BMC
Research Notes

TECHNICAL NOTE   **Open Access**

CoreGenes3.5: a webserver for the determination of core genes from sets of viral and small bacterial genomes

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Abstract

Background: CoreGenes3.5 is a webserver that determines sets of core genes from viral and small bacterial genomes as an automated batch process. Previous versions of CoreGenes have been used to classify bacteriophage genomes and mine data from pathogen genomes.

Findings: CoreGenes3.5 accepts as input GenBank accession numbers of genomes and performs iterative BLASTP analyses to output a set of core genes. After completion of the program run, the results can be either displayed in a new window for one pair of reference and query genomes or emailed to the user for multiple pairs of small genomes in tabular format.

Conclusions: With the number of genomes sequenced increasing daily and interest in determining phylogenetic relationships, CoreGenes3.5 provides a user-friendly web interface for wet-bench biologists to process multiple small genomes for core gene determinations. CoreGenes3.5 is available at http://binf.gmu.edu:8080/CoreGenes3.5.

Keywords: Core genes, Bacteriophage, Taxonomy, Viral genomics, Data mining

Findings

Background

Genes that are common between a set of genomes are known as core genes. Core sets of genes have been used to understand better bacterial genome evolution [1], orthology in viral genomes [2], viral evolutionary complexity [3], and to mine pathogen genomes [4]. Core genes have also been used to investigate the origins of photosynthesis [5], as well as to classify and untangle the taxonomy of bacteriophages [6-8]. With such a myriad of uses for core genes and the growing numbers of whole genome sequences, it is important to provide user-friendly and validated software tools for the determination of these genes from sets of genomes. Originally developed in 2002 [9], CoreGenes, a tool for the identification of shared and unique genes among (small) genomes, has been continually updated and refined in response to user demands [10]. These changes include increased robustness of the tool, as well as the ability to

upload custom and proprietary data not deposited in GenBank. The major update to this version is the ability and versatility to batch process multiple pairs of small genomes, freeing the user from repetitive and time-consuming manual entry of genome sets. This is of benefit to users who have several large sets of genomes that they wish to analyze, for example a family of bacteriophages.

Other software tools have been developed for the determination of core genes including mGenomeSubtractor [11], CEGMA [12], nWayComp [13], and GenomeBlast [14]. mGenomeSubtractor and GenomeBlast both use BLAST-based algorithms to identify core genes. Of these, mGenomeSubtractor is primarily intended for use with bacterial genomes, whilst CEGMA is intended primarily for eukaryotic genomes; nWayComp and GenomeBlast are no longer accessible online, as is another genome comparison tool called GOAT [15]. In contrast, CoreGenes has been continuously available online since 2002, and shown to be invaluable in characterizing and re-determining the taxonomy and relationships of bacteriophages based on coding sequences [6,7,16-19]. It is anticipated that this timely update of CoreGenes will enable the analysis of

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Figure 1 Web interface for CoreGenes3.5. A reference genome accession number is entered into the first text field and query genome accession numbers are entered into the subsequent fields. Options for modifying the BLASTP threshold score and an email address are provided.

shared proteins among viral and small bacterial genomes in a faster and more efficient manner.

Implementation

CoreGenes3.5 is implemented using Java, Javascript, and HTML. It uses an iterative BLASTP algorithm that processes a reference genome and multiple query genomes. This is based on the GeneOrder algorithm described previously [20,21]. Briefly, these genomes are retrieved directly from GenBank, or custom-entered by the user, and the gene translations are parsed from the files. Each protein from the first query genome is analyzed against the reference genome proteins using BLASTP, creating a new reference genome which is a subset of the original reference genome and which contains those proteins that meet or exceed the user-defined similarity threshold

Figure 2 Web interface for batch processing in CoreGenes3.5. Reference and query accession numbers are entered in 'comma delimited' format. An email address is required so that output tables are emailed to the user following completion of the analysis.

The number of homologs in each column is : 19
Total number of genes in genome 1 : 73
% in common, with respect to genome 1: 26.03
Total number of genes in genome 2 : 72
% in common, with respect to genome 2: 26.39

Enterobacteria phage lambda	Enterobacteria phage P22
NC_001416	NC_002371
GI:9626266 PRODUCT:Tail fiber protein	GI:51236728 PRODUCT:head completion protein
GI:9626276 PRODUCT:ea22	GI:51236733 PRODUCT:EaD
GI:9626284 PRODUCT:antitermination protein	GI:9635508 PRODUCT:C3
GI:9626286 PRODUCT:restriction alleviation protein	GI:9635512 PRODUCT:Ral
GI:19263394 PRODUCT:Superinfection exclusion protein B	GI:9635513 PRODUCT:SieB
GI:9626292 PRODUCT:repressor	GI:9635515 PRODUCT:C2
GI:9626294 PRODUCT:cII protein	GI:9635517 PRODUCT:C1

Figure 3 Partial output of the analysis between the Enterobacteria phages lambda and P22, generated using the batch mode input. Links (blue) are provided, pointing to whole genome data and specific proteins archived in GenBank. Statistics of how many homologs are found by CoreGenes are summarized at the top of the table.

