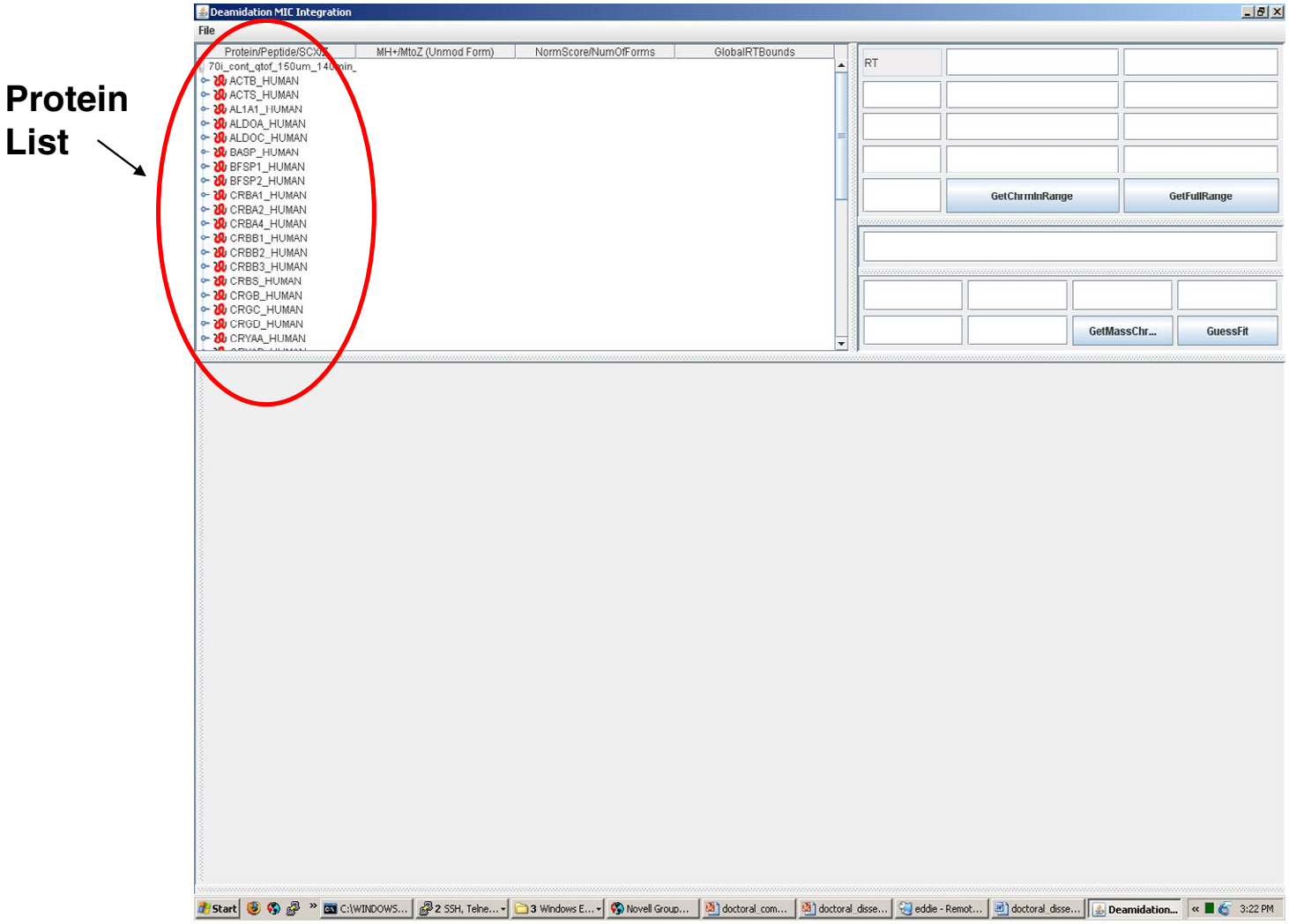
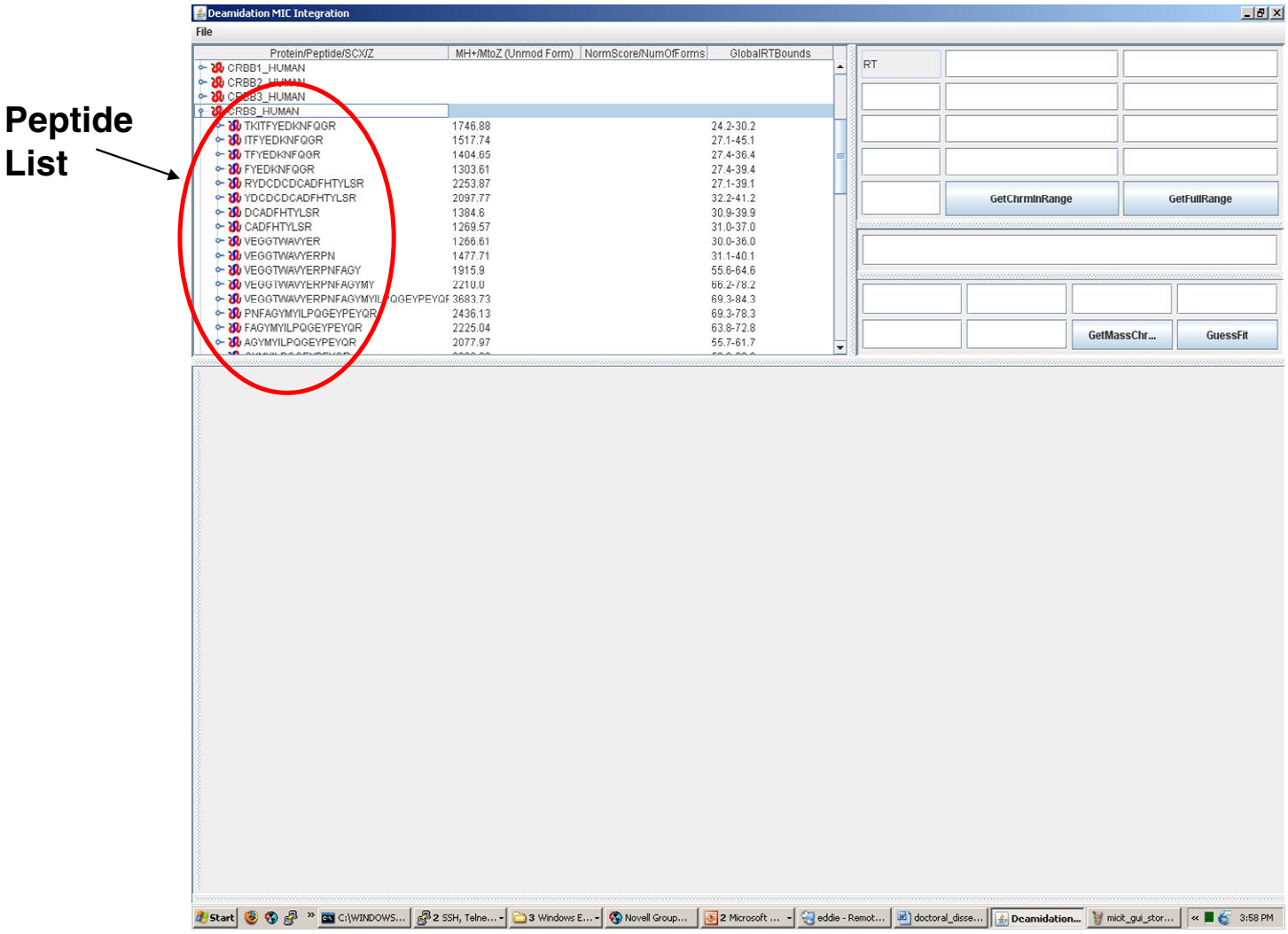
Supplemental File 1. MICIT Workflow Story Boards:

MICIT (Panel 1)



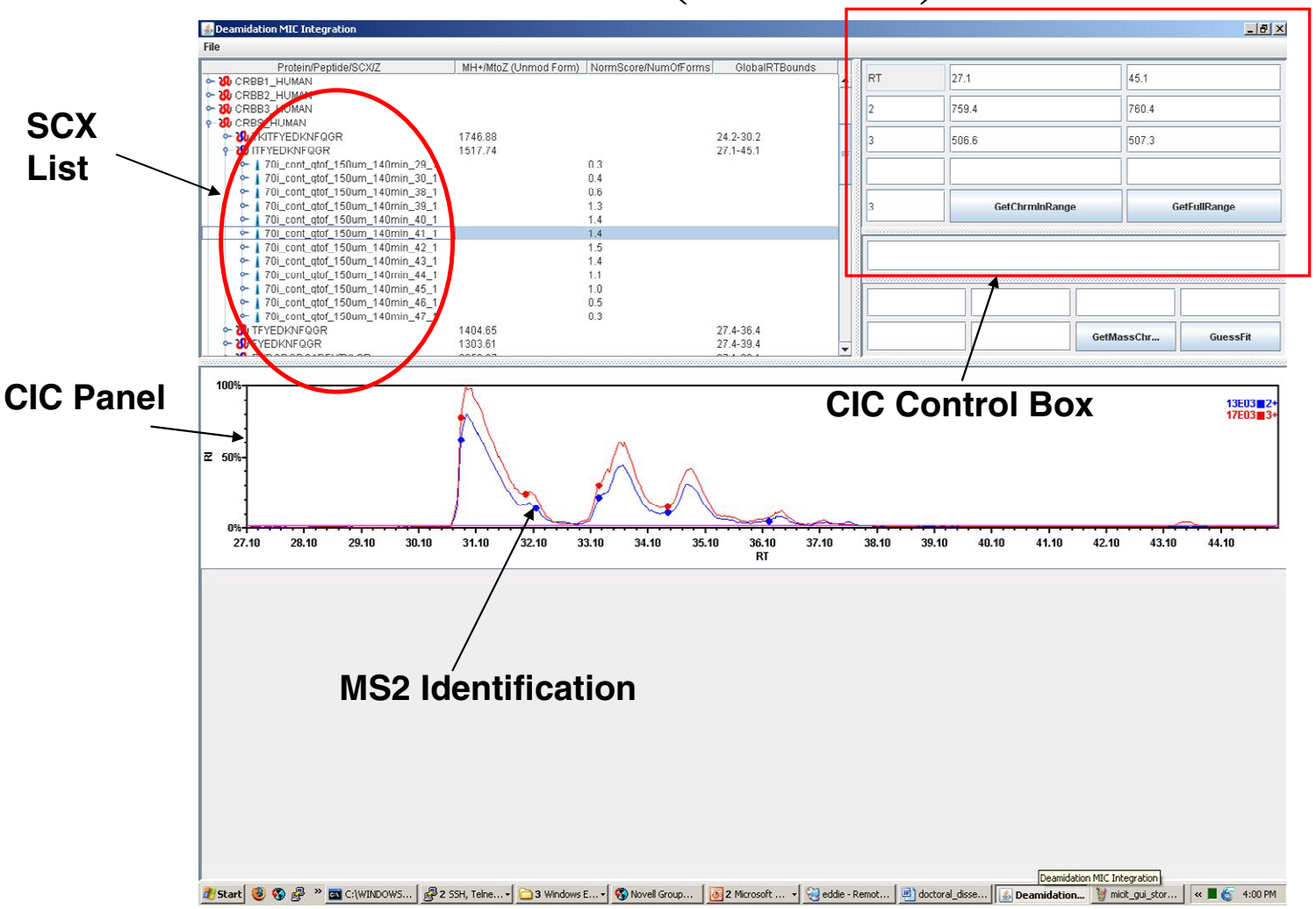
Panel 1.1. Loading Results: MICIT display after protein and peptide identifications are loaded. The protein list shows all protein identifications present in the sample in an expandable tree table.

MICIT (Panel 2)



