User's guide for *ESL* Ver. 0.1

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This is a first draft of the user's guide for the *ESL* (pronounced as /esl/; italicized font) program. This is provided mainly for the peer-review of manuscript titled "Measuring Phylogenetic Information of Incomplete Sequence Data" by Tae-Kun Seo, Olivier Gascuel and Jeffrey Thorne. In the following, this manuscript will be referred to as the SGT manuscript. A more detailed manual will be available soon.

The *ESL* program estimates Effective Sequence Length (ESL; pronounced as / i - es - el/; for the definition of ESL, see pages 12-13 of the SGT manuscript) for given sequence data, tree topology and substitution model.

FORMATS AND OPTIONS

Sequence file : The sequence data format is the phylip (sequential) format as shown in Figure 1.

Option file : All options should be represented as a separate line within the "option.txt" file (Figure 2). In each line that specifies an option, only the first item after the '>' character is recognized as an input. Do not change the string between the '<' and '>' characters.

(1) < seq file >

The sequence file is set here.

(2) < subs model >

```
  12 1000
T1     TTCCGAGGAGCCCTATCACATGGTCGTTTCTAAGATGCCTCCGA...
T2     TTCCGAGGGACTCTCTCACATGATTGTTTCTAAAATGCCTCCGA...
T3     CTCTGAGGGGCCCTGACACATGATTGTTTGTAAAATGCACTCAA...
T4     CTCCGGGAGGCCCTTACATATGACCGTTTGTAAAATTCCTTCAA...
T5     TCCCGAGGGGTACCAGGACATGATAATTTCTAAAATGCCCCTGA...
T6     TCCCGGAGGGTATTGTGACATGATTATTTCTAGAGCGCCTCTGA...
T7     TCCTGGGGGGCCCTACTACATGATAACTCCTAAATTGCCCCTGA...
T8     TCCTGGGGGACCCTATTACATGATAACTCCTAAACTGCCTCTGA...
T9     TTCAGGGGGGCCCCAAGGTATAATTGCTTCCAAAACGCCCTTAG...
T10    TCCAGAAAGGCCCTAAGGCATAATCGCTTCCAAAACGCCCCTAG...
T11    TCCAGGGGAGCCCTGGTTTATAATTGCTTCTGAGGTGCCCCAAG...
T12    CCCAAGGGGGCTCAAGTATATAGTCGCGTCTAAGGTGCCCCCAG...
```

Figure 1: testdata.txt

The model for nucleotide substitution or amino acid replacement is set here. For nucleotide models, JC (Jukes and Cantor 1969), TN93 (Tamura and Nei 1993), and GTR (Tavaré 1986; Yang 1994) can be selected. For amino acid models, WAG (Whelan and Goldman 2001), mtREV24 (Adachi and Hasegawa 1996) , LG (Le and Gascuel 2008), Jones (Jones et al. 1992), and Dayhoff (Dayoff et al. 1978) can be selected.

(3) < freq option >

The option for treating the frequencies of nucleotides or amino acids. When the nucleotide model is selected in (2), only the 'DataFreq' option is valid and empirical nucleotide frequencies are used for rate parameters. When an amino acid model is selected in (2), amino acid frequencies provided by the model can be used by choosing 'ModelFreq'

(4) < rand seed >

A random seed value is required for the bootstrap procedure. Any integer between -2^{63} and $2^{63} - 1$ can be selected for random seed number.

(5) < tree topo >

The tree topology should be provided in the newick format.

(6) < job >

0 should be here to estimate G-Factors and M-Factors. Other job options will be available soon.

(7) < simul length >

```
< seq file >  testdata.txt
< subs model >  GTR // JC, TN93, GTR, WAG, mtREV24, LG, Jones, Dayhoff
< freq option >  DataFreq // DataFreq, ModelFreq
< rand seed >  1
< tree topo >  (((T1,T2),(T3,T4)),((T5,T6),(T7,T8)),((T9,T10),(T11,T12)));
< job >  0
< simul length >  20
< boot iter >  100
< bootstrap type >  0 // 0:RELL, 1:Full
```

Figure 2: esl_options.txt

```
C:\ESL_v01\bin>  java ESL_v01.TestMain   >  scr.txt   [←]
```

Figure 3: How to run

Number of times by which the length of the simulated data is longer than the original data. This corresponds to m of Eq (3) and (4) of the SGT manuscript.

