

## Practical Sessions on RIVPACS III+

A series of step-by-step exercises to demonstrate some of the major options available in RIVPACS III+.

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#### **<u>RIVPACS III+ : INTRODUCTORY NOTES</u>**

#### Loading the software

The RIVPACS software should be loaded in a directory of its own. This does not prevent input and output files being stored elsewhere if necessary (see later exercises).

To load the software, the PC should be switched on and set at the C: drive. To create the required directory for carrying RIVPACS III+, at the C:> prompt type MD RIVPAC3P <<enter>>

The symbols << and >> are used throughout this document to signify a single key which must be pressed.

The PC should now be set to the new directory before loading the software. Therefore, at the C:> prompt type

CD RIVPAC3P

The RIVPACS software is provided on a single disk. To load it, insert the disk in drive A: and type

A:\INSTALLR <<enter>>

The software, which is compressed, will be expanded and loaded and the state of progress will be shown on the screen.

#### Getting going

Your PC should now be set at the C:RIVPAC3P> prompt and the software loaded and ready for action.

To get going type RIVPACS <<enter>>

The first screen is the title page and this is followed by two pages of background text. Each is exited by pressing <<enter>> or any other key.

The main menu panel offers the basic choice of type of run and the seven main options are described in this collection of exercises.

For each of the exercises that follow you need to select one of the main panel options. The exercises successively cover PREDICTION, COMPARE, CLASSIFICATION, DEFAULTS SETUP, AUTO/BATCH SETUP, SAMPLE BIAS SETUP and QUALITY BAND SETUP. HELP boxes with informative text are a menu option at many key stages. EXIT is used to leave RIVPACS and return to the C:\RIVPAC3P> prompt. At each exercise select the appropriate menu option.

#### **General considerations**

Throughout the following exercises the selected response to each on-screen prompt box or menu is determined by highlighting your chosen option, using the up/down, left/right arrows, and pressing <<enter>> or by typing the option number or initial letter of your selection.

Sometimes <<Esc>> can be used to terminate a run and return to the initial main menu. Many prompt boxes/menus offer the option to return to previous screens or return to the start (i.e. main menu panel)

Most prompt boxes allow only a single choice to be made but more than one taxonomic level or season can be selected for each run.

#### **<u>RIVPACS III+ : INTERACTIVE PREDICTIONS (1a)</u>**

#### Introductory note

Many keyboard operations are common to all RIVPACS III+ runs and are tedious to repeat throughout this primer. They are given in italics in this first example only and are assumed for subsequent exercises.

Pressing <<enter>> is not necessary if the initial letter of your chosen option or its menu number is used to select it.

#### BMWP family level - manual data input (Example 1a)

As a simple, introductory example, make the following choices:

Procedure	:	Prediction	< <enter>&gt;</enter>
Country	:	Great Britain	< <enter>&gt;</enter>
Taxonomic level(s)	:	BMWP families & BMWP indices (1)	< <enter>&gt;</enter>
The C: SELECTION COMP	LETED	bar is now highlighted Press << >> o	r < <enter>&gt;</enter>
Biological data on file?	:	NO	< <enter>&gt;</enter>
File input of O-E values?	:	NO	< <enter>&gt;</enter>
Screen input of O-E values?	:	NO	< <enter>&gt;</enter>
Season (s)	:	Spring, summer & autumn combined (7)	< <enter>&gt;</enter>
The C: SELECTION COMP	LETED	bar is now highlighted Press < <c>&gt; o</c>	r < <enter>&gt;</enter>
Environmental variables	:	Option 1	< <enter>&gt;</enter>
Continue/Previous screen	:	Continue	< <enter>&gt;</enter>
Environmental data on file?	:	NO	< <enter>&gt;</enter>
Proceed automatically?	:	NO	< <enter>&gt;</enter>
		[the environmental data to be manually ent	ered]
Predicted taxa on screen?	:	YES	< <enter>&gt;</enter>
Outputs to disk file?	:	NO to each option < <enter>&gt; after each</enter>	ch response
		[in this run see taxa and scores on screen]	
Stop listing taxa?	•	Keep it as 0.1% for BMWP level	< <enter>&gt;</enter>
Continue/Previous screen	:	Continue	< <enter>&gt;</enter>

Now enter the following data (1 site only).

Site reference	:	Frome at East Stoke	< <enter>&gt;</enter>
Grid reference	:	SY 866 867	< <enter>&gt;</enter>
Altitude (m)	:	13	< <enter>&gt;</enter>
Distance from source (km)	:	43	< <enter>&gt;</enter>
Water width (m)	:	18	< <enter>&gt;</enter>
Water depth (cm)	:	64	< <enter>&gt;</enter>
Discharge category	:	6	< <enter>&gt;</enter>
Boulders (% cover)	:	13	< <enter>&gt;</enter>
Gravel (% cover)	:	61	< <enter>&gt;</enter>
Sand (% cover)	:	22	< <enter>&gt;</enter>
Silt (% cover)	:	4	< <enter>&gt;</enter>
Alkalinity (mg $l^{-1}$ CaCO <sub>3</sub> )	:	172	< <enter>&gt;</enter>
Slope (m km <sup>-1</sup> )	:	10	
Are environmental data correct?	:	Y (YES)	< <enter>&gt;</enter>
Enter data for another site?	:	N (NO)	< <enter>&gt;</enter>
Environmental data are then displayed			< <enter>&gt;</enter>
Probabilities of group membership are give	en		< <enter>&gt;</enter>
Predicted BMWP families are listed		Press < <enter>&gt; to scroll</enter>	through the list
BMWP index values are presented		Press < <enter>&gt; to return to</enter>	the main panel

#### **<u>RIVPACS III+ : INTERACTIVE PREDICTIONS (1b)</u>**

#### BMWP family level - data input from files (Example 1b)

Initially, we offer a straightforward example to familiarise you with the use of input/output files and confidence limits for O/E values.