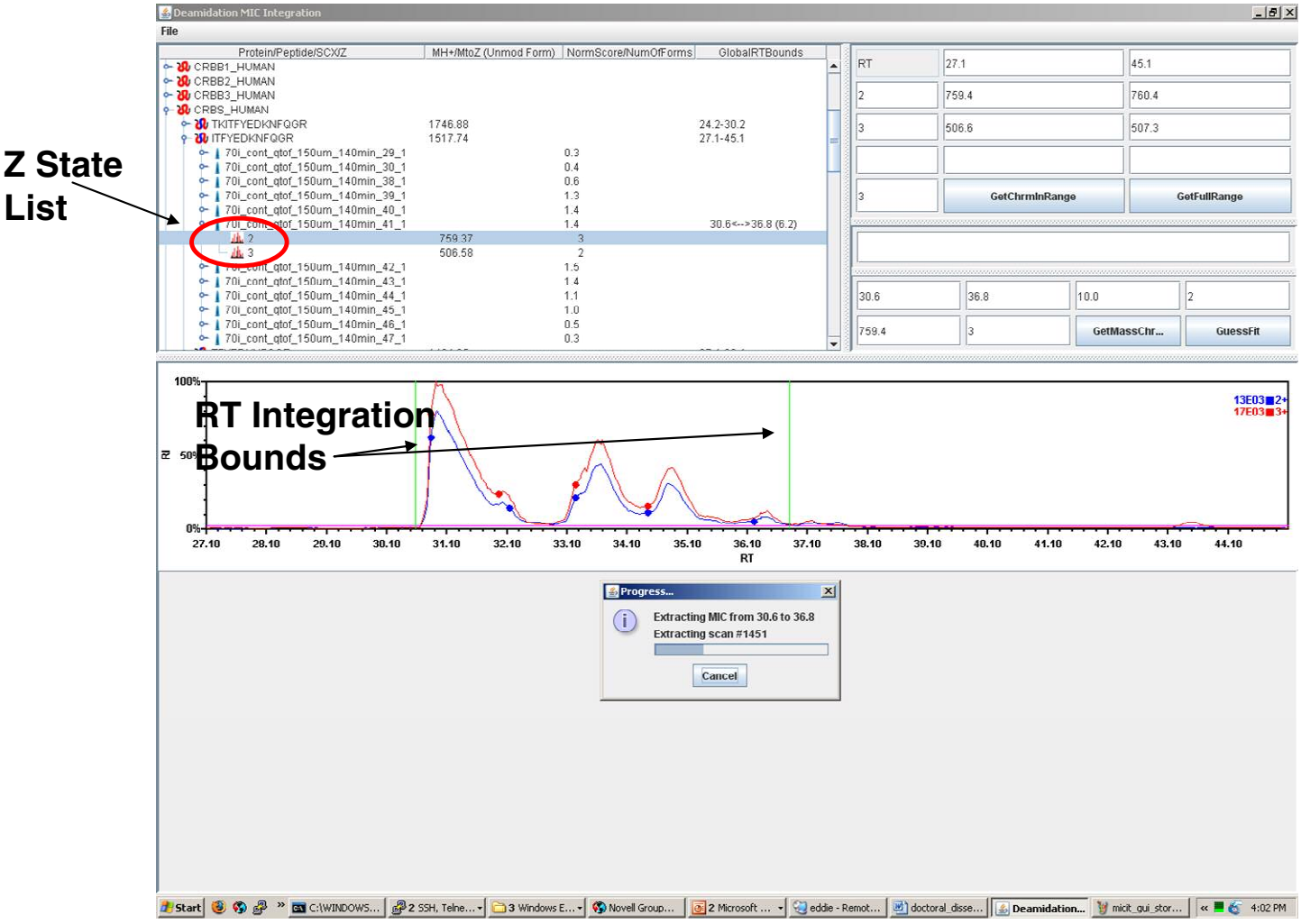
Panel 1.2. Viewing Protein and Peptide Identifications: A protein identification node, when expanded, displays all of its corresponding peptide identifications (Peptide List) as shown above. Plus one ion mass (MH+) and global retention time bounds (determined from the union of MS2 identifications) of each peptide are also displayed.

MICIT (Panel 3)



