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## 1 General

### **Q. What is the Genchek Package?**

**A.** The Genchek package is an integrated comprehensive sequence analysis package consisting of over 50 programs.

### **Q. How can I evaluate Genchek™?**

**A.** You can request Free Evaluation Version by filling the Product Enquiry Form available on our site.

### **Q. Is there any feature which is restricted in the trial version?**

**A.** User cannot create new project. Exporting, saving, copying and printing of analysis and images are not allowed.

### **Q. Is there a live help support for Genchek™?**

**A.** Genchek™ has a dedicated technical support staff to answer your queries. For live help just go to our website and click on Live Help. Here you can chat LIVE with our experts regarding your queries.

### **Q. Is there a Network Version ? How does it work? Or, How can I run Genchek™ on several machines without having to move the copy protection key all the time?**

**A.** We have network compatible programs for all the operating systems. Only one copy protection key is needed and that can be placed on any computer on your network. The key server program monitors the number of users running the Genchek™ program across the network.

### **Q. What are Proxy Settings?**

**A.** In an enterprise that uses the Internet, a proxy server is a server that acts as an intermediary between a workstation/user and the internet so that the enterprise can ensure security, administrative control, and caching service. A proxy server is associated with or part of a gateway server that separates the enterprise network from the outside network as well as a firewall server that protects the enterprise network from outside intrusion. Proxy settings in Genchek™ involves IP address of the proxy server of



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the organization and its port number.

### **Q. When do I need to set proxy?**

**A.** You need to set proxy in case you or your organization is connected to the internet through a proxy server.

### **Q. What is a port? Should the port always be 2001?**

**A.** Ports provide a means by which computers communicate with one another using standard protocols. Each protocol is assigned to a fixed port number so that the computers know which port to check, in order to communicate using the specified protocol. When you start up a program that will communicate with remote computers using a specific protocol, the port assigned to that protocol is the one that it will use to attempt to communicate. The port number being used needs to be the same on each computer attempting to communicate in order that the message packets sent by the one computer are correctly received and recognized by the other. Port numbers fall into three ranges. Ports 0 through 1023 are called "Well Known Ports" and are assigned to their specified purpose by the Internet Assigned Numbers Authority (IANA). Ports 124 through 49151 are available for general use and may be registered with IANA if desired. Ports 49152 through 65535 are used where a dynamic or private port is required. You can protect your computer against some forms of attack over the Internet by restricting the ports that your computer allows other computers to use to communicate with it. For maximum security, you should ensure that access to all ports, except those that you need to use, are blocked.

### **Q. Is Genchek LINUX compatible?**

**A.** Yes. Genchek is developed using advanced Java technologies and that's why it is platform/OS independent. So the user can install it on Windows, LINUX, UNIX, Mac or Solaris.

### **Q. What are the system requirements?**

**A.** Genchek requires normal PC i.e., Pentium 3 and above with minimum 256Mb RAM and minimum 200 Mb free disk space.



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**Q. Does Genchek work with Oracle or SQL server?**

A. Genchek works with Oracle as well as with My SQL.

**Q. Does Genchek Support Network License?**

A. Yes it does. Genchek also facilitates Network license management.

**Q. What is a Project?**

A. A project is a place where different sequences (DNA, Protein, Vector, Phylogenetics and Assembly) along with analysis results can be stored in different experiments.

**Q. What is a Sublist ?**

A. A sublist is a logical grouping of sequences. A sublist may group together sequences that have been imported from a particular location or sequences that have a biological relation or sequences that are a part of a particular project. The grouping of sequence is left to the discretion of the user.

**Q. How many Users does Genchek support?**

A. There is no limit for Users.

**Q. I am not a customer. Can I get support?**

A. Sure. We treat both customers and trial users with the same attention.

## 2 Sequences (Import/Export)

**Q. Is there any restriction on the numbers of sequences to be imported ?**

A. No, Genchek™ has no restriction on the number of sequences that can be imported simultaneously. It will depend on the computer specifications on which Genchek™ is running.

**Q. What data formats does Genchek™ support?**

A. FASTA, GenBank, EMBL, Text, SCF, ABI, DDBJ.

**Q. What are the different ways of importing sequences in Genchek™ ?**

A. User can load sequences into Genchek™ from the local system and if the user has a list of Accession Numbers, then they can use import from NCBI or EBI option available in Genchek™.

