CCLRC
Daresbury Laboratory

MrBUMP - Automated Search Model Generation and Molecular Replacement

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HTPX

The aim of Mr Bump

•An automation framework for Molecular Replacement.
•Particular emphasis on generating a variety of search models.

•Wraps Phaser, Molrep and Amore.
•Also uses a variety of helper applications (e.g. Chainsaw) and bioinformatics tools (e.g. Fasta, Mafft)
•Uses on-line databases (e.g. PDB, Scop)

•In favourable cases, gives “one-button” solution
•In unfavourable cases, will suggest likely search models for manual investigation

Target MTZ
&
Sequence

Target Details

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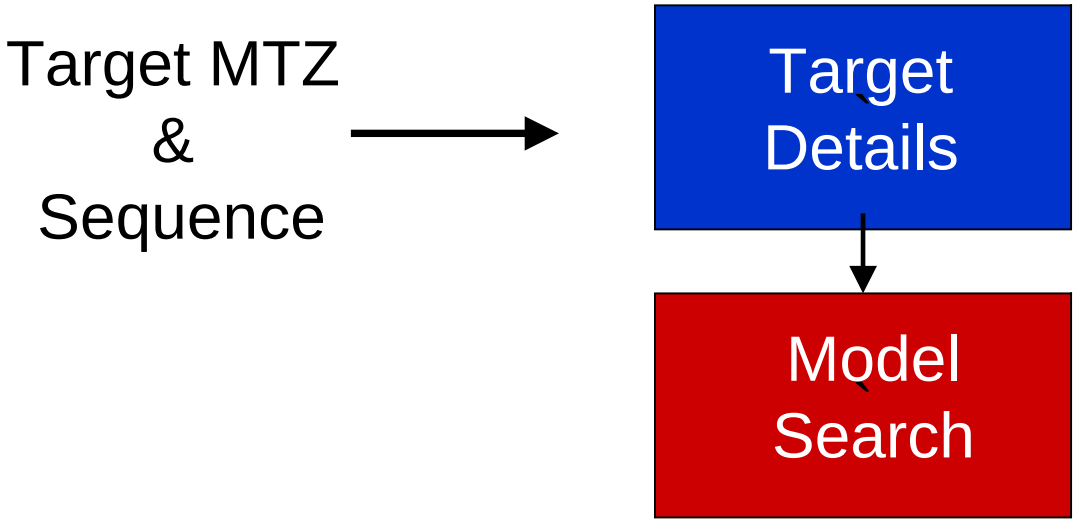
Target Details

- Calculate:
  - Matthews Coefficient.
  - Estimated number of molecules in the a.s.u.
  - Molecular weight.
  - Extraction of any other relevant information.



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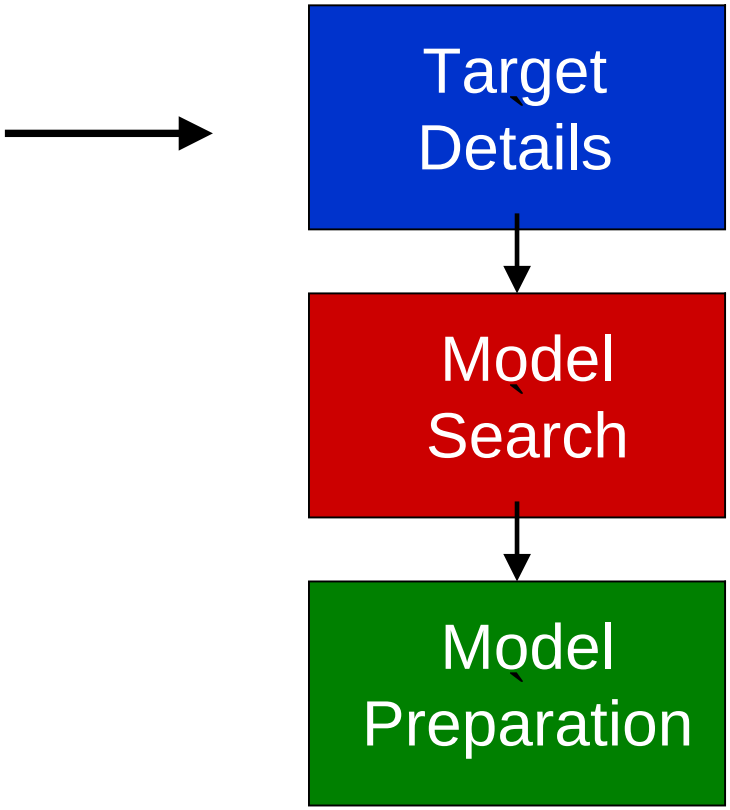
Model Search

- Template search:
  - Fasta sequence-based search using web or a local database of PDB sequences,
  - SSM secondary structure search using EBI webservice,
  - SCOP search (EBI) for domains within structures found in Fasta and SSM searches,
  - PQS search (EBI) for multimers based on structures found in Fasta and SSM searches.
- Multiple Alignment (Mafft or Clustalw) of all sequences with the target sequence and top scoring models are selected for further processing.

