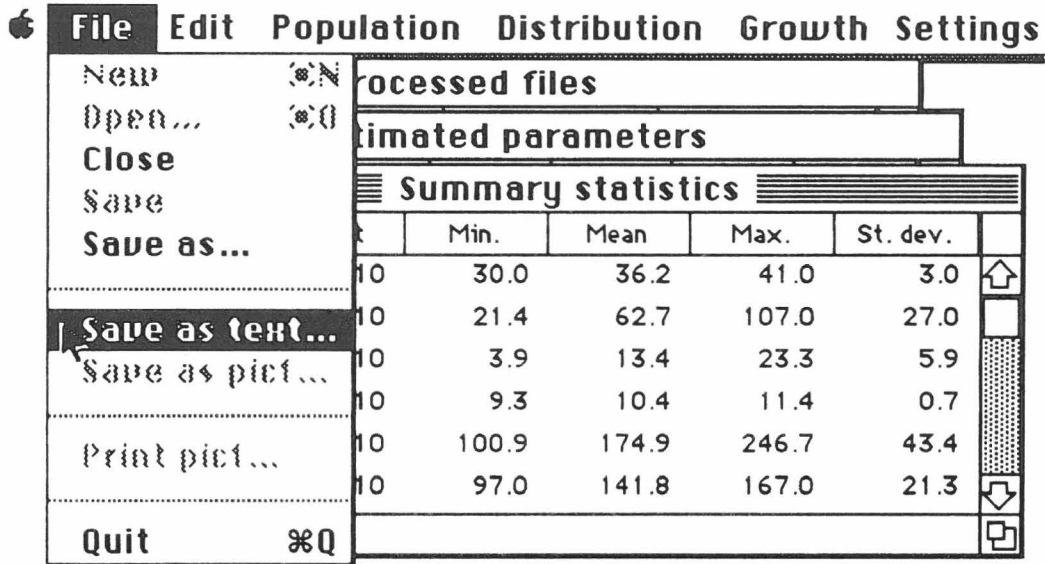


Fig. 24. The "Save as text..." option from the File menu.

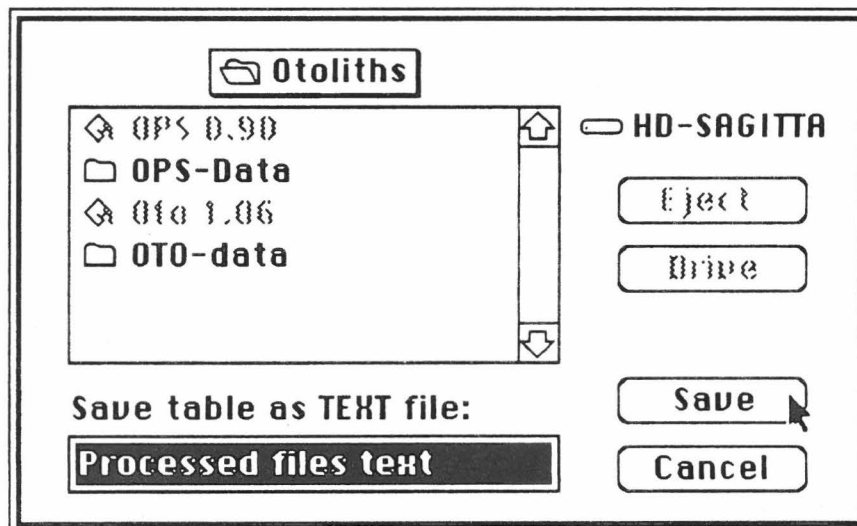


Fig. 25. The "Save as text..." window with a default data file name.

