

## SUPPORTING INFORMATION

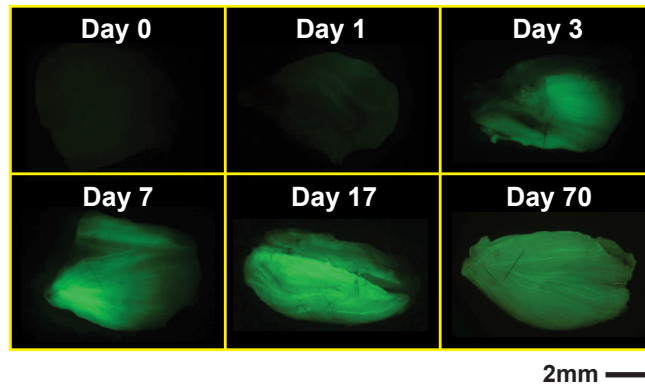
### Supplemental Table 1: Proteomics data.

**Supplemental Figure 1: In vivo transfection of mouse skeletal muscle.** Mouse tibialis anterior (TA) muscles were transfected in vivo with 5  $\mu$ g plasmid encoding eGFP, then harvested at the indicated times post-transfection. Images are of the entire TA muscles, which were photographed at low magnification to visualize eGFP expression.

**Supplemental Figure 2. Selected MS/MS spectra from key ATF4-interacting proteins.** (A) MS/MS for peptide from CEBPB CCAAT/enhancer-binding protein beta (P28033): peptide VLELTAENER with the precursor ion at m/z 587.31<sup>2+</sup>. (B) MS/MS for peptide from CEBPB CCAAT/enhancer-binding protein beta (P28033): peptide APAAEPAIGEHER with the precursor ion at m/z 674.34<sup>2+</sup>. (C) MS/MS for peptide from CEBPD CCAAT/enhancer-binding protein delta (Q00322): peptide GSPGPSLAPGTVR with the precursor ion at m/z 598.33<sup>2+</sup>. (D) MS/MS for peptide from CEBPD CCAAT/enhancer-binding protein delta (Q00322): peptide LVELSAENEK with the precursor ion at m/z 566.30<sup>2+</sup>. (E) MS/MS for peptide from CEBPA CCAAT/enhancer-binding protein alpha (P53566): peptide GLAGHPDLR with the precursor ion at m/z 503.78<sup>2+</sup>. (F) MS/MS for peptide from CEBPG CCAAT/enhancer-binding protein gamma (P53568): peptide QKAQDTLQR with the precursor ion at m/z 535.78<sup>2+</sup>. (G) MS/MS for peptide from MAF MOUSE Transcription factor Maf (P54843): peptide FSDEQLVTMSVR with the precursor ion at m/z 714.35<sup>2+</sup>. (H) MS/MS for peptide from LUZP1 Leucine zipper protein 1 (Q8R4U7): peptide ISSELEMLR with the precursor ion at m/z 539.28<sup>2+</sup>. (I) MS/MS for peptide from BRD4 Bromodomain-containing protein 4 (Q9ESU6): peptide AQAEHAEKEK with the precursor ion at m/z 571.28<sup>2+</sup>. (J) MS/MS for peptide from PTRF Polymerase I and transcript release factor, Cavin-1 (O54724): peptide ATEEPSGTGSDELK with the precursor ion at m/z 767.37<sup>2+</sup>. (K) MS/MS for peptide from SDPR Serum deprivation-response protein, Cavin-2 (Q63918): peptide YQASTSNTVSK with the precursor ion at m/z 593.29<sup>2+</sup>. (L) MS/MS for peptide from MURC Muscle-related coiled-coil protein, Cavin-4 (A2AMM0): peptide VASVVDSVQASQKR with the precursor ion at m/z 491.94<sup>3+</sup>. (M) MS/MS for peptide from SYNCI Syncoilin (Q9EPM5): peptide ALQDTQAEIQEMR with the precursor ion at m/z 774.87<sup>3+</sup>.

**Supplemental Figure 3. Mutation of the ATF-C/EBP site abolishes immobilization-induced Gadd45a reporter activity.** Bilateral mouse TA muscles were transfected with 5  $\mu$ g mutant reporter plasmid (described in Figure 7) plus 0.3  $\mu$ g Renilla plasmid. Three days after transfection, one hindlimb in each mouse was immobilized. Six days post-transfection, bilateral TAs were harvested for analysis of reporter gene activity. In each muscle, the level of reporter gene activity was normalized to the average level in the transfected mobile TAs. Each circle represents one transfected muscle, and bars denote the means. *P*-value was determined with a t-test.

