IsgGui: A New tool for Sequence simulation

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Outline

- Introduction
- Sequence Simulation
- Indel-seq-gen
- IsgGui
- Discussion
- Future work

Introduction - 1

Evolution: The change in the inherited traits (as determined by changes in DNA) of a population of organisms through successive generations.

Studies investigate:

- Biodiversity: species vs. species and within species
- Pathogenicity: one species beneficial and another toxic
- Viruses: Development of vaccines
- Congenital illness: possible gene therapy
- Alternate biochemical pathways
- Drug reaction

Introduction - 2

Evolutionary hypothesis:

- From sequenced DNA
  - Genes identified (predicted or from lab work)
  - Transcripts predicted or isolated in lab
  - Protein sequence decoded and archived
- Multi-sequence alignment
  - Search in databases for similar sequences (Blast)
  - Sequences arranged into multi-sequence-alignment (MSA)
    - Manual alignment by human curator
    - Manually adjusted automated alignments
    - Fully automated alignment
- Phylogeny generation
  - Evolutionary tree predicted from MSA
  - Techniques based on maximum parsimony, maximum likelihood or other computation method.

Multiple sequence alignments (MSA)-1

3_5_exonuc (PF01612): sequence seqment vs. MSA

```
Q9ZDB0_RICPR/2169      NIKIYQNDLPSNFKLEGDIAVDTETMGLNIHRDKLCLLQFSNGNGEAHLVHFNQD
Q55641_SYNY3/9181      QFEVFDYDLPEDVCQQLLACKEVAVDTETMGLNPHRDRLCLVQICDPEGNVTALRIAKGQ
DPO1_AQUAE/3162        FEYVTGEEGLKKAIKRLENSPYLYLDTETTGDRIRLVQIGDEENTYVIDLYEIQDI
DPOL_BPMD2/18212       VEHPEDLDGFRDFIRAHQNCLAVDTETTGLDIYSADFRCRLVQFGTQTESWVLPIEDLE
DPOL_BPML5/18211       VETPEDLEGFRNFIRAHLNCLAVDTETTGLDIYSDTFECRLVQFGTQDEAWVVPVELGD
DPO1_TREPA/409589      LVGFSICFQEAEAFYVPLIVPDVSLHTESTQCTCARSTNVETEKECTEQHGVSASAVQDPAYVQA
DPOL_BPT5/111306       YRPITDPDEAEEYIKMVYNMVIGPVAFDSETSALYCRDGYLLGVSISHQEYQGVYIDSDCLTEV
DPO1_BORBU/321500      YRSITTKIELDDLIESLKKAKYISIDTETSSLDTYTAKLIGISISFKEFEGYYIPIEAKGKI
Q9ZIG3_RHOSI/333513    YRIVRNRQQLDELVARLDGLERLAIDTETTSTEAMWASLVGIAFSWEKGQGYYVPTPLPDGT
DPO1_HAEIN/334520      YETLLTQADLTRWIEKLNAAKLIAVDTETDSLDYMSANLVGISFALENGEAAYLPLQLDYLD
DPO1_ECOLI/330517      YVTILDEETLKAWIAKLEKAPVFAFDTETDSLDNISANLVGLSFAIEPGVAAYIPVAHDYLD
DPO1_CHLAA/339522      VTIVRDATALADLVSRLRNAPAFAFDTECTSLQPVASDLVGISIAIAPDTVCYIPVGHQSET
DPO1_SYNY3/372560      PQIITTIAQLEALVEELKKHTDADFPVAWDTETDSLDPLVANLVGIGCAWGQEPNQVAYIPLKHHQGE
O69858_STRCO/10175     PVIADADALAEVTAAFAAGSGPVAVDAERASGYRYGQRAYLVQLRRAGAGSALIDPVACPD
Q50004_MYCLE/28198     NLSVTPSEIAAAAERLDQGYGPFAVDTERASGFRYSNRAYLIQIRRANAGTVLIDPVSHGN