**Q. I don't see my file when I click Browse button to import file from system ?**

A. Check the file format you want to import and set **Files of Type** field accordingly. If the format is not supported by Genchek™, then an error message pops up.

**Q. I have a sequence data from an automated sequencer. How do I use the data in the Genchek™ Application ?**

A. Genchek™ allows import of electropherogram ( chromatogram ) from a sequencing machine. A user can trace all standard electropherograms (e.g., \*.abi, \*.scf). Genchek™ also allows a researcher to rescale an image, edit a sequence, clip noisy peaks and convert an electropherogram data to text format.

**Q. Does Genchek™ support export of files in image format ?**

A. Yes, Genchek™ supports the export of output files as images as well as in text format.

**Q. Does Genchek Supports other tool formats?**

A. Yes, Genchek™ supports other tool formats. User can import VNTI, Lasergene, GeneJockey and JellyFish formats available at **Sequence > Import Sequence**.

**Q. What is the maximum size of sequence that Genchek support ?**

A. We tested Genchek import facility for maximum up to 22 Mbps FASTA format.

**Q. Can I delete a sequence from data browser or a selected region of a sequence?**

A. Yes, Genchek allows sequence deletion from data browser by using the delete option in the sequence menu. The user can also delete a region of sequence by right click menu options on the sequence editor.

**Q. What is Genchek's capacity for importing sequences?**

A. From our stress testing exercise we could import  $15 \times 10^6$  Bps sequence and also visualize in 3 view window on normal PC(P4 and 256Mb ram). But with better hardware we could import upto  $60 \times 10^6$  Bps sequences which is equivalent to a big bacterial genome. We also successfully imported 3 lacs EST's in to Genchek.

**Q. Why are some menu options disabled though a sequence is selected in the Data Browser?**

A. The menu options in Genchek™ are enabled depending on the type and number of sequences selected in the data browser and as well as based on License and Logged User Role.

**Q. Can I show the Annotations for saved sequences?**

A. Yes, user can view the annotations in Graphical Viewer.

**Q. Does Genchek allow full control to the user over the sequences database?**

A. Yes. Genchek allows import, edit and export of sequences. It also creates new sequence database.

**Q. Is ancestral relationship or parent - child relationship maintained between sequences in Genchek?**

A. We do record parent - child relationships in Genchek. User can view the relation from **Sequence Menu > Ancestral History**.

**Q. Does Genchek support importing tree files exported from Lasergene?**

A. Yes. Genchek supports standard formats like Newick and Nexus exported from Lasergene tool.

**Q. If sequences include U (Uracil), can they still be processed?**

A. Yes, sequences that include U can be processed.

**Q. Can I manually create a sequence in Genchek?**

A. Yes, user can create a sequence manually using **Sequence > New Sequence** menu or New Sequence toolbar button.

**Q. Can I rename the sequence available in Genchek browser?**

A. Yes, user can rename a sequence manually using **Sequence > Rename Sequence** menu or right click on a sequence from Sequence Browser.

**Q. What are the formats in which sequences can be exported?**

A. User can export the sequences in FASTA, Genbank, EMBL and DDBJ formats.

**Q. Can I add annotations manually to sequence?**

A. Yes, user can add annotations manually to sequence using Add Annotation toolbar button from Graphical View.

**Q. Can I view an annotations in a graphical viewer?**

A. Yes, user can view annotations in graphical viewer. User also have the option to hide the Graphical representation of annotations using Features tab in Graphical View by selecting or clearing the checkboxes correspondingly.

**Q. What file formats of chromatogram sequences does Genchek support?**

A. Genchek supports .abi, .abd, .abl and .scf formats.

**Q. Is it possible to view traces of the chromatogram sequences?**

A. Yes, user can view the traces of the chromatogram sequences using Graphical View.

**Q. Can I view the quality values of the chromatogram sequences?**

A. Yes, user can view the quality values of the chromatogram sequences using View Chromatogram toolbar button from Graphical View.