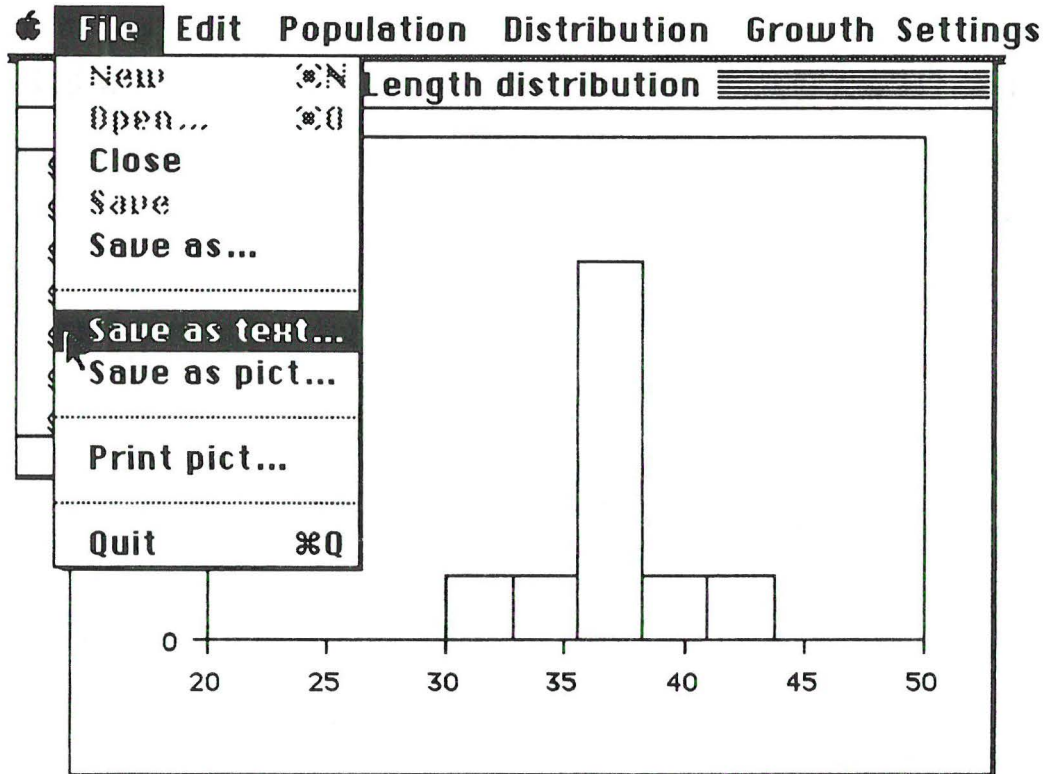


Fig. 26. The "Save as text..." option from the **File** menu.

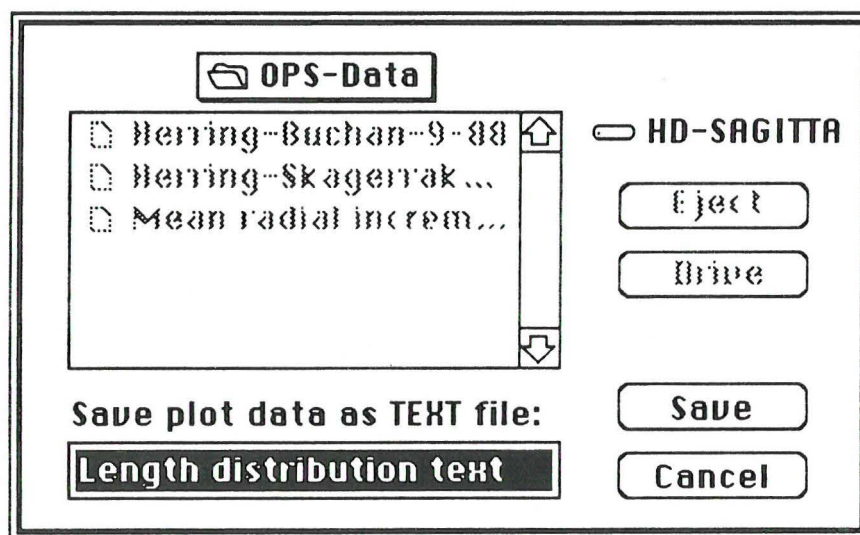


Fig. 27. The window to save a graph as Text file, with a default file name. This default file name can be changed.

File Edit Formula Format Data Options				
A1		x >		
Worksheet1				
Length distribution text				
	A	B	C	D
1	x >	x <	Count	
2	30.000000	32.750000	1	
3	32.750000	35.500000	1	
4	35.500000	38.250000	6	
5	38.250000	41.000000	1	
6	41.000000	43.750000	1	
7	43.750000	46.500000	0	
8	46.500000	49.250000	0	
9	49.250000	52.000000	0	
10	52.000000	54.750000	0	
11	54.750000	57.500000	0	
12	57.500000	60.250000	0	
13				

Fig. 28. The result of "Save as Text..." displayed as a MS Excel worksheet. If the user wants to change the period (.) to a comma (,), it is done by using the "Replace..." function in the "Formula" menu in MS Excel.

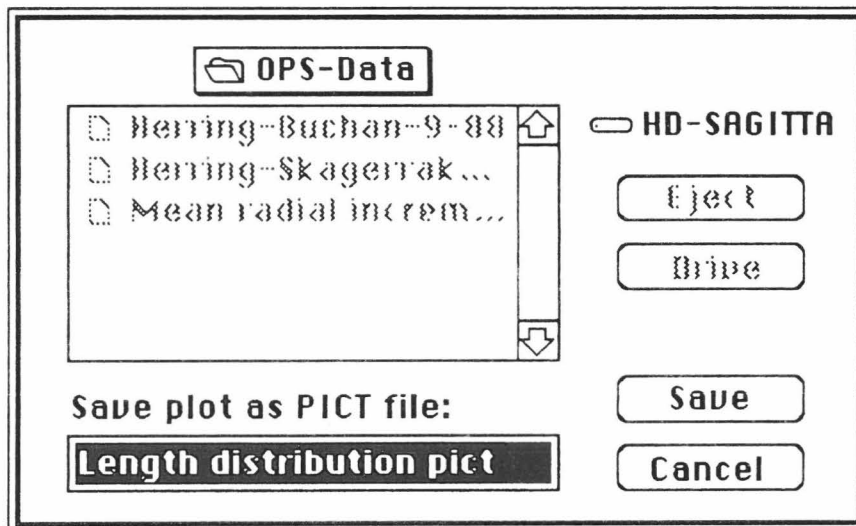


Fig. 29. The window to save a graph as PICT file, with a default file name. This default file name can be changed.

3. SETTINGS

3.1. Age offset

Firstly set the "**Age offset**". This means the number of days missing from the hatching till the first increments. For herring there has been observed in enclosure experiments that approximately 10 increments are missing compared to the actual age of the herring larvae. After choosing "**Age offset...**" from the **Settings** menu (Fig. 30), the user changes the default 0 in the next window (Fig. 31) to a value like 10 as in Fig. 32.