(8) < boot iter >

Number of bootstrap iterations. This corresponds to B of Eq (15) of the SGT manuscript.

(9) < bootstrap type >

RELL-like procedure or full bootstrap procedure. See page 9 of the SGT manuscript.

How to run *ESL*

You must install JAVA runtime environment (JRE; https://java.com/en/download/) to run *ESL*. *ESL* can be run on any OS as long as JRE is pre-installed.

Once JRE is installed, copy the ESL_v01 folder to your favorite location. Put the sequence and option files in the bin folder. Go to the bin folder and type the following commands ('TestMain' is case-sensitive) and hit ENTER. Because the results are displayed in the console, you need to capture them by redirecting messages from screen to the appropriate file. In the example of Figure 3, results are redirected from the console to the scr.txt file.

INTERPRETATION OF THE RESULTS

```
......
Tree topology information
Node 0:  T1
Node 1:  T2
Node 2:6->, <-0, <-1
Node 3:  T3
Node 4:  T4
Node 5:6->, <-3, <-4
......
Tree branch information
br0 :  2 <--> 0
br1 :  2 <--> 1
br2 :  6 <--> 2
......
```

Figure 4: scr.txt

In the beginning of scr.txt, information about tree topology is shown as in Figure 4. "Node W: X->, <-Y, <-Z" means that "Node X is the ancestor of node W, and nodes Y and Z are the descendants of node W". Also, "brX : Y <->Y" means that "branch X connects node Y and node Z".

In the later part of scr.txt, parameterwise M-Factors, G-Factors and global M-Factor and G-Factor are shown as in Figure 5.

The integer within parenthesis after each standard deviation estimate (0's in this example) represent weird M-Factor and G-Factor estimates during bootstrap (see page 9 of SGT manuscript) that have values that are outside the range [0,2]. If this integer is substantially greater than zero, the full version of the bootstrap procedure is recommended in < bootstrap type >option of Figure 2.

After running the *ESL* program, the file named "sequence_file" + "_s-ESL.txt" was generated in the same folder. In our analysis here, "testdata.txt_s-ESL.txt" is generated. Within this file, sitewise and parameter-wise ESL (see pages 12-13 of the SGT manuscript) and R commands for drawing figures are saved (Figure 6). R package "tidyverse" should be pre-installed.

After running *ESL* program, the file named "sequence_file" + "_esl_tree_nex.txt" is generated in the same folder. In our analysis here, "testdata.txt_esl_tree_nex.txt" is generated. Within this file, R commands for drawing Figure 3 of SGT manuscript are saved. Move this file to your favourite folder (say, "c:/temp/") and copy only R commands (not tree information of nexus format) to the R prompt. Then you will get phylogeny similar to Figure 3 of SGT manuscript (Figure 7). If you move the file to a different folder, you

```
......
#### Individual beta (M-Factor) estimate +- std
beta[0] = 0.941930 +- 0.053350 (0)
beta[1] = 0.999968 +- 0.032398 (0)
beta[2] = 1.105290 +- 0.049699 (0)
......
#### Individual Rho (G-Factor) estimate +- std
rho[0] = 0.493388 +- 0.020669 (0)
rho[1] = 0.700998 +- 0.014124 (0)
rho[2] = 0.719371 +- 0.017488 (0)
......
#### globaBeta (M-Factor) = 0.998920 +- 0.023108 ; (TestStat = -0.046738;
pValue from bootstrap = 0.510000) (# of extreme bootstrapped samples = 0,
out of 100)
#### globaRho (G-Factor) = 0.602523 +- 0.015369 ; (# of extreme
bootstrapped samples = 0, out of 100)
......
```

Figure 5: scr.txt

```
......
s_ESL <- c(1.427519, 0.902281, -0.047730, ...
......
sp_ESL <- matrix(c(-0.012685,-0.012966,-0.002481, ....
....
x<-1:dim(sp_ESL)[1]
y<-1:1000
library(ggplot2)
library(reshape2)
z <- sp_ESL
p3 <- persp(x,y,z, col="white", theta=120, xlab="br", ylab="site",
zlab="sp-ESL")
p3
......
```