### 3 Primer Design

**Q. How can I export the sequence of a primer I've just designed?**

A. Select the desired primer from primer viewer window, click **Export** button to export the primer sequence to a disk file or to copy the primer sequence to the Windows clipboard so that you can paste the primer sequence into another application.

**Q. Is there any special tool for PCR to show the Multiplexing of Probes?**

A. Yes, user can copy the probes from Primer Result Viewer and perform Multiplexing from Oligo Evaluation.

**Q. How and why does the primer results differ when different primer design tools are used?**

A. Some of the freely available tools have different set of parameters and differ in criteria for primer selection. Genchek's Primer design tool takes into account all the standard parameters and conditions under which the user can select correct primers.

**Q. Does Genchek have primer design and is able to store results?**

A. Yes, Genchek has primer design tool and is not only able to store the results but is also able to export the results. Algorithm explained.

**Q. How are conserved regions found before degenerate primer design?**

A. In proteins alignment degenerate primers are found based on the frequency of occurrence. While in

the DNA alignment we can also use entropy calculations for finding conserved regions.

**Q. Does Genchek allows to save the product from the primer analysis?**

A. Yes, user has the option to save the Forward and Reverse sequence in the data browser.

**Q. Does Genchek support TaqMan Design?**

A. Yes, TaqMan Design can be used for designing TaqMan probes and user also has the option to design primers for the designed TaqMan probes.

**Q. What are the different primer designs available in Genchek?**

A. TaqMan Design, Exon Junction and SSR Marker Primer Design.

**Q. Does Genchek support SYBR Primer design?**

A. Yes, Genchek supports both SYBR and PCR Primer Design.

**Q. Does Genchek have SSR search program?**

A. Yes, Genchek supports SSR search and also provides option for the user to design primers for the selected SSR.

**Q. It is possible to search SSR based on motif length?**

A. Yes, user has the option to search SSR based on motif length and motif repeat.

**Q. Can I export SSR results from Genchek?**

A. Yes, user can export SSR results in a text file.

## 4 Restriction Enzyme Analysis

### Q. How can I add Restriction Enzymes to the list?

A. To add an enzyme to a sublist, right click on the existing list and select **Create**. A new window opens. Enter the recognition sequence and cutting position and click **Save** button to add the enzyme to the existing list.

### Q. Can the RE map be export in text and picture format and what picture formats?

A. Yes. If the RE analysis is done from the Common toolbar, then the result window provides Export option for both RE map and text results. Currently RE maps are exported in JPEG format but this facility can be extend to any other picture format.

### Q. What database do you use for Restriction enzymes in Genchek and is there are facility to update it automatically?

A. REBASE. Currently Genchek facilitates manual updates only.

### Q. Is it possible to manually create a new Restriction Enzyme specifying the cutting position?

A. Yes, user can create a New Restriction Enzyme manually by selecting **Restriction Enzyme** node from **Sequence** tree and click New toolbar button will open “New Restriction Enzyme” dialog box.

## 5 Stem Loop Analysis

### Q. Is there any program to analyze Stem loops of a sequence?

A. User need to place the UNA Fold program in **Genchek** folder to perform Stem Loop Analysis from Genchek.

## 6 Tools

### Q. What is Local BLAST?

A. Local BLAST manager module demonstrated/explained.

### Q. Why should I create a local database?

A. You need to create a local database only if you want to search your query sequence against sequences that are proprietary or patented.

### Q. Where should Local BLAST Manager be installed?

A. Local BLAST Manager should be installed in the same location where Genchek is installed. Install Local BLAST Manager in **BLAST** folder under Genchek installation folder. By default, this should be installed at **C:\Genchek\BLAST** (for Windows OS), **/usr/local/bin/Genchek/BLAST** (for LINUX & Solaris OS) and **/Genchek/BLAST** (for Mac OS).

### Q. Why do I need to set the Local BLAST settings?

A. Basically, to be able to query your sequence against your local BLAST database.

### Q. How should I start Local BLAST Manager?

A. If default selections are made while installing Local BLAST Manager, select **Start > Programs > Local BLAST Manager - Ocimum Biosolutions > Local BLAST Manager** (for Windows OS), **/usr/bin/Local BLAST Manager - Ocimum Biosolutions/Local BLAST Manager** (for LINUX/Solaris OS) and **Applications > Local BLAST Manager - Ocimum Biosolutions > Local BLAST Manager** (for Mac OS).