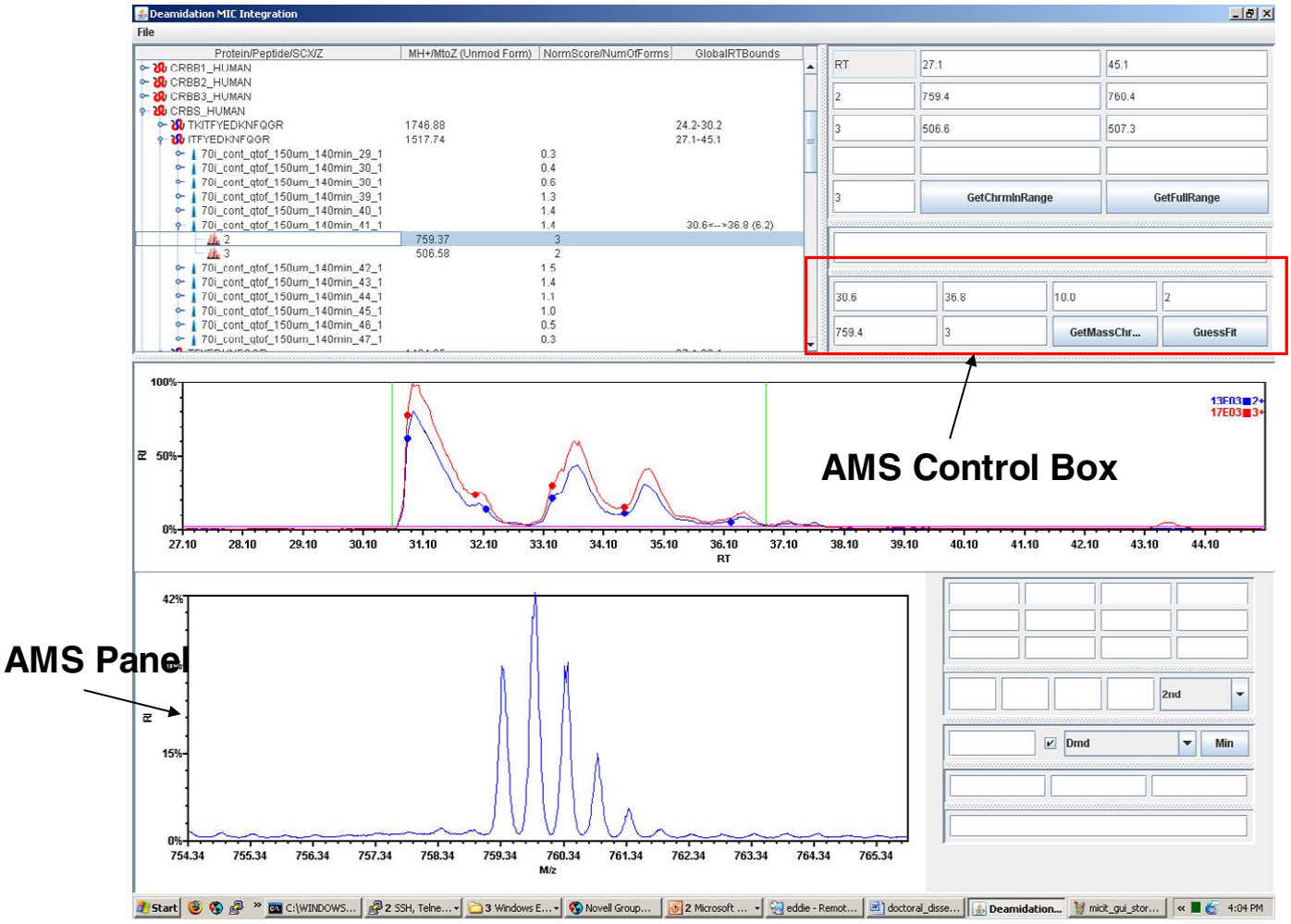
Panel 1.3. Extracting Peptide Ion Chromatograms: A peptide identification node, when expanded, displays all SCX fractions (SCX List) in which corresponding peptide forms (unmodified and deamidated) have been detected. When an SCX fraction is selected, a combined ion chromatogram (CIC) for all peptide forms present in a particular SCX fraction is generated and displayed in CIC panel. Separate CICs are generated for all detected charge states in a SCX fraction and displayed using different colors (red color for 3+ and green color for 2+ as shown in above example). All MS2 identifications of selected peptide (unmodified and deamidated) are overlaid on corresponding CICs. The CIC control box allows control of CIC generation.

MICIT (Panel 4)