Target MTZ
&
Sequence



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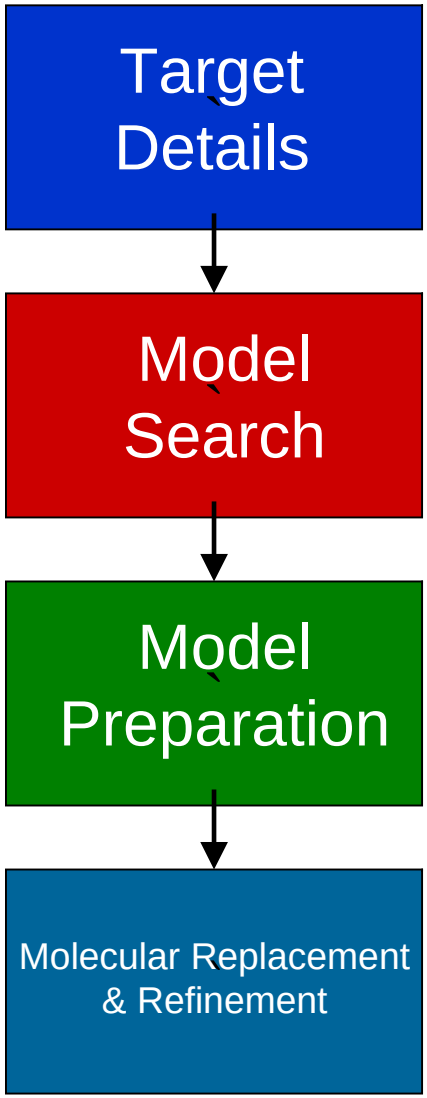
Model Preparation

- PDB files downloaded.
- SCOP results are used to extract domains from the PDB files.
- Multimer PDB files retrieved from PQS server.
- Search models prepared in three ways:
  - PDBclip – original PDB with waters removed, hydrogens removed, most probable confirmations for side chains selected and chain ID's added if missing.
  - Molrep – side-chain pruning
  - Chainsaw – more severe side-chain pruning

Target MTZ
&
Sequence



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Molecular Replacement and Refinement

- search models can be processed with Amore, Molrep or Phaser or combinations of all three,
- an ensemble of the top search models is created as an additional input model to Phaser,
- Refmac used for restrained refinement,
- change in Rfree value during refinement is used to determine how good the resulting model is,
- marginal solutions are high-lighted.

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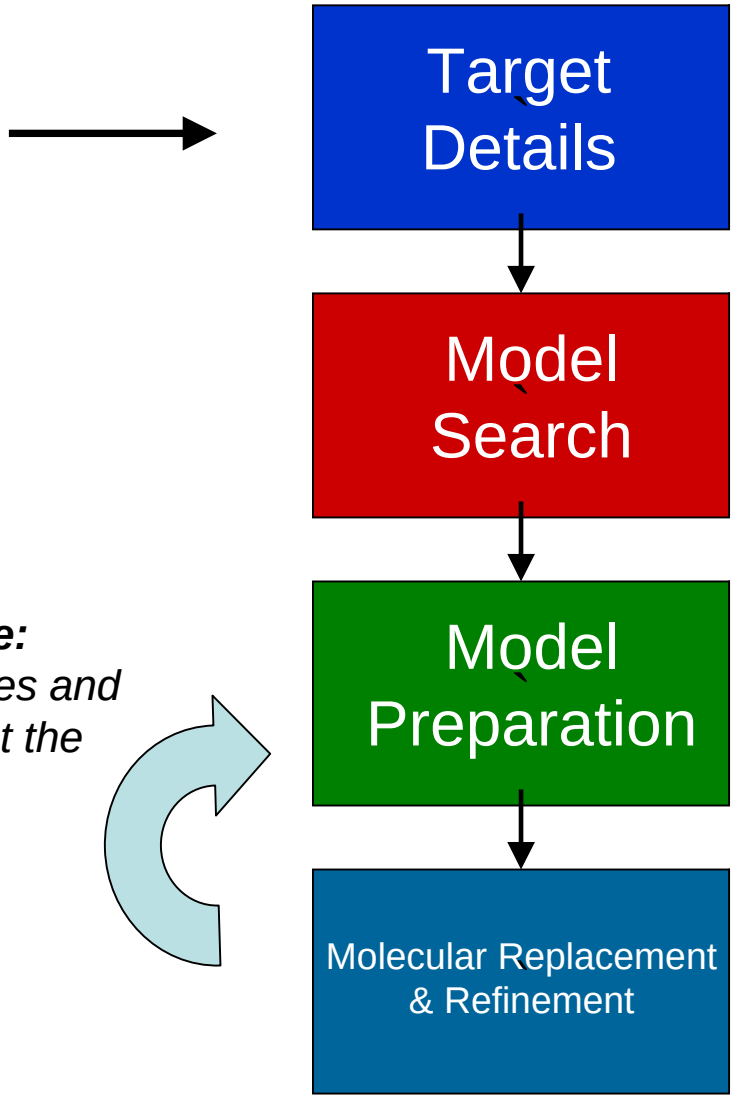
Target MTZ
&
Sequence