### Q. Where is my local database created?

A. Your local database is a collection of flat files containing sequences saved on your local drive where Genchek™ has been installed.



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### **Q. Why is my BLAST server forever sending the sequence?**

**A.** Check your proxy settings in the Preferences option of System in the main menu. The proxy settings may need to be reset.

### **Q. Local BLAST gives zero hits. What should I do?**

**A.** To avoid this, install Local BLAST Manager in the same location where Genchek is installed. Install Local BLAST Manager in **BLAST** folder under Genchek installation folder. By default, this should be installed at **C:\Genchek\BLAST** (for Windows OS), **/usr/local/bin/Genchek/BLAST** (for LINUX & Solaris OS ) and **/Genchek/BLAST** (for Mac OS).

### **Q. “Illegal characters were removed. Database built with errors” message is displayed when I perform Local BLAST. Why do I get this message?**

**A.** Database build might have invalid characters.

### **Q. I tried creating Local BLAST Database using sequences from Genchek Data Browser. I get this message “Unable to format given database.” Why do I get this message?**

**A.** Local BLAST Manager should be installed in the same location where Genchek is installed. Install Local BLAST Manager in **BLAST** folder under Genchek installation folder. By default, this should be installed at **C:\Genchek\BLAST** (for Windows OS), **/usr/local/bin/Genchek/BLAST** (for LINUX & Solaris OS) and **/Genchek/BLAST** (for Mac OS).

### **Q. On selecting Local BLAST option in Select BLAST type dialog, a message 'Start Local BLAST Server from Local BLAST Manager' is seen. How should I start Local BLAST server?**

**A.** Before using Local BLAST module, start Local BLAST Manager. If default selections are made while installing Local BLAST Manager, select **Start > Programs > Local BLAST Manager - Ocimum Biosolutions > Local BLAST Manager** (for Windows OS), **/usr/bin/Local BLAST Manager - Ocimum Biosolutions/Local BLAST Manager** (for LINUX/Solaris OS) and **Applications > Local BLAST Manager - Ocimum Biosolutions > Local BLAST Manager** (for Mac OS).

In “Local BLAST Manager” window, click **Start** button in Local BLAST Manager to start Local



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BLAST Server. Status bar shows message “Server Started”. Now you can go ahead and perform BLAST search on your local database.

### **Q. Can Genchek's Local BLAST perform BLAST locally on NCBI database?**

**A.** Yes. We need to download NCBI sequences in FASTA format and build a local database using Local BLAST Manager.

### **Q. When I try to create a Local BLAST database from Local BLAST Manager, I get a message “Error while creating Local BLAST Database”. Why do I get this message?**

**A.** Local BLAST Manager should be installed in the same location where Genchek is installed. Install Local BLAST Manager in **BLAST** folder under Genchek installation folder. By default, this should be installed at **C:\Genchek\BLAST** (for Windows OS), **/usr/local/bin/Genchek/BLAST** (for LINUX & Solaris OS) and **/Genchek/BLAST** (for Mac OS).

### **Q. 'Internet connection failed. Check your internet connection.' message is displayed when I perform NCBI BLAST. Why do I get this message?**

**A.** This could be due to two reasons. Firstly due to improper internet connection or due to spaces present in the name of the installation folder. To avoid getting this message, ensure proper internet connection and no spaces should be present in the name of the installation folder. By default, Genchek should be installed in **C:\Genchek** (for Windows OS), **/usr/local/bin/Genchek** (for LINUX & Solaris OS) and **Genchek** (for Mac OS).

### **Q. Can I access Local BLAST database from other system?**

**A.** Yes, Genchek facilitates this feature.

### **Q. It is possible to create a database of the sequences loaded in the Genchek and use them for local BLAST?**

**A.** Yes, select sequences from the browser and create a database which can be used for Local BLAST.

**Q. Can I download all the BLAST hit sequences at a time?**

A. Yes, user can download BLAST hit sequences from result viewer using **Save All** button.

**Q. What is a feature?**

A. A feature is anything that can be mapped onto a strand of DNA. It is defined by its start and stop location. A gene is a feature. A protein coding sequence (CDS or PEG) is a feature that, in bacteria, shares the same location on the DNA as its gene and may be represented as an amino acid sequence translated from the nucleotide sequence. Eukaryotic genes also have intron and exon features defined as subsets of the gene feature.