This can be followed by examples of your own using different responses at each asterisk (\*).

The responses for this exercise are as follows:

	Procedure	:	Prediction
*	Country	:	Great Britain
	Taxonomic level(s)	:	BMWP family & BMWP indices (1)
	Biological data on file?	:	YES
*	Season(s)	:	Spring, summer & autumn combined (7)
	Biological file name	:	TSTFAMGB.ASC
*	Environmental variables	:	Option 1
	Environmental data on file?	:	YES
*	Type of file	:	ASCII
	Environmental filename	:	TSTENVGB.ASC
	Allow for sample bias?	:	NO
	Assign to quality bands	:	NO
	Proceed automatically?	:	YES
	Output to disk file?	:	YES
	Output listing filename	:	WKBMWP1B.LST
*	Include predicted taxa?	:	YES

\* Do not create additional output files on this run

\* Stop listing taxa? : 0.1%

The three Great Britain test file sites are flagged as the prediction proceeds, before the main panel reappears.

Examine your output file WKBMWP1.LST as follows:

Exit from main panel At the prompt C:\RIVPAC3P> type EDIT WKBMWP1.LST <<enter>>

To close the DOS editor press the following three keys in sequence <<Alt>> <<F>> <<X>>

Then re-enter RIVPACS from the C:\RIVPAC3P> prompt, type RIVPACS <<enter>>

Now do another run and make your own choice at each asterisk. When making your own choices, refer to the list of example input files in Appendix 3 of the RIVPACS III+ manual to be sure of entering the correct file names.

#### **<u>RIVPACS III+ : INTERACTIVE PREDICTIONS (1c)</u>**

#### BMWP family level - file input O-E values, assign samples to quality bands (Example 1c)

This example introduces the option to use previously calculated observed(O) and expected(E) BMWP index values for the samples as an input file and the facility to assign samples probabilistically to quality bands. The O-E file to be used as input in RIVPACS III+ could have been derived as an output file from a run of RIVPACS III which used the biological and environmental data files for the samples.

Use the procedure detailed under example (1b) to view the contents of O-E input file TSTOET2.DAT to be used in this example.

Procedure	:	Prediction
Country	:	Great Britain
Taxonomic level(s)	:	BMWP family & BMWP indices (1)
Biological data on file?	:	NO
Input O-E values from file	?:	YES
O-E input data filename	:	TSTOET2.DAT
Allow for sample bias?	:	No
Assign to quality bands	:	YES use default GQA limits (2)
Proceed automatically?	:	NO
Output to disk file?	:	YES
Output listing filename	. :	WKBMWP1C.LST
Output O-E type 1 file?	:	NO

The confidence limits for the O/E ratios for a sample and the subsequent estimated probabilities of belonging to each quality band are presented, for convenience, on one output screen. As the band given to a sample based on the GQA banding system is the lower of its bands based on each of O/E for number of taxa and ASPT, no information is given for O/E based on BMWP score.

Use the procedure detailed under example (1b) to view the contents of output file WKBMWP1C.LST

#### **<u>RIVPACS III+ : INTERACTIVE PREDICTIONS (1d)</u>**

### <u>BMWP family level - screen input of O-E values, allow for bias, assign to quality bands</u> (Example 1d)

This example introduces the ability to allow for the effect of sample bias on an assessment of O/E values and on the assignment of samples to quality bands. It demonstrates the ability to input the O and E values, the seasons involved and the sample biases from the screen.

	Procedure	:	Prediction
	Country	:	Great Britain
	Taxonomic level(s)	:	BMWP family & BMWP indices (1)
	Biological data on file?	:	NO
	Input O-E values from file?	:	NO
	Screen input of O-E values?	:	YES
*	Store in O-E data filename	:	WKOET1D.DAT
*	Sample code	:	Tumbledown
*	Observed BMWP score	:	139
*	Observed number of taxa	:	25
	Observed ASPT calcu	ulated a	utomatically as 139/25 = 5.56
*	Expected BMWP score	:	190.1
*	Expected number of taxa	:	30.2
*	Expected ASPT	:	6.29
*	Seasons involved Spring	:	N
*	Summer	:	Y
*	Autumn	:	Y
*	Specify sample biases?	:	Y
*	Bias for Summer	:	1.9
*	Autumn	:	2.4
*	Enter another sample?	:	N
*	Assign to quality bands	:	YES use default GQA limits (2)
*	Proceed automatically?	:	NO
*	Output to disk file?	:	YES
*	Output listing filename	:	WKBMWP1D.LST
*	Output O-E type 1 file?	:	YES
*	O-E output filename	:	WKBMWP1D.OE1

The confidence limits for the O/E ratios for a sample and the estimated probabilities of belonging to each quality band are presented, for convenience, on one output screen with the results uncorrected for bias on the left-hand side and results corrected for bias on the right-hand side.