(BLASTP score). The second query genome is then BLASTP-analyzed against this new reference genome, creating another new reference genome. This iterative process continues until no further query genomes remain. The final output consists of proteins that are common to all the input genomes.

Batch processing of genomes in CoreGenes3.5 is implemented using Javascript and provides the facility to perform comparisons between many pairs of reference and query genomes. Given two lists of genome accession numbers, the script iteratively submits genomes in pairs to the main CoreGenes program such that each reference genome is individually compared with each query genome. When two identical input lists of genome accession numbers are provided, the script results in an 'all versus all' comparison, where all genomes are individually compared against each other as both reference and query.

Additionally, each genome is compared against itself, providing an internal control where the number of paired proteins should equal the number of protein encoding genes denoted within the genome annotation.

As the BLASTP comparisons are performed *ab-initio* and not pre-computed, CoreGenes3.5 is limited to genome sizes of 2 Mb or less. While CoreGenes3.5 can take larger genomes as input, the time taken to process them also increases. Therefore, it is recommended that users submit genomes with the aforementioned limit.

Results and discussion

The input to CoreGenes3.5 consists of a reference genome and a query genome. These are entered into the text fields on the web interface as GenBank accession numbers (Figure 1). As noted, the BLASTP threshold score can be changed from its default of "75". An email address must be entered to which results are sent. For genomes that are not available in GenBank, a link is provided to a custom genome entry page where the user can upload proprietary and unpublished data (these data and results are scrubbed from the server nightly to provide a level of confidentiality). In the batch entry mode (Figure 2), the input consists of two "comma-delineated" lists of GenBank accession numbers in addition to the threshold field. Additionally, CoreGenes3.5 works best with the Mozilla Firefox web browser.

The output of CoreGenes3.5 is a list of core genes in tabular format (Figure 3). Each gene is linked to its corresponding entry in GenBank. This allows users to identify particular genes of interest for further investigation, provide valuable insights for annotation of function and inform the design of wet-bench studies. In batch processing mode, results tables are emailed to the user in HTML format which can be viewed using any web browser.

The advent and continued development of next generation technologies has substantially increased the throughput and fidelity of genome sequence data. With reducing costs, the number of viral and bacterial genomes deposited in the International Nucleotide Sequence Databases/ GenBank has grown rapidly (and continues to do so). It is therefore crucial to continue the development and improvement of novel and existing software tools that can efficiently mine this expanding wealth of sequence data and facilitate comparisons of multiple closely or distantly related genomes.

CoreGenes3.5 is the latest and most versatile update to a user-friendly tool for locating and identifying core genes from viral and small bacterial genomes. Like previous versions of CoreGenes, this newest version will be continually updated in response to demands from the user community. The ability of CoreGenes to deal with larger bacterial genomes is actively being addressed.

Conclusions

The batch processing feature of CoreGenes3.5 enables researchers to analyze multiple small genomes expeditiously using a web interface. This allows users to data mine the increasing numbers of genomes in sequence databases and to determine quickly the phylogenetic relationships amongst them.

Availability and requirements

Project name: CoreGenes3.5
Project home page: http://binf.gmu.edu:8080/Core Genes3.5
Operating system(s): Platform independent
Programming language: Java
Any restrictions to use by non-academics: License required for commercial usage