**Q. Is it possible to extract annotations or features of a sequence in a Genchek?**

A. Yes, we can extract by using the Feature Extraction module.

## 7 Sequence Alignment

**Q. Can we perform the Multiple Sequence Alignment for all types of Sequences?**

A. Yes.

**Q. What are different methods to perform Tree Analysis for Multiple Alignment Sequences?**

A. Neighbor-Joining.

**Q. Can we export the Consensus Sequence?**

A. Yes. We can export the Contig sequence in ACE file format which consists of quality of Contigs and fragments present in Contig.

**Q. Does the MSA module have print and export options?**

A. Yes. Genchek's MSA editor has printing and exporting options. The alignment files as well as publication quality pictures can be exported.

**Q. Does the MSA editor allow to add annotation map for a region of alignment such as conserved regions?**

A. Yes. If the user right clicks on the editor at consensus area pop up menu allows the user to add an annotation. After this the user can select any region of the alignment and label it.

**Q. Does your multiple alignment provide overall alignment score and % identity?**

A. User can select a pair of sequences to view the % identity and overall alignment score.

**Q. Can Genchek compare protein sequences from different organisms and find common regions?**

A. Yes. Genchek's multiple alignment tool and viewer performs this analysis.

**Q. Difference between pairwise alignment and PCA?**

A. Pairwise alignment finds similarity but PCA uses the distances between the aligned sequences and groups them in such a way so as to maximize the variance in one direction in the 3 D space.

**Q. Can the tree visualization tool show trees as rooted or unrooted or circular form ?**

A. User can view the tree in rooted format only.

**Q. Can you integrate Phylip into Genchek?**

A. Yes. We have customized third party tools into Genchek.

**Q. Can I export tree map and in text and shuffle leaves on a tree nodes?**

A. Yes. The tree visualizer allows user to save tree map in various picture formats such JPEG, EPS, TIFF, BMP. Also the user can export the tree in Newick format is text file.

**Q. What are the different algorithms available to perform multiple sequence alignment in Genchek?**

A. ClustalW and MAP.

**Q. What are the different pairwise alignment algorithms available in Genchek?**

A. Local Alignment, Global Alignment and Dot Plot.

**Q. What type of scoring matrix used for multiple sequence alignment for protein and DNA in Genchek?**

A. For DNA, IDENTITY, EDNAFULL and MATCH scoring matrix and for Protein, BLOSUM and PAM scoring matrix are used.

**Q. In what formats can a user export multiple sequence alignment? Or what output formats does ClustalW-MSA/MSA produce?**

A. User can save aligned sequences in Clustal (.aln), .pfam, .msf, .pir, .blc, Phylip (.ph), NEXUS formats and user can also save image in .jpg, .tif, .png and .bmp file formats.

**Q. In what formats can the tree be exported?**

A. Newick, EPS and PNG formats.

**Q. What are the default parameters used by ClustalW in Genchek?**

A. \* DNA Gap Open Penalty = 15.0 \* DNA Gap Extension Penalty = 6.66 \* DNA Matrix = Identity \* Protein Gap Open Penalty = 10.0 \* Protein Gap Extension Penalty = 0.2 \* Protein matrix = Gonnet

**Q. How can I get the color version of the alignment?**

A. The alignment will appear on the results page in black and white. There is an option **Color**, it displays alignment in color according to the physiochemical characteristics of the amino acids.

**Q. How can I create a local alignment of two sequences?**

A. A local alignment will identify the small parts of two sequences where there is some similarity. Genchek's pairwise alignment tool performs this analysis.

## 8 Vector Trimming

**Q. Can I perform the Vector trimming for all type of Vectors which are existing in database?**

**A.** Yes, user perform vector trimming from available vectors in database. User can also perform vector trimming using nr-UniVec database available at NCBI.