Use the procedure detailed under example (1b) to examine the contents of output file WKBMWP1D.LST.

Examine O-E type 1 output file WKBMWP1C.OE1 (one long line for each of the two samples) and check the meaning of the output statistics by referring to section 6.11 of the RIVPACS III+ User Manual. (Such files could be read into EXCEL for further analysis.)

Now do other runs and make your own choice at each asterisk.

#### **<u>RIVPACS III+</u>** INTERACTIVE PREDICTIONS (1e)

#### Abundance for all families and the abundance index (Example 1e)

This example involves making predictions of mean expected abundances (weighted arithmetic means of log categories) and of the experimental abundance index Q14.

The required responses for this exercise are:

Procedure	:	Prediction
Country	:	Great Britain
Taxonomic level(s)	:	Abundance for all families and abundance index (2)
Biological data on file?	:	YES
Season(s)	:	Spring (1)
Biological filename	:	TSTSPRGB.ASC
Environmental variables	:	Option 1
Environmental data on file?	:	YES
Type of file?	:	ASCII
Environmental filename	:	TSTENVGB.ASC
Proceed automatically?	:	YES
Output to disk file?	:	YES
Output filename	:	WKSPABUN.LST
Include predicted taxa?	:	YES

:

Do not create additional output files on this run

Stop listing taxa?

\*

Retain 0.0 log abundance.

As in the previous example (b), the three test sites are flagged as the prediction proceeds, before the main panel reappears.

Use the procedure detailed under example (b) to view contents of WKSPABUN.LST

Alternatively the full prediction may be viewed at leisure on screen if you answer NO at this point and then request that predicted taxa are to be listed on screen. You still retain the option of creating output files if required.

#### **<u>RIVPACS III+ : INTERACTIVE PREDICTIONS (1f)</u>**

#### Multiple taxonomic levels (and/or seasons) on a single run (Example 1f)

In theory it is possible to request predictions at 5 taxonomic levels (1-5) and for 7 seasons/seasonal combinations (1-7). However, when using BMWP families and indices (taxonomic level 1) with error assessment, no other taxonomic level should be selected. Also, we recommend that you exclude taxonomic level 5 (Existing customisation) until you have become familiar with this option (see next section on setting up a customisation).

Merely excluding taxonomic levels 1 and 5 could still generate 17 predictions for each site if you request all of taxonomic levels 2-4 and seasons 1-7 (NB taxonomic level 2, abundance for all families only operates on seasons 1-3 (i.e. single seasons) at present).

Hence, we suggest that you are selective in your initial choice. Bear in mind that there are 3 sites in the environmental data example files and that if you request output files they will be substantial if you ask for the inclusion of predicted taxa.

As one example, try the following:

Procedure	:	Prediction
Country	:	Great Britain
Taxonomic level(s)	:	Choose 3 & 4
Biological data on file?	:	NO - on this initial run.
		[ On a later run, reply YES, but bear in
		mind the need to input the appropriate
		example files when requested]
Season(s)	:	Choose 1 & 7
Environmental variables	:	Option 1
Environmental data on file?	:	YES
Type of file	:	ASCII
Environmental filename	:	TSTENVGB.ASC
Proceed automatically?	:	YES
Output to disk file?	:	YES
Output biological filename	:	WKSPMULT.LST
Include predicted taxa?	:	YES
Do not create additional out	put file	s

Stop listing taxa?	:	Retain as 0.1 for all families
		Change to 50.0 for species

Follow the previous instructions for example (b) to examine the output file.

For each of the 3 example sites, the output sequence is,

Site name/code			
Environmental data			
Probabilities of grou	p membership		
Predicted families	Spring	То	0.1%
	Spring, summer and autumn combined	То	0.1%
Predicted species	Spring	То	50.0%
	Spring, summer and autumn combined	То	50.0%

#### RIVPACS III+ : INTERACTIVE PREDICTIONS (1g)

#### Setting up and using a customisation.

At the outset, go through the sequence of operations to familiarise yourself with the procedure. Do not attempt to set up a useful customisation. This can be done later.

In the simple (and unlikely!) example below, we want a prediction in which all members of the Tricladida (flatworms) are identified at generic level. The remaining invertebrates are predicted at major taxonomic group only.

Procedure	:	Prediction				
Country	:	Great Britain				
Taxonomic level(s)	:	Chose new customisation (6)				
Name?	:	CUSTOM1				
Description?	:	Triclads to genus				
Country	:	Great Britain (G)				
Current taxon	:	Tricladida.	Select? N			
		Planariidae.	Select? N			
		<i>Planaria</i> sp.	Select? Y			
		Polycelis sp.	Select? Y			
		Phagocata sp.	Select? Y			
		Crenobia sp.	Select? Y			
		Dugesia sp.	Select? Y			
		Dendrocoelidae	Select? N			
		Bdellocephala sp.	Select? Y			
		Dendrocoelum sp.	Select? Y			
Current taxon	:	Gastropoda	Select? Y			
		Answer Y to all sub	sequent questions.			

