Command Reference for MrBayes ver. 3.1

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   1. Command summary
Commands that are available from the command
line or from a MrBayes block include:
About
               -- Describes the program
Acknowledgments -- Shows program acknowledgments
Charset -- Assigns a group of sites to a set
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              -- Shows status of characters
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              -- Sets run conditions and defines active data partition
Showmatrix -- Shows current character matrix
Showmodel -- Shows model settings
              -- Shows user tree
Showtree
```

-- Summarizes parameters from MCMC analysis

Sump

-- Assigns a group of taxa to a set Unlink -- Unlinks parameters across character partitions Usertree -- Defines a single user tree Version -- Shows program version Commands that should be in a NEXUS file (data block or trees block) include: Begin -- Denotes beginning of block in file Dimensions -- Defines size of character matrix End -- Denotes end of a block in file Endblock -- Alternative way of denoting end of a block Format -- Defines character format in data block Matrix -- Defines matrix of characters in data block Translate -- Defines alternative names for taxa -- Defines a tree from MCMC analysis Note that this program supports the use of the shortest unambiguous spelling of the above commands (e.g., "exe" instead of "execute"). ______ * 2. MrBayes commands About This command provides some general information about the program. ______ Acknowledgments This command shows the authors' acknowledgments. ______ Charset This command defines a character set. The format for the charset command is charset <name> = <character numbers> For example, "charset first_pos = $1-720\3$ " defines a character set called "first_pos" that includes every third site from 1 to 720. The character set name cannot have any spaces in it. The slash (\) is a nifty way of telling the program to assign every third (or second, or fifth, or whatever) character to the character set. This option is best used not from the command line, but rather as a line in the mrbayes block of a file. Note that you can use "." to stand in for the last character (e.g., charset $1-.\3$).

-- Summarizes trees from MCMC analysis

-- Shows status of taxa

Sumt

Taxastat

Charstat

This command shows the status of all the characters. The correct usage is

charstat

After typing "charstat", the character number, whether it is excluded or included, and the partition identity are shown. The output is paused every 100 characters. This pause can be turned off by setting autoclose to "yes" (set autoclose=yes).

Citations

This command shows a thorough list of citations you may consider using when publishing the results of a MrBayes analysis.

Comparetree

This command compares the trees in two files, called "filename1" and "filename2". It will output a bivariate plot of the split frequencies as well as plots of the tree distance as a function of the generation. The plots can be used to get a quick indication of whether two runs have converged onto the same set of trees. The "Comparetree" command will also produce a ".parts" file and a ".dists" file (these file endings are added to the end of the "Outputname"). The ".parts" file contains the paired split frequencies from the two tree samples; the ".dists" file contains the tree distance values. Note that the "Sumt" command provides a different set of convergence diagnostics tools that you may also want to explore. Unlike "Comparetree", "Sumt" can compare more than two tree samples and will calculate consensus trees and split frequencies from the pooled samples.

Parameter	Options	Current Setting
Filename1 Filename2 Outputname Burnin	<name> <name> <name> <name> <number></number></name></name></name></name>	temp.t temp.t temp.comp

Constraint

This command defines a tree constraint. The format for the constraint command is

constraint <constraint name> probability> = of taxa>

A list of taxa can be specified using a taxset, taxon names, or taxon numbers. A probability must also be specified. For now, MrBayes ignores this probability value and treats the constraint as an absolute requirement of trees. That is, trees that are not compatible with the constraint have zero prior (and hence zero posterior) probability.

Future releases of MrBayes will use the probability value to determine how

much more probable a tree is that contains the constraint than a tree without the constraint. For example, the following command

constraint example 100 = taxon_2 taxon_3

defines a constraint called "example" that includes two taxa. In future releases of MrBayes, trees that contain a clade with those two taxa together will have a prior probability that is 100 times that of trees without the constraint. In the current version, the probability value will be ignored and trees without the two taxa together will not be sampled.

If you are interested in inferring ancestral states for a particular node, you need to constrain that node first using the 'constraint' command. For more information on how to infer ancestral states, see the help for the 'report' command.

It is important to note that simply defining a constraint using this command is not sufficient for the program to actually implement the constraint in an analysis. You must also specify the constraints using 'prset topologypr = constraints (<name of constraint>)'. For more information, see the help on the 'prset' command.

Ctype

This command sets the character ordering for standard-type data. The correct usage is:

ctype <ordering>:<characters>

The available options for the <ordering> specifier are:

unordered -- Movement directly from one state to another is

allowed in an instant of time.

ordered -- Movement is only allowed between adjacent characters. For example, perhaps only between 0 <-> 1 and 1 <-> 2 $\,$

for a three state character ordered as 0 - 1 - 2.

irreversible -- Rates of change for losses are 0.

The characters to which the ordering is applied is specified in manner that is identical to commands such as "include" or "exclude". For example,

ctype ordered: 10 23 45

defines charactes 10, 23, and 45 to be of type ordered. Similarly,

ctype irreversible: 54 - 67 71-92

defines characters 54 to 67 and characters 71 to 92 to be of type irreversible. You can use the "." to denote the last character, and "all" to denote all of the characters. Finally, you can use the specifier "\" to apply the ordering to every n-th character or you can use predefined charsets to specify the character.

Only one ordering can be used on any specific application of ctype.

If you want to apply different orderings to different characters, then you need to use ctype multiple times. For example,

ctype ordered: 1-50

ctype irreversible: 51-100

sets characters 1 to 50 to be ordered and characters 51 to 100 to be irreversible.

The ctype command is only sensible with morphological (here called "standard") characters. The program ignores attempts to apply character orderings to other types of characters, such as DNA characters.

Databreaks

This command is used to specify breaks in your input data matrix. Your data may be a mixture of genes or a mixture of different types of data. Some of the models implemented by MrBayes account for nonindependence at adjacent characters. The autocorrelated gamma model, for example, allows rates at adjacent sites to be correlated. However, there is no way for such a model to tell whether two sites, adjacent in the matrix, are actually separated by many kilobases or megabases in the genome. The databreaks command allows you to specify such breaks. The correct usage is:

databreaks <break 1> <break 2> <break 3> ...

For example, say you have a data matrix of 3204 characters that include nucleotide data from three genes. The first gene covers characters 1 to 970, the second gene covers characters 971 to 2567, and the third gene covers characters 2568 to 3204. Also, let's assume that the genes are not directly adjacent to one another in the genome, as might be likely if you have mitochondrial sequences. In this case, you can specify breaks between the genes using:

databreaks 970 2567;

The first break, between genes one and two, is after character 970 and the second break, between genes two and three, is after character 2567.

Delete

This command deletes taxa from the analysis. The correct usage is:

delete <name and/or number and/or taxset> ...

A list of the taxon names or taxon numbers (labelled 1 to ntax in the order in the matrix) or taxset(s) can be used. For example, the following:

delete 1 2 Homo_sapiens

deletes taxa 1, 2, and the taxon labelled Homo_sapiens from the analysis. You can also use "all" to delete all of the taxa. For example,

delete all

deletes all of the taxa from the analysis. Of course, a phylogenetic analysis that does not include any taxa is fairly uninteresting. ______ ______ Deroot This command deroots the user tree. If the tree is already unrooted, a warning is issued. The correct usage is "deroot". ______ Disclaimer This command shows the disclaimer for the program. In short, the disclaimer states that the authors (John Huelsenbeck and Fredrik Ronquist) are not responsible for any silly things you may do to your computer or any unforseen but possibly nasty things the computer program may inadvertently ______ ______ Exclude This command excludes characters from the analysis. The correct usage is exclude <number> <number> <number> or exclude <number> - <number> or exclude <charset> or some combination thereof. Moreover, you can use the specifier "\" to exclude every nth character. For example, the following exclude $1-100\3$ would exclude every third character. As a specific example, exclude 2 3 10-14 22 excludes sites 2, 3, 10, 11, 12, 13, 14, and 22 from the analysis. Also, exclude all excludes all of the characters from the analysis. Excluding all characters does not leave you much information for inferring phylogeny. ______ ______ Execute This command executes a file called <file name>. The correct usage is: execute <file name>

For example,

execute replicase.nex

would execute the file named "replicase.nex". This file must be in the same directory as the executable.

Help

This command provides useful information on the use of this program. The correct usage is

help

which gives a list of all available commands with a brief description of each or

help <command>

which gives detailed information on the use of <command>.

Include

This command includes characters that were previously excluded from the analysis. The correct usage is

include <number> <number> <number>

or

include <number> - <number>

or

include <charset>

or some combination thereof. Moreover, you can use the specifier " $\$ " to include every nth character. For example, the following

include $1-100\3$

would include every third character. As a specific example,

include 2 3 10-14 22

includes sites 2, 3, 10, 11, 12, 13, 14, and 22 from the analysis. Also,

include all

includes all of the characters in the analysis. Including all of the characters (even if many of them are bad) is a very total-evidence-like thing to do. Doing this will make a certain group of people very happy. On the other hand, simply using this program would make those same people unhappy.