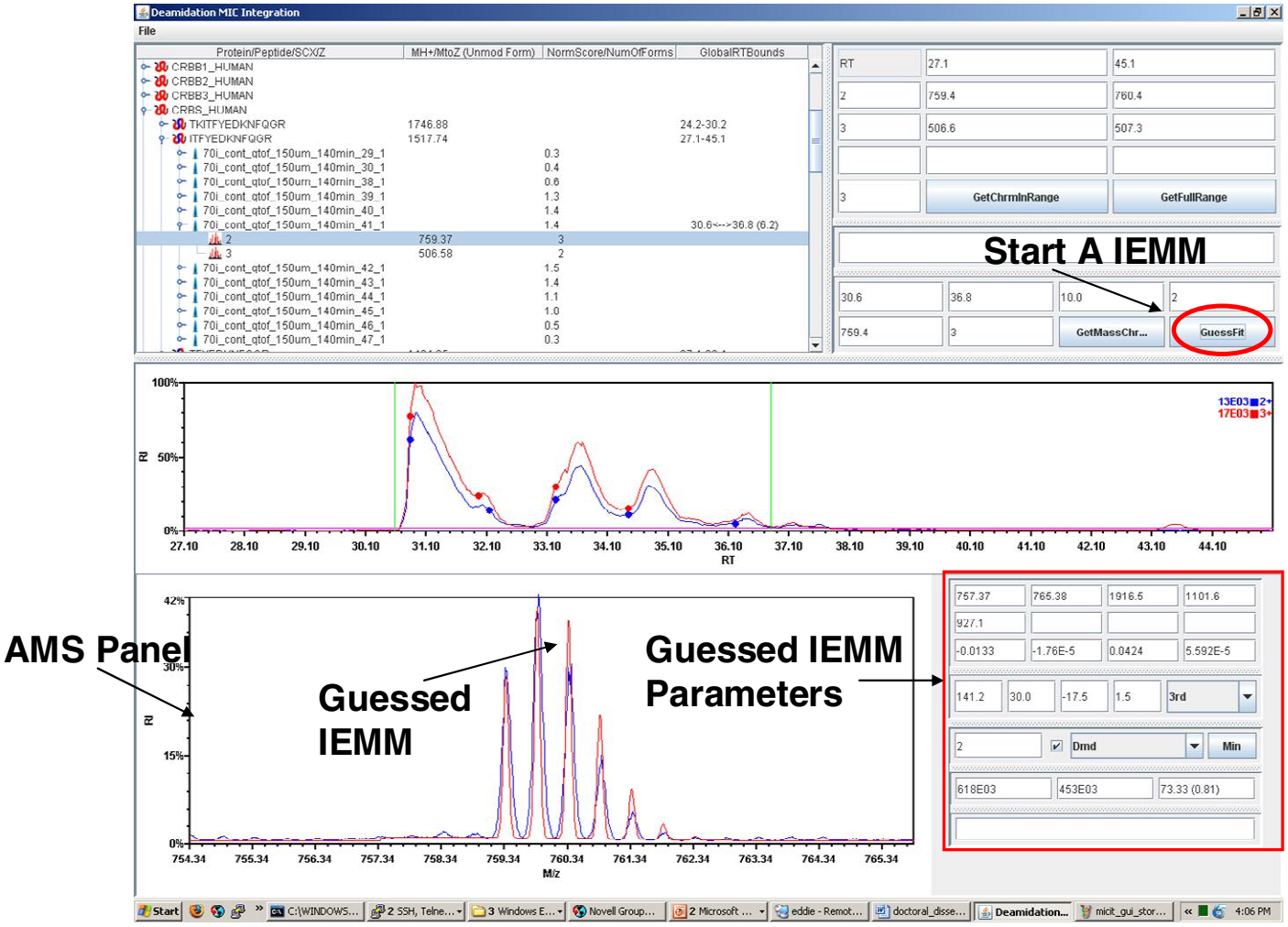
Panel 1.4. Generating Averaged Mass Spectrum (AMS): Retention time integration bounds (RT Integration Bounds) for a SCX fraction are selected using a left button depressed mouse drag on corresponding combined chromatogram. The selected SCX fraction is expanded to display all detected charge states of the peptide (Z State List). Selection of a specific charge state results in generation of a mass spectrum by averaging all MS-scans triggered between the selected integration bounds.

MICIT (Panel 5)



