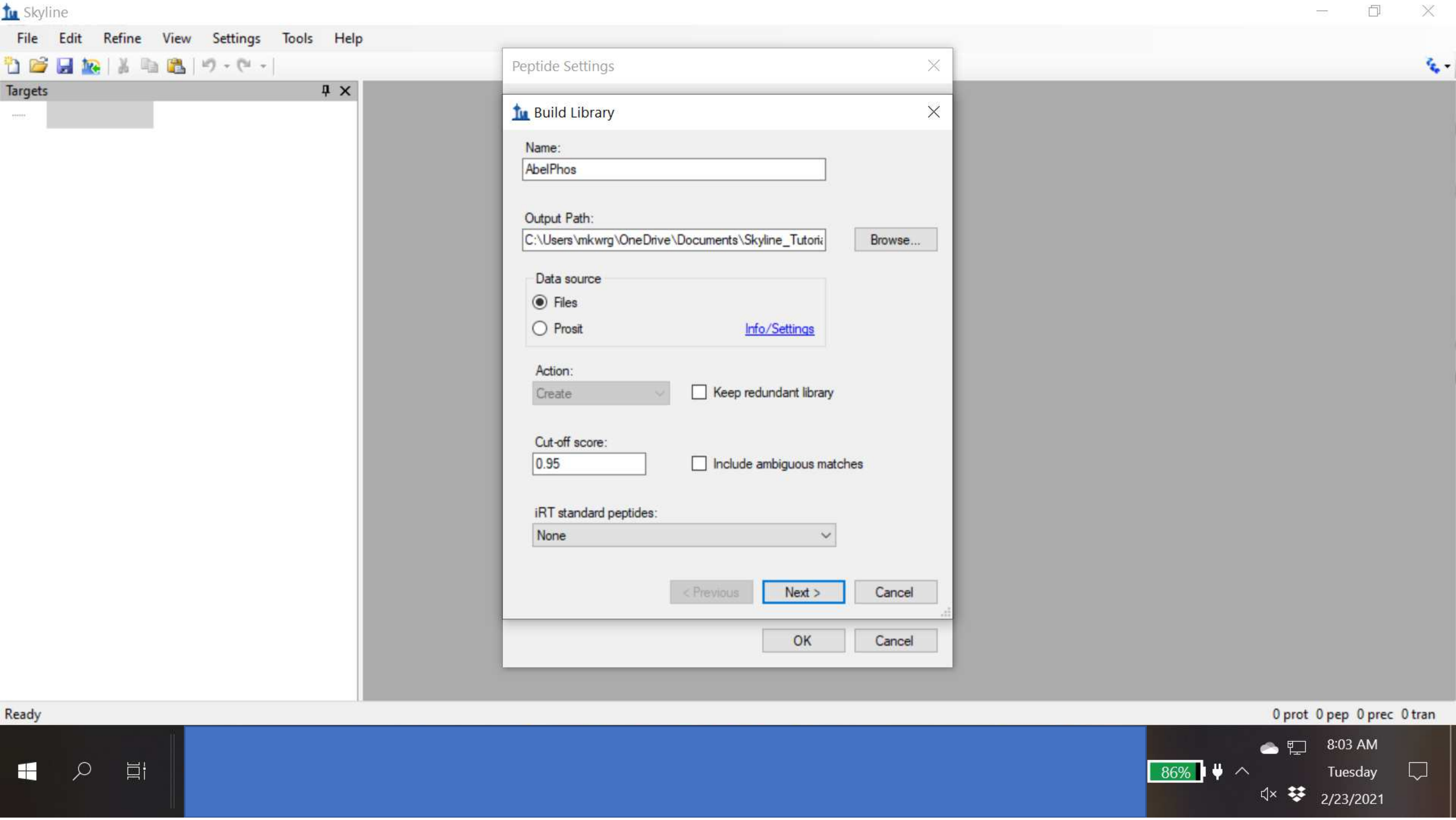


Build Library

Name: AbelPhos

Output Path: C:\Users\mkwrg\OneDrive\Documents\Skyline_Tutori

Data source
● Files
○ Prosit

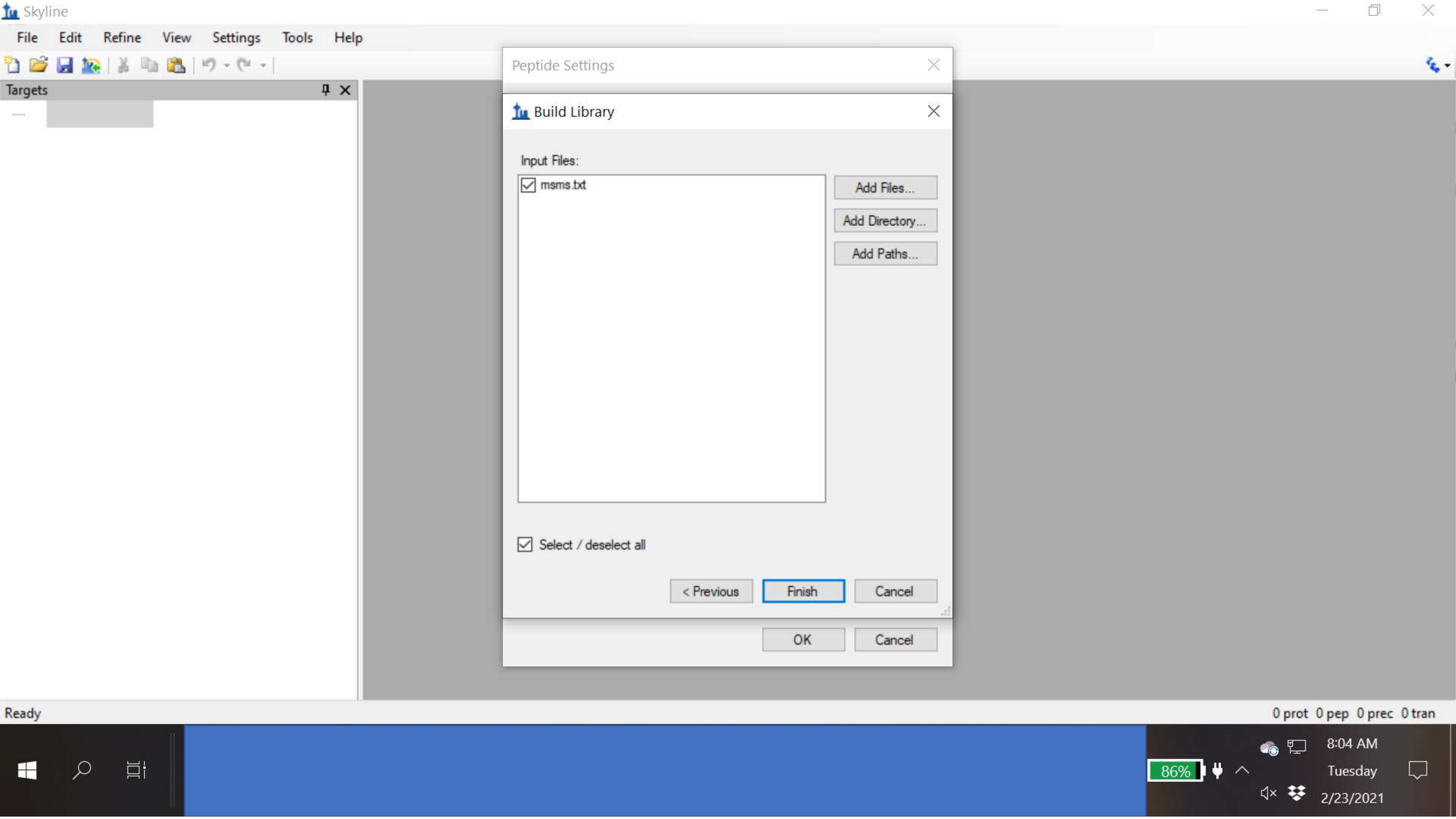