At the end, the screen indicates that this customisation generates 31 taxa. The necessary files are then created before the taxonomic level panel reappears. The customisation is thus saved to file and can be accessed in subsequent runs as follows:

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Taxonomic level(s)

Existing customisation (5)

On the next screen choose the UP or DOWN options in the upper menu box as often as necessary to scroll to the required customisation (CUSTOM1) in the lower menu box. After highlighting the required customisation, choose SELECT. Choose SELECT again on the subsequent screen to re-enforce your choice. [This screen also offers the option of deleting unwanted customisations. Use it on a subsequent occasion to delete CUSTOM1]

Biological data on file?	:	NO
Season(s)	:	Spring (1)
Environmental variables	:	Option 1
Environmental data on file?	:	YES
Type of file?	:	ASCII
Environmental filename	:	TSTENVGB.ASC
Proceed automatically?	:	YES
Output to disk file?	:	YES
Output biological filename	:	WKSPCUST.BIO
Include predicted taxa?	:	YES
Do not create additional out	put file	S

Stop listing taxa?

Retain 0.1%

Follow previous instructions in example (b) to examine output file.

## **RIVPACS III+ : INTERACTIVE COMPARE (2a)**

## <u>Compare two samples - screen input of O-E values, assess differences in O/E values and likelihood of differences in quality bands (Example 2a)</u>

This example introduces main menu procedure Compare. This is used to compare two samples (or multiple pairs of samples) by assessing the statistical significance of the differences in their O/E values and by assessing the likelihood of a difference in quality bands.

The two samples can be the same site at different times, or two different sites; the season(s) involved can be the same or different. The only information needed for each sample is:

- (i) the previously calculated observed(O) and expected(E) values of BMWP score, number of taxa and ASPT for the two samples.
- (ii) the seasons involved
- (iii) the sample biases applicable to each season involved (optional)
- (iv) whether the expected values of both samples were based on the same values of the environmental variables.

In this example, the information is input from the screen; in later examples it is read from files.

	Procedure	:	Compare	
	Input O-E values from file?	:	NO	
*	Store in O-E data filename	:	WKOET2A.DAT	
*	Sample code	:	Tumbledown	Pickmeup Place
*	Observed BMWP score	:	139	83
*	Observed number of taxa	:	25	17
Observed AS	PT calculated automatically a	s: 13	9/25 = 5.56	83/17 = 4.88
*	Expected BMWP score	:	190.1	175.6
*	Expected number of taxa	:	30.2	27.1
*	Expected ASPT	:	6.29	6.48
*	Seasons involved Spring	:	Ν	Ν
*	Summer		Y	Y
*	Autumn	:	Y	Ν
*	Specify sample biases?	:	Y	
*	Bias for Summer	:	1.9	2.5
*	Autumn	:	2.4	
*	Enter another sample?	:	Ν	
*	Same values for variables	:	Ν	
	Assign to quality bands	:	YES use default G	QA limits (2)
*	Proceed automatically?	:	NO	
	Output to disk file?	:	YES	
	Output listing filename	:	WKCOMP2A.LST	
*	Tables of change in band	:	Taxa, ASPT and O	verall table
	Output O-E type 1 file?	:	No	
	Output O-E type 2 file?	:	No	

The assessment of the O/E values and probabilistic banding for each of the two individual samples is given first; this part of the output to the screen and/or listing file and O-E type 1 file is exactly the same as for procedure Prediction for the individual samples. Next is a statistical assessment for differences in O/E values between the two samples and finally an assessment of the likelihood of differences in quality band when based on O/E for number of taxa, ASPT or the overall GQA band.

Use the procedure detailed under example (1b) to view the contents of O-E input file WKOET2A.DAT made from the screen input and the output listing file WKCOMP2A.LST.

#### <u>RIVPACS III+ : INTERACTIVE COMPARE (2b)</u>

# <u>Compare two samples - file input of O-E values, assess differences in O/E values and likelihood of differences in quality bands both ignoring and corrected for sample biases (Example 2b)</u>

This example demonstrates the potential effect of allowing for sample biases in the comparison of two samples. The observed(O) and expected(E) values for the samples are also read from an O-E input file; this input file could have been derived as an output file, from a previous run of RIVPACS III+ or RIVPACS III, based on the biological and environmental data files for the samples.

Use the procedure detailed under example (1b) to view the contents of O-E input file TSTOET2.DAT to be used in this example.

	Procedure	:	Compare
	Input O-E values from file?	:	YES
	Single or pair of files	:	1
	O-E input data filename	:	TSTOET2.DAT
*	Same values for variables	:	No
*	Allow for sample bias?	:	YES
	Obtain biases from?	:	bias file (1)
	Bias filename	:	TSTBIAS.DAT
	Assign to quality bands	:	YES use default GQA limits (2)
*	Proceed automatically?	:	NO
	Output to disk file?	:	YES
	Output listing filename	:	WKCOMP2B.LST
*	Tables of change in band	:	Overall table only
	Output O-E type 1 file?	:	Yes
*	O-E type 1 output filename	:	WKCOMP2B.OE1
	Output O-E type 2 file?	:	Yes
*	O-E type 1 output filename	:	WKCOMP2B.OE2

When correction for sample bias is requested all assessments are first given uncorrected for bias and then corrected for bias.