**Figure S1**



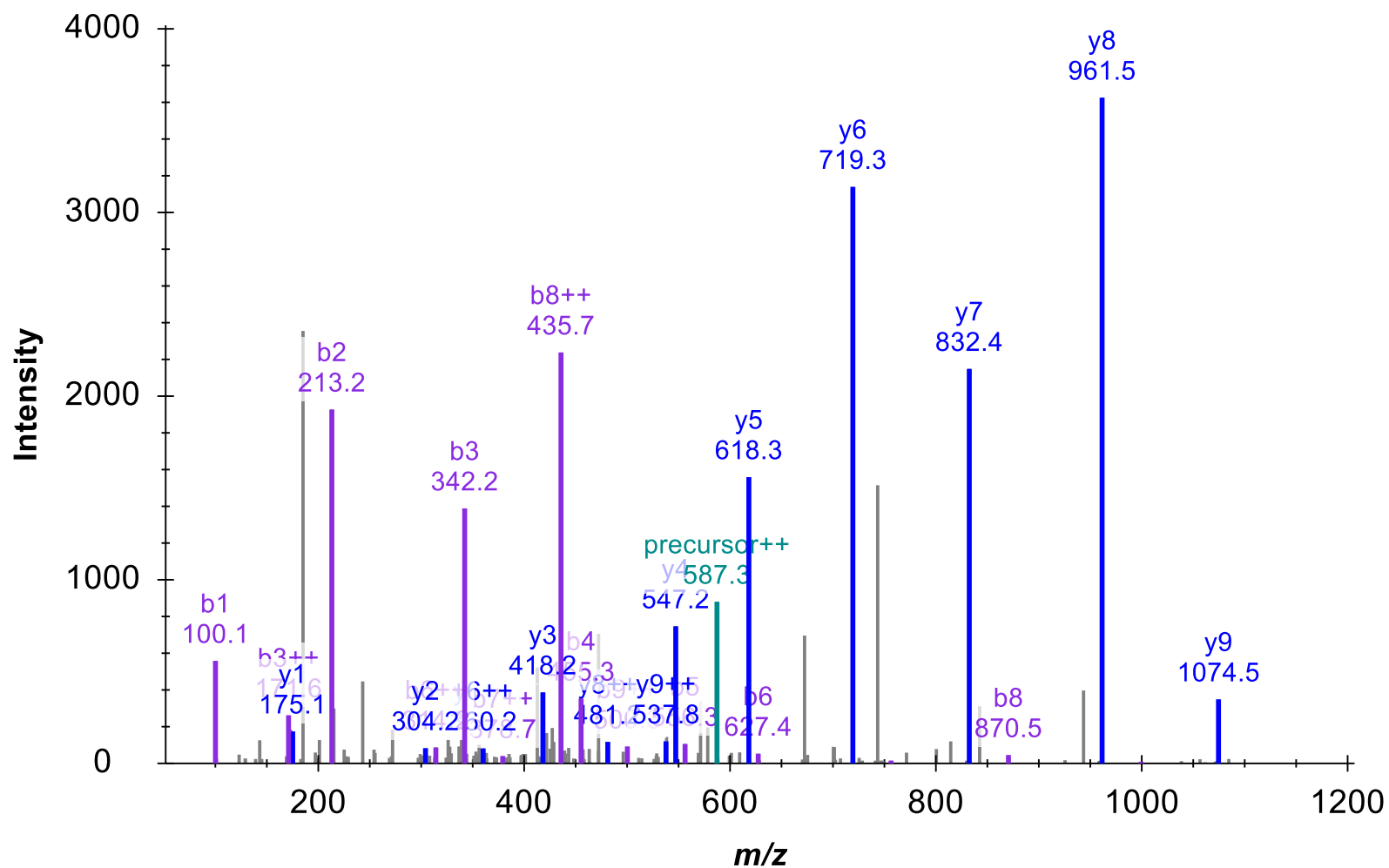
Suppl  
Fig 2A

# CEBPB CCAAT/enhancer-binding protein beta

P28033

VLELTAENER [253, 262]

m/z 587.31<sup>2+</sup>



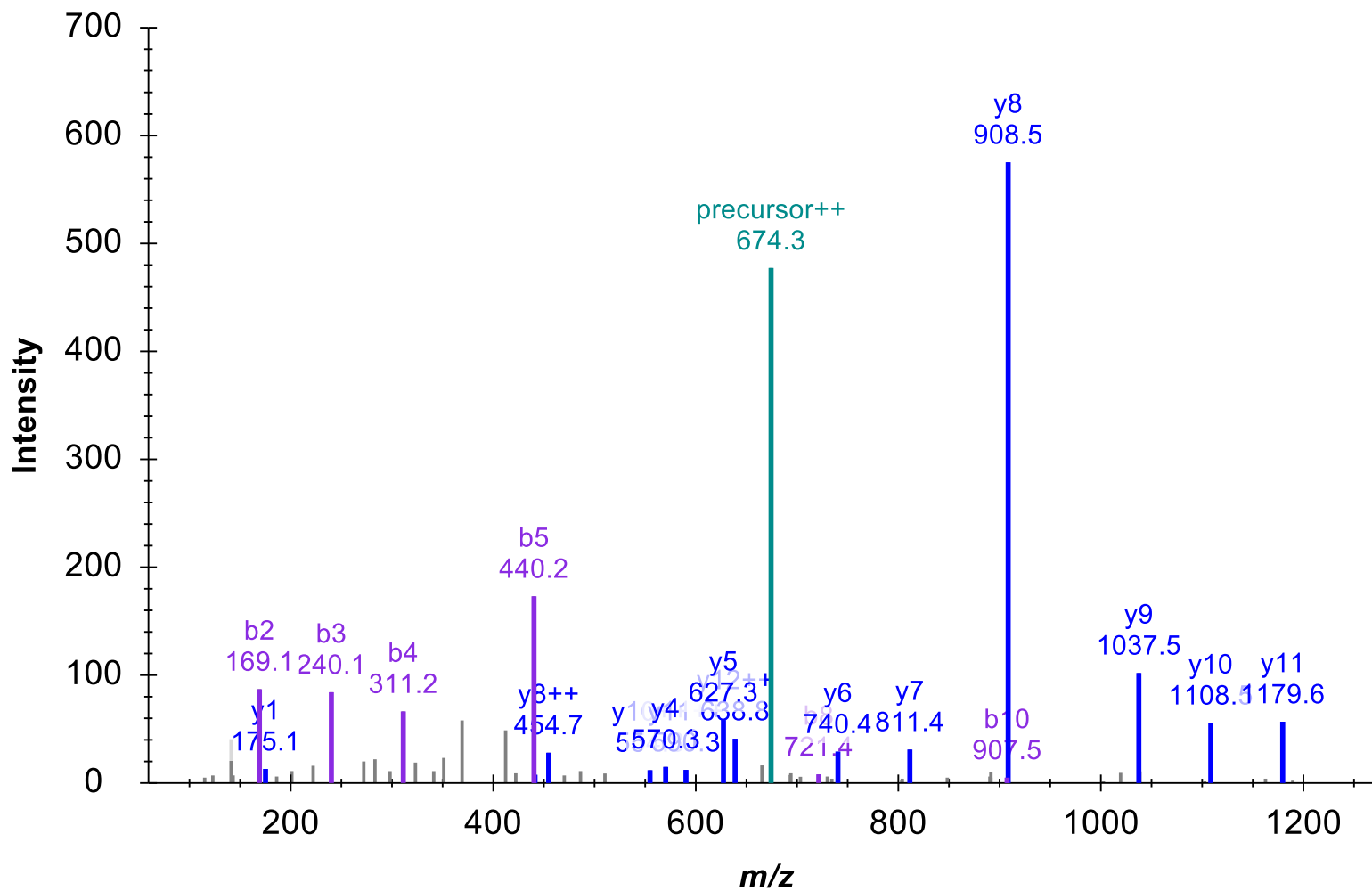
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Fig 2B