Action: Create

☐ Keep redundant library

Cut-off score: 0.95

☐ Include ambiguous matches

iRT standard peptides: None

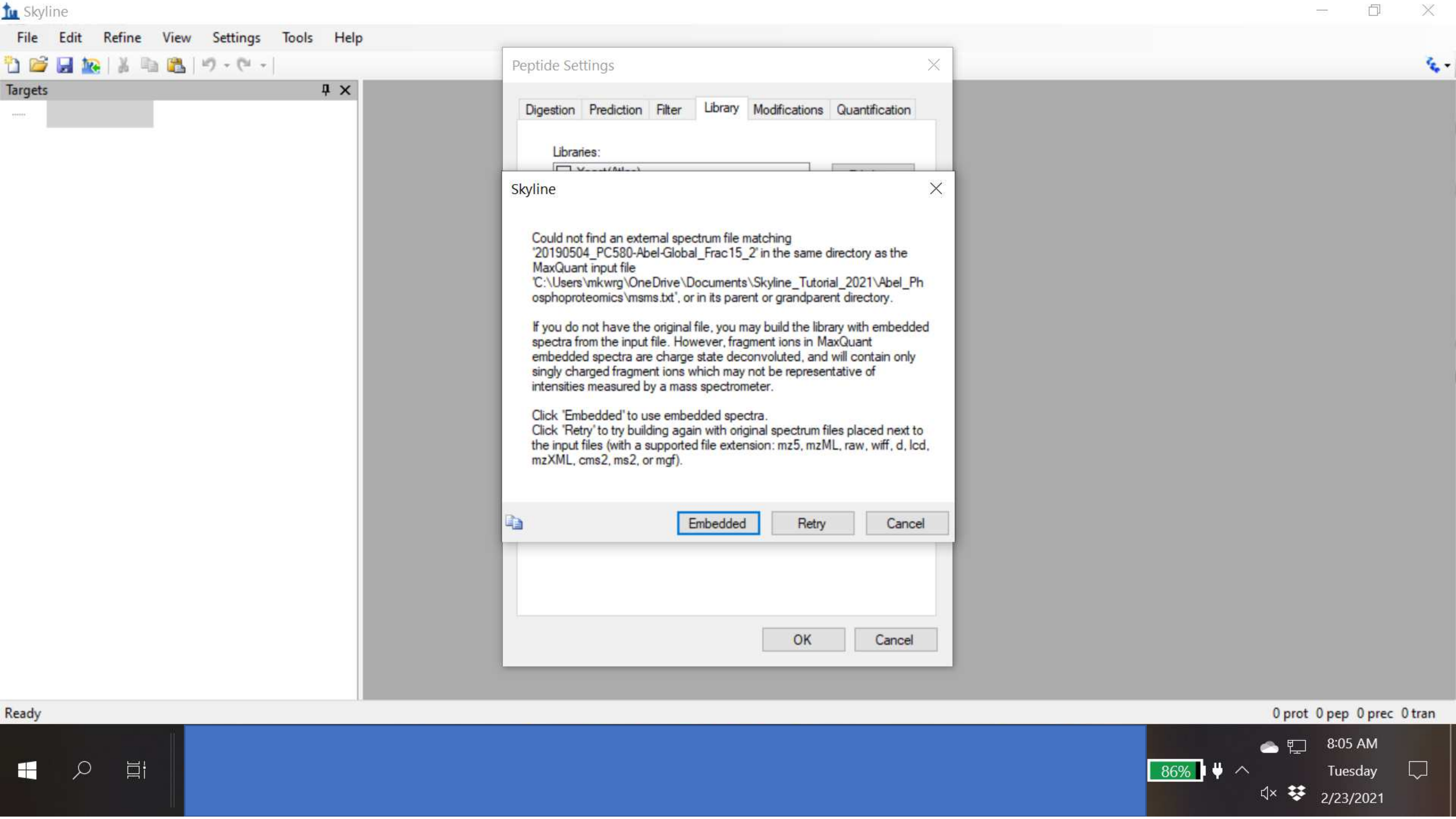


Input Files:

☑ msms.txt

☑ Select / deselect all

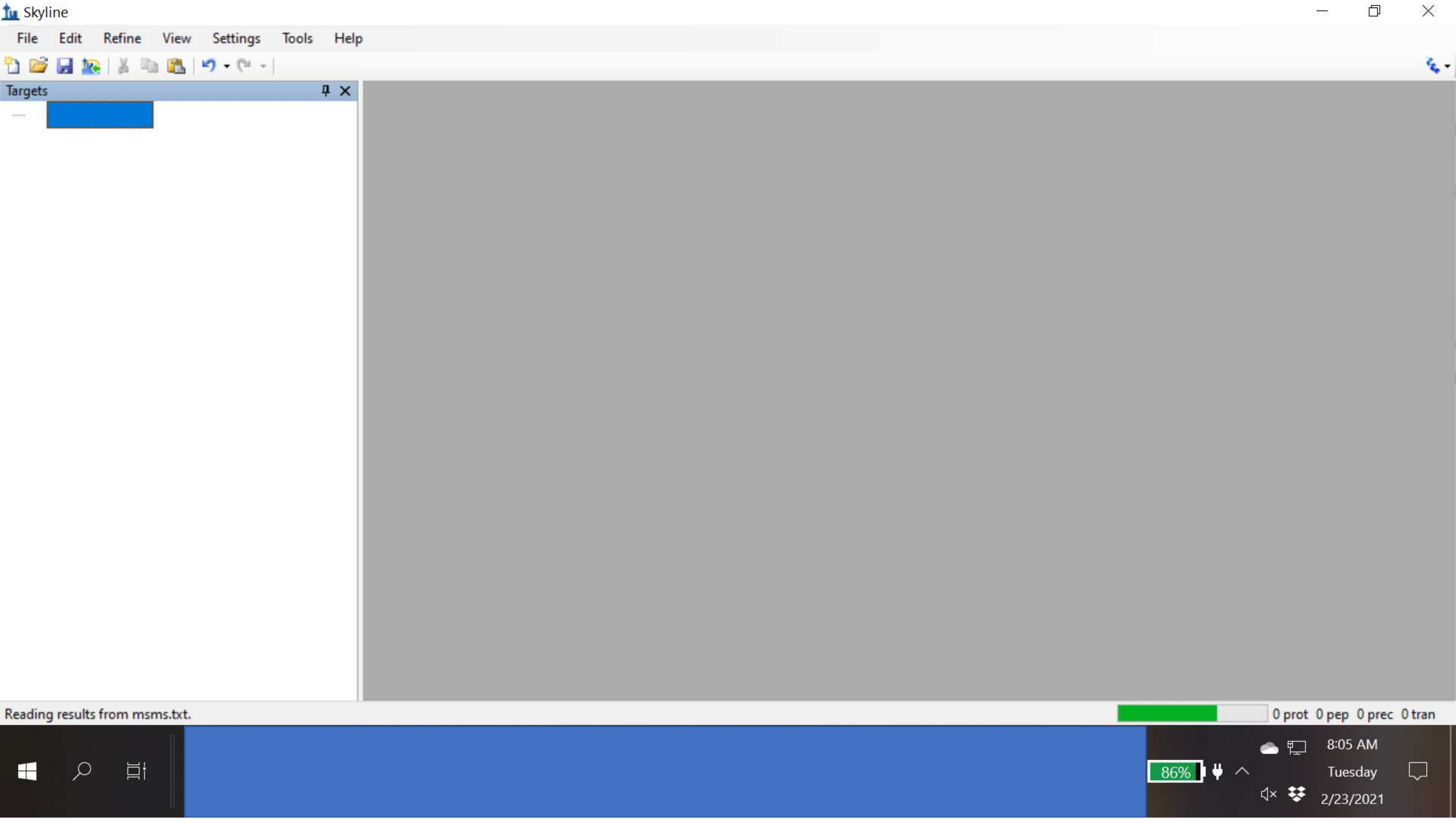
When I try to complete this task, the program returns with this error saying a file is missing. I go ahead and click the embed feature and the program moves ahead and spits out MS/MS spectra. Should I be concerned here?



Could not find an external spectrum file matching '20190504_PC580-Abel-Global_Frac15_2' in the same directory as the MaxQuant input file 'C:\Users\mkwrg\OneDrive\Documents\Skyline_Tutorial_2021\Abel_Phosphoproteomics\msms.txt', or in its parent or grandparent directory.