The assessment of the O/E values and probabilistic banding for each of the two individual samples is given first; this part of the output to the screen and/or listing file and O-E type 1 file is exactly the same as for procedure Prediction for the individual samples. Next is a statistical assessment for differences in O/E values between the two samples. The assessment of the likelihood of differences in quality band is given in a sequence of output screens based on (in order) O/E for number of taxa, ASPT or the overall GQA band, uncorrected for bias, then corrected for bias.

Use the procedure detailed under example (1b) to view the contents of output listing file WKBMWP2B.LST

Examine O-E type 1 output file WKBMWP2B.OE1 (one long line for each of the two samples) and O-E type 2 output file WKCOMP2B.OE2 (one long line for sample comparison statistics ignoring bias and a second corrected for bias). Check the meaning of the output statistics in both files by referring to sections 6.11 and 6.12 of the RIVPACS III+ User Manual. (Such files could be read into EXCEL for further analysis).

#### <u>RIVPACS III+ : INTERACTIVE BIOLOGICAL CLASSIFICATION (3)</u>

#### Species level classification

The classification procedures are simple to operate and depend upon the biological data being held on file. Only one practical example is given here.

A new site may only be classified using species level data (three seasons combined) or BMWP family data (three seasons combined).

Bear in mind that when the reference sites were used to develop the 35 group Great Britain and seven group Northern Ireland classifications, information on both species composition and family abundances was utilised. Use of this same procedure for classification of new sites would have been very cumbersome and is unlikely to have been used in practice. The simplified options of species or BMWP family classification have, thus, been retained from RIVPACS II.

We recommend that classification results are used, with caution, to allocate sites to a restricted area of the classification. It is unwise to place great importance to the individual classification group with which the site has most in common.

As an example, use the following procedure:

Procedure	:	<b>Biological classification</b>
Country	:	Great Britain
Taxonomic level	:	Species
Type of file?	:	ASCII
Biological filename	:	TSTSPRGB.ASC
Output to disk file?	:	NO
Proceed automatically?	:	NO

Then view the results on screen.

#### **RIVPACS III+ : DEFAULTS FILES (4a)**

#### Using defaults files

Defaults files contain the paths for the user's input and output files.

At installation, the RIVPACS III+ program is instructed to find all of the input files (eg the example environmental and biological files) in the RIVPACS III+ software directory and all output files are initially written to this directory. RIVPACS III+ is supplied with a main "defaults" file entitled DEFAULTS.DAT which contains this instruction.

The user may make alternative defaults files, if required, instructing the program to input and output files to alternative directories or sub-directories. This example shows how defaults files are created and used.

In this exercise you will:

- i) create a new directory and suitable sub-directories for holding input and output files
- ii) place example environmental and biological files in the input sub-directories
- iii) use the input files in a prediction
- iv) examine the results in the new output sub-directory

#### Creating directories and sub-directories

Exit RIVPACS at the main panel Return to the main C:> prompt by typing CD.. <<enter>> Create a directory called RUBBISH by typing MD RUBBISH <<enter>> Move to your new directory by typing CD RUBBISH <<enter>> Create six new sub-directories (BIO, ENV and OUT) by typing

MD BIO <<enter>> MD ENV <<enter>> MD OE1 <<enter>> MD BIAS <<enter>> MD BAND <<enter>> MD OUT <<enter>>

Put example input files in the new sub-directories

Whilst at the existing prompt of C:\RUBBISH>, type

COPY C:\RIVPAC3P\TSTENVGB.ASC C:\RUBBISH\ENV

This copies a test file of environmental data to sub-directory C:\RUBBISH\ENV

Whilst at the existing prompt of C:\RUBBISH>, type

COPY C:\RIVPAC3P\TSTFAMGB.ASC C:\RUBBISH\BIO

This copies a test file of family level biological data to sub-directory C:\RUBBISH\BIO

Similarly, whilst at the existing prompt of C:\RUBBISH>, type

COPY C:\RIVPAC3P\TSTOET2.DAT C:\RUBBISH\OE1 COPY C:\RIVPAC3P\TSTBIAS.DAT C:\RUBBISH\BIAS COPY C:\RIVPAC3P\TSTBAND.DAT C:\RUBBISH\BAND

#### **<u>RIVPACS III+ : DEFAULTS FILES (4b)</u>**

#### Returning to RIVPACS III+

At the prompt C:\RUBBISH> type CD C:\RIVPAC3P <<enter>> At the prompt C:\RIVPAC3P> type RIVPACS <<enter>>

#### Creating a new defaults file

In RIVPACS III+ select Defaults setup from the main panel

The screen shows a series of questions which must be answered successively by typing the necessary information in screen boxes. In this example the following actions are required:

The first screen box appears with the file name DEFAULTS.DAT already entered. Enter the new defaults file name by over-typing

DEF3.DAT <<enter>>

The next screen box highlights the instruction E: Edit this file. Select this option by pressing either <<enter>> or <<E>>.