Q9ZD81_RICPR/1169      MQIIDKQKTLEEFCNKLANHTSLSIDTEFERRYTYYAQLSIIQVKAEGYCGIIDTLSNLD
RND_HAEIN/30198        FRVVTDNTALLEVCNLAQQKSAVALDTEFMRVSTYFPKLGLIQLYDGEHVSLIDPLAITD
P92030_DROME/522715    IANVKQSVFVTDIILKYAAKNESIVVSLDCEGINLGLKGEITLIEIGTTRGEAFLFDVQSCPA
WRN_XENLA/51225        YESNDCSLLSEDIRSSLLEEDVLGFDIEWPPVYTKGKTGKVALIQVCVSEKKCYLFHISPMAGFP
WRN_MOUSE/51226        YEASDCSFLSEDISMRLSDGDVVGFDMEWPPIYKPGKRSRVAVIQLCVSENKCYLFHISSMSVFP
MUT7_CAEEL/404605      IHMVKTESEMNYLCSEIKSLSDEPAPVYVGFDSEWKPSNLTAVHDSKIAIIQLFFKNCVWLVDCVELEKANMADD
YO63_CAEEL/422620      IRIVQNEQDLEILLSELGELEEGMYIGYDSEFKPYHLIDVSTSRLAIIQLFFKDKAWLINCVAIDNLASRD
```

```
                          10        20        30        40        50        60
O9ZDB0_RICPR/2-169    NIKIYQNDLPSNFKLEG----------DIAVDTETMGLNI--H-RDKLCLLQFSNG-----NGEAI
O55641_SYNY3/9-181    QFEVFDYDLPEDVCQQLLACK------EVAVDTETMGLNP--H-RDRLCLVQICDP-----EGNVT
DPO1_AQUAE/3-162      FEYVTGEEGLKKAIKRLENSP------YLYLDTETTGDRI---------RLVQIGDE-----ENTYV
DPOL_BPMD2/18-212     VEHPEDLDGFRDFIRAHQN---------CLAVDTETTGLDIYSA-DFRCRLVQFGTQ-----TESWV
DPOL_BPML5/18-211     VETPEDLEGFRNFIRAHLN---------CLAVDTETTGLDIYSD-TFECRLVQFGTQ-----DEAWV
DPO1_TREPA/409-589    LVGFSICFQEAEAFYVPLIVP------DVSLHTESTQCTCARS-TNVETEKECTEQH--GVSASAV
DPOL_BPT5/111-306     YRPITDPDEAEEYIKMVYNMVI----GPVAFDSETSALYCRDG-YLLGVSISHQEY-----QGVYI
DPO1_BORBU/321-500    YRSITTKIELDDLIESLKKAK------YISIDTETSSLDTYTA-KLIGISISFKEF-----EGYYI
O9ZIG3_RHOSI/333-513  YRIVRNRQQLDELVARLDGLE------RLAIDTETTSTEAMWA-SLVGIAFSWEKG-----QGYYV
DPO1_HAEIN/334-520    YETLLTQADLTRWIEKLNAAK------LIAVDTETDSLDYMSA-NLVGISFALENG-----EAAYI
DPO1_ECOLI/330-517    YVTILDEETLKAWIAKLEKAP------VFAFDTETDSLDNISA-NLVGLSFAIEPG-----VAAYI
DPO1_CHLAA/339-522    VTIVRDATALADLVSRLRNAP------AFAFDTECTSLQPVAS-DLVGISIAIAPD-----TVCYI
DPO1_SYNY3/372-560    PQIITTIAQLEALVEELKKHTDA--DFPVAWDTETDSLDPLVA-NLVGIGCAWGQEP--N-QVAYI
O69858_STRCO/10-175   PVIADADALAEVTAAFAAGSG------PVAVDAERASGYR--Y-GQRAYLVQLRRAG-----AGSAI
Q50004_MYCLE/28-198   NLSVTPSEIAAAAERLDQGYG------PFAVDTERASGFR--Y-SNRAYLIQIRRAN-----AGTVI
O9ZD81_RICPR/1-169    MQIIDKQKTLEEFCNKLANHT------SLSIDTEFERRYT--Y-YAQLSIIQVKAE-----GYCGI
RND_HAEIN/30-198      FRVVTDNTALLEVCNLAQQKS------AVALDTEFMRVST--Y-FPKLGLIQLYDG-----EHVSI
P92030_DROME/522-715  IANVKQSVFVTDIILKYAAKNES---IVVSLDCEGINLGL----KGEITLIEIGTTR-----GEAFI
WRN_XENLA/51-225      YESNDCSLLSEDIRSSLLEED------VLGFDIEWPPVYTKGK-TGKVALIQVCVSE--KKCYLFI
WRN_MOUSE/51-226      YEASDCSFLSEDISMRLSDGD------VVGFDMEWPPIYKPGK-RSRVAVIQLCVSE--NKCYLFI
MUT7_CAEEL/404-605    IHMVKTESEMNYLCSEIKSLSDEPAPVYVGFDSEWKPSNLTAVHDSKIAIIQLFFKNCVWLVDCVI
YO63_CAEEL/422-620    IRIVQNEQDLEILLSELGELEEG---MYIGYDSEFKPYHLIDVSTSRLAIIQLFFKDKAWLINCVI
```