Serial mode:
Check Scores and exit or select the next model

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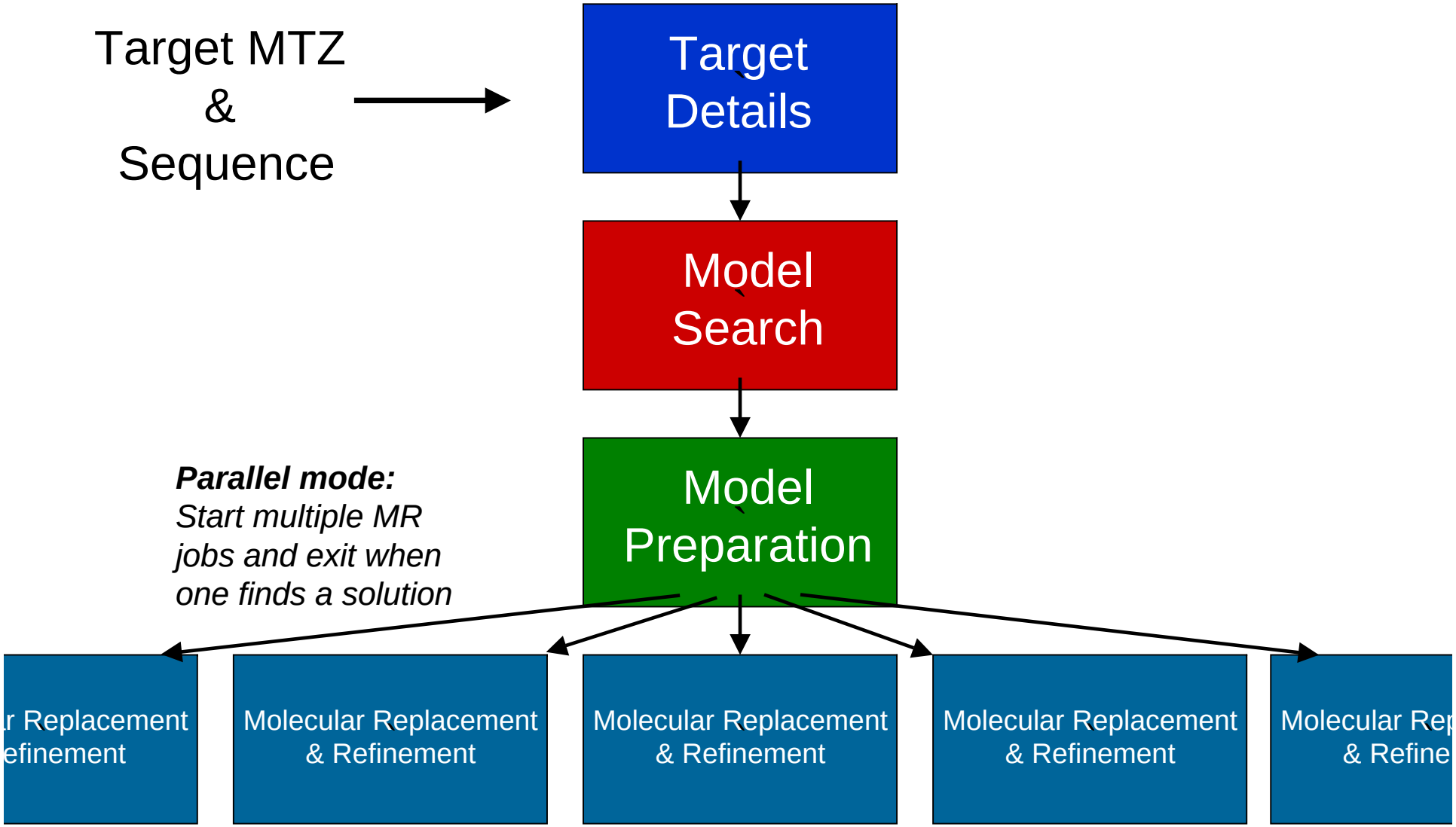
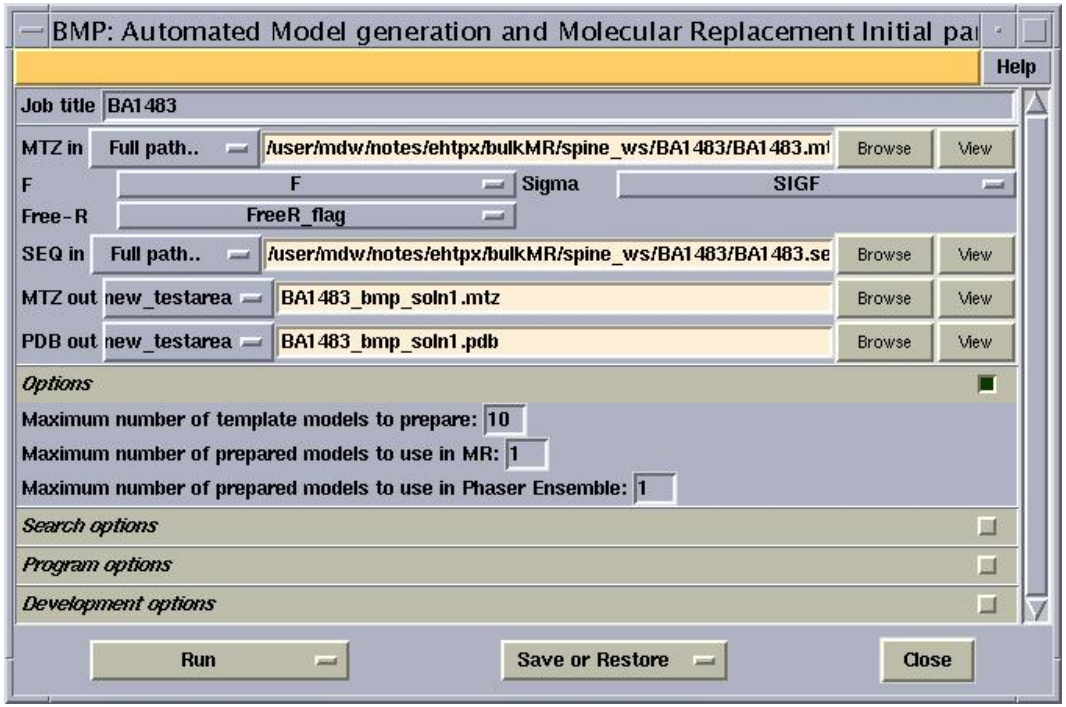