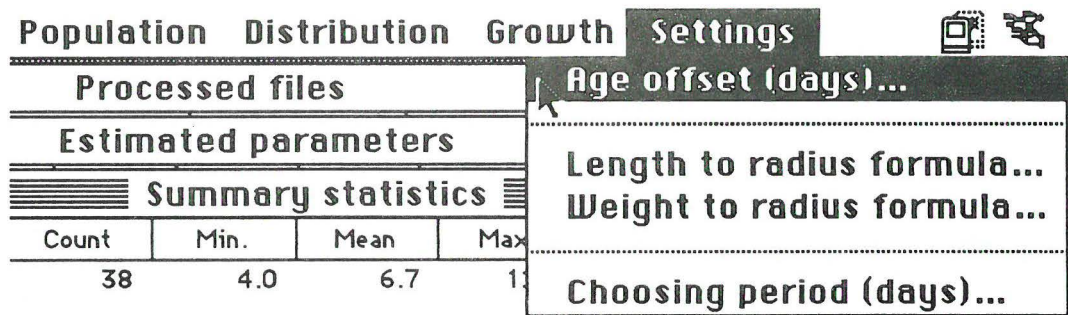


Fig. 30. Choosing "**Age offset...**" from the **Settings** menu

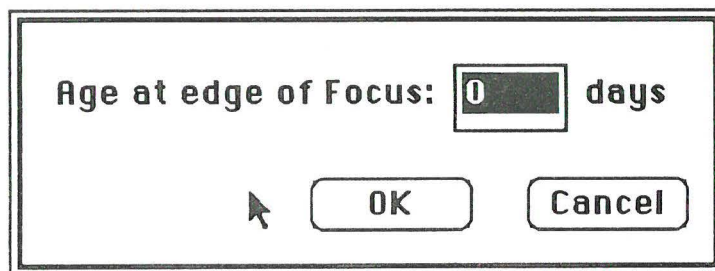


Fig. 31. The default values in the "**Age offset**" window.

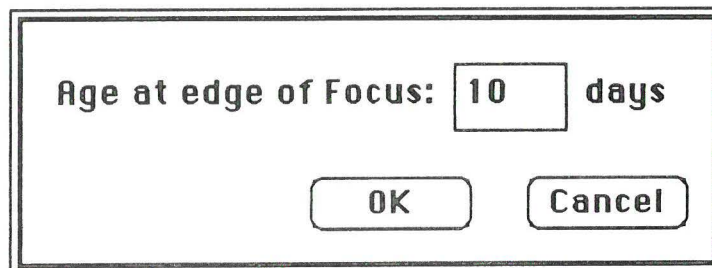


Fig. 32. Changing the default values in the "**Age offset**" window.

The user is now ready to estimate the average increment size of the selected OTO files and does so by choosing **Increment size (μm) at age** from the **Growth** menu (Fig. 39).

3.2. Fish size to otolith radius relationship

To go from increment size to daily growth rate in length or weight, the user needs to establish a relationship between the radius of the otolith and the length or the weight (wet or dry) of the fish species. This relationship can be entered by choosing "**Length to radius formula...**" (Fig. 33) or "**Weight to radius formula...**" (Fig. 35) from the **Settings** menu. The formula (relationship) is entered in window for length (Fig. 34) or for weight (Fig. 36). The formulas (regressions) entered can only use the same mathematical expressions as used in MS Excel. The symbol = is not allowed. Each formula can be saved as a unique data file (see Fig. 2 and Figs. 34 and 36). The graph showing the daily length increment and daily weight increment are chosen by selecting "**Length increment (mm/d) at age**" and "**Weight increment at age**" respectively from the **Growth** menu. The graphs showing the length and weight at age, are made by selecting "**Length (mm) at age**" and "**Weight at age**" from the **Growth** menu.

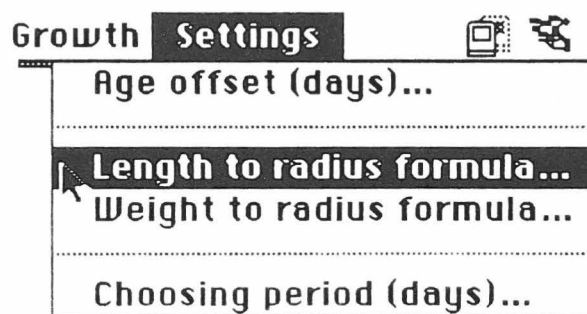


Fig. 33. Choosing "**Length to radius formula...**" from the **Settings** menu to enter a new relationship between the length of the fish and the radius of their otolith.