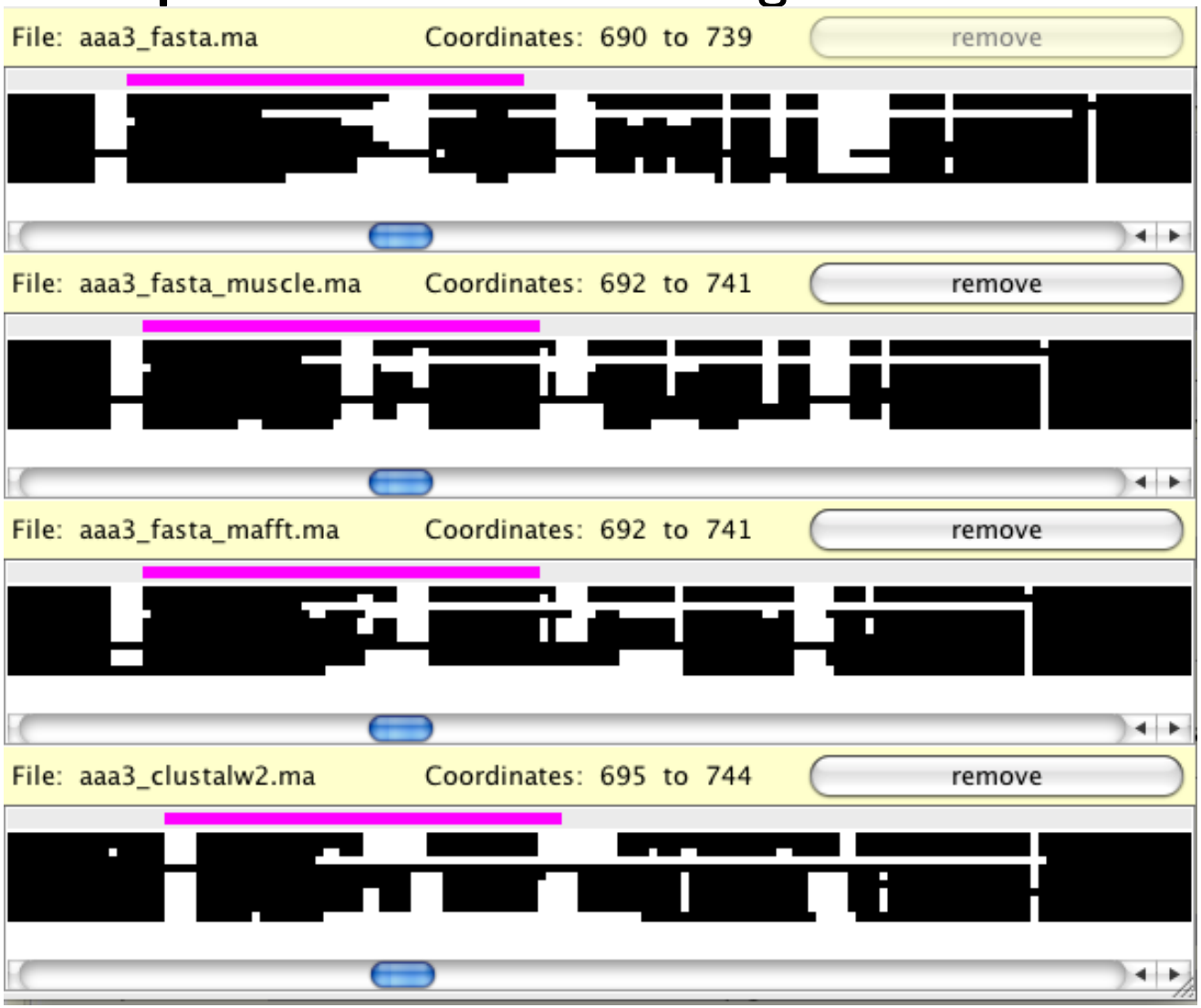
Multiple sequence alignments (MSA)-2

Three methods

- ClustalW
  - Does alignment between of all paits
  - Generates distance Matix
  - Using nearest Neighbor, builds guide tree
  - Performs alignment using guide tree
- Mafft
  - Converts each amino acid to a vector of volume and polarity)
  - Uses Fast Fourier Transform to calculate correlation between two amino acids
  - Detects areas of Similarity by peaks in FFT
  - Aligns homologous areas
- Muscle
  - Devises a distance measure using ‘k-mers’, based on compressed alphabets,
  - Uses a “log-expectancy” function to align profiles.

Multiple sequence alignments (MSA)-3

Pixel plots of 4 alternative alignments



Evolutionary hypothesis -2

Weakness in flow

- For extant species, exact phylogenies are not known.
- Manually generated alignments or phylogenies are best guess only.
- Evolution invoked in laboratories with mutagens provide further insight but are limited in scope [D.Hillis, 1992]
- Convergent evolution disallows phenotype guide trees.
- Automated alignments methods agree in conserved segments but vary in area of divergence.

Sequence Simulation - 1

Using Sequence Simulation

- Can generate a set of homologous sequences (all)
- Can generate the "true" MSA (most)
- Can generate the "true" phylogeny (some)
- Can generate a set of heterogeneous protein sequences (few)
- Can mix pseudogene, non-coding (intron) and coding (exon) areas in one sequence (one)

Using simulation results:

- With the "true" MSA can evaluate alignment programs and the effects of "disagreement" on the results phylogeny
- With the exact or "true" phylogenies, can evaluate phylogenetic programs
- Have a family of heterogeneous proteins

Sequence Simulation - 2

Parameters for simulation:

- Root sequence (DNA or protein)
- Guide tree