Target MTZ
&
Sequence
Target Details
Model Search
Model Preparation
Parallel mode:
Start multiple MR jobs and exit when one finds a solution
r Replacement
efinement
Molecular Replacement & Refinement
Molecular Replacement & Refinement
Molecular Replacement & Refinement
Molecular Rep
& Refine

Testing and pre-release version of Mr BUMP January '06



- Tested against list of SPINE targets in July '05
- Announced at CCP4 study weekend and on CCP4 BB.
- Comes with CCP4 GUI .
- Good deal of interest and one report of a user using it to solve a structure that they hadn't been able to solve since 2002.

Pre-release version available from:

www.ccp4.ac.uk/martyn/BMP/BMP.php

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MrBUMP and e-HTPX

e-HTPX – an e-science resource for High Throughput Protein Crystallography

Reasons for e-HTPX

The high volume of data coming from structural genome projects has generated a demand for new methods to obtain structural and functional information about biological proteins and macromolecules. This has led to a demand for high throughput techniques to determine the structure of important proteins.

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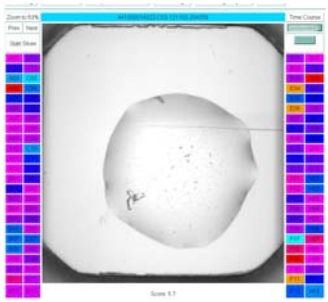
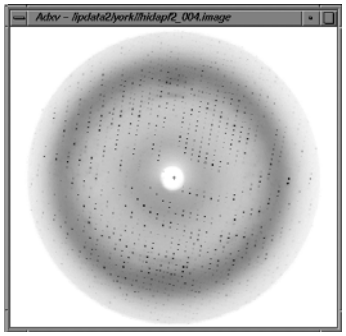
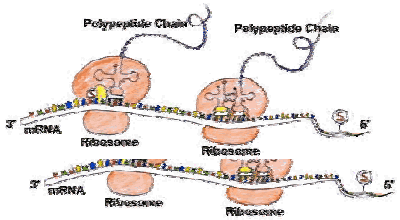