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References

1. Liang W, Zhao Y, Chen C, Cui X, Yu J, Xiao J, Kan B: **Pan-Genomic analysis provides insights into the genomic variation and evolution of Salmonella Paratyphi A.** *PLoS One* 2012, **7:**e45346.
2. Garavaglia MJ, Miele SAB, Iserte JA, Belaich MN, Ghiringhelli PD: **The ac53, ac78, ac101 and ac103 are newly discovered core genes in the family Baculoviridae.** *J Virol* 2012, **86:**12069–12079.
3. Yutin N, Koonin EV: **Hidden evolutionary complexity of Nucleo-Cytoplasmic Large DNA viruses of eukaryotes.** *Virol J* 2012, **9:**161.
4. Mahadevan P, King JF, Seto D: **Data mining pathogen genomes using GeneOrder and CoreGenes and CGUG: gene order, synteny and in silico proteomes.** *Int J Comput Biol Drug Des* 2009, **2:**100–114.
5. Mulkidjanian AY, Koonin EV, Makarova KS, Mekhedov SL, Sorokin A, Wolf YI, Dufresne A, Partensky F, Burd H, Kaznadzey D, Haselkorn R, Galperin MY: **The cyanobacterial genome core and the origin of photosynthesis.** *Proc Natl Acad Sci USA* 2006, **103:**13126–13131.
6. Lavigne R, Darius P, Summer EJ, Seto D, Mahadevan P, Nilsson AS, Ackermann HW, Kropinski AM: **Classification of Myoviridae bacteriophages using protein sequence similarity.** *BMC Microbiol* 2009, **9:**224.
7. Lavigne R, Seto D, Mahadevan P, Ackermann H-W, Kropinski AM: **Unifying classical and molecular taxonomic classification: analysis of the Podoviridae using BLASTP-based tools.** *Res Microbiol* 2008, **159:**406–414.
8. Mahadevan P, Seto D: **Taxonomic parsing of bacteriophages using core genes and in silico proteome-based CGUG and applications to small bacterial genomes.** *Adv Exp Med Biol* 2010, **680:**379–385.
9. Zafar N, Mazumder R, Seto D: **CoreGenes: a computational tool for identifying and cataloging "core" genes in a set of small genomes.** *BMC Bioinforma* 2002, **3:**12.
10. Mahadevan P, King JF, Seto D: **CGUG: in silico proteome and genome parsing tool for the determination of "core" and unique genes in the analysis of genomes up to ca. 1.9 Mb.** *BMC Res Notes* 2009, **2:**168.
11. Shao Y, He X, Harrison EM, Tai C, Ou H-Y, Rajakumar K, Deng Z: **mGenomeSubtractor: a web-based tool for parallel in silico subtractive hybridization analysis of multiple bacterial genomes.** *Nucleic Acids Res* 2010, **38:**W194–200.
12. Parra G, Bradnam K, Korf I: **CEGMA: a pipeline to accurately annotate core genes in eukaryotic genomes.** *Bioinformatics* 2007, **23:**1061–1067.
13. Yao J, Lin H, Doddapaneni H, Civerolo EL: **nWayComp: a genome-wide sequence comparison tool for multiple strains/species of phylogenetically related microorganisms.** *In Silico Biol (Gedrukt)* 2007, **7:**195–200.
14. Lu G, Jiang L, Helikar RMK, Rowley TW, Zhang L, Chen X, Moriyama EN: **GenomeBlast: a web tool for small genome comparison.** *BMC Bioinforma* 2006, **7**(Suppl 4):S18.
15. Kaluszka A, Gibas C: **Interactive gene-order comparison for multiple small genomes.** *Bioinformatics* 2004, **20:**3662–3664.
16. Chibeu A, Lingohr EJ, Masson L, Manges A, Harel J, Ackermann H-W, Kropinski AM, Boerlin P: **Bacteriophages with the ability to degrade uropathogenic Escherichia coli biofilms.** *Viruses* 2012, **4:**471–487.
17. Kropinski AM, Van den Bossche A, Lavigne R, Noben J-P, Babinger P, Schmitt R: **Genome and proteome analysis of 7-7-1, a flagellotropic phage infecting Agrobacterium sp H13-3.** *Virol J* 2012, **9:**102.
18. Lehman SM, Kropinski AM, Castle AJ, Svircev AM: **Complete genome of the broad-host-range Erwinia amylovora phage phiEa21-4 and its relationship to Salmonella phage felix O1.** *Appl Environ Microbiol* 2009, **75:**2139–2147.
19. Villegas A, She Y-M, Kropinski AM, Lingohr EJ, Mazzocco A, Ojha S, Waddell TE, Ackermann H-W, Moyles DM, Ahmed R, Johnson RP: **The genome and proteome of a virulent Escherichia coli O157:H7 bacteriophage closely resembling Salmonella phage Felix O1.** *Virol J* 2009, **6:**41.
20. Celamkoti S, Kundeti S, Purkayastha A, Mazumder R, Buck C, Seto D: **GeneOrder3.0: software for comparing the order of genes in pairs of small bacterial genomes.** *BMC Bioinforma* 2004, **5:**52.
21. Mahadevan P, Seto D: **Rapid pair-wise synteny analysis of large bacterial genomes using web-based GeneOrder4.0.** *BMC Res Notes* 2010, **3:**41.