

PROTA-2X™

FT-IR of Biomolecules
Specifications - Applications

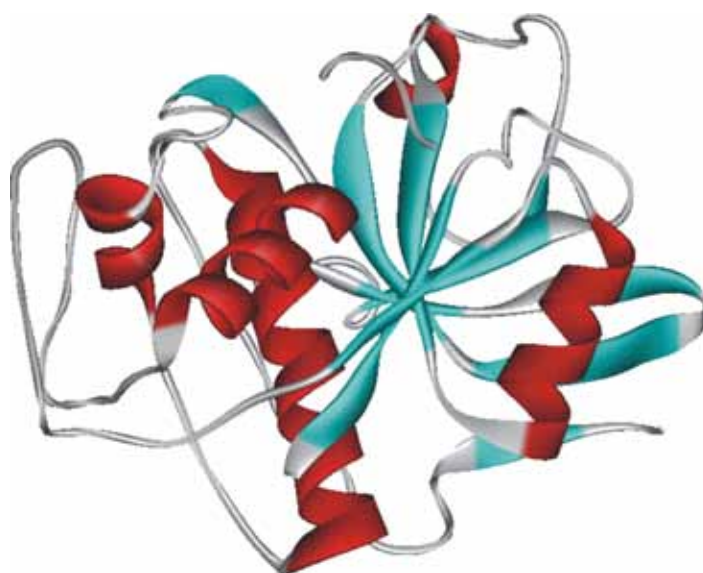
FT-IR Spectroscopy of Biologics

BioIR™ is a family of dedicated solutions for the biopharmaceutical industry based on FT-IR instrumentation and comprised of specialized software and databases.

The first such system, PROTA™, was introduced in 1998 and has since rapidly become the industry's preferred choice. PROTA™ provides a fast, cost-effective and sensitive way to determine secondary structure of a protein or to follow structural changes due to perturbations. This turnkey system is designed to be used by both spectroscopists and non-spectroscopists. It's user-friendly, intuitive software guides users through data acquisition and analysis. PROTA™ includes all of the functions, in one integrated package, required to link IR spectral data and protein structure.

APPLICATIONS

- » Formulation studies (liquids and solids) - effects of excipients, pH and buffers
- » Determination of secondary structures of de novo proteins
- » Conformational stability and dynamics
- » Structure in aggregates
- » Mutation studies
- » Structural characterization upon environmental effects
- » Stability studies (thermal and chemical)
- » Drug delivery
- » Protein-protein, protein-DNA/RNA and protein-drug interactions
- » Crystallization condition screening



PROTA-2X™ Instrument

FT-IR SPECTROMETER SYSTEM

- » ABB MB3000 FT-IR spectrometer consisting of:
 - Arid-Zone sample compartment with countercurrent purge flow in telescopic purge tubes
 - Non-hygroscopic ZnSe beamsplitter
 - High-sensitivity DTGS detector
- » Resolution: 1 cm^{-1} - 128 cm^{-1}
- » Spectral range: 6,500 - 500 cm^{-1}

ADDITIONAL HARDWARE

- » Pre-loaded PC
- » *BioCell*™ with CaF_2 windows
- » Purge Control Kit

SOFTWARE

- » Windows-based integrated software for data acquisition and data analysis
- » Complete Grams A/I software
- » Protein FT-IR databases
 - Transmission
 - ATR
 - Excipients

AFTER-SALES SERVICE

- » Installation and training on FT-IR spectroscopy of proteins by experienced scientists
- » BioTools combined service and technical protein application support for one year

OPTIONS

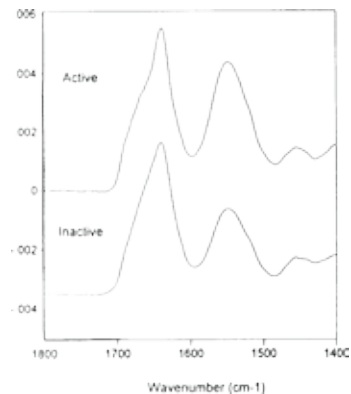
- » Variable Temperature cell and automatic temperature controller with computer interface
- » KBr and ATR accessories
- » Microscope
- » A dry air purge supply is recommended to achieve optimal performance



PROTA™ Applications

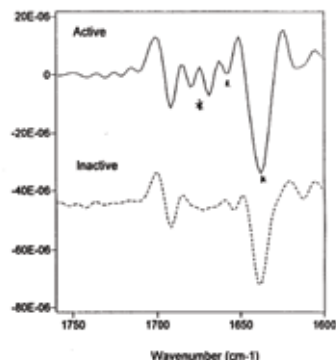
APPLICATION EXAMPLES

1) Low Protein Concentration Studies



Protein samples with concentrations as low as 3 mg/ml in H₂O can be routinely measured with PROTA™. Conditions: 20 min. acquisition time, NO water vapor subtraction, buffer subtracted using the automated buffer subtraction function.

2) Formulation Studies



FT-IR derivative spectrum of 'intact' immunogenic antibody compared to a lyophilisate, that yields inactive, non-immunogenic form after reconstitution with water. Obvious differences are observed between the two spectra. These spectral differences identify structural changes that a protein undergoes upon lyophilization. This information may streamline and improve the formulation process.

3) Protein Secondary Structure Determination

| Protein | % α-helix | % β-sheet | % bend | % turn | % other | sum |
|----------------------|-----------|-----------|--------|--------|---------|-----|
| A-chymotrypsin | 12 | 32 | 12 | 11 | 33 | 100 |
| Carbonic Anhydrase B | 14 | 33 | 14 | 13 | 26 | 100 |
| Citrate Synthase | 58 | 4 | 9 | 11 | 20 | 102 |
| Carboxypeptidase | 43 | 15 | 8 | 13 | 23 | 102 |
| Catalase | 44 | 11 | 11 | 12 | 24 | 102 |
| Γ-chymotrypsin | 12 | 35 | 12 | 13 | 26 | 98 |
| Chymotrypsinogen | 27 | 24 | 12 | 13 | 25 | 101 |
| Concanavalin A | 0 | 43 | 15 | 13 | 25 | 97 |
| Other Proteins | | | | | | |

PROTA™ CAN BE USED TO DETERMINE QUANTITATIVE PROTEIN SECONDARY STRUCTURE USING FACTOR ANALYSIS, COMPARED TO THE 50-PROTEIN DATABASE. TYPICAL RESULTS ARE SHOWN ABOVE.