Crystallisation

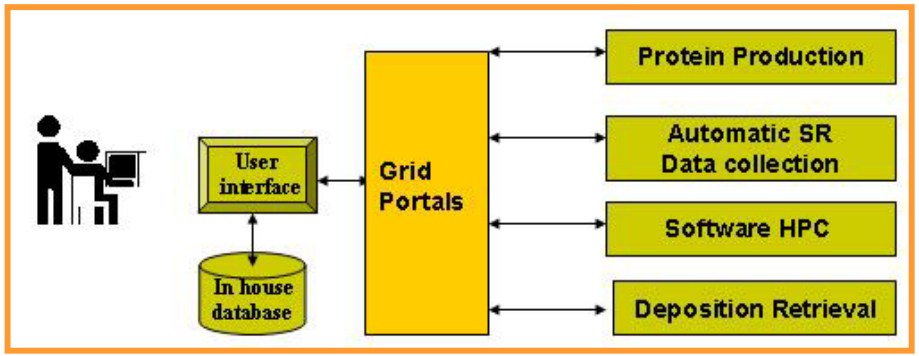


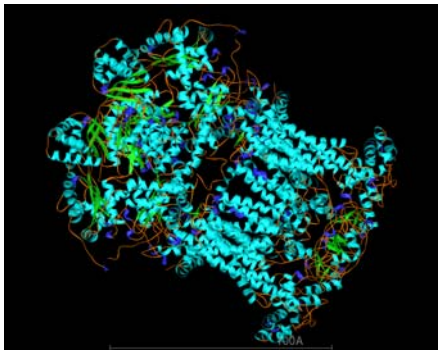
Data Collection

Phasing



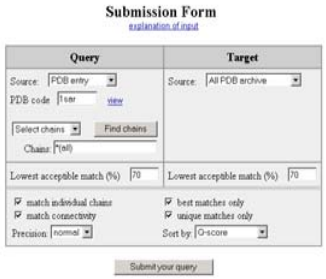
Protein Production



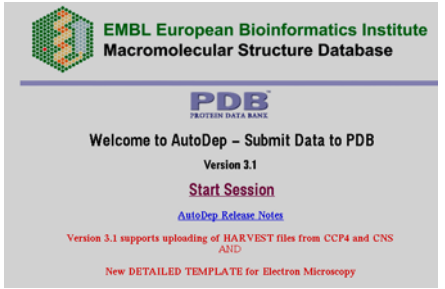


Protein Structure

Target Selection



Structure analysis



Deposition

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e-HTPX System Architecture – Portal Architectures

- Two portal architectures have been designed in order to suit different research environment needs.
  - Service site portal – portal is hosted by the synchrotron, minimizes the requirements placed on the user. (web-browser is the only requirement).
  - Client site portal – portal is installed and managed at the client institution.
- Both implementations are equivalent clients to the underlying web services.

Service-Site Architecture

• e-HTPX portal is hosted at the synchrotron site and acts as the client interface to the SRS services.

Advantages:

• Client access is centralised through a known / single point of entry (a gateway server). Allows service site firewall administrators to implement IP-recognition methods via defined ports.

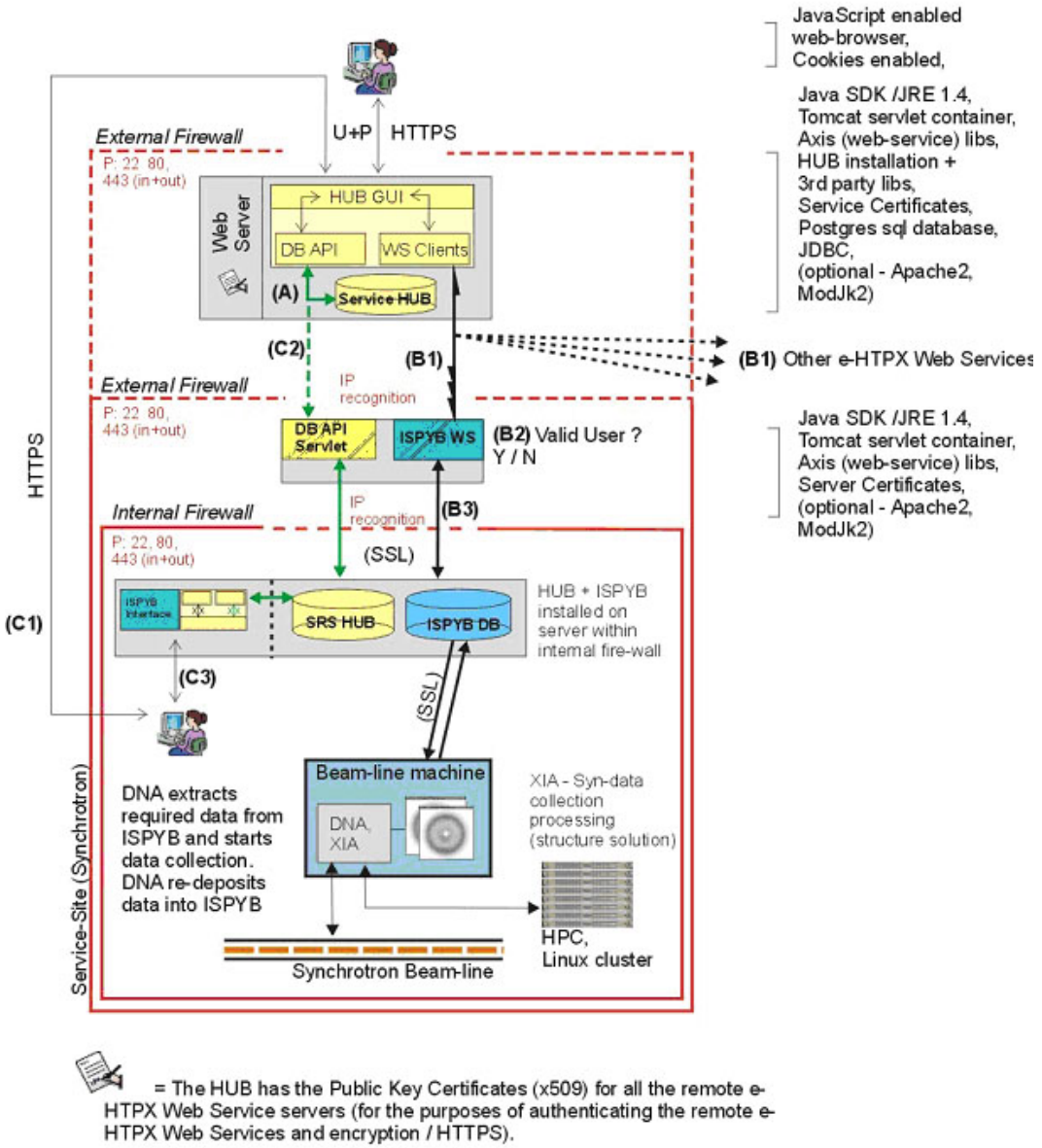
• No requirements placed on client.