## 9 Vector Cloning

**Q. How do you perform Cloning in Genchek?**

**A.** Cloning modules demonstrated.

**Q. Does Genchek facilitates whole or partial linkers creation?**

**A.** Yes. Genchek facilitates this utility through **Vector Construction Tools** menu option.

**Q. Can we export Cloned Gene Vector?**

**A.** Yes.

**Q. Are the fragments generated automatically in Graphical Viewer while performing the Cloning?**

**A.** Yes, they will be generated based on the selected region between 2 Res.

**Q. How Genchek classifies the vector sequences into expression or destination vector or Donor or Entry clones?**

**A.** Genchek is able to classify vectors based on Genbank file information. Import automatically recognizes vector type if Genbank file is imported and assigns vector in data browser subtype column.

**Q. What are the different cloning algorithm available in Genchek?**

**A.** TA Cloning, Restriction Enzyme Cloning.

## 10 Protein Analysis

**Q. Does Genchek support DNA to protein translation and how does one know that which frame of translation is correct?**

**A.** Yes. One can translate a DNA sequence into protein in all six reading frames and the translated sequences can be BLASTed against the NR/swissprot database. BLAST results can be analyzed to find correct reading frame.

**Q. Can I add new genetic codons in the Reverse translation tool?**

**A.** We have provided big list of codon usage tables which is currently static. Based on the client requirements we can facilitate addition of new codon tables.

**Q. Can I change color scheme for Amino Acid frequency Bar chart?**

**A.** Currently user cannot change the color scheme.

**Q. Can this software support for searching the transcription Sequence sites**

**A.** Yes, we can perform by using Pattern Search tool.

**Q. Is Prosite Pattern search tool available in Genchek?**

**A.** Yes. User can download the Prosite patterns in one click and use them for pattern search.

## 11 Assembly

**Q. Can I add reference sequences while assembling fragments?**

**A.** Yes.

**Q. What assembly programs does Genchek support?**

**A.** Phred Phrap, PolyPhred and CAP 3 programs.

**Q. Is it possible to trim low quality regions in chromatogram sequences?**

**A.** Yes, we can trim the sequence by using a End Clipping module.

**Q. How can I find out if my samples contain information about Quality Values (QV)?**

**A.** With Genchek, it is very easy to get this information. Double click on SCF sequence (from browser) to open it in Graphical Viewer and click **View Chromatogram** button. If you see blue columns above each base in chromatogram, then your file contains Quality Value information.

## 12 Mutagenesis

**Q. How do you find mutations in Genchek?**

**A.** PolyPhred, SNP BLAST and Mutagenesis modules demonstrated.

## 13 Oligo Analysis

**Q. Can I append RE sites to the oligo and analyzed oligo properties?**

**A.** Yes. Demonstrated fragments generation tools and oligo analysis.

**Q. What are the different oligo analysis available in Genchek?**

**A.** Multiplex, batch analysis and oligo search.

## 14 User Management

**Q. How do we integrate third party tool like ClustalW, Phred Phrap into Genchek?**

**A.** By using a.exe files.

**Q. Is it possible to upgrade the URLs of NCBI Blast, RE Base and Prosite by a user?**

**A.** Yes, user can upgrade NCBI Blast, RE Base and Prosite by selecting **Preferences > URL > Management** menu.

**Q. What are Role Privileges?**

**A.** Role Privileges define the role of the user. The role can be Admin, Power, Normal and Applications. Users associated with these roles have defined authority.

**Q. What are Authority Privileges?**

**A.** Authority Privileges are associated with roles. These define authority to access modules in Genchek.

**Q. Can we create a user for a specific time period?**

**A.** Yes, while creating a user, admin can specify expiry date for the user.

**Q. It is possible to activate or deactivate a user?**

**A.** Yes, admin can activate or deactivate a user using **Activate** checkbox option in User Management.

**Q. Can I create a role privilege having only DNA analysis privileges?**

**A.** Yes, admin need to create a role privilege having only DNA analysis options selected and should then associate the user with that role privilege.

## 15 Web Interface

**Q. How is the Web Interface module helpful?**

**A.** Web Interface allows users to set web links for online analysis tools. Through Web Interface module, the user is able to send sequence for analysis to the selected web tool.