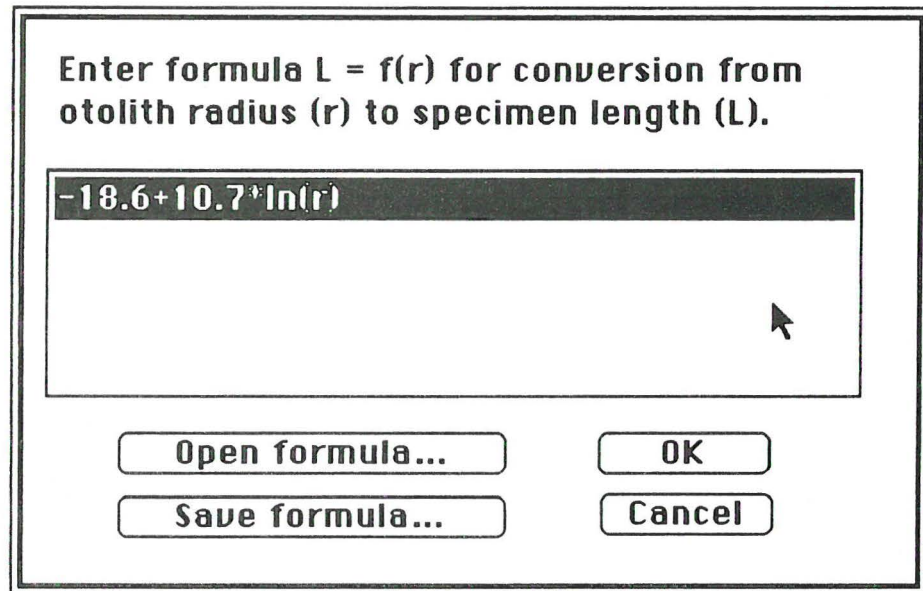


Fig. 34. Enter a new relationship between the length of the fish and the radius of their otolith. Open an existing formula by choosing "Open formula" or save a new formula by choosing "Save formula". A formula is accepted by choosing "OK".

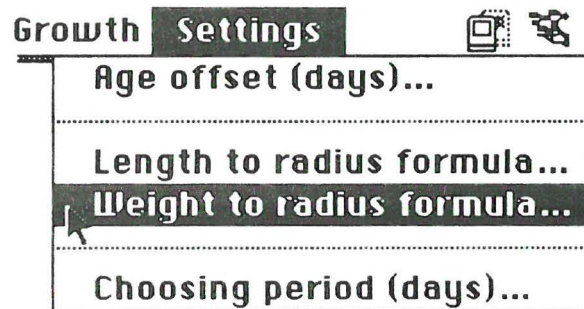


Fig. 35. Choosing "Weight to radius formula..." from the **Settings** menu to enter a new relationship between the length of the fish and the radius of their otolith.

Enter formula $W = f(r)$ for conversion from otolith radius (r) to specimen length (W).

0.090116+0.0030975*r+0.00094577*r²-0.00008876*r³+0.000000027399*r⁴

Open formula... OK

Save formula... Cancel

Fig. 36. Enter a new relationship between the weight (wet or dry) of the fish and the radius of their otolith.

3.3. Choosing period

All the measurements in the otoliths are done on a daily basis. However, there are situations where the user will want to compare growth between cohorts over a period of several days. This can be done in the programme by selecting the **choosing period** option in the **Settings** menu (Fig. 37). This gives the user the option to average increment size over whatever period of days he/she likes (Fig. 38).

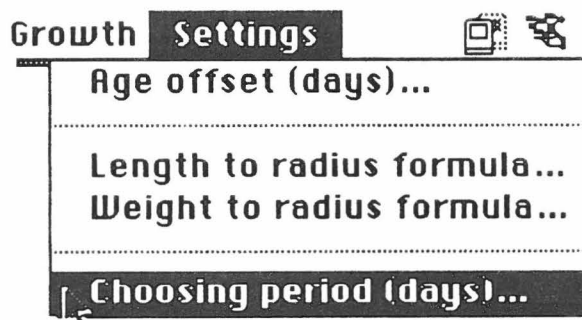


Fig. 37. Selecting the **choosing period** option in the **Settings** menu.



Fig. 38. Giving the number of days to average increment size data over. The default number is 1.

4. CALCULATION FROM THE TIME OF HATCHING OF THE FISH

4.1. Calculation of increment size

The calculation of average increment size (**Increment size (μm) at age**), daily length increments (**Length increment (mm/d) at age**) and daily weight increments (**Weight increment at age**; both wet and dry weight) are all processed from the **Growth** menu. However, when using this menu, the user should be aware that all calculations are done from day 0 or the first increment. The length (LI) and weight increments (WI) are done according to the formulae:

$LI = (L_2 - L_1)/(t_2 - t_1)$ and $WI = (W_2 - W_1)/(t_2 - t_1)$; where L_1 , L_2 , W_1 , W_2 are length and weight at day t_1 and t_2 .

The first to be done is to set the "**Age offset**". This means the number of days missing from the hatching till the first increments. This is done in the **Settings** menu. It is also recommended to set the length and weight to radius relationship.

The user is now ready to estimate the average increment size of the selected OTO files and does so by choosing **Increment size (μm) at age** from the **Growth** menu (Fig. 39). While the OPS-programme processes the graph a "Progress" window (Fig. 40) will appear on the screen till all data from all the OTO files have been processed. The resulting graph containing average values (thick line) and standard deviation (two thin

lines) will be shown in a new window (Fig. 41). The axes of the graph can be altered by double clicking on one of the axes and a new window will appear as shown in Fig. 42. The scale can be changed by changing the axis values in the window.

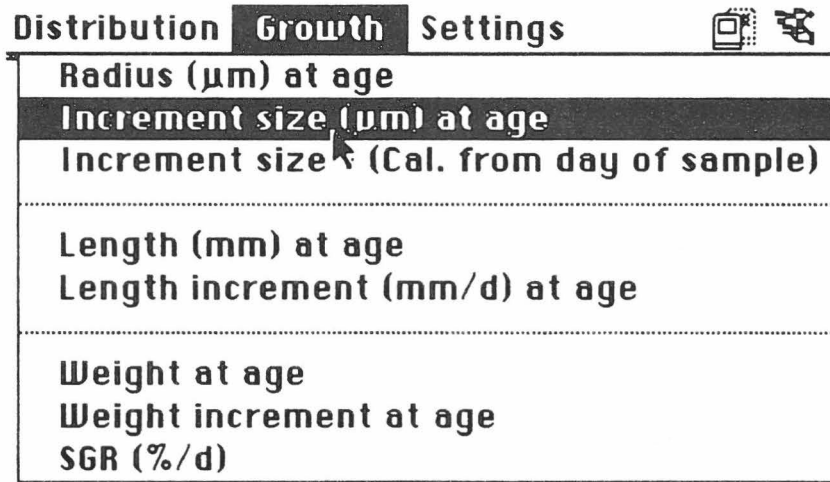


Fig. 39. Choosing the **Increment size (μm) at age** from the **Growth** menu to estimate the average increment size with standard deviation.