# CEBPB CCAAT/enhancer-binding protein beta

P28033

APAAEPAIGEHER (46 - 58)

m/z 674.34<sup>2+</sup>



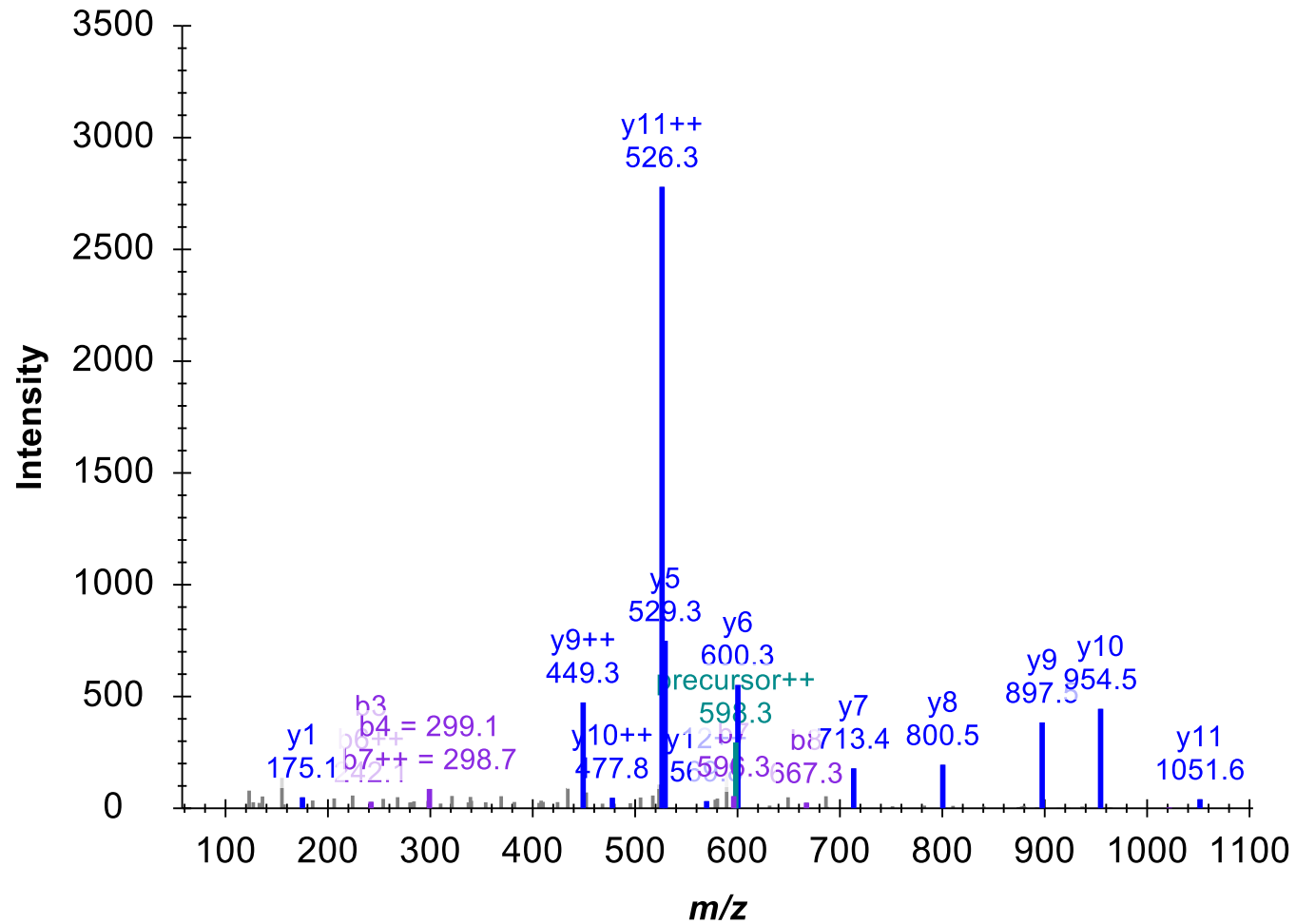
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Fig 2C

# CEBPD CCAAT/enhancer-binding protein delta

Q00322

GSPGPSLAPGTVR [165, 177]

