GOHTAM: a website for ‘Genomic Origin of Horizontal Transfers, Alignment and Metagenomics’
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ABSTRACT
Motivation: This website allows the detection of horizontal transfers based on a combination of parametric methods and proposes an origin by researching neighbors in a bank of genomic signatures. This bank is also used to research an origin to DNA fragments from metagenomics studies.
Results: Different services are provided like the possibility of inferring a phylogenetic tree with sequence signatures or comparing two genomes and displaying the rearrangements that happened since their separation.
Availability and implementation: http://gohtam.rpbs.univ-paris-diderot.fr/
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1 INTRODUCTION
Horizontal transfers (HTs) are a major force of evolution (Keeling and Palmer, 2008; Ochman et al., 2000) and this website proposes methods for their detection. The genomic signature was demonstrated to be species-specific (Deschavanne et al., 1999; Sandberg et al., 2001) and allows HT detection in terms of tetranucleotide frequencies (Dufraigne et al., 2005). Parametric methods were designed to work only with the information contained in genomic sequences. They rely either on the whole set of genes or on local variations of genomic signature (Dufraigne et al., 2005; Mallet et al., 2010). Recently, a benchmark has determined the most efficient parametric methods in different conditions and has proposed to use a combination of methods to analyze HTs in genomes (Becq et al., 2010). Up to now, these methods were never proposed for online genome analysis (Fig. 1A).

2 GOHTAM SERVICES
2.1 HT detection
The two methods proposed can be used alone or in combination. The first is a window-based signature method as described in Dufraigne et al. (2005), except that the distance used is the Jensen–Shannon divergence, a symmetric version of the Kullback–Leibler divergence (Azad and Lawrence, 2007; Becq et al., 2010). Either sensitivity or specificity can be increased by adjustable classification process (Azad and Lawrence, 2007). A gene-based method is also proposed with the same distance (Becq et al., 2010). Up to now, these methods were never proposed for online genome analysis (Fig. 1A).

2.2 Bank of genomic signatures
A key feature of GOHTAM is the biggest bank of genomic signatures to date. Instead of using only complete genomes (van Passel et al., 2005; Teeling et al., 2004), this bank is based on the whole set of sequences of Genbank (release 188, only sequences <1 kbp were discarded) and contains ~248 000 tetranucleotide species signatures. The bank is updated at each major release.

2.3 Origin of transferred regions
Each detected region signature is compared with the signatures of the bank and the 10 closest neighbors are displayed with a confidence rating depending on the length of both query and reference sequences and the distance between the two signatures (Fig. 1B).
2.4 Metagenomics
In the case of a metagenomics study, a sequence or a set of sequences (multi-Fasta) is loaded; the signatures of these sequences are compared as above to propose a species of origin.

2.5 Oligonucleotide content
The whole set of tetranucleotides of a sequence represents the signature of a sequence (Deschavanne et al., 1999). This signature of the 256 possible tetranucleotides is under the form of a $16 \times 16$ frequency matrix and can be displayed as a signature image (Fig. 1D).

2.6 Phylogenetic tree of sequence signatures
It was shown that the species specificity of genomic signatures could be used to infer phylogenetic trees (Chapus et al., 2005). Loading a multi-Fasta file of sequences leads to build a neighbor-joining phylogenetic tree (Fig. 1C; Felsenstein, 2005).

2.7 Genome alignment
The website uses maximum unique matches (MUMs) to align genomes. All rearrangements superior to 1 kb between two genomes are graphically displayed with the possibility to choose a region or modify the length of MUMs (Fig. 1E; Delcher et al. 1999).

3 IMPLEMENTATION
Except for use of programs like the Phylip package (http://evolution.gs.washington.edu/phylip.html) or Mummer (http://mummer.sourceforge.net/), the original programs are written in Python, Perl or R and available at: http://gohtam.rpbs.univ-paris-diderot.fr:8080/Data/bin/GOHTAM_bin.tgz

An online help is available. Some analyses require time: HT detection lasts ~6 min and the research for neighbors ~2 min depending on the server load and the sequence length.

This site provides some unique features in terms of HT detection, origin of HT regions, metagenomics studies as well as for phylogenetic analyses of homologous or non-homologous sequences due to its extended reference database and improves the analyses proposed by other sites of genome analysis (van Passel et al., 2005; Teeling et al., 2004).

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