A new screen is now displayed

The next two screen boxes allow the input paths to be entered for the biological and environmental data respectively. Type the following

C:\RUBBISH\BIO <<enter>> (The input path for the biological data) C:\RUBBISH\ENV <<enter>> (The input path for the environmental data) C:\RUBBISH\OE1 <<enter>> (The input path for the O-E data) C:\RUBBISH\BIAS <<enter>> (The input path for the sample bias file) C:\RUBBISH\BAND <<enter>> (The input path for the quality band limit file)

The next box allows the output path to be defined. Type the following

C:\RUBBISH\OUT <<enter>>

Press <<enter>> again to move on to the next screen

At the next box type Y <<enter>> to initiate the saving of the file. The next box displays your chosen defaults file name, DEF3.DAT Type <<enter>> to confirm this.

You will be asked if you want to overwrite the existing file. In this exercise respond YES. This prompt is a useful reminder to ensure you don't lose an existing file which you want to keep.

Now press <<F3>> in order to save the file.

#### **<u>RIVPACS III+</u>** : DEFAULTS FILES (4c)

#### Selecting a defaults file

In order to use your new defaults file in interactive mode you first need to select it as follows:

Select Defaults setup at the main panel

In the first screen box, replace DEFAULTS.DAT as the defaults file name, by typing DEF3.DAT

At the next screen box either

Select U: Use as defaults file by pressing <<enter>>, or Press <<U>> Either option automatically returns you to the main panel.

Your new defaults file is set and you can proceed with whatever procedure run you require.

#### Using your defaults file

Having created and selected your new defaults file, DEF3.DAT, try the following exercise which uses biological and environmental data only from file.

Make a simple family level prediction for Great Britain by choosing option 1 from the taxonomic selection menu and option 7, spring, summer and autumn, from the season menu.

Select the options that state that you hold environmental data on file in ASCII format.

When requested, give the appropriate biological and environmental file names.

Biological filename	:	TSTFAMGB.ASC
Environmental filename	:	TSTENVGB.ASC

Your defaults file, DEF3.DAT, will find these files for you using the paths you have stipulated.

When prompted, request that the prediction proceeds automatically without user intervention.

When requested send the results, including the probabilities of taxon occurrences, to output file WKSPDFIL.LST

Your defaults file will automatically send this to the required sub-directory.

To see the contents of the output file exit RIVPAC3P at the main panel and type: EDIT C:\RUBBISH\OUT\WKSPDFIL.LST

#### **Further information**

After exiting RIVPACS III+, the next use of the system will automatically revert to using DEFAULTS.DAT as the defaults file.

If you wish to use DEF3.DAT again it needs to be re-selected using the procedure above. If you want to change the defaults directory semi-permanently, then you must edit the file DEFAULTS.DAT as required.

For a fuller explanation of the defaults file procedures it is important to study section 2.5 of the RIVPACS III+ Manual.

#### <u>**RIVPACS III+ : BATCH MODE (5a)</u>**</u>

The making, use and format of batch files is described in the Sections 2.6, 2.9.3 and 6.2 of the RIVPACS III+ User Manual.

#### Using the Auto/Batch setup menu option (Example 5a)

At the main panel choose Auto/Batch setup. Type in the following responses:

Batch file name	:	NEWAUTO.DAT
Edit batch instructions set number	:	1 (first instruction set of new file)
Edit or delete this set	:	E (to allow entry of new instructions)
Defaults file name	:	DEFAULTS.DAT
		[This means that the input and output
		files will be in the same directory as the
		RIVPACS III+ software]
Procedure	:	Prediction (1)
Country	:	Great Britain (1)
Taxonomic level(s)	•	BMWP families (1)
Biological data files?	:	YES (Y)
Season(s)	:	All three seasons combined (7)
Predictive variables	:	Option 1
Biological filename	:	TSTFAMGB.ASC
Environmental file format	:	ASCII (1)
Environmental filename	:	TSTENVGB.ASC
Allow for sample bias?	:	1: Yes (fixed)
Bias filename	:	TSTBIAS.DAT
Assign to quality bands	:	1: Yes (use EA GQA defaults)
Output listing file	:	WKBATCH1.LST
Include predicted taxa?	:	Y
Stop listing taxa at % probability?	:	0.1%
Additional output files		
Environmental data	:	WKBATCH1.ENV
BMWP O-E type 1	:	WKBATCH1.OE1
Overwrite existing files?	:	YES (Y)
Save changes?	:	YES (Y), to file NEWAUTO.DAT

Press <<F3>> to save the batch file and to return to the main panel.

Exit the main panel and use the procedure detailed under example (1b) to view the contents of batch file NEWAUTO.DAT created above. The file includes the batch mode instruction line and a list of the specified files.

#### Running a simple batch mode prediction

To run RIVPACS III+ in batch mode, at the C:\RIVPAC3P> prompt type

RIVPACS AUTO NEWAUTO.DAT <<enter>>

The program only displays the name of the sample(s) currently being processed.

On completion of the batch run, examine the various output files, of the form WKBATCH1.\*\*\*, using the procedure detailed under example (1b).

#### RIVPACS III+ : BATCH MODE (5b)

#### Multiple runs in batch mode (Example 5b)

This example runs a prediction using O-E input files, then runs a comparison using the output O-E type 1 file for the three samples in example 5a with the first three samples in another O-E input file called TSTOET1.DAT.