Disadvantages:

• Main Architectural weaknesses:-

a) increased service side responsibility for storing users project data in portal/hub database;

b) Interoperability with clients LIMS system.



Client-Site Architecture

• e-HTPX portal is hosted at the client site:
a) Client site responsibility for storing potentially sensitive data (especially relevant for industrial clients).

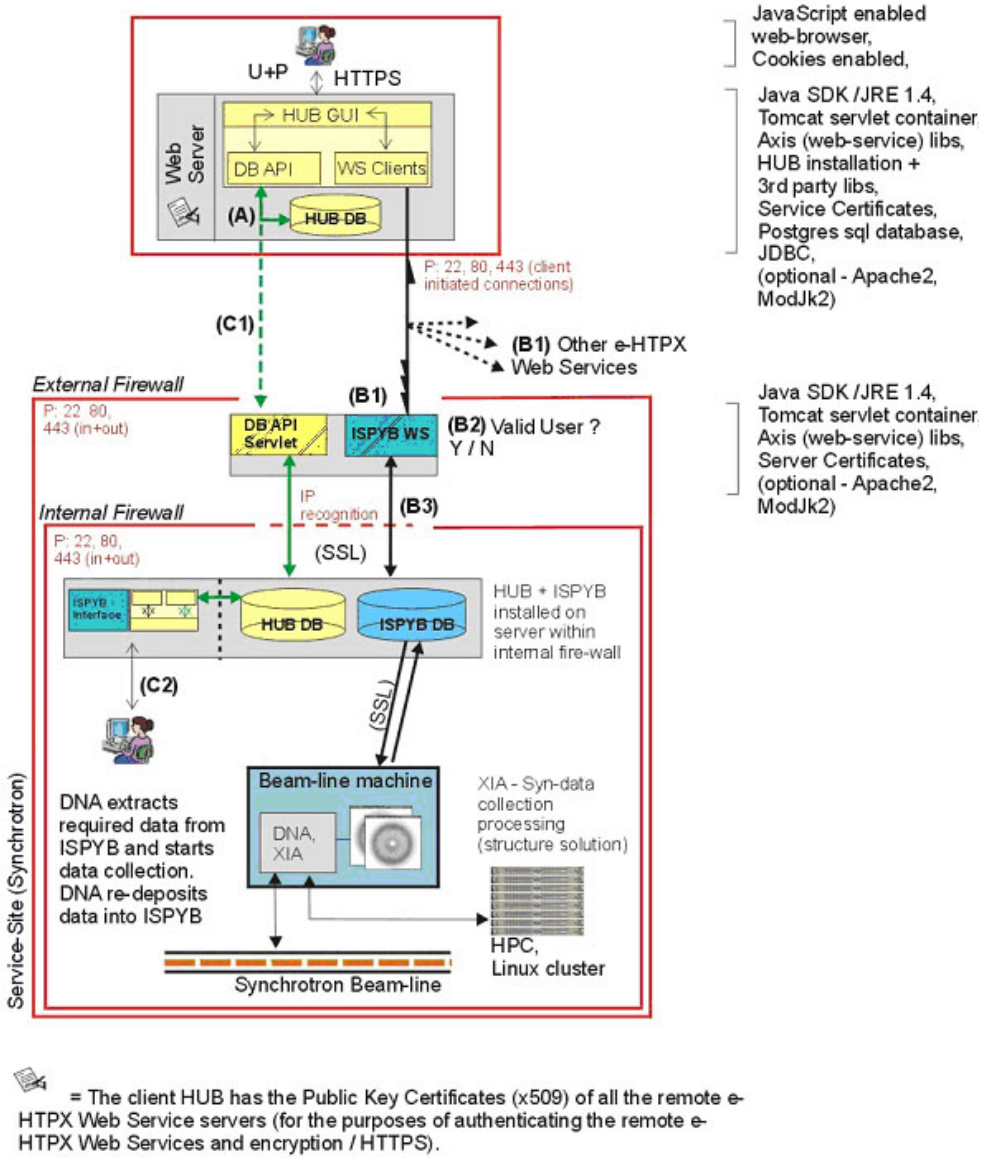
•b) Interaction with LIMS (laboratory information management system).