- Model of evolution
  - Substitution matrix (PAM, JTT, ... etc)
  - Distributions for substitution events
  - Distribution for indel events (insertion and deletion)
  - Functional constraints for proteins (clade and motif parameters)

Results from simulation :

- Family of taxon sequences including ancestors
- "True" multiple sequence alignment
- "True" phylogeny of sequences including mapping of indel events
- Record of indel events

iSGv2: indel-seq-gen (1)

Overview of iSGv2:

- Developed by Cory Strope, 2010
- Modified iSGv1 to allow generating realistic protein families.
- Can parameterize and simulate heterogeneous domains.
- Can generate divergent protein sequences without destroying functional properties.
- Combines all features offered in other simulation programs
- Offers improved indel event generation.

iSGv2: indel-seq-gen (2)

Description of iSGv2:

- A command-line program
- Simulation objective can require many lengthy parameters

indel-seq-gen -m JTT -j des -o f -e lipocalin_out -d 011110 -s 70 -n 5 -w -b 1.5 -a 1.4 -g 16 -i 0.3 -z 6543 -f 0.045,0.05,0.05,0.05,0.05,0.06,0.05,0.04,0.05,0.05,0.05,0.03,0.05, 0.07,0.05,0.05,0.05,0.015,0.025,0.05 < lipocalin.tree

- Advanced parameters have dependencies
- Can be time consuming to set up and check parameters

iSGv2: indel-seq-gen (3)

Description of iSGv2:

- Support file needed:
  - substitution rates (protein or DNA)
  - indel occurrence rates
  - indel length distribution rates
  - lineage file (clades and motifs)
- Guide tree file:

[:lipocalin_ma(1:56,6,c)]"motif_1"{5,0.1,idLD}(((taxon1:0.32,taxon2:0.24)Clad
taxon3:0.7)Clade1:0.36,((taxon4:0.35,taxon5:0.55)Clades4:0.250,taxon6:0.31)C
[200]"no_motif'#b1.5#{2,0.03}(((taxon1:0.32,taxon2:0.24)Clade2:0.42,
taxon3:0.7)Clade1:0.36,((taxon4:0.35,taxon5:0.55)Clades4:0.250,taxon6:0.31)C

IsgGui (1)