Procedure	:	Auto/Batch setup
Batch file name	:	MULTAUTO.DAT
Edit batch instructions set number	:	1
Edit or delete this set	:	Е
Defaults file name	:	DEFAULTS.DAT
Procedure	:	Prediction (1)
Country	:	Great Britain (1)
Taxonomic level(s)	:	BMWP families (1)
Biological data files?	:	N
O-E type 1 file as input?	:	Y
O-E type 1 input filename	:	TSTOET2.DAT
Allow for sample bias?	:	1: Yes (fixed)
Bias filename	:	TSTBIÀS.DÁT
Assign to quality bands	:	2: No
Output listing file	:	WKBATCH2.LST
Additional output files		
BMWP O-E type 1	:	WKBATCH2.OE1
Overwrite existing files?	:	YES (Y)
Save changes?	:	YES (Y), to file MULTAUTO.DAT
Press < <f3>&gt; to save the batch file and re</f3>	turn to	the main panel.
Procedure	:	Auto/Batch setup
Batch file name	:	MULTAUTO.DAT
Edit batch instructions set number	:	2
Edit or delete this set	:	E
Defaults file name	:	DEFAULTS.DAT
Procedure	:	Compare (2)
No. of files containing O-E values	:	2
Same values of variables	:	N
O-E filename for samples A	:	WKBATCH1.OE1
O-E filename for samples A	:	TSTOET1.DAT
Allow for sample bias?	:	1: Yes (fixed)
Bias filename	:	TSTBIAS.DAT
Assign to quality bands	:	1: Yes (use EA GQA defaults)
Output listing file	:	WKBATCH3.LST
Tables of change in band	:	Overall table only
Additional output files		
BMWP O-E type 1	:	WKBATCH3.OE1
BMWP O-E type 2	:	WKBATCH3.OE2
Overwrite existing files?	:	YES (Y)
Save changes?	:	YES (Y), to file MULTAUTO.DAT
Press < <f3>&gt; to save the batch file and re</f3>	turn to	the main panel.

#### Now run MULTAUTO.DAT from the C:\RIVPAC3P> prompt by typing RIVPACS AUTO MULTAUTO.DAT <<enter>>

After the batch run, examine the output files as directed previously.

Batch files can also be used to run classifications or a mixed package of both predictions, comparisons and classifications. Defaults files other than the standard DEFAULTS.DAT can be used to input and output files from and to other directories.

Try some other examples.

#### RIVPACS III+ : SAMPLE BIAS FILE SETUP (6)

To allow for sample bias, RIVPACS requires an appropriate estimate of the average bias for single season samples from each of the three RIVPACS seasons. In interactive runs of RIVPACS samples, these biases can either be entered from the screen, or read from a sample bias file. In batch mode, the biases must be read from a sample bias file.

The format of bias files is described in Section 6.9 of the RIVPACS III+ User Manual.

A bias file could be made using an ASCII file editor (such as EDIT in MS-DOS)

This example demonstrates how to make a sample bias file using the sample bias setup option of interactive RIVPACS. The bias file will contain biases for each pair of samples (A and B) to be compared using RIVPACS procedure Compare. However, this bias file can also be used in RIVPACS prediction runs; in which case all the samples are assumed to have the biases specified for samples A.

#### Using the Sample bias setup menu option (Example 6)

Procedure		:	Sample bias setup
Bias filename		:	WKBIAS.DAT
Single or Paired sam	ples	:	2 (paired)
Sample A bias for :	Spring	:	2.7
-	Summer	:	2.3
	Autumn	:	2.1
Sample B bias for :	Spring	:	1.9
-	Summer	:	1.9
	Autumn	:	1.7
Save changes?		÷	YES (Y), to file WKBIAS.DAT

Press  $\langle F_3 \rangle$  to save the batch file and return to the main panel.

#### RIVPACS III+ : QUALITY BAND FILE SETUP (7)

To assign samples to quality bands, RIVPACS requires the lower limit of O/E for each band for each of BMWP score, number of taxa and ASPT. In interactive runs of RIVPACS samples, the band lower limits can either be entered from the screen, or read from a quality band file. In batch mode, the limits must be read from a quality band file.

The format of quality band files is described in Section 6.10 of the RIVPACS III+ User Manual.

A quality band file could be made using an ASCII file editor (such as EDIT in MS-DOS)

This example demonstrates how to make a quality band file using the Quality band setup option of interactive RIVPACS.

#### Using the Quality band setup menu option (Example 7)

Procedure		:	Quality band setup
Bias filename		:	WKBAND.DAT
Number of bands		:	4
Name for first band		:	Fine
Lower limit for :	Score O/E	:	0.80
	Taxa O/E	:	0.85
	ASPT O/E	:	0.90
Name for second ba	nd	:	Good
Lower limit for :	Score O/E	;	0.60
	Taxa O/E	:	0.70
	ASPT O/E	:	0.80
Name for third band	l	:	Fair
Lower limit for :	Score O/E	:	0.40
	Taxa O/E	:	0.55
	ASPT O/E	:	0.70
Name for fourth band			Poor
Lower limits for last	t band are auto	matic	ally set to 0.00

Save changes?