Figure 6: testdata.txt_s-ESL.txt

should change the path within "testdata.txt_esl_tree_nex.txt". Colors, thickness of branches, and font sizes can be adjusted by modifying R commands within "testdata.txt_esl_tree_nex.txt".
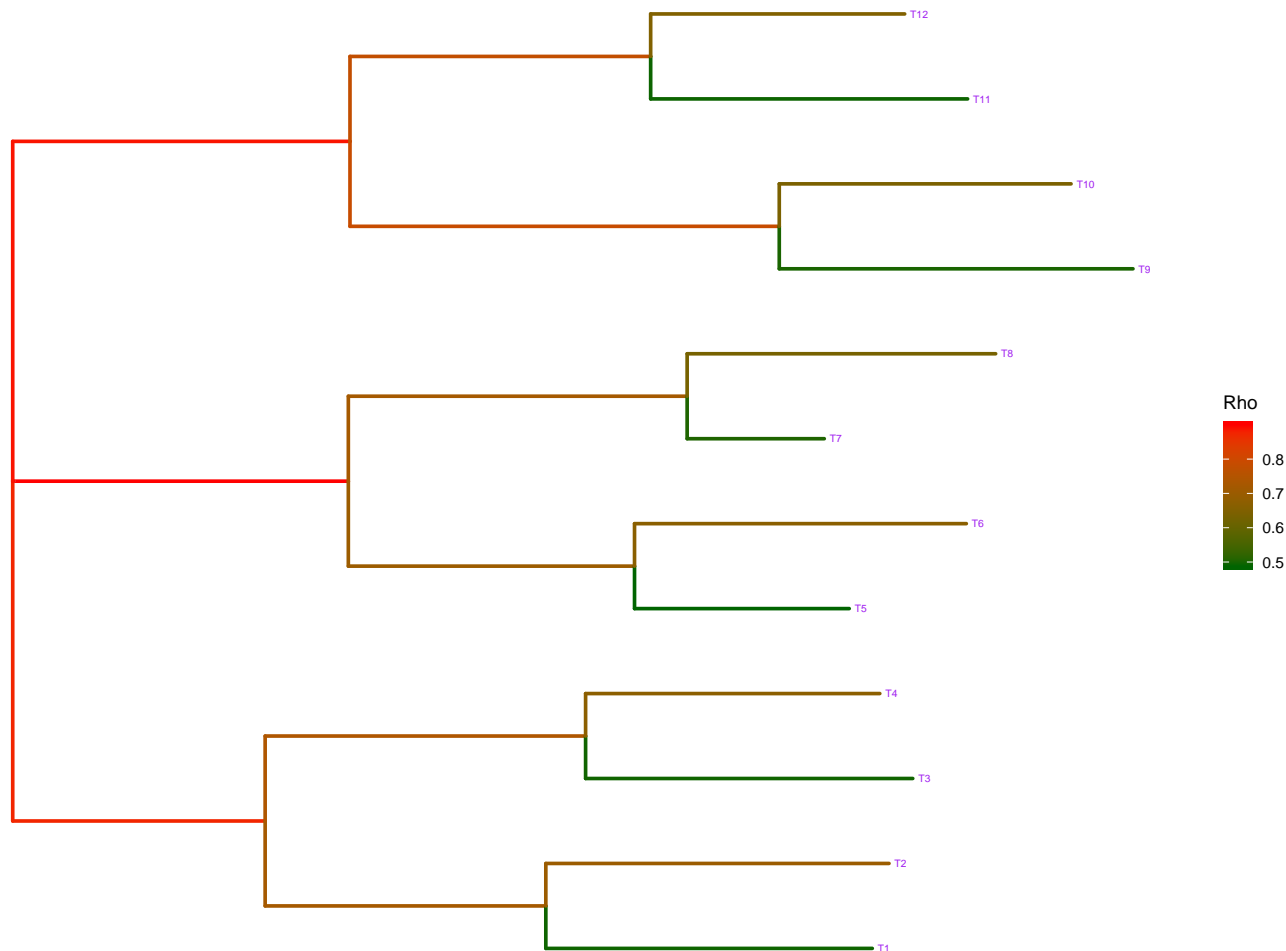

Figure 7: G-Factor (ρ_i) estimates of testdata.txt; For R commands see "testdata.txt_esl_tree_nex.txt"

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