IsgGui : a graphical user interface for iSGv2

- Developed by Cate Anderson
- Programmed in java SE 6
- Works on top of installed indel-seq-gen-2.1
- Packaged in an executable jar file with needed libraries
- Set up entails the selection of various directories

IsgGui (2)

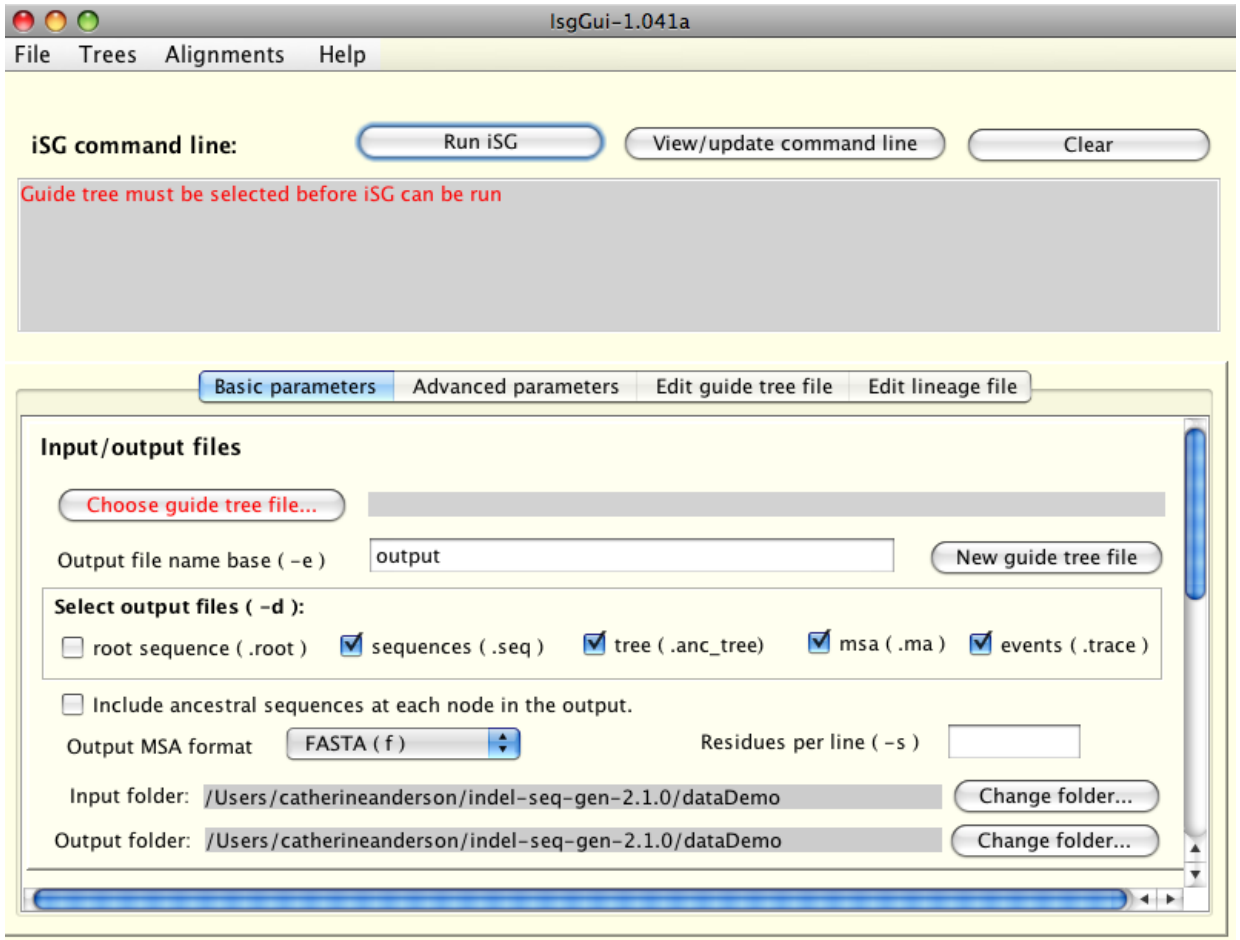
Advantages

- Allows for quick addition of both global and guide tree parameters.
- Provides parameter format error checking.
- Provides parameter compatability checking.
- Displays command-line text from GUI.
- Generate graphical representation of results.