```
Link
This command links model parameters across partitions of the data. The
correct usage is:
   link <parameter name> = (<all> or <partition list>)
The list of parameters that can be linked includes:
   Tratio
                  -- Transition/transversion rate ratio
   Revmat
                  -- Substitution rates of GTR model
   Omega
                  -- Nonsynonymous/synonymous rate ratio
   Statefreq -- Character state frequencies
Shape -- Gamma shape parameter
   Shape
                  -- Gamma shape parameter
  Shape -- Gamma Shape parameter

Pinvar -- Proportion of invariable sites

Correlation -- Correlation parameter of autodiscrete gamma

Switchrates -- Switching rates for covarion model

The largeths of tree
   Brlens
                  -- Branch lengths of tree
                  -- Topology of tree
   Speciationrates -- Speciation rates for birth-death process
   Extinctionrates -- Extinction rates for birth-death process
           -- Parameter for coalescence process
   Growthrate -- Growth rate of coalescence process
For example,
   link shape=(all)
links the gamma shape parameter across all partitions of the data.
You can use "showmodel" to see the current linking status of the
characters. For more information on this command, see the help menu
for link's converse, unlink ("help unlink");
______
Log
This command allows output to the screen to also be output to a file.
The useage is:
   log start/stop filename=<name> append/replace
The options are:
Start/Stop
            -- Starts or stops logging of output to file.
Append/Replace -- Either append to or replace existing file.
Filename -- Name of log file (currently, the name of the log
                 file is "log.out").
______
```

This command sets the parameters of the likelihood model. The likelihood function is the probability of observing the data conditional on the phylogenetic model. In order to calculate the likelihood, you must assume a model of character change. This command lets you tailor the biological assumptions made in the phylogenetic model. The correct usage is

lset <parameter>=<option> ... <parameter>=<option>

For example, "lset nst=6 rates=gamma" would set the model to a general model of DNA substition (the GTR) with gamma-distributed rate variation across sites.

Options:

-- This option allows you to apply the lset commands to specific partitions. This command should be the first in the list of commands specified in lset. Moreover, it only makes sense to be using this command if the data have been partitioned. A default partition is set on execution of a matrix. If the data are homogeneous (i.e., all of the same data type), then this partition will not subdivide the characters. Up to 30 other partitions can be defined, and you can switch among them using "set partition=<partition name>". Now, you may want to specify different models to different partitions of the data. Applyto allows you to do this. For example, say you have partitioned the data by codon position, and you want to apply a nst=2 model to the first two partitions and nst=6 to the last. This could be implemented in two uses of lset:

lset applyto=(1,2) nst=2

lset applyto=(3) nst=6

The first applies the parameters after "applyto" to the first and second partitions. The second lset applies nst=6 to the third partition. You can also use applyto=(all), which attempts to apply the parameter settings to all of the data partitions. Importantly, if the option is not consistent with the data in the partition, the program will not apply the lset option to that partition.

Nucmodel

-- This specifies the general form of the nucleotide substitution model. The options are "4by4" [the standard model of DNA substitution in which there are only four states (A,C,G,T/U)], "doublet" (a model appropriate for modelling the stem regions of ribosomal genes where the state space is the 16 doublets of nucleotides), and "codon" (the substitution model is expanded around triplets of nucleotides--a codon).

Nst

-- Sets the number of substitution types: "1" constrains all of the rates to be the same (e.g., a JC69 or F81 model); "2" allows transitions and transversions to have potentially different rates (e.g., a K80 or HKY85 model); "6" allows all rates to be different, subject to the constraint of time-reversibility (e.g., a GTR model).

Code

-- Enforces the use of a particular genetic code. The default is the universal code. Other options include "vertmt" for vertebrate mitocondrial DNA, "mycoplasma", "yeast", "ciliates", and "metmt" (for metazoan mitochondrial DNA except vertebrates).

Ploidy

-- Specifies the ploidy of the organism. Options are "Haploid" or "Diploid". This option is used when a coalescence prior is used on trees.

Rates

-- Sets the model for among-site rate variation. In general, the

rate at a site is considered to be an unknown random variable. The valid options are:

- -- No rate variation across sites. * equal
- -- Gamma-distributed rates across sites. The rate * gamma at a site is drawn from a gamma distribution. The gamma distribution has a single parameter that describes how much rates vary.
- -- Autocorrelated rates across sites. The marg-* adgamma inal rate distribution is gamma, but adjacent sites have correlated rates.
- * propinv -- A proportion of the sites are invariable.
- * invgamma -- A proportion of the sites are invariable while the rate for the remaining sites are drawn from a gamma distribution.

Note that MrBayes versions 2.0 and earlier supported options that allowed site specific rates (e.g., ssgamma). In versions 3.0 and later, site specific rates are allowed, but set using the 'prset ratepr' command for each partition.

- Ngammacat -- Sets the number of rate categories for the gamma distribution. The gamma distribution is continuous. However, it is virtually impossible to calculate likelihoods under the continuous gamma distribution. Hence, an approximation to the continuous gamma is used; the gamma distribution is broken into neat categories of equal weight (1/ncat). The mean rate for each category represents the rate for the entire cateogry. This option allows you to specify how many rate categories to use when approximating the gamma. The approximation is better as neat is increased. In practice, "ncat=4" does a reasonable job of approximating the continuous gamma.
- Nbetacat -- Sets the number of rate categories for the beta distribution. A symmetric beta distribution is used to model the stationary frequencies when morphological data are used. This option specifies how well the beta distribution will be approximated.
- -- Allows the nonsynonymous/synonymous rate ratio (omega) to vary across codons. Ny98 assumes that there are three classes, with potentially different omega values (omega1, omega2, omega3): omega2 = 1; 0 < omega1 <1; and omega3 > 1. Like the Ny98 model, the M3 model has three omega classes. However, their values are less constrained, with omega1 < omega2 < omega3. The default (omegavar = equal) has no variation on omega across sites.
- -- This forces the use of a covarion-like model of substitution Covarion for nucleotide or amino acid data. The valid options are "yes" and "no". The covarion model allows the rate at a site to change over its evolutionary history. Specifically, the site is either on or off. When it is off, no substitutions are possible. When the process is on, substitutions occur according to a specified substitution model (specified using the other lset options).
- Coding -- This specifies how characters were sampled. If all site patterns had the possibility of being sampled, then "all" should be specified (the default). Otherwise "variable" (only variable characters had the possibility of being sampled), "noabsence" (characters for which all taxa were coded as absent were not sampled), and "nopresence" (characters for which all taxa were coded as present were not sampled. "All" works for all data types. However, the others only work for

- Omegavar

morphological (all/variable) or restriction site (all/variable/noabsence/nopresence) data.

Parsmodel -- This forces calculation under the so-called parsimony model described by Tuffley and Steel (1998). The options are "yes" or "no". Note that the biological assumptions of this model are anything but parsimonious. In fact, this model assumes many more parameters than the next most complicated model implemented in this program. If you really believe that the parsimony model makes the biological assumptions described by Tuffley and Steel, then the parsimony method is miss-named.

Default model settings:

Parameter	Options	Current Setting
Nucmodel	4by4/Doublet/Codon	4by4
Nst	1/2/6	1
Code	Universal/Vertmt/Mycoplasma/	
	Yeast/Ciliates/Metmt	Universal
Ploidy	Haploid/Diploid	Diploid
Rates	Equal/Gamma/Propinv/Invgamma/Adgamma	Equal
Ngammacat	<number></number>	4
Nbetacat	<number></number>	5
Omegavar	Equal/Ny98/M3	Equal
Covarion	No/Yes	No
Coding	All/Variable/Noabsencesites/	
	Nopresencesites	All
Parsmodel	No/Yes	No

Manual

This command allows you to generate a text file containing help information on all the available commands. This text file can be used as an up-to-date command reference. You can set the name of the text file using the "filename" option; the default is "commref_mb<version>.txt".

Parameter	Uptions	Current Setting
Filename	<name></name>	commref_mb3.1.txt
Mcmc		

This command starts the Markov chain Monte Carlo (MCMC) analysis. The posterior probability of phylogenetic trees (and other parameters of the substitution model) cannot be determined analytically. Instead, MCMC is used to approximate the posterior probabilities of trees by drawing (dependent) samples from the posterior distribution. This program can implement a variant of MCMC called "Metropolis-coupled Markov chain Monte Carlo", or MCMCMC for short. Basically, "Nchains" are run, with Nchains - 1 of them heated. The chains are labelled 1, 2, ..., Nchains. The heat that is applied to the i-th chain is B = 1 / (1 + temp X i). B is the power to which the posterior probability is raised. When B = 0, all trees have equal probability and the chain freely visits trees. B = 1 is

the "cold" chain (or the distribution of interest). MCMCMC can mix better than ordinary MCMC; after all of the chains have gone through one cycle, two chains are chosen at random and an attempt is made to swap the states (with the probability of a swap being determined by the Metropolis et al. equation). This allows the chain to potentially jump a valley in a single bound. The correct usage is

mcmc <parameter> = <value> ... <parameter> = <value>

For example,

mcmc ngen=100000 nchains=4 temp=0.5

performs a MCMCMC analysis with four chains with the temperature set to 0.5. The chains would be run for 100,000 cycles.

Options:

Swapfreq

Seed	Sets the seed number for the random number generator. The random number seed is initialized haphazardly at the beginning of each MrBayes session. This option allows you to set the seed to some specific value, thereby allowing you to exactly repeat an analysis. If the analysis uses swapping between cold and heated chains, you must also set the swap seed (see below) to exactly repeat the analysis.
Swapseed	Sets the seed used for generating the swapping sequence when Metropolis-coupled heated chains are used. This seed is initialized haphazardly at the beginning of each MrBayes session. This option allows you to set the seed to some specific value, thereby allowing you to exactly repeat a swap sequence. See also the 'Seed' option.
Ngen	This option sets the number of cycles for the MCMC alg- orithm. This should be a big number as you want the chain to first reach stationarity, and then remain there for enough time to take lots of samples.
Nruns	How many independent analyses are started simultaneously.
Nchains	How many chains are run for each analysis for the MCMCMC variant. The default is 4: 1 cold chain and 3 heated chains. If Nchains is set to 1, MrBayes will use regular MCMC sampling, without heating.
Temp	The temperature parameter for heating the chains. The higher the temperature, the more likely the heated chains are to move between isolated peaks in the posterior distribution. However, excessive heating may lead to very low acceptance rates for swaps between different chains. Before changing the default setting, however, note that the acceptance rates of swaps tend to fluctuate during the burn-in phase of the run.