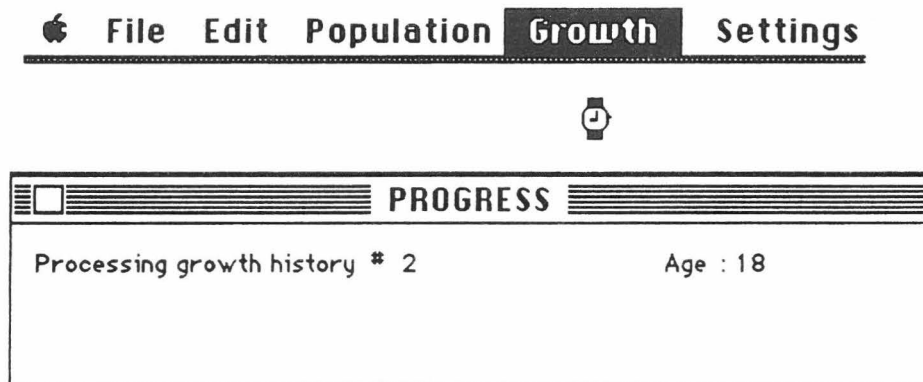


Fig. 40. The window appearing on the screen while the OPS programme processes the data.

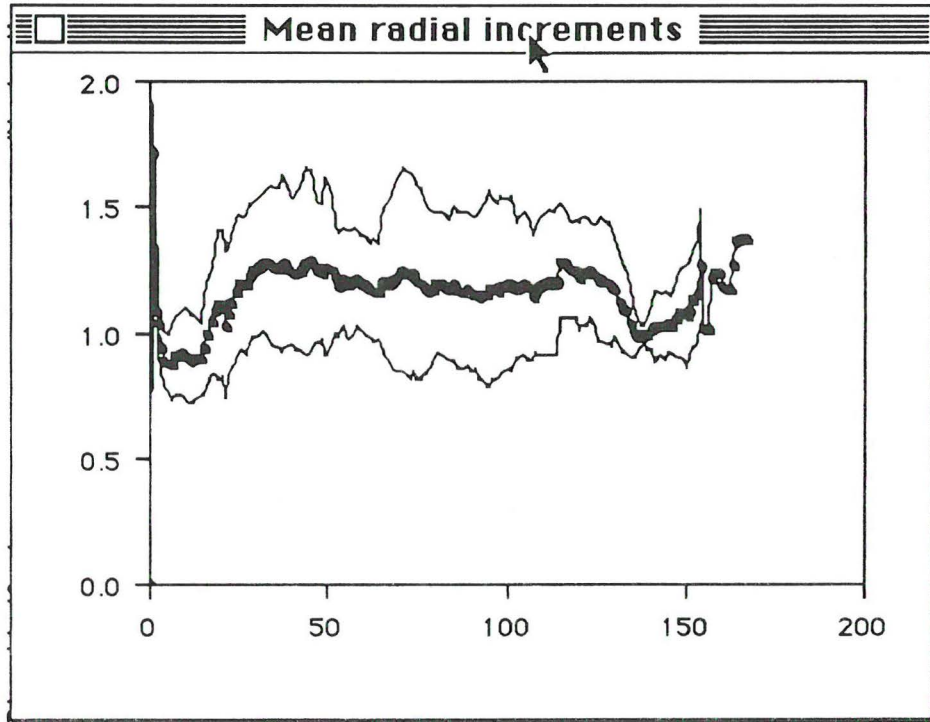


Fig. 41. An example of a graph produced by the "Increment size (μm) at age" option.

Population Distribution Growth Settings

Processed files

Mean radial increments

Y-axis scaling

Minimum

Maximum

Stepsize

X-axis scaling

Minimum

Maximum

Stepsize

Fig. 42. The scale of the axes can be altered by double clicking on one of the axes.

4.2. Calculation of daily growth rate in length and weight

To go from increment size to daily growth rate in length or weight, the user needs to establish a relationship between the radius of the otolith and the length or the weight (wet or dry) of the fish species. This relationship can be entered by choosing "**Length to radius formula**" (Fig. 33) or "**Weight to radius formula**" (Fig. 35) in the **Settings** menu.

The graph showing the daily length increment and daily weight increment is chosen by selecting "**Length increment (mm/d) at age**" and "**Weight increment at age**" respectively from the **Growth** menu. The graphs showing the length and weight at age, are made by selecting "**Length (mm) at age**" and "**Weight at age**" from the **Growth** menu.