IsgGui (3)

Main Window

Go to live demo



iSGv2: indel-seq-gen (4)

Lineage File

```
MOTIFS =
{
        root:
                MARKER=b;
                NAME=PS00213: Lipocalin signature;
                PATTERN=[DENG]-{A}-[DENQGSTARK]-x(0,2)-[DENQARK]-[LIVFY]
                                -{CP}-G-{C}-W-[FYWLRH]-{A}-[LIVMTA];

      root:
              MARKER=7;
              NAME=lipo_ma: Lipocalin partial template;
              PATTERN=x(5,20)-x(10,30);

}
```

iSGv2: indel-seq-gen (5)

Root sequence multiple alignment file

```
          0000000000000000000000000000000000000000000000000000000000
Taxon1    EGP-LDT-----AVEEE--QISGNWLGMRSTYHV--VS--L-FNQGILQ-----EV
Taxon2    ERS-LEMT---VVGDSSGDAIDGVWLFLQYINEVGFLRQRLKF-----AALGTIKL
Taxon3    DGP-IDTIQTNIVLDSS--DIAGRWYVMDLIGDAMFRRTMKGL-----KNVLSGPL
Taxon4    PFPNLATKLIGVQPDQD--EIIGQWYELSHHSKSAIFGDTS-M-----KLV-TK--
PS00213   **************bbbbbbbbbbbbbb*****************************
lipo_ma   *7777777777777777777777777777777777.....7777777
```

Root sequence file

```
50
00000000000000000000000000000000000000000000000000
ASPISTIQAATVPDSS--EVAGKWYIVALASNTSFLREKGKMKMVMARIL
**************bbbbbbbbbbbbbb**********************
*00000000000000000000000000000000000000000000000000
```

Implementation of IsgGui (4)

4 components of the Main Window

- Basic parameters - entering basic global parameter.
- Advanced parameters - entering more specific controls
- Edit guide tree - allows additional partitions to be added or deleted from guide tree file
- Edit lineage file - allows changes to subtree (clade) parameters and motifs

Implementation of IsgGui (5)

3 display options

- Alignment display -
  - Can display any MSA in fasta format
  - Can display events within in alignment
  - Provides facility to compare alternate alignments
- Phylogeny display
  - Allows for the display of any tree in Newick format
  - Allows for the display of indel events on phylogeny
  - Allows for the display and editing of guide trees for iSGv2.
- Pixel-plot display
  - Allows for a larger area view of alignment
  - Allows for the comparison of up to four alternate alignments