m/z 598.33 <sup>2+</sup>



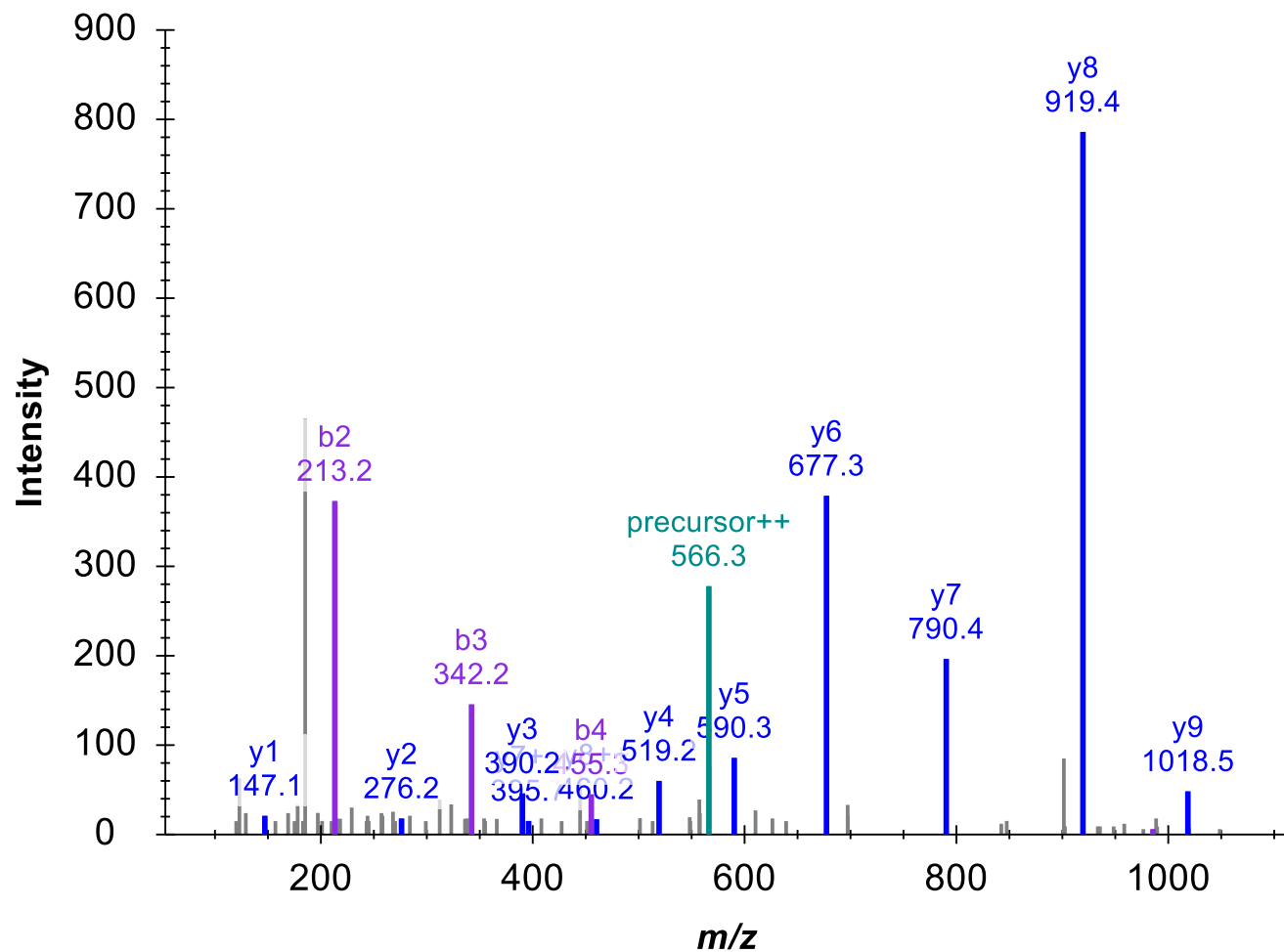
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Fig 2D

# CEBPD CCAAT/enhancer-binding protein delta

Q00322

LVELSAENEK (223-232)

m/z 566.30<sup>2+</sup>



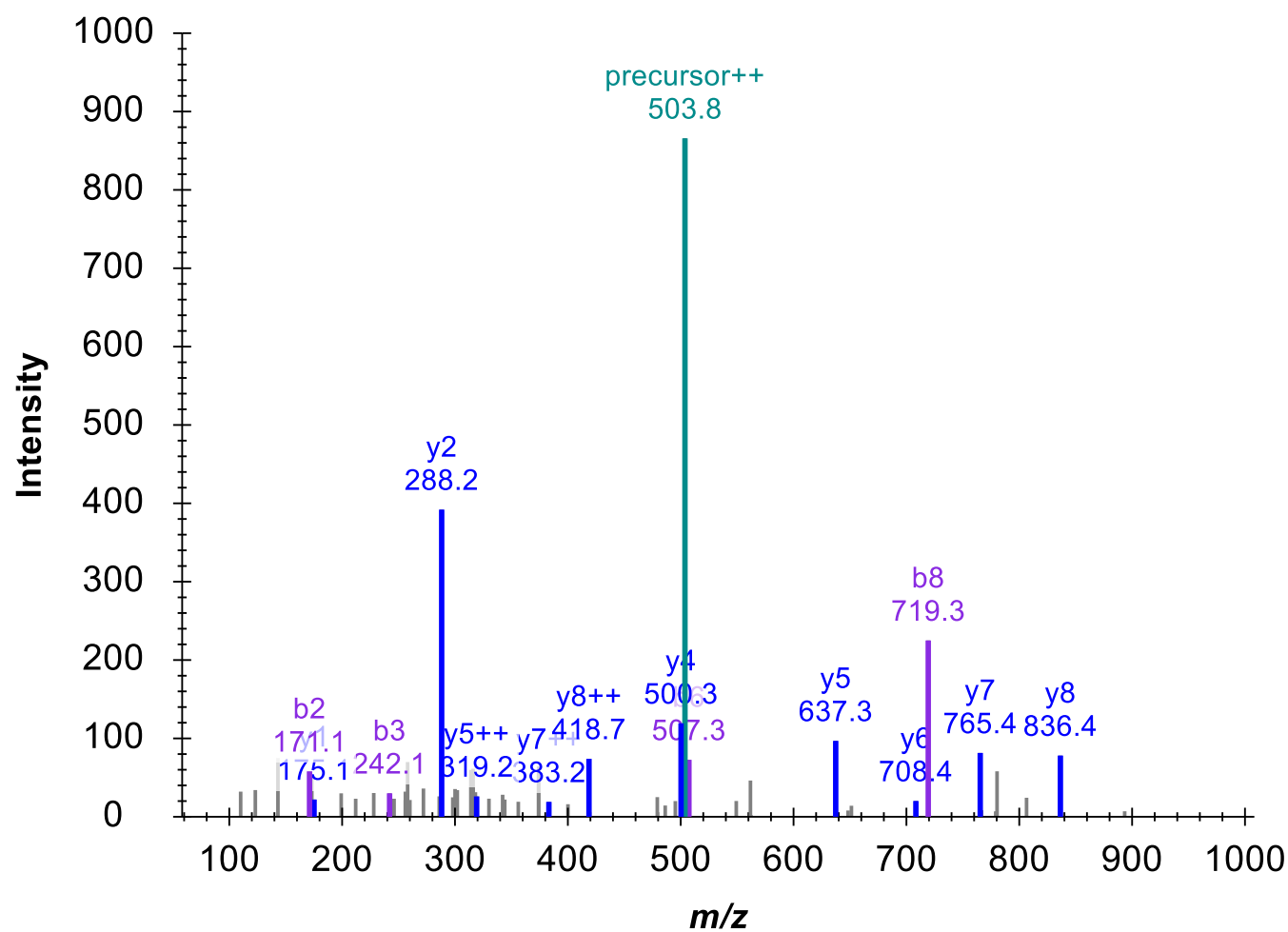
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Fig 2E