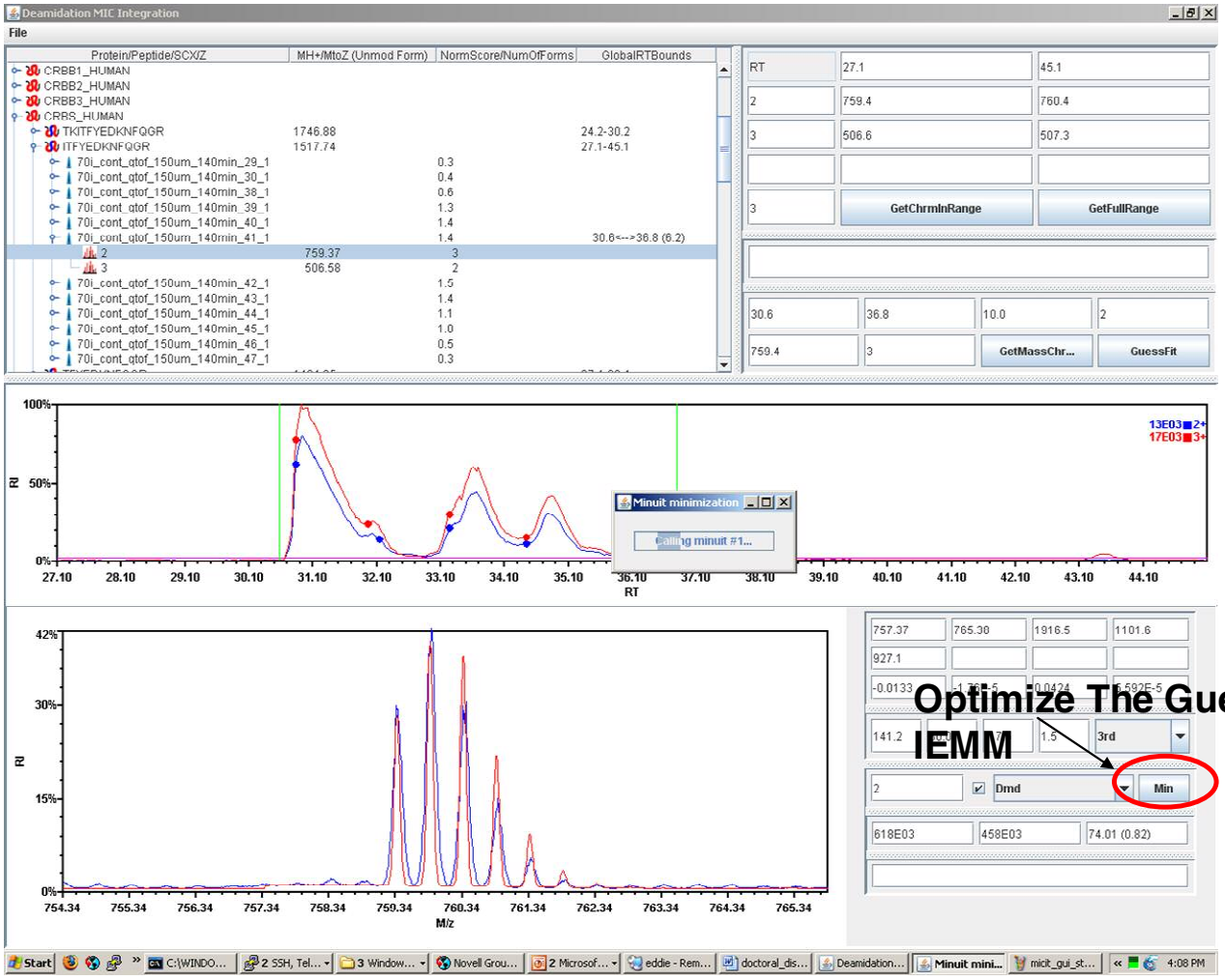
Panle 1.5. Viewing Averaged Mass Spectrum: Averaged experimental mass Spectrum (AMS) for a peptide is displayed in AMS panel below combined ion chromatogram. The AMS view is automatically chosen to display ± 5.0 Dalton around the isotopic distribution of the selected peptide. Control of the AMS panel is available using the AMS control box.

MICIT (Panel 6)