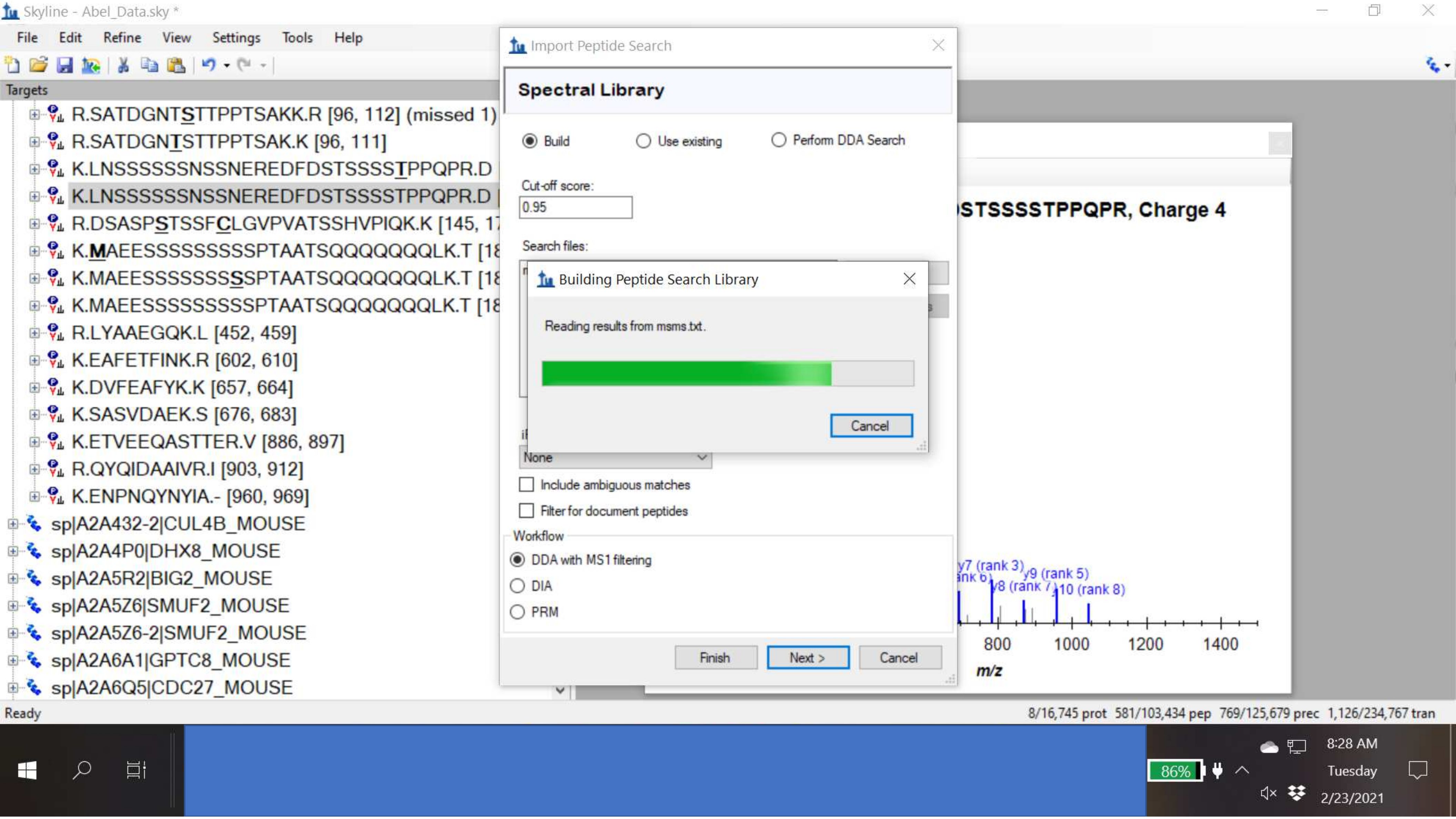
If you do not have the original file, you may build the library with embedded spectra from the input file. However, fragment ions in MaxQuant embedded spectra are charge state deconvoluted, and will contain only singly charged fragment ions which may not be representative of intensities measured by a mass spectrometer.

Click 'Embedded' to use embedded spectra.
Click 'Retry' to try building again with original spectrum files placed next to the input files (with a supported file extension: mz5, mzML, raw, wiff, d, lcd, mzXML, cms2, ms2, or mgf).



Reading results from msms.txt.

0 prot 0 pep 0 prec 0 tran



Skyline - Abel_Data.sky *

R.SATDGNT**S**TTPPTSAKK.R [96, 112] (missed 1)
R.SATDGN**T**STTPPTSAK.K [96, 111]
K.LNSSSSSSNSSNEREDFDSTSSSS**T**PPQPR.D
K.LNSSSSSSNSSNEREDFDSTSSSSTPPQPR.D
R.DSASP**S**TSSF**C**LGVPVATSSHVPIQK.K [145, 1
K.**M**AEESSSSSSSSSPTAATSQQQQQQQLK.T [1
K.MAEESSSSSSS**S**SPTAATSQQQQQQQLK.T [1
K.MAEESSSSSSSSSPTAATSQQQQQQQLK.T [1
R.LYAAEGQK.L [452, 459]
K.EAFETFINK.R [602, 610]
K.DVFEAFYK.K [657, 664]
K.SASVDAEK.S [676, 683]
K.ETVEEQASTTER.V [886, 897]
R.QYQIDAAIVR.I [903, 912]
K.ENPNQYNYIA.- [960, 969]
sp|A2A432-2|CUL4B_MOUSE
sp|A2A4P0|DHX8_MOUSE
sp|A2A5R2|BIG2_MOUSE
sp|A2A5Z6|SMUF2_MOUSE
sp|A2A5Z6-2|SMUF2_MOUSE
sp|A2A6A1|GPTC8_MOUSE
sp|A2A6Q5|CDC27_MOUSE

Import Peptide Search

Spectral Library

● Build ○ Use existing ○ Perform DDA Search

Cut-off score: 0.95

Search files:

Building Peptide Search Library

Reading results from msms.txt.