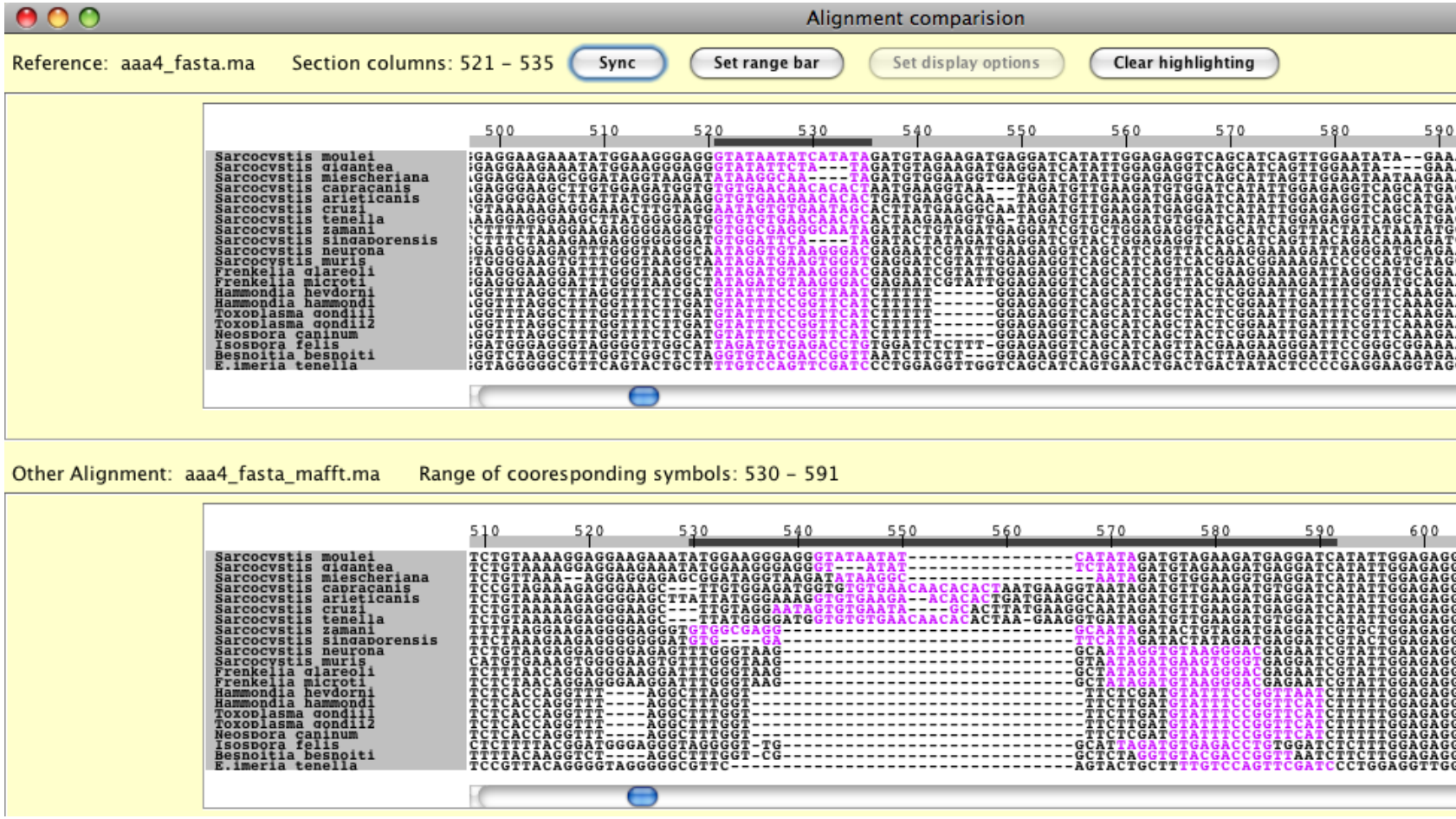
Discussion

Uses for IsgGui

- For evolution simulation
  - Used to learn indel-seq-gen
  - Used to debug parameters
  - Used to debug support files
- For alignment comparisons
  - Compare two full alignments with highlighting to indicate conserved areas between alignments.
  - Compare up to 4 alignments in Pixel Plot format

Discussion

For alignment comparisons



Future Work

Gui

- Multi-thread capability
- Faster image processing

Functionality

- Provide scoring of alternate MSAs based on "true" reference alignment
- Evaluate effect on Phylogeny for inconsistent alignment
- Identify the sequence of indel events that cause most difficulty, and adapt alignment method to "consider" these possibilities

References - Alignment programs and curated alignments

- *"CLUSTAL W: improving the sensitivity of progressivemultiple sequence alignment through sequence weighting,position-specific gap penalties and weight matrix choice"*

  Higgins D., Thompson J., Gibson T., Thompson J.D., Higgins D.G., Gibson T.J.(1994)

- *"MUSCLE: a multiple sequence alignment method with reduced time and space complexity."*

  Edgar R.C. (2004)

- *"MAFFT: a novel method for rapid multiple sequence alignment based on fast Fourier transform"*

  Kazutaka Katoh, Kazuharu Misawa, Kei-ichi Kuma and Takashi Miyata (2002)

- Manually curated MSA's http://hem.fyristorg.com/acacia/alignments.htm

References - iSGv2 and IsgGui

- *"indel-Seq-Gen: A New Protein Family Simulator Incorporating Domains, Motifs, and Indels"*

  Cory L. Strope, Stephen D. Scott and Etsuko N. Moriyama (2009

- *"indel-Seq-Gen v2.0.5 Manual"*

  Cory L. Strope, Kevin Abel, Stephen D. Scott and Etsuko N. Moriyama(2010)

- *"IsgGui: A Graphical User interface to enhance the use of iSGv2'*

  Catherine Anderson, Cory L. Strope, and Etsuko N. Moriyama (2010

- *"IsgGui 1.00 User's manual"*

  Catherine Anderson, Cory L. Strope and Etsuko N. Moriyama (2010)

Questions

Thank you for your attention!

Are there any questions?