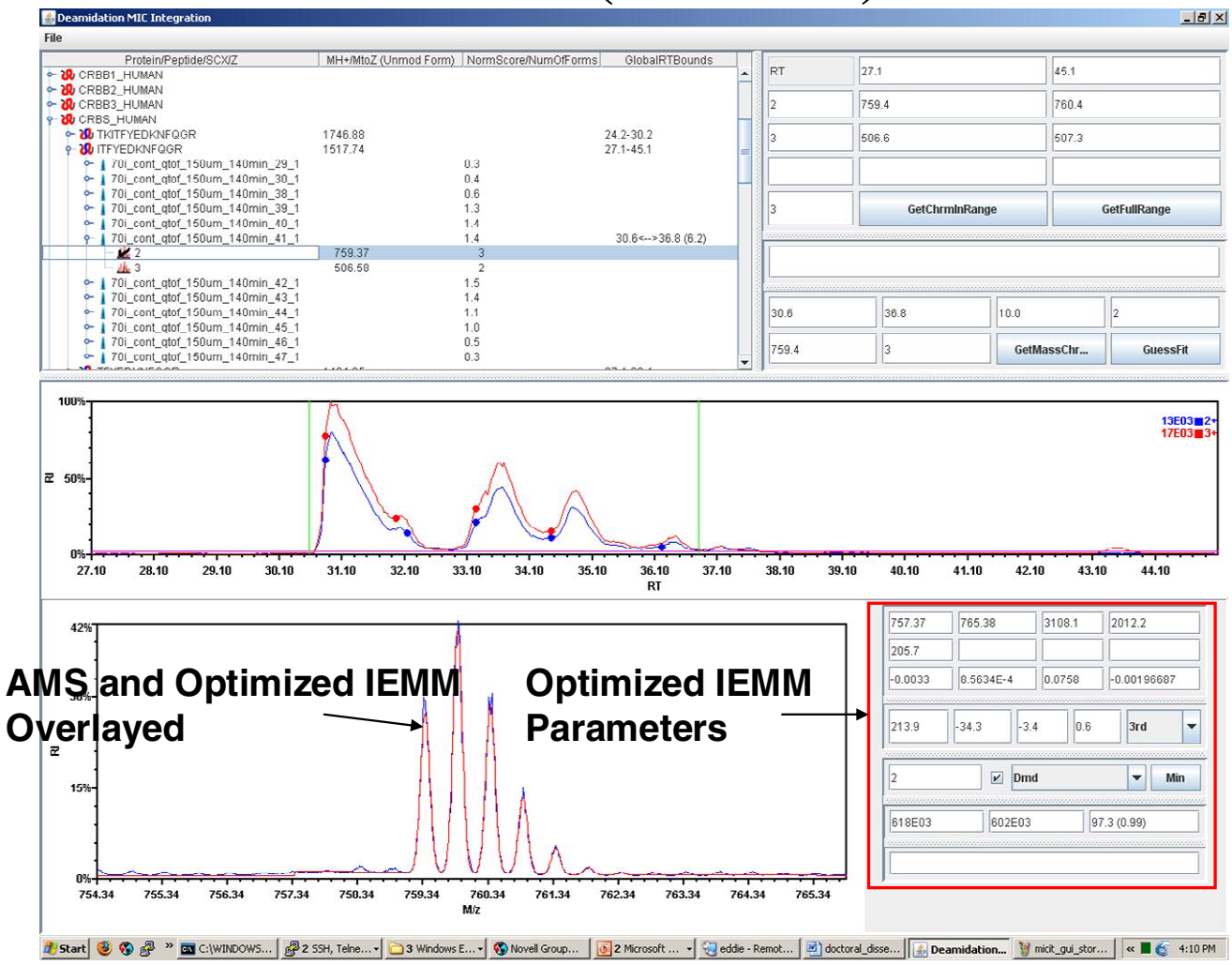
Panel 1.6. Initializing an Isotopic Envelope Mixture Model (IEMM): After generation of an averaged experimental AMS, an initial theoretical isotopic envelope mixture model (IEMM) is generated by clicking on "GuessFit" button in AMS control panel. Starting IEMM is overlaid on AMS in red color. The parameters of the initial IEMM (peptide intensities, peak shape, and background) are displayed in a control box next to AMS panel and are user adjustable.

MICIT (Panel 7)



Panel 1.7. Optimizing the Guessed IEMM: The initial IEMM is optimized by clicking on the "Min" button. The least squares optimization is carried out using the Minuit function minimization package (version 1.7, CERN, Geneva, Switzerland). A dialog box provides a real time update during the optimization procedure.

MICIT (Panel 8)



Panel 1.8. AMS and Optimized IEMM display: Optimized IEMM is overlaid on top of the averaged experimental AMS in the AMS panel after minimization. The initial IEMM parameters are updated with final optimized IEMM parameters. The final peptide intensity parameters are used to quantify the different forms of the peptides present in the IEMM. The result of the optimization can be saved to a XML formatted file in real time.