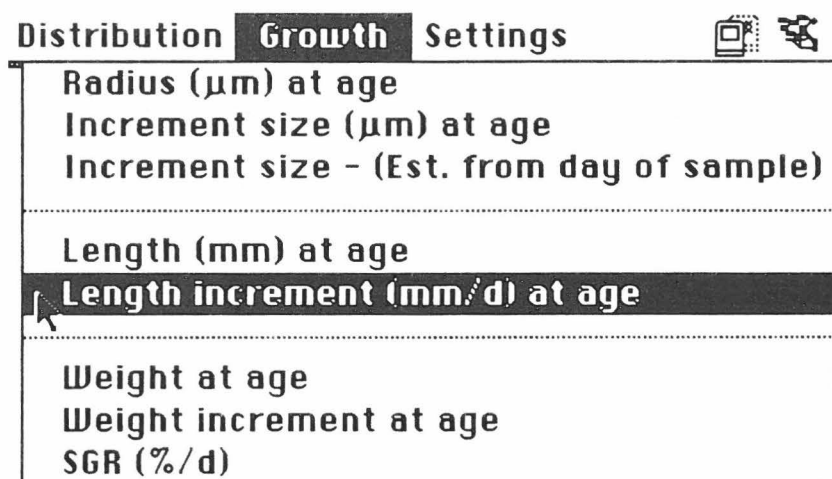


Fig. 43. Choosing "**Length increment (mm/d) at age**" from the **Growth** menu to have the average length increments of the sampled fish backcalculated.

4.3. Calculation of specific growth rate (%/d)

The backcalculation of specific growth rate (%/d) is based upon the relationship between the otolith radius and the weight of the fish being known. This relationship has to be entered by using the **Settings** menu (see ch. 3.2).

The specific growth rate, SGR, is calculated according to the formula (Houde and Schekter, 1981):

$$\text{SGR} = (\exp((\ln (Wt_2) - \ln (Wt_1)) / (t_2 - t_1)) - 1) * 100; \text{ where } Wt_1 \text{ and } Wt_2$$

are wet weights of fishes at days t_1 and t_2 .

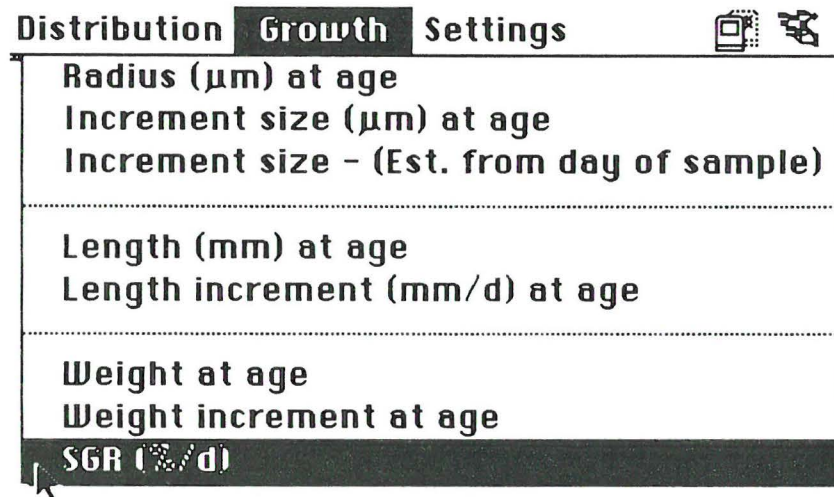


Fig. 44. Choosing "**SGR (%/d)**" from the **Growth** menu to have backcalculated the specific growth rate (%/d) of the sampled fish.

5. CALCULATION FROM THE TIME OF CAPTURE OF THE FISH

This option gives the user the possibility to calculate the average increment size from the moment of capture of the fish and back to the moment of hatching. The option is useful when the user wants to compare recent growth between cohorts.

5.1. Calculation of increment size

The average increment size from the moment of capture of the fish and back to the moment of hatching is calculated by choosing the **Increment size - (Cal. from day of sample)** option in the **growth** menu. By selecting the **Choosing period (days)** option in the **Settings** menu, the user will get calculated values over periods of more than one day.

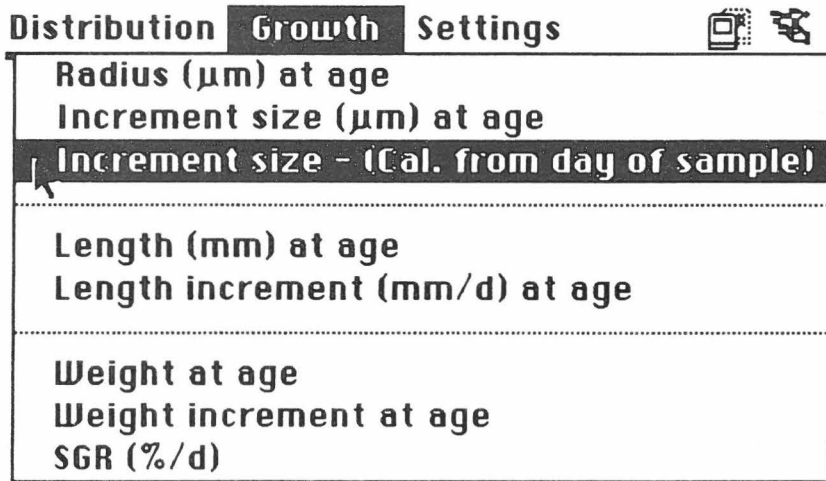


Fig. 45. Choosing "**Increment size - (Cal. from day of sample)**" from the **Growth** menu to have the specific growth rate (%/d) of the sampled fish backcalculated. The calculation is done from the moment of capture and back to the moment of hatching.