• Web-Service polling methodology required to circumvent client firewall restrictions.

• Clearly separates web-service client code from web-service host code.

• Host server certificates required on hub to authenticate client institution. Similar approach to authenticating clients (x509 digital certificates).

• To increase service site security, requests may be optionally routed through a single gateway server. A number of methodologies to implement this:- a) Reverse proxy server (apache mod_security); b) Multiple actors on the SOAP path; c) Place web-services on gateway.



MrBUMP and e-HTPX

- Dedicated machine for running e-HTPX structure solution jobs submitted via the e-HTPX portal interface.
- MrBUMP service is made available as a web service with a WSDL description.

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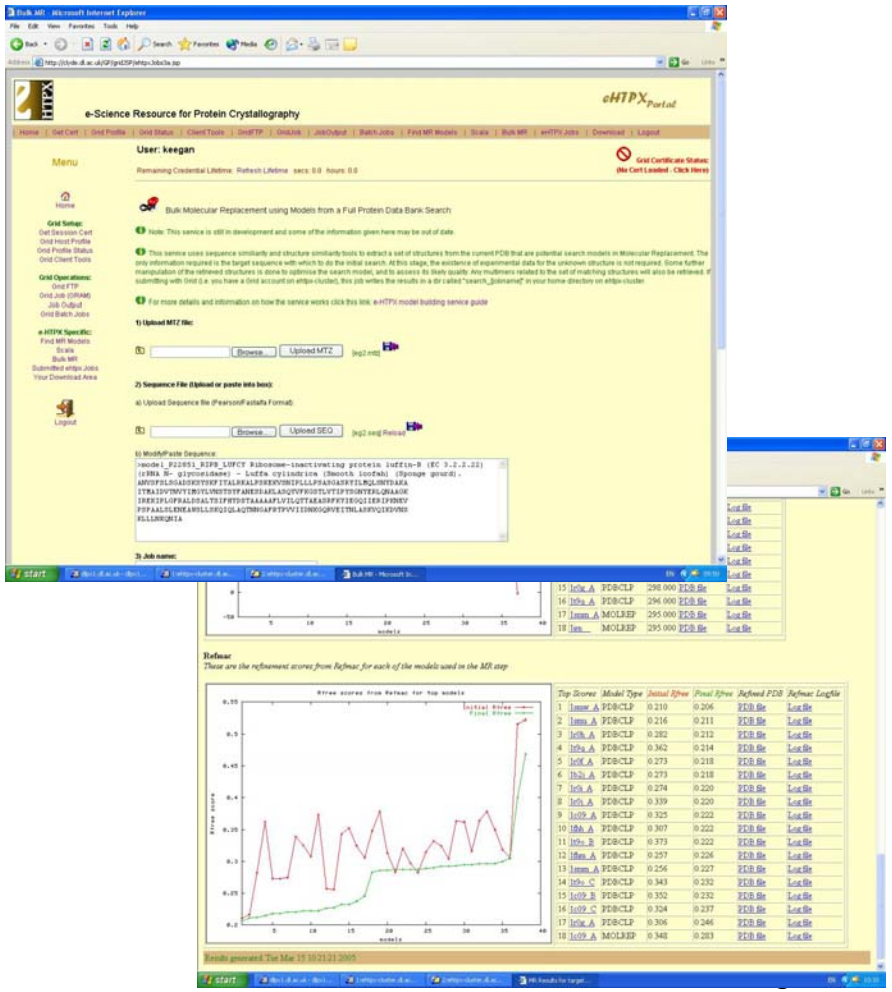
e-HTPX MrBUMP Portal

- The WSDL can be parsed by a user so as to incorporate the service into their own application.

- Alternatively, a remote user can access the service via the stand-a-lone interface provided by e-HTPX.