YES (Y), to file WKBAND.DAT

Press <<F3>> to save the batch file and return to the main panel.

:

## RIVPACS III+

## STATISTICAL NOTES ON METHODS USED TO ASSESS ERRORS IN O/E MEASURES OF BIOLOGICAL QUALITY

## RIVPACS III+

## STATISTICAL METHODS USED TO ASSESS ERRORS IN O/E MEASURES OF BIOLOGICAL QUALITY

Details of exact procedures are given in Section 7 of the RIVPACS III+ User Manual.

Background research in :

Furse, M.T., Clarke, R.T., Winder, J.M., Symes, K.L., Blackburn, J.H., Grieve, N.J. & Gunn, R.J.M. (1995).

Biological assessment methods: controlling the quality of biological data. package 1: The variability of data used for assessing the biological condition of rivers. Final report to the National Rivers Authority. **R&D Note 412**. 114 pp.

BMWP indices : ASPT, number of taxa and BMWP score

O = Actual or 'face' value of OBSERVED BMWP index for sample

E = Actual EXPECTED BMWP index value for the sample

Index of biological quality = Q = O / E

Computer simulation of possible values of O/E for the sample based on estimates of :

- (i) typical sampling variation
- (ii) sample sorting and identification errors
- (iii) errors in the values of the environmental predictor variables

Frequency distribution of simulated values provides confidence limits for the sample's O/E index of quality

## SAMPLING VARIATION

Estimated from Furse et al (1995) :

Number of seasons in co	>1	2	3	
Square root of observed number of BMWP taxa	Sampling standard deviation (SD <sub>%T</sub> )	0.228	0.164	0.145
Observed ASPT	Sampling standard deviation (SD <sub>A</sub> )	0.249	0.161	0.139

Examples of 95% sampling confidence intervals (95%CL) for observed values of number of taxa based on single or combined season samples

No. seasons in the combined sample	Observed value	95% CL	
		Lower	Upper
1	5	3	7
2	5	4	7
3	5	4	6
1	15	12	19
2	15	13	18
3	15	13	17
1	30	25	35
2	30	27	34
3	30	27	33

## SAMPLE SORTING AND IDENTIFICATION ERRORS

Results derived from Furse et al (1995) analyses of sample audit data:

- Bias = Net under-estimation of number of taxa in the sample
  - = Net "gains" estimated from a quality control procedure or audit scheme

Assume a Poisson distribution for the sample bias :



Need to specify  $B_1$ ,  $B_2$  and  $B_3$ , the average biases for each of spring, summer and autumn single season samples

Average bias for a two seasons combined sample

=

51% (sum of the biases in the two individuals seasons)

Average bias for a three seasons combined sample

= 37% (sum of the biases in the three individuals seasons)

## ERRORS IN THE EXPECTED (E) VALUES

## DUE TO ERRORS IN MEASURING THE ENVIRONMENTAL PREDICTOR VARIABLES

Remember : ALL RIVPACS predictions must be based on the average values of the environmental variables measured in each of the three RIVPACS seasons. Ideally they should be the average of several years three season averages.

Results derived by Furse et al (1995):

Size of error in E values does not depend significantly on either :

- (i) the number of seasons involved in the biological sample
- or (ii) the type of site

Estimates are :

Standard deviation for Expected number of taxa	=	0.53
Standard deviation for Expected ASPT	=	0.081

Note : may under-estimate size of errors in the Expected value, especially for long-term average predictions

## **GENERATING THE SIMULATED VALUES OF O/E**

Q <sub>r</sub>	=	O/E for $r^{th}$ simulation =	$\frac{O + R_{O} + B}{E + R_{E}}$
O R <sub>o</sub> B	-	Actual OBSERVED value random element of sampling va random Poisson bias term	riation
E R <sub>E</sub>	= =	Actual EXPECTED value random variation due to errors in v	values for environmental variables

## COMPARING TWO SAMPLES

 $Q_{r1}$  and  $Q_{r2}$  denote the O/E values for samples 1 and 2 (or samples A and B) for the r<sup>th</sup> simulation

 $D_r = Q_{r2} - Q_{r1} = difference in O/E values for the r<sup>th</sup> simulation$ 

Frequency distribution and confidence limits for D<sub>r</sub> indicate whether the O/E values for the two samples are statistically significantly different.

#### Figure 1:

Frequency histogram of 1000 RIVPACS III+ simulated values of O/E for number of taxa for two samples. The histogram is divided into bands (A, B, C,...) based on the Environment Agency's GQA biological quality band system. The percentage of values (%prob) falling in each band estimates the probability that the sample belongs to each band.

X denotes the observed or 'face' value of O/E for each sample



#### Figure 2: RIVPACS III+ comparison of two samples

Cross-classification table showing the probability sample A is in one biological quality band whilst sample B in is in another (perhaps the same or different) band based on their O/E values for number of taxa.

Table plots the distribution of the O/E values for number of taxa for the two samples for each of 1000 RIVPACS III+ simulations (shown as dots). The plot is divided into cells denoting bands (A, B, C,...) based on the Environment Agency's GQA biological quality banding system. The percentage of simulations falling in each cell of the twoway table estimates the probability that samples A and B have the indicated bands.

X denotes the observed or 'face' value of O/E for each of the two samples