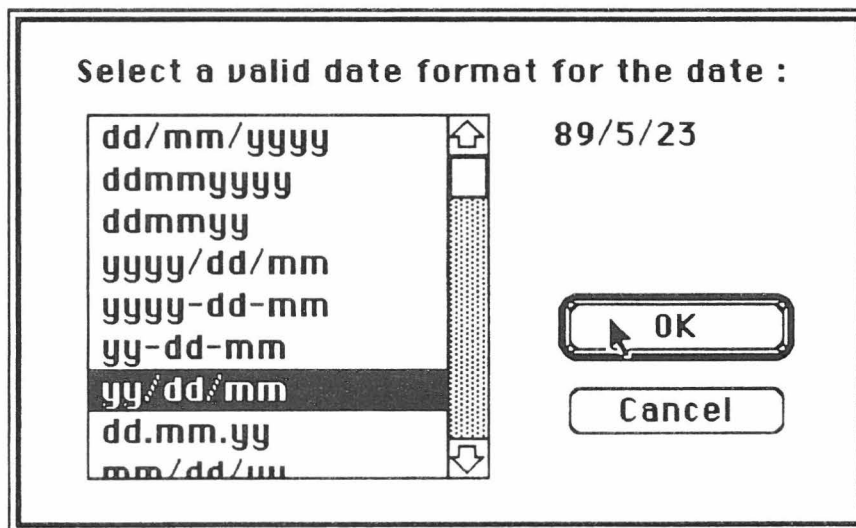


Fig. 46. Selecting date format.



Fig. 47. Appearing window if the hatching chart can not be made.

6. SAVE TABLES AND CHARTS

The graphs can either be saved as Text files or as PICT files. The "Save as text..." (Fig. 48) option saves the mean, standard deviation, number of count at age (days), sum and sums of squares. An example is shown in Fig. 49, with a text file open from MS Excel. The comma (,) can be changed to a period (.) by using the command **Replace** in MS Excel. By choosing **Save as pict...** the figure will be save as PICT-file, which can later be read by programmes such as MacDraw.

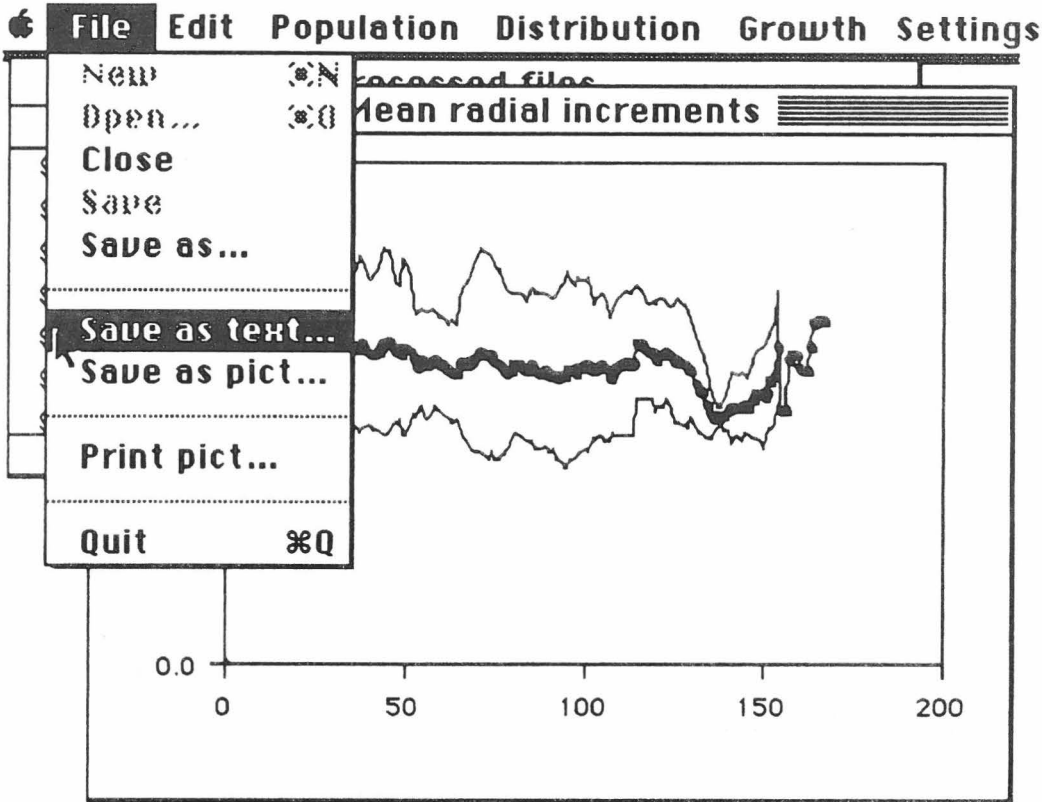


Fig. 48. The "Save as text..." option from the File menu.

File Edit Formula Format Data Options Macro Windo									
A1		Age							
Radial increments us. age text									
	A	B	C	D	E	F	G	H	
1	Age	Mean	St.dev.	Min.	Max.	Count	Sum	SumSqr	
2	0	0	0	0	0	0	0	0	
3	1	0,896	0,1057	0,727	1,0489	10	8,95614	8,121765	
4	2	0,877	0,0985	0,727	1,0165	10	8,77214	7,782377	
5	3	0,872	0,0976	0,727	1,0218	10	8,72202	7,693075	
6	4	0,871	0,1172	0,727	1,0345	10	8,70624	7,703424	
7	5	0,856	0,1114	0,7195	0,9962	10	8,56172	7,442056	
8	6	0,851	0,1232	0,7002	1,1173	10	8,50756	7,374527	
9	7	0,867	0,1534	0,6929	1,2423	10	8,6722	7,73254	
10	8	0,904	0,1634	0,691	1,1648	10	9,03643	8,406099	
11	9	0,925	0,1798	0,6588	1,1652	10	9,25451	8,855492	
12	10	0,929	0,1813	0,6584	1,1821	10	9,29159	8,929078	

Fig. 49. The result of "Save as Text..." displayed as an MS Excel worksheet. If the user wants to change the comma (,) to period (.), this is done by using the "Replace..." function in the "Format" menu in MS Excel.