# CEBPA CCAAT/enhancer-binding protein alpha

P53566

GLAGAHPLDLR (251, 260)

m/z 503.78<sup>2+</sup>



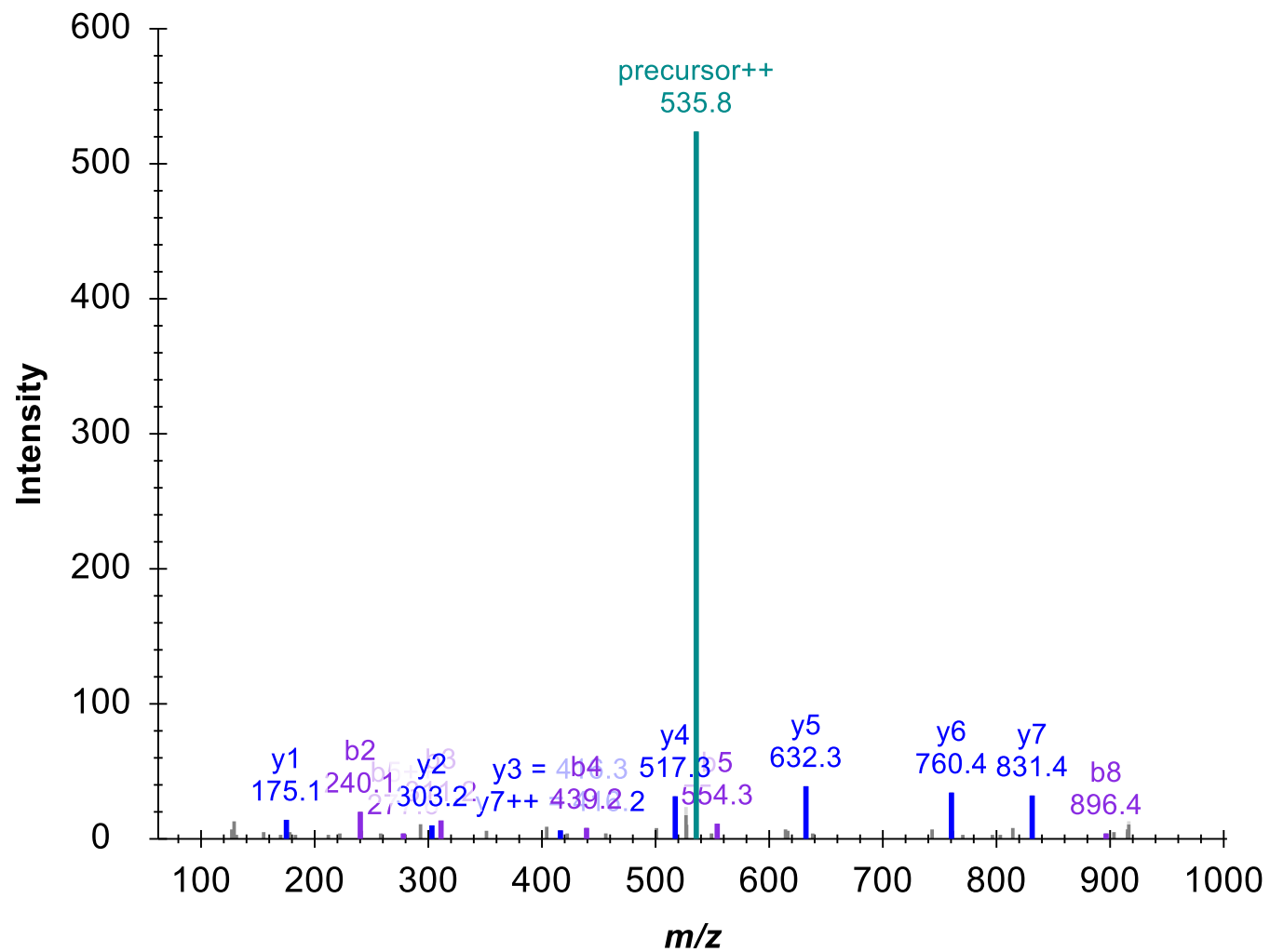
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Fig 2F

# CEBPG CCAAT/enhancer-binding protein gamma

P53568

QKAQDTLQR (85, 93)