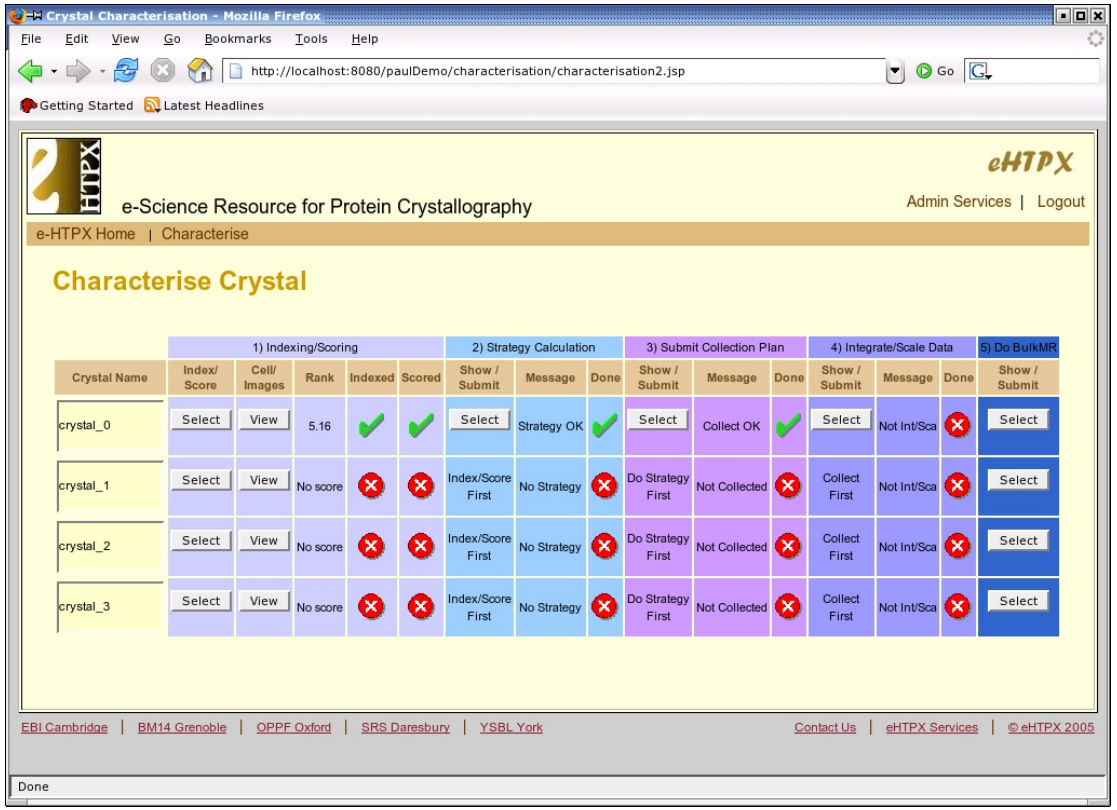
- Users upload their X-ray experiment diffraction data and invoke the service on the HPC resources provided at Daresbury Laboratory.

- Information is transferred in XML format using SOAP.



MrBUMP as part of e-HTPX workflow

- MrBUMP can also be accessed on the dedicated cluster as part of the e-HTPX workflow.
- Portal contains a top-level interface connecting with each of the web-services hosted at each of the sites involved.
- Allows the remote user to monitor the progress of their experiment and also to access the data processing facilities to process the collected data.



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Future developments

- additional model building methods such as removal of parts of the main chain (e.g. by position or by B-factors),
- support for complexes,
- develop web-service version to allow CCP4i users to run jobs on e-HTPX/CCP4 clusters,
- simplified CCP4 GUI.
- Incorporate PISA multimer determining service (EBI).

Acknowledgements

- e-HTPX:
  - Dave Meredith, Graeme Winter, Michael Gleaves, Daresbury Laboratory.
  - Ian Berry, OPPF, Oxford.
  - Paul Young, YSBL, York University.
  - Oleg Dolomanov, EBI, Hinxton.
  - Ludovic Launer, BM14, ESRF.
  - http://www.e-htpx.ac.uk
- MrBUMP:
  - Martyn Winn, CCP4.
  - http://www.ccp4.ac.uk/BMP/BMP.html

e-HTPX Project

A distributed computing infrastructure for protein crystallographic structure determination.

- Project integrates a number of key services provided by UK e-Science, protein manufacture and synchrotron laboratories.
- Remote access to these services is implemented by a collection of Web services, each developed at the corresponding service-site.
- Client access to the web services is through the e-HTPX portal/hub
  - Easy to use interface
  - Hides user from complexity of underlying distributed computing infrastructure

Stage 1 – Select protein target

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Stage 2 – Crystallization of Protein

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Stage 3 – Data Collection (X-ray diffraction images, Scaling and Integration)

↓

Stage 4 – Structure Solution (HPC data processing to derive digital protein model)

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Stage 5 – Submit model into public database

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