None

☐ Include ambiguous matches

☐ Filter for document peptides

Workflow

● DDA with MS1 filtering

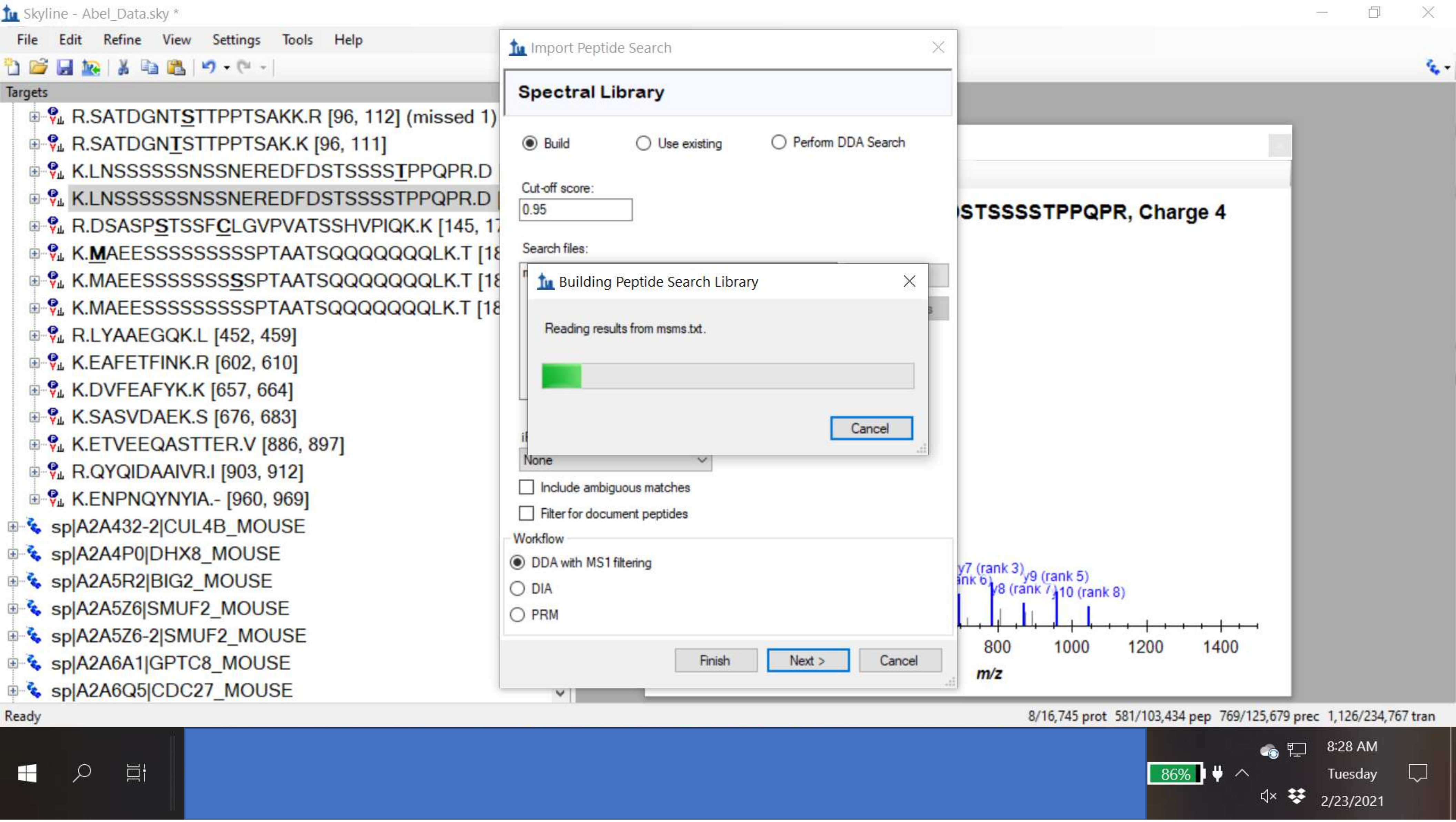
○ DIA

○ PRM

STSSSSTPPQPR, Charge 4

Ready

8/16,745 prot 581/103,434 pep 769/125,679 prec 1,126/234,767 tran



Import Peptide Search

Spectral Library

● Build ○ Use existing ○ Perform DDA Search

Cut-off score: 0.95

Search files:

None

☐ Include ambiguous matches

☐ Filter for document peptides

Workflow

● DDA with MS1 filtering

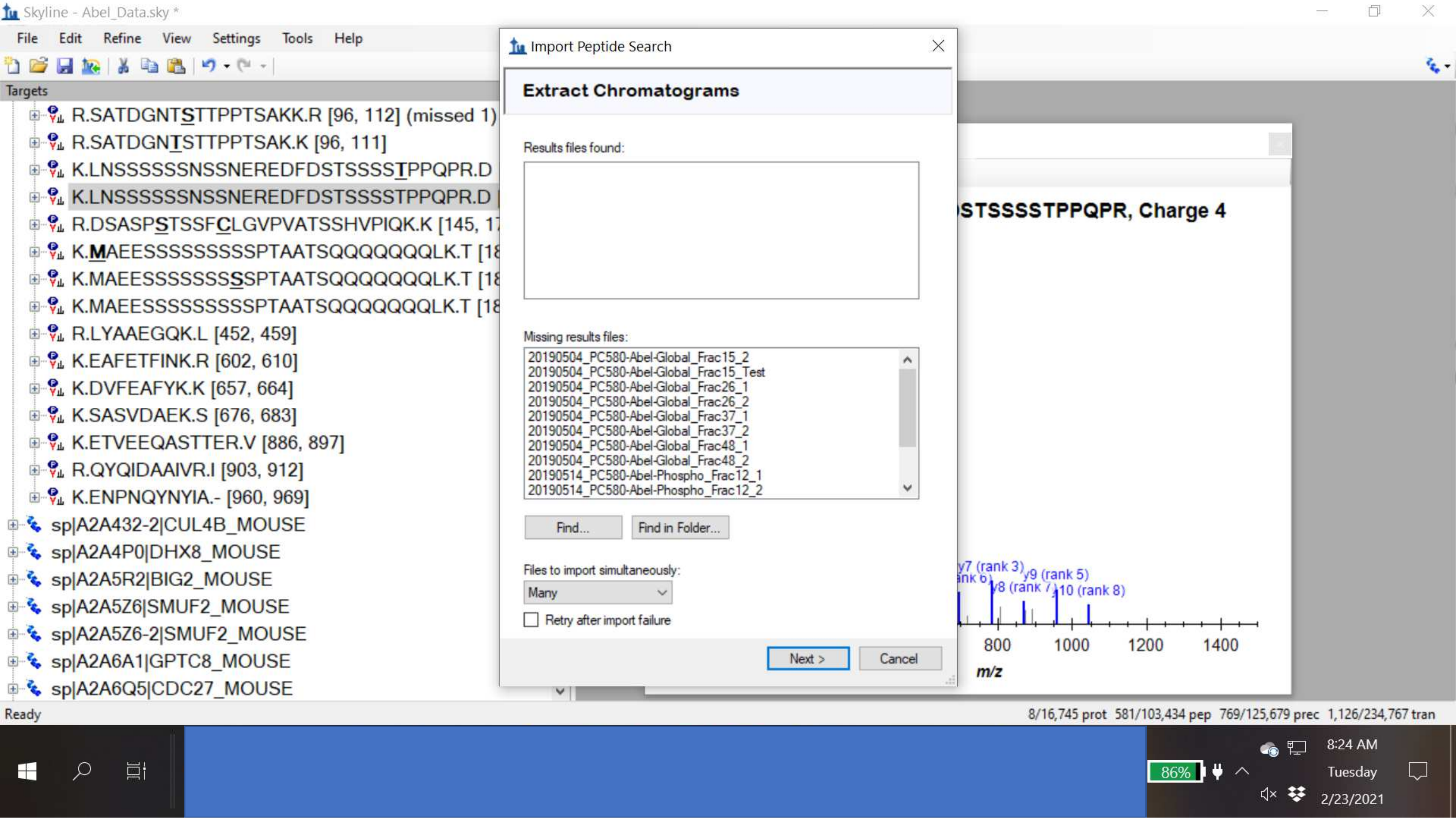
○ DIA

○ PRM

Building Peptide Search Library

Reading results from msms.txt.

When I try to complete this task, the program returns with this error saying I have missing files in the folder. The files listed in the folder are the files provided to me by an external MS facility. Should I request the facility send all the MS files contained in the msms.txt file?



R.SATDGNTSTTPPTSAKK.R [96, 112] (missed 1)
R.SATDGNTSTTPPTSAK.K [96, 111]
K.LNSSSSSSNSSNEREDFDSTSSSSTPPQPR.D
K.LNSSSSSSNSSNEREDFDSTSSSSTPPQPR.D
R.DSASPSTSSFCLGVPVATSSHVPIQK.K [145, 17
K.MAEESSSSSSSSSPTAATSQQQQQQLK.T [18
K.MAEESSSSSSSSSPTAATSQQQQQQLK.T [18
K.MAEESSSSSSSSSPTAATSQQQQQQLK.T [18
R.LYAAEGQK.L [452, 459]
K.EAFETFINK.R [602, 610]
K.DVFEAFYK.K [657, 664]
K.SASVDAEK.S [676, 683]
K.ETVEEQASTTER.V [886, 897]
R.QYQIDAAIVR.I [903, 912]
K.ENPNQYNYIA.- [960, 969]
sp|A2A432-2|CUL4B_MOUSE
sp|A2A4P0|DHX8_MOUSE
sp|A2A5R2|BIG2_MOUSE
sp|A2A5Z6|SMUF2_MOUSE
sp|A2A5Z6-2|SMUF2_MOUSE
sp|A2A6A1|GPTC8_MOUSE
sp|A2A6Q5|CDC27_MOUSE

Extract Chromatograms

Results files found:

Missing results files:

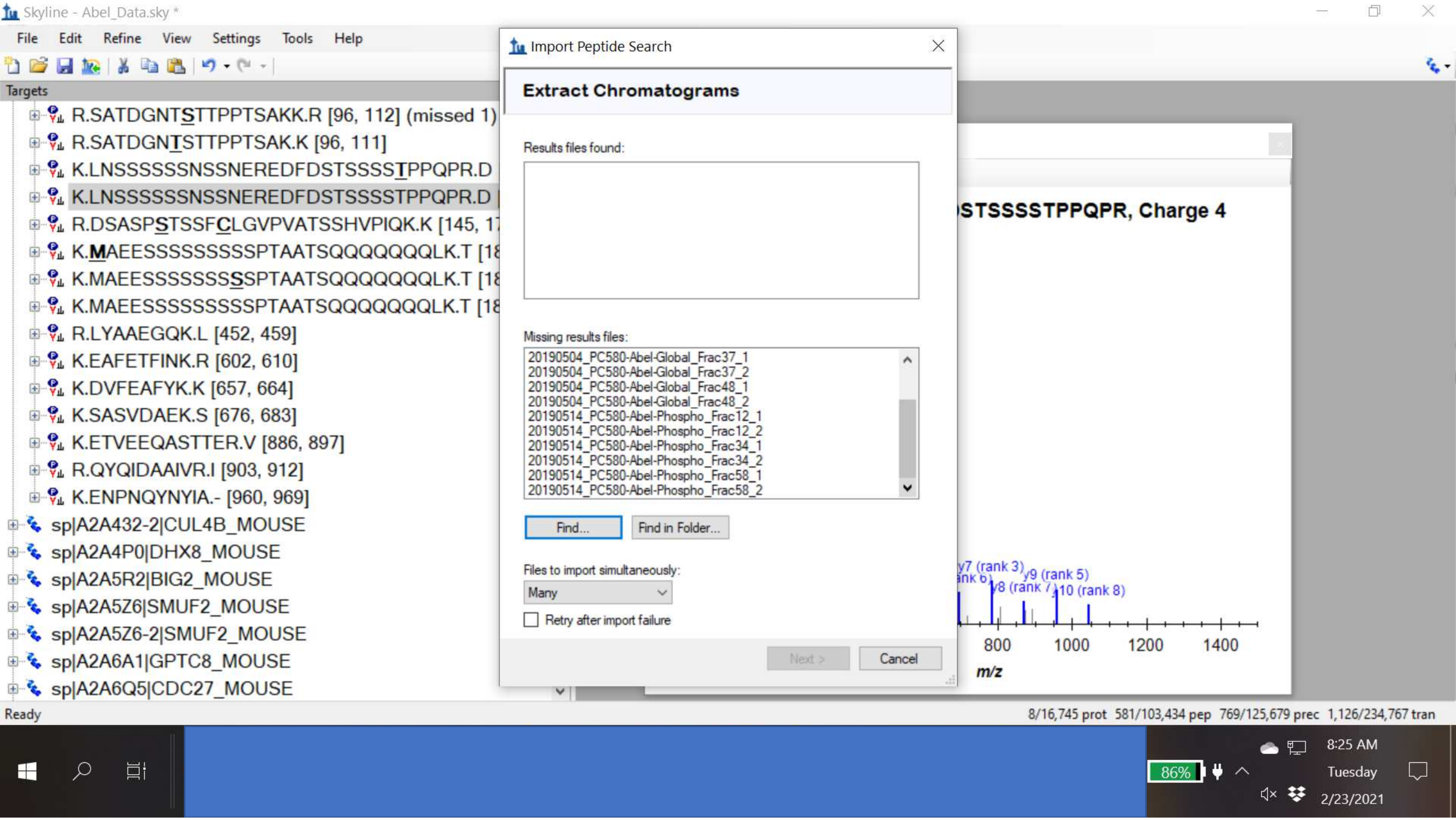
20190504_PC580-Abel-Global_Frac15_2
20190504_PC580-Abel-Global_Frac15_Test
20190504_PC580-Abel-Global_Frac26_1
20190504_PC580-Abel-Global_Frac26_2
20190504_PC580-Abel-Global_Frac37_1
20190504_PC580-Abel-Global_Frac37_2
20190504_PC580-Abel-Global_Frac48_1
20190504_PC580-Abel-Global_Frac48_2
20190514_PC580-Abel-Phospho_Frac12_1
20190514_PC580-Abel-Phospho_Frac12_2

Files to import simultaneously: Many

☐ Retry after import failure

Ready

8/16,745 prot 581/103,434 pep 769/125,679 prec 1,126/234,767 tran



R.SATDGNTSTTPPTSAKK.R [96, 112] (missed 1)
R.SATDGNTSTTPPTSAK.K [96, 111]
K.LNSSSSSSNSSNEREDFDSTSSSSTPPQPR.D
K.LNSSSSSSNSSNEREDFDSTSSSSTPPQPR.D
R.DSASPSTSSFCLGVPVATSSHVPIQK.K [145, 17
K.MAEESSSSSSSSSPTAATSQQQQQQQLK.T [18
K.MAEESSSSSSSSSPTAATSQQQQQQQLK.T [18
K.MAEESSSSSSSSSPTAATSQQQQQQQLK.T [18
R.LYAAEGQK.L [452, 459]
K.EAFETFINK.R [602, 610]
K.DVFEAFYK.K [657, 664]
K.SASVDAEK.S [676, 683]
K.ETVEEQASTTER.V [886, 897]
R.QYQIDAAIVR.I [903, 912]
K.ENPNQYNYIA.- [960, 969]
sp|A2A432-2|CUL4B_MOUSE
sp|A2A4P0|DHX8_MOUSE
sp|A2A5R2|BIG2_MOUSE
sp|A2A5Z6|SMUF2_MOUSE
sp|A2A5Z6-2|SMUF2_MOUSE
sp|A2A6A1|GPTC8_MOUSE
sp|A2A6Q5|CDC27_MOUSE

Extract Chromatograms

Results files found:

Missing results files:

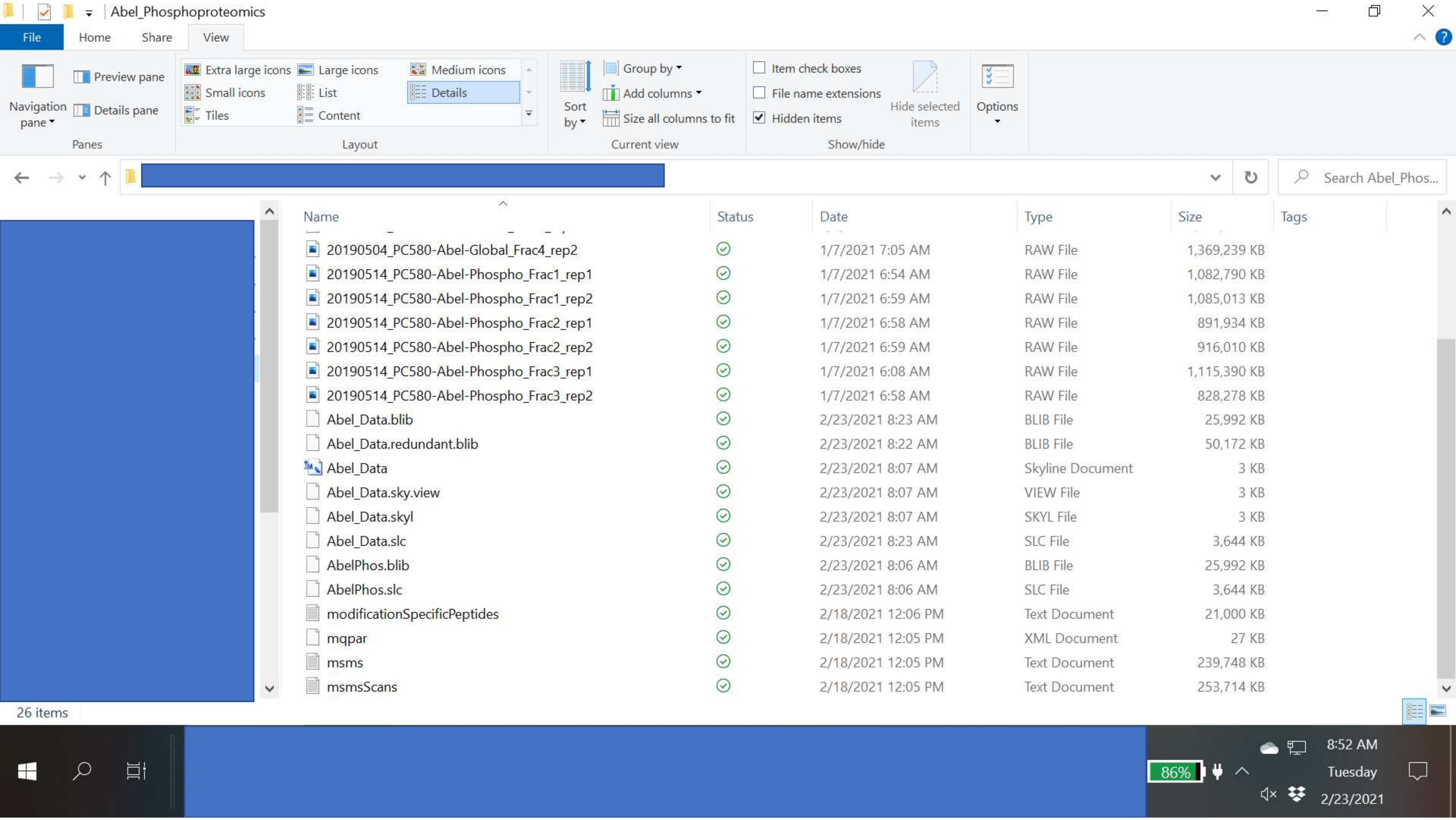
20190504_PC580-Abel-Global_Frac37_1
20190504_PC580-Abel-Global_Frac37_2
20190504_PC580-Abel-Global_Frac48_1
20190504_PC580-Abel-Global_Frac48_2
20190514_PC580-Abel-Phospho_Frac12_1
20190514_PC580-Abel-Phospho_Frac12_2
20190514_PC580-Abel-Phospho_Frac34_1
20190514_PC580-Abel-Phospho_Frac34_2
20190514_PC580-Abel-Phospho_Frac58_1
20190514_PC580-Abel-Phospho_Frac58_2

Files to import simultaneously: Many

☐ Retry after import failure

Ready

8/16,745 prot 581/103,434 pep 769/125,679 prec 1,126/234,767 tran



[redacted]

[redacted]

Name	Status	Date	Type	Size	Tags
20190504_PC580-Abel-Global_Frac4_rep2	✓	1/7/2021 7:05 AM	RAW File	1,369,239 KB	
20190514_PC580-Abel-Phospho_Frac1_rep1	✓	1/7/2021 6:54 AM	RAW File	1,082,790 KB	
20190514_PC580-Abel-Phospho_Frac1_rep2	✓	1/7/2021 6:59 AM	RAW File	1,085,013 KB	
20190514_PC580-Abel-Phospho_Frac2_rep1	✓	1/7/2021 6:58 AM	RAW File	891,934 KB	
20190514_PC580-Abel-Phospho_Frac2_rep2	✓	1/7/2021 6:59 AM	RAW File	916,010 KB	
20190514_PC580-Abel-Phospho_Frac3_rep1	✓	1/7/2021 6:08 AM	RAW File	1,115,390 KB	
20190514_PC580-Abel-Phospho_Frac3_rep2	✓	1/7/2021 6:58 AM	RAW File	828,278 KB	
Abel_Data.blib	✓	2/23/2021 8:23 AM	BLIB File	25,992 KB	
Abel_Data.redundant.blib	✓	2/23/2021 8:22 AM	BLIB File	50,172 KB	
Abel_Data	✓	2/23/2021 8:07 AM	Skyline Document	3 KB	
Abel_Data.sky.view	✓	2/23/2021 8:07 AM	VIEW File	3 KB	
Abel_Data.skyl	✓	2/23/2021 8:07 AM	SKYL File	3 KB	
Abel_Data.slc	✓	2/23/2021 8:23 AM	SLC File	3,644 KB	
AbelPhos.blib	✓	2/23/2021 8:06 AM	BLIB File	25,992 KB	
AbelPhos.slc	✓	2/23/2021 8:06 AM	SLC File	3,644 KB	
modificationSpecificPeptides	✓	2/18/2021 12:06 PM	Text Document	21,000 KB	
mqpar	✓	2/18/2021 12:05 PM	XML Document	27 KB	
msms	✓	2/18/2021 12:05 PM	Text Document	239,748 KB	
msmsScans	✓	2/18/2021 12:05 PM	Text Document	253,714 KB	

26 items

[redacted]