m/z 535.78 <sup>2+</sup>





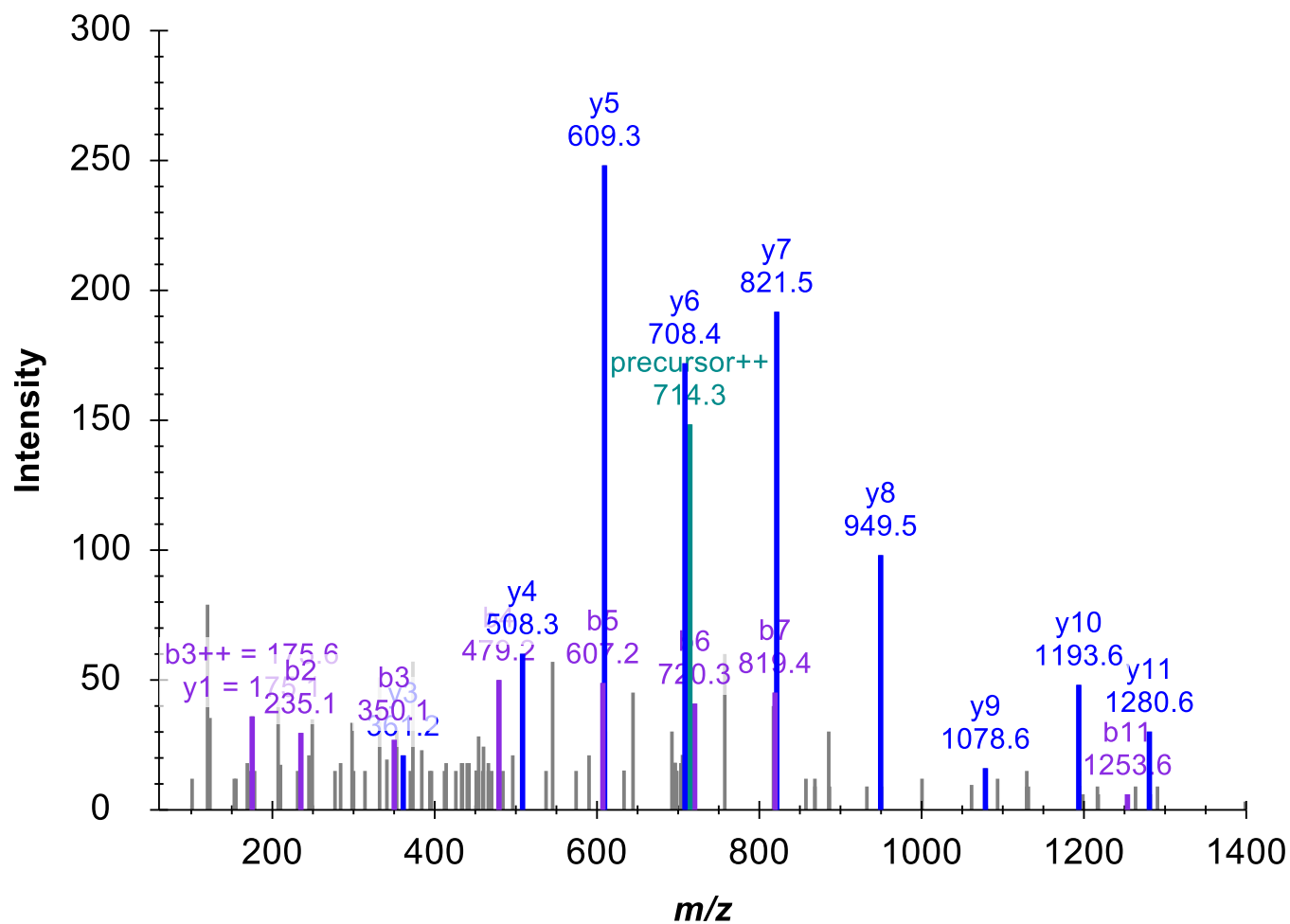
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Fig 2G

# MAF Transcription factor Maf

P54843

FSDEQLVTMSVR [257, 268]

m/z 714.35<sup>2+</sup>



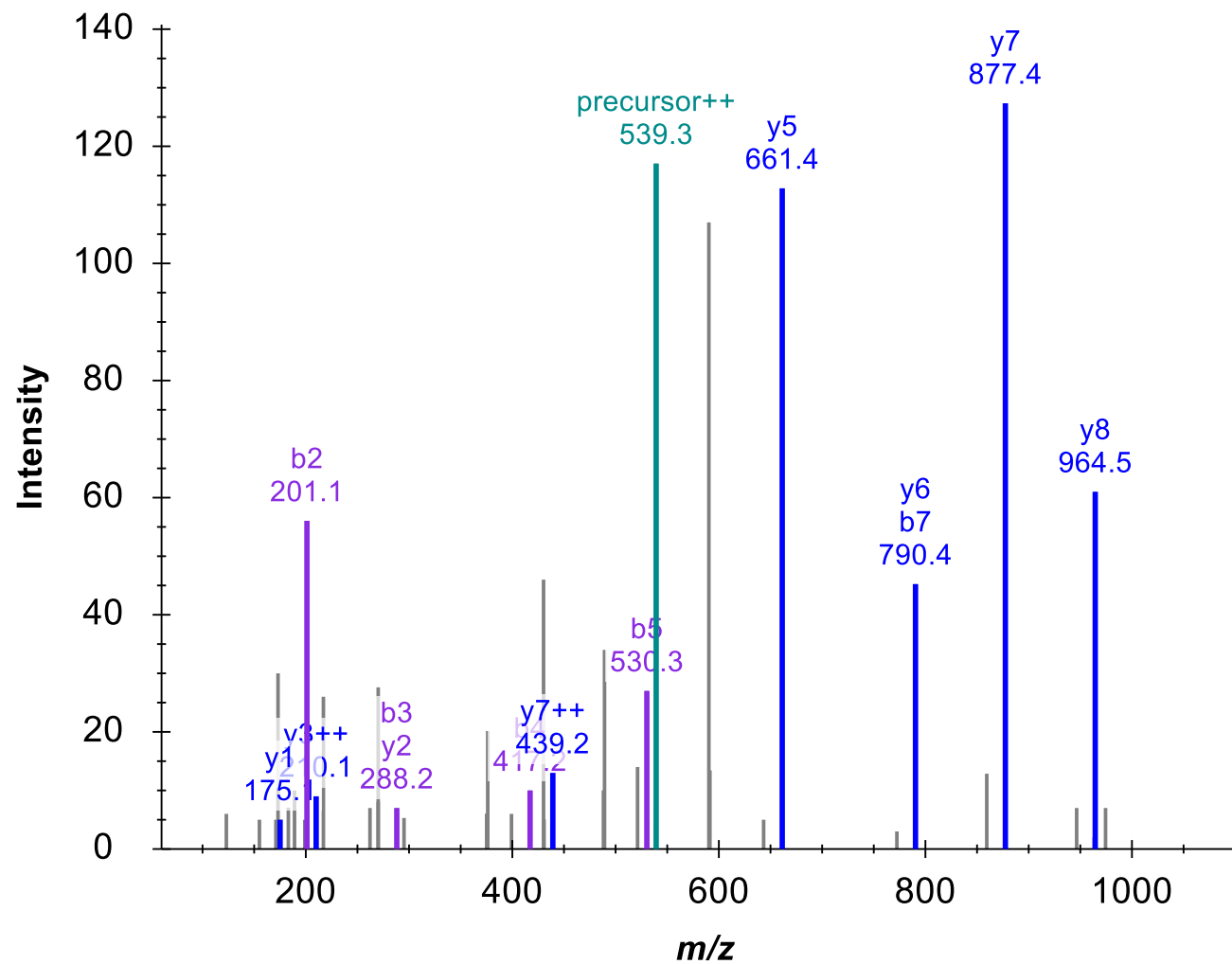
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Fig 2H