Reweight	Here, you specify three numbers, that respectively represent the percentage of characters to decrease in weight, the percentage of characters to increase in weight, and the increment. An increase/decrease in weight is acheived by replicating/removing a character in the matrix. This is only done to non-cold chains. The format for this parameter is "reweight=(<number>,<number>)" or "reweight=(<number>,"</number></number></number>

-- This specifies how often swaps of states between chains are

attempted. You must be running at least two chains for this

<number>,<number>)".

option to be relevant. The default is Swapfreq=1, resulting in Nswaps (see below) swaps being tried each generation of the run. If Swapfreq is set to 10, then Nswaps swaps will be tried every tenth generation of the run. Nswaps -- The number of swaps tried for each swapping generation of the chain (see also Swapfreq). Samplefreq -- This specifies how often the Markov chain is sampled. You can sample the chain every cycle, but this results in very large output files. Thinning the chain is a way of making these files smaller and making the samples more independent. Printfreq -- This specifies how often information about the chain is printed to the screen. Printall -- If set to NO, only cold chains in a MCMC analysis are printed to screen. If set to YES, both cold and heated chains will be output. This setting only affects the printing to screen, it does not change the way values are written to file. Printmax -- The maximum number of chains to print to screen. Mcmcdiagn -- Determines whether acceptance ratios of moves and swaps will be printed to file. The file will be named similarly to the '.p' and '.t' files, but will have the ending '.mcmc'. If more than one independent analysis is run simultaneously (see Nruns below), convergence diagnostics for tree topology will also be printed to this file. The convergence diagnostic used is the average standard deviation in partition frequency values across independent analyses. The Burnin setting (see below) determines how many samples will be discarded as burnin before calculating the partition frequencies. The Minpartfreq setting (see below) determines the minimum partition frequency required for a partition to be included in the calculation. As the independent analyses approach stationarity (converge), the value of the diagnostic is expected to approach zero. Diagnfreq -- The number of generations between the calculation of MCMC diagnostics (see Mcmcdiagn above). Minpartfreq -- The minimum frequency required for a partition to be included in the calculation of the topology convergence diagnostic. The partition is included if the minimum frequency is reached in at least one of the independent tree samples that are compared. -- If this option is set to YES, acceptance ratios for moves are Allchains recorded for all chains, cold or heated. By default, only the acceptance ratios for the cold chain are recorded. -- If this option is set to YES, topological convergence diag-Allcomps nostics are calculated over all pairwise comparisons of runs. If it is set to NO, only the overall value is reported. -- If this option is set to YES, then a proportion of the Relburnin sampled values will be discarded as burnin when calculating the convergence diagnostic. The proportion to be discarded is set with Burninfrac (see below). By default, the Relburnin option is set to NO, resulting in a specific number of samples being discarded instead. This number is set by Burnin (see below).

-- Determines the number of samples (not generations) that will

Burnin

be discarded when convergence diagnostics are calculated. The value of this option is only relevant when Relburnin is set to NO.

BurninFrac -- Determines the fraction of samples that will be discarded when convergence diagnostics are calculated. The value of this option is only relevant when Relburnin is set to YES.

Example: A value for this option of 0.25 means that 25

4525774f

the samples will be discarded.

Stoprule -- If this option is set to NO, then the chain is run the number of generations determined by Ngen. If it is set to YES, and topological convergence diagnostics are calculated (Mcmcdiagn is set to YES), then the chain will be stopped before the

pre-

determined number of generations if the convergence

diagnostic

falls below the stop value.

Stopval -- The critical value for the topological convergence diagnostic.

Only used when Stoprule and Mcmcdiagn are set to yes, and more than one analysis is run simultaneously (Nruns > 1).

Filename -- The name of the files that will be generated. Two files are generated: "<Filename>.t" and "<Filename>.p".

The .t file contains the trees whereas the .p file contains the sampled values of the parameters.

Startingtree -- The starting tree for the chain can either be randomly selected or user-defined. It might be a good idea to start from randomly chosen trees; convergence seems likely if independently run chains, each of which started from different random trees, converge to the same answer.

Nperts -- This is the number of random perturbations to apply to the user starting tree. This allows you to have something between completely random and user-defined trees start the chain.

Savebrlens -- This specifies whether branch length information is saved on the trees.

Ordertaxa -- Determines whether taxa should be ordered before trees are printed to file. If set to 'Yes', terminals in the sampled trees will be reordered to match the order of the taxa in the data matrix as closely as possible. By default, trees will be printed without reordering of taxa.

Parameter	Options	Current Setting
Seed Swapseed Ngen Nruns Nchains Temp	<number> <number> <number> <number> <number> <number> <number> <number></number></number></number></number></number></number></number></number>	1116355510 1116355510 1000000 2 4 0.200000
Reweight Swapfreq Nswaps Samplefreq Printfreq Printall	<number>,<number> <number> <number> <number> <number> <number> <number> Yes/No</number></number></number></number></number></number></number></number>	0.00 v 0.00 ^ 1 1 100 100 Yes

Printmax	<number></number>	8
Mcmcdiagn	Yes/No	Yes
Diagnfreq	<number></number>	1000
Minpartfreq	<number></number>	0.10
Allchains	Yes/No	No
Allcomps	Yes/No	No
Relburnin	Yes/No	Yes
Burnin	<number></number>	0
Burninfrac	<number></number>	0.25
Stoprule	Yes/No	No
Stopval	<number></number>	0.01
Filename	<name></name>	temp.out. <p t=""></p>
Startingtree	Random/User	Random
Nperts	<number></number>	0
Savebrlens	Yes/No	Yes
Ordertaxa	Yes/No	No

Mcmcp

This command sets the parameters of the Markov chain Monte Carlo (MCMC) analysis without actually starting the chain. This command is identical in all respects to Mcmc, except that the analysis will not start after this command is issued. For more details on the options, check the help menu for Mcmc.

Parameter	Options	Current Setting
Seed Swapseed	<number></number>	1116355510 1116355510
Ngen	<number></number>	1000000
Nruns	<number></number>	2
Nchains	<number></number>	4
Temp	<number></number>	0.200000
Reweight	<number>,<number></number></number>	0.00 v 0.00 ^
Swapfreq	<number></number>	1
Nswaps	<number></number>	1
Samplefreq	<number></number>	100
Printfreq	<number></number>	100
Printall	Yes/No	Yes
Printmax	<number></number>	8
Mcmcdiagn	Yes/No	Yes
Diagnfreq	<number></number>	1000
Minpartfreq	<number></number>	0.10
Allchains	Yes/No	No
Allcomps	Yes/No	No
Relburnin	Yes/No	Yes
Burnin	<number></number>	0
Burninfrac	<number></number>	0.25
Stoprule	Yes/No	No
Stopval	<number></number>	0.01
Filename	<name></name>	temp.out. <p t=""></p>
Startingtree	Random/User	Random
Nperts	<number></number>	0
Savebrlens	Yes/No	Yes
Ordertaxa	Yes/No	No

This command assigns a taxon to the outgroup. The correct usage is:

outgroup <number>/<taxon name>

For example, "outgroup 3" assigns the third taxon in the matrix to be the outgroup. Similarly, "outgroup Homo_sapiens" assings the taxon "Homo_sapiens" to be the outgroup (assuming that there is a taxon named "Homo_sapiens" in the matrix). Only a single taxon can be assigned to be the outgroup.

Pairs

This command is used to specify pairs of nucleotides. For example, your data may be RNA sequences with a known secondary structure of stems and loops. Substitutions in nucleotides involved in a Watson-Crick pairing in stems are not strictly independent; a change in one changes the probability of a change in the partner. A solution to this problem is to expand the model around the pair of nucleotides in the stem. This command allows you to do this. The correct usage is:

pairs <NUC1>:<NUC2>, <NUC1>:<NUC2>,..., <NUC1>:<NUC2>;

For example,

pairs 30:56, 31:55, 32:54, 33:53, 34:52, 35:51, 36:50;

specifies pairings between nucleotides 30 and 56, 31 and 55, etc. Only nucleotide data (DNA or RNA) may be paired using this command. Note that in order for the program to actually implement a "doublet" model involving a 16 X 16 rate matrix, you must specify that the structure of the model is 16×16 using "lset nucmodel=doublet".

Partition

This command allows you to specify a character partition. The format for this command is

partition <name> = <num parts>:<chars in first>, ...,<chars in last>

For example, "partition by_codon = 3:1st_pos,2nd_pos,3rd_pos" specifies a partition called "by_codon" which consists of three parts (first, second, and third codon positions). Here, we are assuming that the sites in each partition were defined using the charset command. You can specify a partition without using charset as follows:

partition by_codon = 3:1 4 6 9 12,2 5 7 10 13,3 6 8 11 14

However, we recommend that you use the charsets to define a set of characters and then use these predefined sets when defining the partition. Also, it makes more sense to define a partition as a line in the mrbayes

block than to issue the command from the command line (then again, you may be a masochist, and want to do extra work).

Plot

This command plots specified parameters in the .p file created by the program. The program prints two files during a MCMC analysis: a tree file and a parameter file. The parameter file has the extension ".p". This command, plot, makes an x-y graph of the parameter over the course of the chain. The command can be useful for visually diagnosing convergence for many of the parameters of the phylogenetic model. The parameter to be plotted is specified by the "parameter" option. Several parameters can be plotted at once by using the "match" option, which has a default value of "perfect". For example, if you were to set "parameter = pi" and "match = consistentwith", then all of the state frequency parameters would be plotted. You can also set "match=all", in which case all of the parameters are plotted.

Parameter	Options	Current Setting
Filename Burnin Parameter Match	<name> <name> <name> Perfect/Consistentwith/All</name></name></name>	temp.p 0 lnL Perfect

Prset

This command sets the priors for the phylogenetic model. Remember that in a Bayesian analysis, you must specify a prior probability distribution for the parameters of the likelihood model. The prior distribution represents your prior beliefs about the parameter before observation of the data. This command allows you to tailor your prior assumptions to a large extent.

Options:

Applyto

-- This option allows you to apply the prset commands to specific partitions. This command should be the first in the list of commands specified in prset. Moreover, it only makes sense to be using this command if the data have been partitioned. A default partition is set on execution of a matrix. If the data are homogeneous (i.e., all of the same data type), then this partition will not subdivide the characters. Up to 30 other partitions can be defined, and you can switch among them using "set partition=<partition name>". Now, you may want to specify different priors to different partitions of the data. Applyto allows you to do this. For example, say you have partitioned the data by codon position, and you want to fix the statefregs to equal for the first two partitions but apply a flat Dirichlet prior to the statefreqs of the last. This could be implemented in two uses of prset:

```
prset applyto=(1,2) statefreqs=fixed(equal)
prset applyto=(3) statefreqs=dirichlet(1,1,1,1)
```

The first applies the parameters after "applyto" to the first and second partitions. The second prset applies a flat Dirichlet to the third partition. You can also use applyto=(all), which attempts to apply the parameter settings to all of the data partitions. Importantly, if the option is not consistent with the data in the partition, the program will not apply the prset option to that partition.

Tratiopr

-- This parameter sets the prior for the transition/transversion rate ratio (tratio). The options are:

```
prset tratiopr = beta(<number>, <number>)
prset tratiopr = fixed(<number>)
```

The program assumes that the transition and transversion rates are independent gamma-distributed random variables with the same scale parameter when beta is selected. If you want a diffuse prior that puts equal emphasis on transition/transversion rate ratios above 1.0 and below 1.0, then use a flat Beta, beta(1,1), which is the default. If you wish to concentrate this distribution more in the equal-rates

region,

then use a prior of the type beta(x,x), where the magnitude of x determines how much the prior is concentrated in the equal rates region. For instance, a beta(20,20) puts more probability on rate ratios close to 1.0 than a beta(1,1). If you think it is likely that the transition/transversion rate ratio is 2.0, you can use a prior of the type beta(2x,x), where x determines how strongly the prior is concentrated on tratio values near 2.0. For instance, a beta(2,1) is much more diffuse than a beta(80,40) but both have the expected tratio 2.0 in the absence of data. The parameters of the Beta can be interpreted as counts: if you have observed x transitions and y transversions, then a beta(x+1,y+1) is a good representation of this information. The fixed option allows you to fix the tratio to a particular value.

Revmatpr

-- This parameter sets the prior for the substitution rates of the GTR model for nucleotide data. The options are:

The program assumes that the six substitution rates are independent gamma-distributed random variables with the same scale parameter when dirichlet is selected. The six numbers in brackets each corresponds to a particular substitution type. Together, they determine the shape of the

prior.

The six rates are in the order A<->C, A<->G, A<->T, C<->G, C<->T, and G<->T. If you want an uninformative prior you can use dirichlet(1,1,1,1,1,1), also referred to as a 'flat' Dirichlet. This is the default setting. If you wish a prior

where the C<->T rate is 5 times and the A<->G rate 2 times higher, on average, than the transversion rates, which are all the same, then you should use a prior of the form dirichlet(x,2x,x,x,5x,x), where x determines how much the prior is focused on these particular rates. For more info, see tratiopr. The fixed option allows you to fix the substitution rates to particular values.

Aamodelpr

-- This parameter sets the rate matrix for amino acid data. You can either fix the model by specifying aamodelpr= fixed(<model name>), where <model name> is 'poisson' (a glorified Jukes-Cantor model), 'jones', 'dayhoff', 'mtrev', 'mtmam', 'wag', 'rtrev', 'cprev', 'vt', 'blosum', 'equalin' (a glorified Felsenstein 1981 model), or 'gtr'. You can also average over the first ten models by specifying aamodelpr= mixed. If you do so, the Markov chain will sample each model according to its probability. The sampled model is reported as an index: poisson(0), jones(1), dayhoff(2), mtrev(3), mtmam(4), wag(5), rtrev(6), cprev(7), vt(8), or blosum(9). The 'Sump' command summarizes the MCMC samples and

calculates

the posterior probability estimate for each of these models.

Aarevmatpr -- This parameter sets the prior for the substitution rates of the GTR model for amino acid data. The options are:

The options are the same as those for 'Revmatpr' except that they are defined over the 190 rates of the time-reversible GTR model for amino acids instead of over the 6 rates of the GTR model for nucleotides. The rates are in the order A<->R, A<->N, etc to Y<->V. In other words, amino acids are listed in alphabetic order based on their full name. The first

amino

acid (Alanine) is then combined in turn with all amino acids following it in the list, starting with amino acid 2 (Arginine) and finishing with amino acid 20 (Valine). The second amino acid (Arginine) is then combined in turn with all

amino

acids following it, starting with amino acid 3 (Asparagine)
and finishing with amino acid 20 (Valine), and so on.
-- This parameter specifies the prior on the nonsynonymous/
synonymous rate ratio. The options are:

Omegapr

prset omegapr = uniform(<number>,<number>)
prset omegapr = exponential(<number>)
prset omegapr = fixed(<number>)

This parameter is only in effect if the nucleotide substitution model is set to codon using the lset command (lset nucmodel=codon). Moreover, it only applies to the case when there is no variation in omega across sites (i.e., "lset omegavar=equal").

Ny98omega1pr

-- This parameter specifies the prior on the nonsynonymous/ synonymous rate ratio for sites under purifying selection. The options are:

```
prset Ny98omegalpr = beta(<number>,<number>)
prset Ny98omegalpr = fixed(<number>)
```

This parameter is only in effect if the nucleotide substitution model is set to codon using the lset command (lset nucmodel=codon). Moreover, it only applies to the case where omega varies across sites using the model of Nielsen and Yang (1998) (i.e., "lset omegavar=ny98"). If fixing the parameter, you must specify a number between 0 and 1.

Ny98omega3pr

-- This parameter specifies the prior on the nonsynonymous/ synonymous rate ratio for positively selected sites. The options are:

```
prset Ny98omega3pr = uniform(<number>,<number>)
prset Ny98omega3pr = exponential(<number>)
prset Ny98omega3pr = fixed(<number>)
```

This parameter is only in effect if the nucleotide substitution model is set to codon using the lset command (lset nucmodel=codon). Moreover, it only applies to the case where omega varies across sites according to the NY98 model. Note that if the NY98 model is specified that this parameter must be greater than 1, so you should not specify a uniform(0,10) prior, for example.

M3omegapr

-- This parameter specifies the prior on the nonsynonymous/ synonymous rate ratios for all three classes of sites for the M3 model. The options are:

```
prset M3omegapr = exponential
prset M3omegapr = fixed(<number>,<number>,<number>)
```

This parameter is only in effect if the nucleotide substitution model is set to codon using the lset command (lset nucmodel=codon). Moreover, it only applies to the case where omega varies across sites using the M3 model of Yang et al. (2000) (i.e., "lset omegavar=M3"). Under the exponential prior, the four rates (dN1, dN2, dN3, and dS) are all considered to be independent draws from the same exponential distribution (the parameter of the exponential does not matter, and so you don't need to specify it). The rates dN1, dN2, and dN3 are taken to be the order statistics with dN1 < dN2 < dN3. These three rates are all scaled to the same synonymous rate, dS. The other option is to simply fix the three rate ratios to some values.

Codoncatfreqs -- This parameter specifies the prior on frequencies of sites under purifying, neutral, and positive selection. The options are:

```
prset codoncatfreqs = dirichlet(<num>,<num>,<num>)
prset codoncatfreqs = fixed(<number>,<number>,<number>)
```

This parameter is only in effect if the nucleotide substitution model is set to codon using the 1set command (lset nucmodel=codon). Moreover, it only applies to the case where omega varies across sites using the models of

Nielsen and Yang (1998) (i.e., "lset omegavar=ny98") or Yang et al. (2000) (i.e., "lset omegavar=M3") Note that the sum of the three frequencies must be 1. -- This parameter specifies the prior on the state freq-Statefreqpr uencies. The options are: prset statefreqpr = dirichlet(<number>) prset statefreqpr = dirichlet(<number>,...,<number>) prset statefreqpr = fixed(equal) prset statefreqpr = fixed(empirical) prset statefreqpr = fixed(<number>,...,<number>) For the dirichlet, you can specify either a single number or as many numbers as there are states. If you specify a single number, then the prior has all states equally probable with a variance related to the single parameter passed in. Shapepr -- This parameter specifies the prior for the gamma shape parameter for among-site rate variation. The options are: prset shapepr = uniform(<number>,<number>) prset shapepr = exponential(<number>) prset shapepr = fixed(<number>) -- This parameter specifies the prior for the proportion of Pinvarpr invariable sites. The options are: prset pinvarpr = uniform(<number>,<number>) prset pinvarpr = fixed(<number>) Note that the valid range for the parameter is between 0 and 1. Hence, "prset pinvarpr=uniform(0,0.8)" is valid while "prset pinvarpr=uniform(0,10)" is not. The default setting is "prset pinvarpr=uniform(0,1)". -- This parameter specifies the prior for the autocorrelation Ratecorrpr parameter of the autocorrelated gamma distribution for among-site rate variation. The options are: prset ratecorrpr = uniform(<number>,<number>) prset ratecorrpr = fixed(<number>) Note that the valid range for the parameter is between -1 and 1. Hence, "prset ratecorrpr=uniform(-1,1)" is valid while "prset ratecorrpr=uniform(-11,10)" is not. The default setting is "prset ratecorrpr=uniform(-1,1)". Covswitchpr -- This option sets the prior for the covarion switching rates. The options are: prset covswitchpr = uniform(<number>,<number>) prset covswitchpr = exponential(<number>) prset covswitchpr = fixed(<number>,<number>) The covarion model has two rates: a rate from on to off and a rate from off to on. The rates are assumed to have independent priors that individually are either uniformly

or exponentially distributed. The other option is to fix the switching rates, in which case you must specify

both rates. (The first number is off->on and the second is on->off).

Symdirihyperpr - This option sets the prior for the stationary frequencies of the states for morphological (standard) data. There can be as many as 10 states for standard data. However, the labelling of the states is somewhat arbitrary. For example, the state "1" for different characters does not have the same meaning. This is not true for DNA characters, for example, where a "G" has the same meaning across characters. The fact that the labelling of morphological characters is arbitrary makes it difficult to allow unequal characterstate frequencies. MrBayes gets around this problem by assuming that the states have a dirichlet prior, with all states having equal frequency. The variation in the dirichlet can be controlled by this parameter--symdirihyperpr. Symdirihyperpr specifies the distribution on the variance parameter of the dirichlet. The valid options are:

```
prset Symdirihyperpr = uniform(<number>,<number>)
prset Symdirihyperpr = exponential(<number>)
prset Symdirihyperpr = fixed(<number>)
prset Symdirihyperpr = fixed(infinity)
```

Topologypr

If "fixed(infinity)" is chosen, the dirichlet prior is fixed such that all character states have equal frequency. -- This parameter specifies the prior probabilities of phylogenies. The options are:

```
prset topologypr = uniform
prset topologypr = constraints(<list>)
```

If the prior is selected to be "uniform", the default, then all possible trees are considered a priori equally probable. The constraints option allows you to specify complicated prior probabilities on trees (constraints are discussed more fully in "help constraint"). Note that you must specify a list of constraints that you wish to be obeyed. The list can be either the constraints' name or number. Also, note that the constraints simply tell you how much more (or less) probable individual trees are that possess the constraint than trees not possessing the constraint.

Brlenspr

-- This parameter specifies the prior probability distribution on branch lengths. The options are:

```
prset brlenspr = unconstrained:uniform(<num>,<num>)
prset brlenspr = unconstrained:exponential(<number>)
prset brlenspr = clock:uniform
prset brlenspr = clock:birthdeath
prset brlenspr = clock:coalescence
```

Trees with unconstrained branch lengths are unrooted whereas clock-constrained trees are rooted. The option after the colon specifies the details of the probability density of branch lengths. If you choose a birth-death or coalescence prior, you may want to modify the details of the parameters of those processes.

Treeheightpr -- This parameter specifies the prior probability distribution on the tree height, when a clock model is specified. The options are:

> prset treeheightpr = Gamma(<num>,<num>) prset treeheightpr = Exponential(<number>)

(And, yes, we know the exponential is a special case of the gamma distribution.) The tree height is the expected number of substitutions on a single branch that extends from the root of the tree to the tips. This parameter does not come into play for the coalescence prior. It insures that the prior probability distribution for unconstrained and birth-death models is proper.

Ratepr

-- This parameter allows you to specify the site specific rates model. First, you must have defined a partition of the characters. For example, you may define a partition that divides the characters by codon position, if you have DNA data. Second, you must make that partition the active one using the set command. For example, if your partition is called "by_codon", then you make that the active partition using "set partition=by_codon". Now that you have defined and activated a partition, you can specify the rate multipliers for the various partitions. The options are:

```
prset ratepr = fixed
prset ratepr = variable
prset ratepr = dirichlet(<number>,<number>,...,<number>)
```

If you specify "fixed", then the rate multiplier for that partition is set to 1 (i.e., the rate is fixed to the average rate across partitions). On the other hand, if you specify "variable", then the rate is allowed to vary across partitions subject to the constraint that the average rate of substitution across the partitions is 1. You must specify a variable rate prior for at least two partitions, otherwise the option is not activated when calculating likelihoods. The variable option automatically associates the partition rates with a dirichlet(1, ..., 1)prior. The dirichlet option is an alternative way of setting a partition rate to be variable, and also gives accurate control of the shape of the prior. The parameters of the Dirichlet are listed in the order of the partitions that the ratepr is applied to. For instance, "prset applyto=(1,3,4) ratepr = dirichlet(10,40,15)" would set the Dirichlet parameter 10 to partition 1, 40 to partition 3, and 15 to partition 4.

Speciationpr -- This parameter sets the prior on the speciation rate. The options are:

```
prset speciationpr = uniform(<number>,<number>)
prset speciationpr = exponential(<number>)
prset speciationpr = fixed(<number>)
```

This parameter is only relevant if the birth-death process is selected as the prior on branch lengths. prset extinctionpr = uniform(<number>,<number>,
prset extinctionpr = exponential(<number>)
prset extinctionpr = fixed(<number>)

This parameter is only relevant if the birth-death process is selected as the prior on branch lengths.

-- This parameter sets the fraction of species that are sampled in the analysis. This is used with the birth-death prior on trees (see Yang and Rannala, 1997).

-- This parameter sets the prior on the coalescence para-

-- This parameter sets the prior on the coalescence parameter. The options are:

prset thetapr = uniform(<number>,<number>)
prset thetapr = exponential(<number>)
prset thetapr = fixed(<number>)

This parameter is only relevant if the coalescence process is selected as the prior on branch lengths.

Default model settings:

Sampleprob

Thetapr

Parameter	Options	Current Setting
Tratiopr	Beta/Fixed	Beta(1.0,1.0)
Revmatpr	Dirichlet/Fixed	
Dirichlet(1.0,1.0,1	.0,1.0,1.0,1.0)	
Aamodelpr	Fixed/Mixed	Fixed(Poisson)
Aarevmatpr	Dirichlet/Fixed	Dirichlet(1.0,1.0,)
Omegapr	Dirichlet/Fixed	Dirichlet(1.0,1.0)
Ny98omega1pr	Beta/Fixed	Beta(1.0,1.0)
Ny98omega3pr	Uniform/Exponential/Fixed	<pre>Exponential(1.0)</pre>
M3omegapr	Exponential/Fixed	Exponential
Codoncatfreqs	Dirichlet/Fixed	Dirichlet(1.0,1.0,1.0)
Statefreqpr	Dirichlet/Fixed	Dirichlet
Treeheightpr	Exponential/Gamma	<pre>Exponential(1.0)</pre>
Ratepr	Fixed/Variable=Dirichlet	Fixed
Shapepr	Uniform/Exponential/Fixed	Uniform(0.0,50.0)
Ratecorrpr	Uniform/Fixed	Uniform(-1.0,1.0)
Pinvarpr	Uniform/Fixed	Uniform(0.0,1.0)
Covswitchpr	Uniform/Exponential/Fixed	Uniform(0.0,100.0)
Symdirihyperpr	Uniform/Exponential/Fixed	Fixed(Infinity)
Topologypr	Uniform/Constraints	Uniform
Brlenspr	Unconstrained/Clock	<pre>Unconstrained:Exp(10.0)</pre>
Speciationpr	Uniform/Exponential/Fixed	Uniform(0.0,10.0)
Extinctionpr	Uniform/Exponential/Fixed	Uniform(0.0,10.0)
Sampleprob	<number></number>	1.00
Thetapr	Uniform/Exponential/Fixed	Uniform(0.0,10.0)

Props

This command allows the user to change the details of the MCMC mechanism that updates the state of the chain. The useage is:

props

On typing "props", you will get a list of parameters to change. The program works as follows: On typing "mcmc", MrBayes figures out which model parameters need to be updated. For example, if you include a transition/transversion rate parameter, then the program needs to update this parameter along with others, such as the tree and branch lengths. Once MrBayes figures out which moves are needed, it figures out the probability of making each move on every cycle of the chain. MrBayes updates parameters in blocks; it decides which parameter to update, changes the parameter, and then accepts or rejects the move according to the Metropolis-Hastings equation. The probability of making a move is calculated as the proposal rate for the move divided by the sum of the proposal rates for all of the other parameters that need to be updated. This command also allows you to change the details of each proposal mechanism. Many of the moves change parameters using sliding windows centered on the current value of the parameter. If you increase or decrease the window size, you will respectively decrease or increase the acceptance rate of the move. Some of the other moves update using a dirichlet or beta distribution, centered on the current values. You can change the variance parameter of the dirichlet or beta distribution. Finally, a few of the topology moves have a tuning parameter which influences the degree to which branch lengths are modified. If you increase this tuning parameter, you will make more radical changes to the branch lengths.

One word of warning: You should be extremely careful when modifying any of the chain parameters using "props". It is quite possible to completely wreck any hope of achieving convergence by inappropriately setting the chain parameters. Please exercise this command with caution.

Quit

This command quits the program. The correct usage is:

quit

It is a very easy command to use properly.

Report

This command allows you to control how the posterior distribution is reported. For rate parameters, it allows you to choose among several popular parameterizations. The report command also allows you to request printing of some model aspects that are usually not reported. For instance, if a node is constrained in the analysis, MrBayes can print the probabilities of the ancestral states at that node. Similarly, if there is rate variation in the model, MrBayes can print the inferred site rates, and if there is omega varia-

tion, MrBayes can print the inferred omega (positive selection) values for each codon. In a complex model with several partitions, each partition is controlled separately using the same 'Applyto' mechanism as in the 'Lset' and 'Prset' commands.

Options:

Applyto -- This option allows you to apply the report commands to specific partitions. This command should be the first in the list of commands specified in 'report'. For example,

report applyto=(1,2) tratio=ratio

report applyto=(3) tratio=dirichlet

would result in the transition and transversion rates of the first and second partitions in the model being reported as a ratio and the transition and transversion rates of the third partition being reported as proportions of the rate sum (the Dirichlet parameterization).

-- This specifies the report format for the transition and transversion rates of a nucleotide substituion model with nst=2. If 'ratio' is selected, the rates will be reported as a ratio (transition rate/transversion rate). If 'dirichlet' is selected, the transition and transversion rates will instead be reported as proportions of the rate sum. For example, if the transition rate is three times the transversion rate and 'ratio' is selected, this will reported as a single value, '3.0'. If 'dirichlet' is selected instead, the same rates will be reported using two values, '0.75 0.25'. The sum of the Dirichlet values is always

> Although the Dirichlet format may be unfamiliar to some users, it is more convenient for specifying priors than the ratio format.

-- This specifies the report format for the substitution rates of a GTR substitution model for nucleotide or amino acid data. If 'ratio' is selected, the rates will be reported scaled to the G-T rate (for nucleotides) or the Y-V rate (for amino acids). If 'dirichlet' is specified instead, the rates are reported as proportions of the rate sum. For instance, assume that the C-T rate is twice the A-G rate and four times the transversion rates, which are equal. If the report format is set to 'ratio', this would be reported as '1.0 2.0 1.0 1.0 4.0 1.0' since the rates are reported in the order rAC, rAG, rAT, rCG, rCT, rGT and

relative to the last rate, the G-T rate. If 'dirichlet' is

ted instead, the same rates would have been reported as '0.1 0.2 0.1 0.1 0.4 0.1' since the rates are now scaled so that they sum to 1.0. The Dirichlet format is the parameterization used for formulating priors on the rates.

Ratemult -- This specifies the report format used for the rate multiplier of different model partitions. Three formats are available. If 'scaled' is selected, then rates are scaled such that the mean rate per site across partitions is 1.0. If 'ratio' is chosen, the rates are scaled relative to the rate of the first partition. Finally, if 'dirichlet' is chosen, the rates are given as proportions of the rate sum. The latter is the format used when formulating priors on the rate multiplier.

Ancstates -- If this option is set to 'yes', MrBayes will print the probability of the ancestral states at all constrained nodes. Typically, you are interested in the ancestral states of only a few

Tratio

1.

Revmat

scaled

selec-

characters and only at one node in the tree. To perform such an analysis, first define and enforce a topology constraint using 'constraint' and 'prset topologypr = constraints (...)'. Then put the character(s) of interest in a separate partition

and

set MrBayes to report the ancestral states for that partition. For instance, if the characters of interest are in partition 2, use 'report applyto=(2) ancestates=yes' to force MrBayes to print the probability of the ancestral states of those characters at the constrained node to the '.p' file.

Siterates -- If this option is set to 'yes' and the relevant model has rate variation across sites, the mean site rate in the posterior will be reported for each site to the '.p' file.

Possel -- If this option is set to 'yes' and the relevant model has omega variation across sites, the mean omega value for each model site (codon in this case) will be written to the '.p' file.

Current settings:

Parameter	Options	Current Setting
Tratio	Ratio/Dir	Ratio
Revmat Ratemult	Ratio/Dir Scaled/Ratio/Dir	Dirichlet Scaled
Ancstates Siterates	Yes/No Yes/No	No No
Possel	Yes/No	No

Restore

This command restores taxa to the analysis. The correct usage is:

restore <name and/or number and/or taxset> ...

A list of the taxon names or taxon numbers (labelled 1 to ntax in the order in the matrix) or taxset(s) can be used. For example, the following:

restore 1 2 Homo_sapiens

restores taxa 1, 2, and the taxon labelled Homo_sapiens to the analysis. You can also use "all" to restore all of the taxa. For example,

restore all

restores all of the taxa to the analysis.

Root

This command roots the tree. If the tree is already rooted, a warning is issued. The tree is rooted at the outgroup species. the correct usage is "root".

Set

This command is used to set some general features of the model or program behavior. The correct usage is

```
set <parameter>=<value> ... <parameter>=<value>
```

Only four parameters can be changed using "set". First, you can set the autoclose feature:

```
set autoclose=<yes/no>
```

If autoclose is set to yes, then the program will not prompt you during the course of executing a file. Second, you can set the partition that is in effect:

```
set partition=<partition id>
```

A valid partition ID is either a number or a partition name. This command enforces use of a specific partitioning of the data. When the program executes, a default partition (that may not divide the data at all) is created called "Default". You can always go back to the original or default partition by typing

```
set partition=default
```

or

set partition=1

Third, you can set the nowarnings feature:

```
set nowarnings=<yes/no>
```

If nowarnings is set to yes, then the program will not prompt you when overwriting an output file that is already present. If nowarnings=no (the default setting) then the program prompts the user before overwriting output files.

Fourth, you can set the quitonerror feature:

```
set quitonerror=<yes/no>
```

If quitonerror is set to yes, then the program will quit when an error is encountered, after printing an error message. If quitonerror=no (the default setting) then the program will wait for additional commands from the command line after printing the error message.

Showmatrix

This command shows the character matrix currently in memory.

Showmodel

This command shows the current model settings. The correct usage is

showmodel

After typing "showmodel", the modelling assumptions are shown on a partition-by-partition basis.

Showtree

This command shows the current user tree. The correct usage is "showtree".

Sump

During a MCMC analysis, MrBayes prints the sampled parameter values to a tabdelimited text file. This file has the extension ".p". The command 'Sump' summarizes the information in the parameter file. By default, the name of the parameter file is assumed to be the name of the last matrix-containing nexus file, but with a '.p' extension. You can set 'Sump' to summarize the information in any other parameter file by setting the 'filename' option to the appropriate file name. The 'Sump' command does not require a matrix to be read in first. When you invoke the 'Sump' command, three items are output: (1) a generation plot of the likelihood values; (2) estimates of the marginal likelihood of the model; and (3) a table with the mean, variance, and 95 percent credible interval for the sampled parameters. Each of these items can be switched on or off using the options 'Plot', 'Marglike', and 'Table'. By default, all three items are output but only to the screen. If output to a file is also desired, set 'Printtofile' to 'Yes'. The name of the output file is specified by setting the 'Outputname' option. When a new matrix is read in or when the 'Mcmc' output filename or 'Sump' input filename is changed, the 'Sump' outputname is changed as well. If you want to output to another file than the default, make sure you specify the outputname every time you invoke 'Sump'. If the specified outputfile already exists, you will be prompted about whether you like to overwrite it or append to it. This behavior can be altered using 'Set nowarn=yes'; see the help for the 'Set' command. When running 'Sump' you typically want to discard a specified number of samples from the beginning of the chain as the burn in. Note that the 'Burnin' value of the 'Sump' command is set separately from the 'Burnin' values of the 'Sumt' and 'Mcmc' commands. That is, if you issue

sump burnin = 4000
sumt burnin = 2000
sump

the burnin of the last 'Sump' command is 4000 and not 2000. The burnin values are reset to 0 every time a new matrix is read in. Similarly, 'Plot', 'Marglike' and 'Table' are all set to 'Yes' and 'Printtofile' to 'No' (the default values) when a new matrix is processed. If you have run several independent MCMC analyses, you may want to summarize and compare the samples from each of these runs. To do this, set 'Nruns' to the number of runs you want to compare and make sure that the '.p' files are named using the MrBayes convention (<filename>.run1.p, <filename>.run2.p, etc). When you run several independent analyses simultaneously in MrBayes, the 'Nruns' and 'Filename' options are automatically set such that 'Sump' will summarize all the resulting output files.

Options:

Burnin	Determines the number of samples that will be discarded from the input file before calculating summary statistics. If
there	
Nruns	<pre>are several input files, the same number of samples will be discarded from each. Note that the burnin is set separately for the 'sump', 'sumt', and 'mcmc' commands Determines how many '.p' files from independent analyses that</pre>
	will be summarized. If Nruns > 1 then the names of the files are derived from 'Filename' by adding '.run1.p', '.run2.p', etc. If Nruns=1, then the single file name is obtained by adding '.p' to 'Filename'.
Filename	The name of the file to be summarized. This is the base of
the	
	file name to which endings are added according to the current setting of the 'Nruns' parameter. If 'Nruns' is 1, then only '.p' is added to the file name. Otherwise, the endings will be '.run1.p', '.run2.p', etc.
Printtofile	Determines whether results will be printed to file.
Outputname	Name of the file to which 'sump' results will be printed if 'Printtofile' is set to YES.
Plot	Determines whether a likelihood plot should be output.
Marglike	Determines whether estimates of marginal model likelihoods should be calculated. The marginal model likelihoods are use-
Table	ful in Bayesian model testing.Determines whether the table summarizing the parameter value samples should be output.

Current settings:

Parameter	Options	Current Setting
Burnin	<number></number>	0
Nruns	<number></number>	1
Filename	<name></name>	temp.p<.p>
Printtofile	Yes/No	No
Outputname	<name></name>	temp.p.stat
Plot	Yes/No	Yes
Marglike	Yes/No	Yes
Table	Yes/No	Yes

Sumt

This command is used to produce summary statistics for trees sampled during a Bayesian MCMC analysis. You can either summarize trees from one individual analysis, or trees coming from several independent analyses. In either case, all the sampled trees are read in and the proportion of the time any single taxon bipartition is found is counted. The proportion of the time that the bipartition is found is an approximation of the posterior probability of the bipartition. (Remember that a taxon bipartition is defined by removing a branch on the tree, dividing the tree into those taxa to the left and right of the removed branch. This set is called a taxon bipartition.) The branch length of the bipartition is also recorded, if branch lengths have been saved to file. The result is a list of the taxon bipartitions found, the frequency

with which they were found, the posterior probability of the bipartition and, if the branch lengths were recorded, the mean and variance of the length of the branch.

The partition information is output to a file with the suffix ".parts". A consensus tree is also printed to a file with the suffix ".con" and printed to the screen as a cladogram, and as a phylogram if branch lengths have been saved. The consensus tree is either a 50 percent majority rule tree or a majority rule tree showing all compatible partitions. If branch lengths have been recorded during the run, the ".con" file will contain a consensus tree with branch lengths and interior nodes labelled with support values. This tree can be viewed in a program such as TreeView.

Finally, MrBayes produces a file with the ending ".trprobs" that contains a list of all the trees that were found during the MCMC analysis, sorted by their probabilities. This list of trees can be used to construct a credible set of trees. For example, if you want to construct a 95 percent credible set of trees, you include all of those trees whose cumulated probability is less than or equal to 0.95. You have the option of displaying the trees to the screen using the "Showtreeprobs" option. The default is to not display the trees to the screen; the number of different trees sampled by the chain can be quite large. If you are analyzing a large set of taxa, you may actually want to skip the calculation of tree probabilities entirely by setting "Calctreeprobs" to NO.

When calculating summary statistics you probably want to skip those trees that

were sampled in the initial part of the run, the so-called burn-in period. The

number of skipped samples is controlled by the "burnin" setting. The default is 0 but you typically want to override this setting.

If you are summarizing the trees sampled in several independent analyses, such as those resulting from setting the "Nruns" option of the "Mcmc" command to a value larger than 1, MrBayes will also calculate convergence diagnostics for the sampled topologies and branch lengths. These values can help you determine whether it is likely that your chains have converged.

The "Sumt" command expands the "Filename" according to the current values of the "Nruns" and "Ntrees" options. For instance, if both "Nruns" and "Ntrees" are set to 1, "Sumt" will try to open a file named "<Filename>.t". If "Nruns" is set to 2 and "Ntrees" to 1, then "Sumt" will open two files, "<Filename>.run1.t" and "<Filename>.run2.t", etc. By default, the "Filename" option will be set such that "Sumt" automatically summarizes all the results from your immediately preceding "Mcmc" command. You can also use the "Sumt" command to summarize tree samples in older analyses. If you want to do that, remember to first read in a matrix so that MrBayes knows what taxon names to expect in the trees. Then set the "Nruns", "Ntrees" and "Filename" options appropriately.

Options:

Burnin

-- Determines the number of samples that will be discarded from the input file before calculating summary statistics. If there are several input files, the same number of samples will be discarded from each. Note that the burnin is set separately for the 'sumt', 'sump', and 'mcmc' commands.

Nruns		Determines how many '.t' files from independent analyses
that		
		will be summarized. If Nruns > 1 then the names of the files are derived from 'Filename' by adding '.run1.t', '.run2.t', etc. If Nruns=1 and Ntrees=1 (see below), then only '.t' is added to 'Filename'.
Ntrees		Determines how many trees there are in the sampled model. If 'Ntrees' > 1 then the names of the files are derived from 'Filename' by adding '.tree1.t', '.tree2.t', etc. If there are both multiple trees and multiple runs, the filenames
will		
		be ' <filename>.tree1.run1.t', '<filename>.tree1.run2.t',</filename></filename>
etc.		
Filename		The name of the file(s) to be summarized. This is the base
of		
		the file name, to which endings are added according to the current settings of the 'Nruns' and 'Ntrees' options.
Displaygeq		The minimum probability of partitions to display.
Contype		Type of consensus tree. 'Halfcompat' results in a 50 major-
		ity rule tree, 'Allcompat' adds all compatible groups to
such		
		a tree.
Calctreeprob	s	Determines whether tree probabilities should be calculated.
Showtreeprob	s	Determines whether tree probabilities should be displayed on

Current settings:

Parameter	Options	Current Setting
Burnin		0
Nruns	<pre><number></number></pre>	1
Ntrees	<number></number>	1
Filename	<name></name>	temp.t<.t>
Displaygeq	<number></number>	0.05
Contype	Halfcompat/Allcompat	Halfcompat
Calctreeprobs	Yes/No	Yes
Showtreeprobs	Yes/No	No

screen.

Taxastat

This command shows the status of all the taxa. The correct usage is

taxastat

After typing "taxastat", the taxon number, name, and whether it is excluded or included are shown.

Taxset

This command defines a taxon set. The format for the taxset command is

taxset <name> = <taxon names or numbers>

For example, "taxset apes = Homo Pan Gorilla Orang gibbon" defines a taxon set called "apes" that includes five taxa (namely, apes). You can assign up to 30 taxon sets. This option is best used not from the command line but rather as a line in the mrbayes block of a file.

Unlink

This command unlinks model parameters across partitions of the data. The correct usage is:

```
unlink <parameter name> = (<all> or <partition list>)
```

A little background is necessary to understand this command. Upon execution of a file, a default partition is set up. This partition referenced either by its name ("default") or number (0). If your data are all of one type, then this default partition does not actually divide up your characters. However, if your datatype is mixed, then the default partition contains as many divisions as there are datatypes in your character matrix. Of course, you can also define other partitions, and switch among them using the set command ("set partition=<name/number>"). Importantly, you can also assign model parameters to individual partitions or to groups of them using the "applyto" option in lset and prset. When the program attempts to perform an analysis, the model is set for individual partitions. If the same parameter applies to differpartitions and if that parameter has the same prior, then the program will link the parameters: that is, it will use a single value for the parameter. The program's default, then, is to strive for parsimony. However, there are lots of cases where you may want unlink a parameter across partitions. For example, you may want a different transition/ transversion rate ratio to apply to different partitions. This command allows you to unlink the parameters, or to make them different across partitions. The converse of this command is "link", which links together parameters that were previously told to be different. The list of parameters that can be unlinked includes:

```
Tratio
                -- Transition/transversion rate ratio
                -- Substitution rates of GTR model
Revmat
                -- Nonsynonymous/synonymous rate ratio
Omega
Statefreq
               -- Character state frequencies
                -- Gamma shape parameter
Shape
Pinvar -- Proportion of invariable sites
Correlation -- Correlation parameter of autodiscrete gamma
Pinvar
                -- Proportion of invariable sites
Switchrates
                -- Switching rates for covarion model
                -- Branch lengths of tree
Brlens
                -- Topology of tree
Topology
Speciationrates -- Speciation rates for birth-death process
Extinctionrates -- Extinction rates for birth-death process
               -- Parameter for coalescence process
Growthrate -- Growth rate of coalescence process
```

For example,

unlink shape=(all)

unlinks the gamma shape parameter across all partitions of the data. You can use "showmodel" to see the current linking status of the Usertree This command allows you to specify a user tree. The user tree can then be used as a starting tree for a MCMC analysis. The format for the command is usertree = <tree in Newick format> For example, usertree = (A,B,(C,D))specifies an unrooted tree of four species. Note that the program requires that trees are binary (i.e., strictly bifurcating). Hence, there can be only one three-way split, as shown in the example. If the tree is not binary, the program will return an error. ______ Version This command shows the release version of the program. ______ 3. 'Data' or 'tree' block commands (in #NEXUS file) ******************** ______ Begin This command is used to format data or commands in the program. The correct usage is begin <data or mrbayes>; The two valid uses of the "begin" command, then, are

begin data;
begin mrbayes;

The "data" specifier is used to specify the beginning of a data block; your character data should follow. For example, the following is an example of a data block for four taxa and ten DNA sites:

```
begin data;
  dimensions ntax=4 nchar=10;
  format datatype=dna;
  matrix
  taxon_1 AACGATTCGT
  taxon_2 AAGGATTCCA
  taxon_3 AACGACTCCT
```

```
taxon_4 AAGGATTCCT
;
end;
```

The other commands -- dimensions, format, and matrix -- are discussed in the appropriate help menu. The only thing to note here is that the block begins with a "begin data" command. The "mrbayes" command is used to enter commands specific to the MrBayes program into the file. This allows you to automatically process commands on execution of the program. The following is a simple mrbayes block:

```
begin mrbayes;
  charset first = 1-10\3;
  charset second = 2-10\3;
  charset third = 3-10\3;
end;
```

This mrbayes block sets off the three "charset" commands, used to predefine some blocks of characters. The mrbayes block can be very useful. For example, in this case, it would save you the time of typing the character sets each time you executed the file. Also, note that every "begin <data or mrbayes>" command ends with an "end". Finally, you can have so-called foreign blocks in the file. An example of a foreign block would be "begin paup". The program will simply skip this block. This is useful because it means that you can use the same file for MrBayes, PAUP* or MacClade (although it isn't clear why you would want to use those other programs).

Dimensions

This command is used in a data block to define the number of taxa and characters. The correct usage is

dimensions ntax=<number> nchar=<number>

The dimensions must be the first command in a data block. The following provides an example of the proper use of this command:

```
begin data;
  dimensions ntax=4 nchar=10;
  format datatype=dna;
  matrix
  taxon_1 AACGATTCGT
  taxon_2 AAGGATTCCA
  taxon_3 AACGACTCCT
  taxon_4 AAGGATTCCT
;
end;
```

Here, the dimensions command tells MrBayes to expect a matrix with four taxa and 10 characters.

End

This command is used to terminate a data or mrbayes block. The correct

```
usage is
  end;
For more information on this, check the help for the "begin" command.
______
______
Endblock
This is an older, deprecated version of "End", see that command.
Format
This command is used in a data block to define the format of the char-
acter matrix. The correct usage is
  format datatype=<name> ... <parameter>=<option>
The format command must be the second command in a data block. The following
provides an example of the proper use of this command:
  begin data;
     dimensions ntax=4 nchar=10;
     format datatype=dna gap=-;
     matrix
     taxon 1 AACGATTCGT
     taxon 2 AAGGAT--CA
     taxon_3 AACGACTCCT
     taxon_4 AAGGATTCCT
  end;
Here, the format command tells MrBayes to expect a matrix with DNA char-
acters and with gaps coded as "-".
The following are valid options for format:
         -- This parameter MUST BE INCLUDED in the format command. More-
Datatype
            over, it must be the first parameter in the line. The
            datatype command specifies what type of characters are
            in the matrix. The following are valid options:
               Datatype = Dna: DNA states (A,C,G,T,R,Y,M,K,S,W,H,B,
                         V,D,N)
               Datatype = Rna: DNA states (A,C,G,U,R,Y,M,K,S,W,H,B,
                         V,D,N)
               Datatype = Protein: Amino acid states (A,R,N,D,C,Q,E,
                         G,H,I,L,K,M,F,P,S,T,W,Y,V)
               Datatype = Restriction: Restriction site (0,1) states
               Datatype = Standard: Morphological (0,1) states
               Datatype = Continuous: Real number valued states
               Datatype = Mixed(<type>:<range>,...,<type>:<range>): A
```

mixture of the above datatypes. For example, "datatype=mixed(dna:1-100,protein:101-200)" would specify a mixture of DNA and amino acid characters with the DNA characters occupying the first 100 sites and the amino acid char-

acters occupying the last 100 sites.

```
Interleave -- This parameter specifies whether the data matrix is in
    interleave format. The valid options are "Yes" or "No",
    with "No" as the default. An interleaved matrix looks like
```

```
format datatype=dna gap=- interleave=yes;
matrix
taxon_1    AACGATTCGT
taxon_2    AAGGAT--CA
taxon_3    AACGACTCCT
taxon_4    AAGGATTCCT

taxon_1    CCTGGTAC
taxon_2    CCTGGTAC
taxon_3    ---GGTAG
taxon_4    ---GGTAG;
;
```

Gap -- This parameter specifies the format for gaps. Note that gap character can only be a single character and that it cannot correspond to a standard state (e.g., A,C,G,T,R,Y, M,K,S,W,H,B,V,D,N for nucleotide data).

Missing -- This parameter specifies the format for missing data. Note that the missing character can only be a single character and cannot correspond to a standard state (e.g., A,C,G,T,R,Y, M,K,S,W,H,B,V,D,N for nucleotide data). This is often an unnecessary parameter to set because many data types, such as nucleotide or amino acid, already have a missing character specified. However, for morphological or restriction site data, "missing=?" is often used to specify ambiguity or unobserved data.

Matchchar -- This parameter specifies the matching character for the matrix. For example,

The only non-standard NEXUS format option is the use of the "mixed", "restriction", "standard" and "continuous" datatypes. Hence, if

you use any of these datatype specifiers, a program like PAUP* or MacClade will report an error (as they should because MrBayes is not strictly NEXUS compliant).

Matrix

This command specifies the actual data for the phylogenetic analysis. The character matrix should follow the dimensions and format commands in a data block. The matrix can have all of the characters for a taxon on a single line:

```
begin data;
      dimensions ntax=4 nchar=10;
     format datatype=dna gap=-;
     matrix
     taxon_1 AACGATTCGT
      taxon_2 AAGGAT--CA
      taxon_3 AACGACTCCT
     taxon_4 AAGGATTCCT
   end;
or be in "interleaved" format:
   begin data;
     dimensions ntax=4 nchar=20;
      format datatype=dna gap=- interleave=yes;
     matrix
      taxon_1 AACGATTCGT
      taxon_2 AAGGAT--CA
      taxon_3 AACGACTCCT
      taxon_4 AAGGATTCCT
     taxon_1 TTTTCGAAGC
      taxon_2 TTTTCGGAGC
      taxon 3 TTTTTGATGC
     taxon_4 TTTTCGGAGC
   end;
```

Note that the taxon names must not have spaces. If you really want to indicate a space in a taxon name (perhaps between a genus and species name), then you might use an underline ("_"). There should be at least a single space after the taxon name, separating the name from the actual data on that line. There can be spaces between the characters.

If you have mixed data, then you specify all of the data in the same matrix. Here is an example that includes two different data types:

```
begin data;
  dimensions ntax=4 nchar=20;
  format datatype=mixed(dna:1-10,standard:21-30) interleave=yes;
  matrix
  taxon_1  AACGATTCGT
  taxon_2  AAGGAT--CA
```

```
taxon_3 AACGACTCCT
taxon_4 AAGGATTCCT

taxon_1 0001111111
taxon_2 0111110000
taxon_3 1110000000
taxon_4 1000001111
;
end;
```

The matrix command is terminated by a semicolon.

Finally, just a note on data presentation. It is much easier for others to (1) understand your data and (2) repeat your analyses if you make your data clean, comment it liberally (using the square brackets), and embed the commands you used in a publication in the mrbayes block. Remember that the data took a long time for you to collect. You might as well spend a little time making the data file look nice and clear to any that may later request the data for further analysis.

Translate

This command is used by MrBayes to specify the mapping between taxon names and taxon numbers in a Nexus tree file. For instance,

```
translate
  1 Homo,
  2 Pan,
  3 Gorilla,
  4 Hylobates;
```

establishes that the taxon labeled 1 in the trees that follow is Homo, the taxon labeled 2 is Pan, etc.

Tree

This command is used by MrBayes to write trees to a nexus tree file. Trees are written in the Newick format. For instance,

```
tree ((1,2),3,4);
```

describes an unrooted tree with taxa 1 and 2 being more closely related to each other than to taxa 3 and 4. If branch lengths are saved to file, they are given after a colon sign immediately following the terminal taxon or the interior node they refer to. An example of an unrooted tree with branch lengths is:

```
tree ((1:0.064573,2:0.029042):0.041239,3:0.203988,4:0.187654);
```

Trees that are rooted (clock trees) are written with a basal dichotomy instead of a basal trichotomy. If the tree described above had been rooted on the branch leading to taxon 4, it would have been represented as:

tree (((1,2),3),4);