Radial increments vs. time text									
	A	B	C	D	E	F	G	H	I
1	Age	Mean	St.dev.	Min.	Max.	Count	Sum	SumSqr	
2	0	0	0	0	0	0	0	0	
3	1	0,8213	0	0,821	0,82	1	0,82132	0,67456	
4	2	0,8213	0	0,821	0,82	1	0,82132	0,67456	
5	3	0,7952	0	0,795	0,8	1	0,79518	0,63231	
6	4	0,7705	0	0,771	0,77	1	0,77052	0,5937	
7	5	0,7697	0	0,77	0,77	1	0,76971	0,59246	
8	6	0,7693	0	0,769	0,77	1	0,76928	0,59179	
9	7	0,849	0	0,849	0,85	1	0,84901	0,72082	
10	8	0,9751	0	0,975	0,98	1	0,97509	0,9508	
11	9	0,9832	0	0,983	0,98	1	0,98318	0,96664	
12	10	0,9878	0	0,988	0,99	1	0,98781	0,97576	
13	11	0,9818	0	0,982	0,98	1	0,98178	0,96389	
14	12	0,9753	0	0,975	0,98	1	0,97527	0,95116	

Fig. 50. The result of "Save as Text..." displayed as an MS Excel worksheet. If the user wants to change the comma (,) to a period (.), this is done by using the "Replace..." function in "Format" menu in MS Excel.

6.1. Using comma or period when saving Text files

The user can choose between comma or period when exporting data as text-files. This is done by setting the Format configuration to US format when period is to be used and Norwegian format when a comma is to be used. If the user wants to change format when in the programme, the computer has to be rebooted to get the new format.

7. PRINT TABLES AND CHARTS

Only Graphs can be printed. This is done by choosing "Print pict..." from the **File** menu.

8. COPY AND PASTE TABLES AND CHARTS

The tables produced by the OPS programme cannot be copied to the Scrapbook or to another programme. It is recommended to save the Tables as Text files by using the "Save as text..." option in the **File** menu. The graphs can all be copied to the Scrapbook or to programmes like MS Word, MacDraw or SuperPaint. However, parts of the graphs can be lost when you use the copy function. It is therefore recommended to save the

graphs as PICT files for use in programmes like SuperPaint and MacDraw.

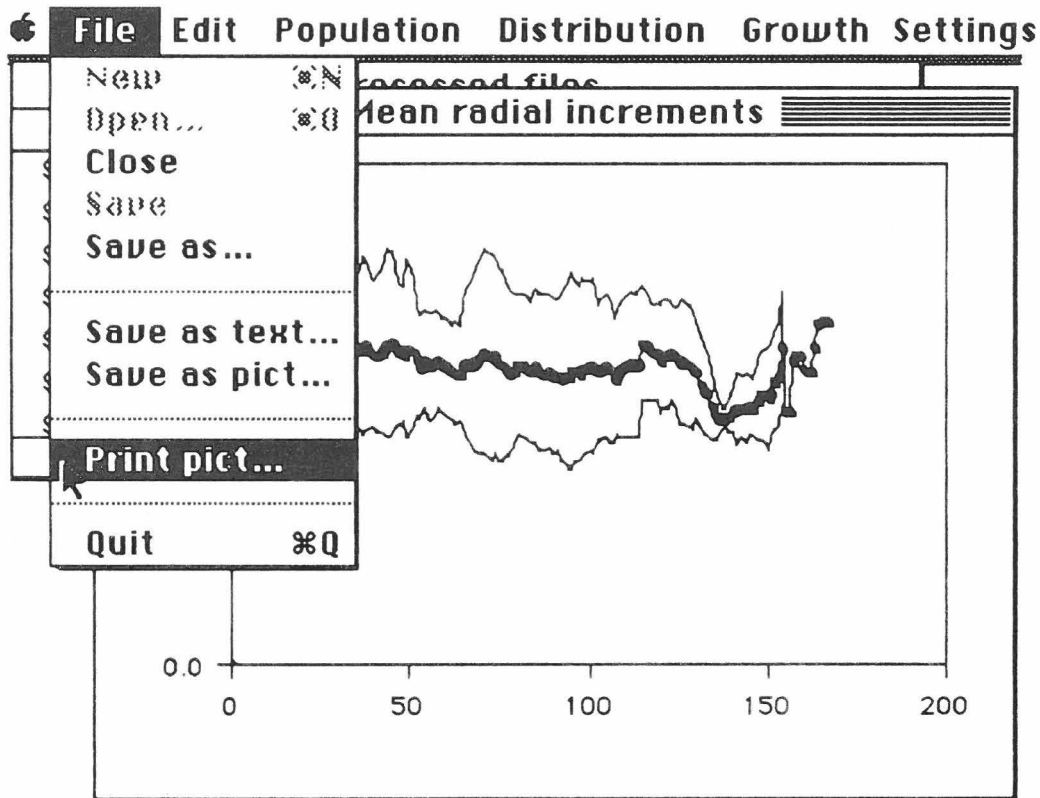


Fig. 51. The "**Print pict...**" option from the File menu.

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