# LUZP1 Leucine zipper protein 1

Q8R4U7

ISSELEMLR [150, 158]

m/z 539.28<sup>2+</sup>



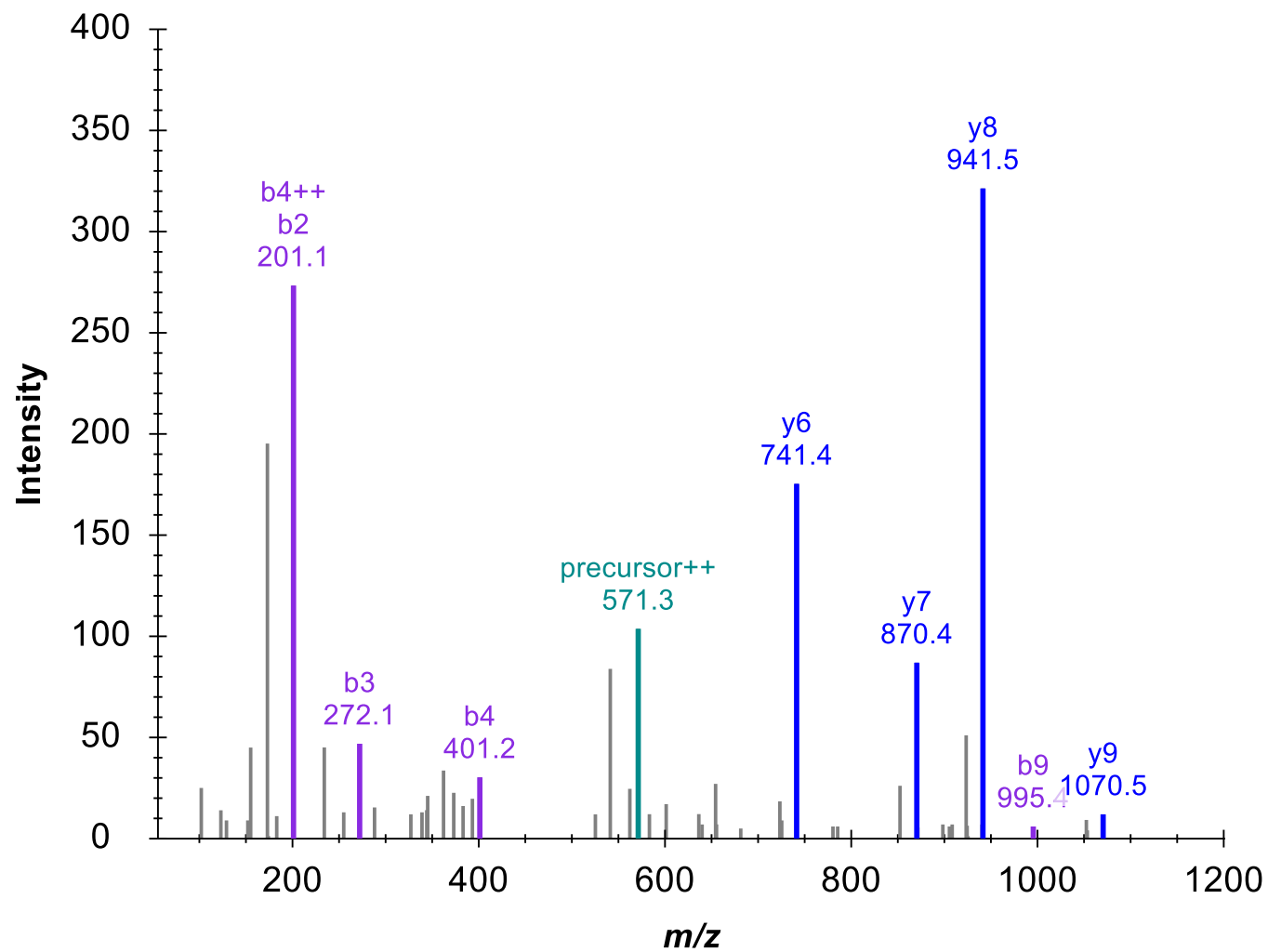
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Fig 2I

# BRD4 Bromodomain-containing protein 4

Q9ESU6

AQAAEHAEKEK [1278, 1287]

m/z 571.28<sup>2+</sup>



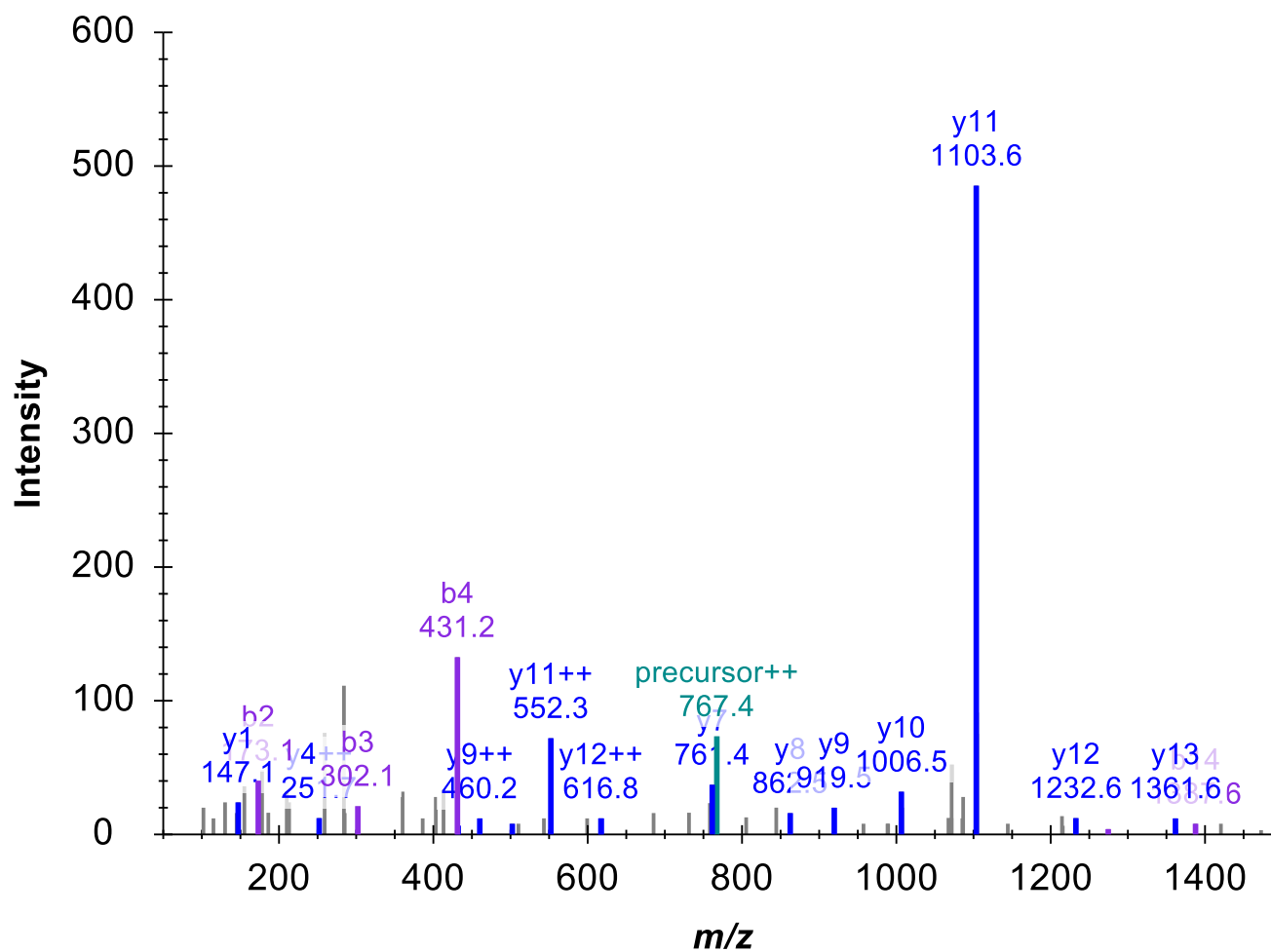
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Fig 2J

# PTRF Polymerase I and transcript release factor Cavin-1

O54724

ATEEPSGTGSDLIK [32, 46]

m/z 767.37<sup>2+</sup>



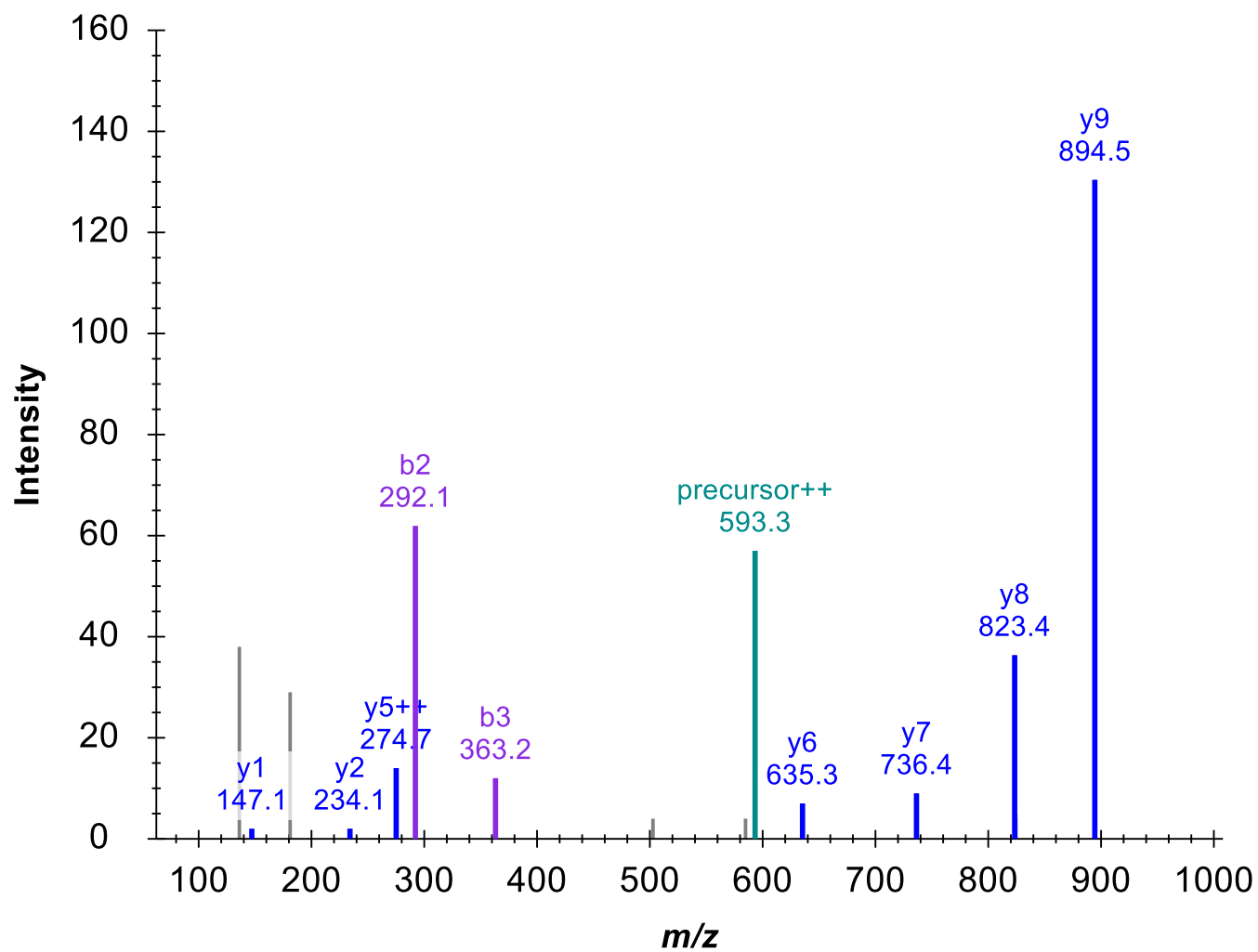
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Fig 2K

# SDPR Serum deprivation-response protein Cavin-2

Q63918

YQASTSNTVSK [102, 112]

m/z 593.29<sup>2+</sup>



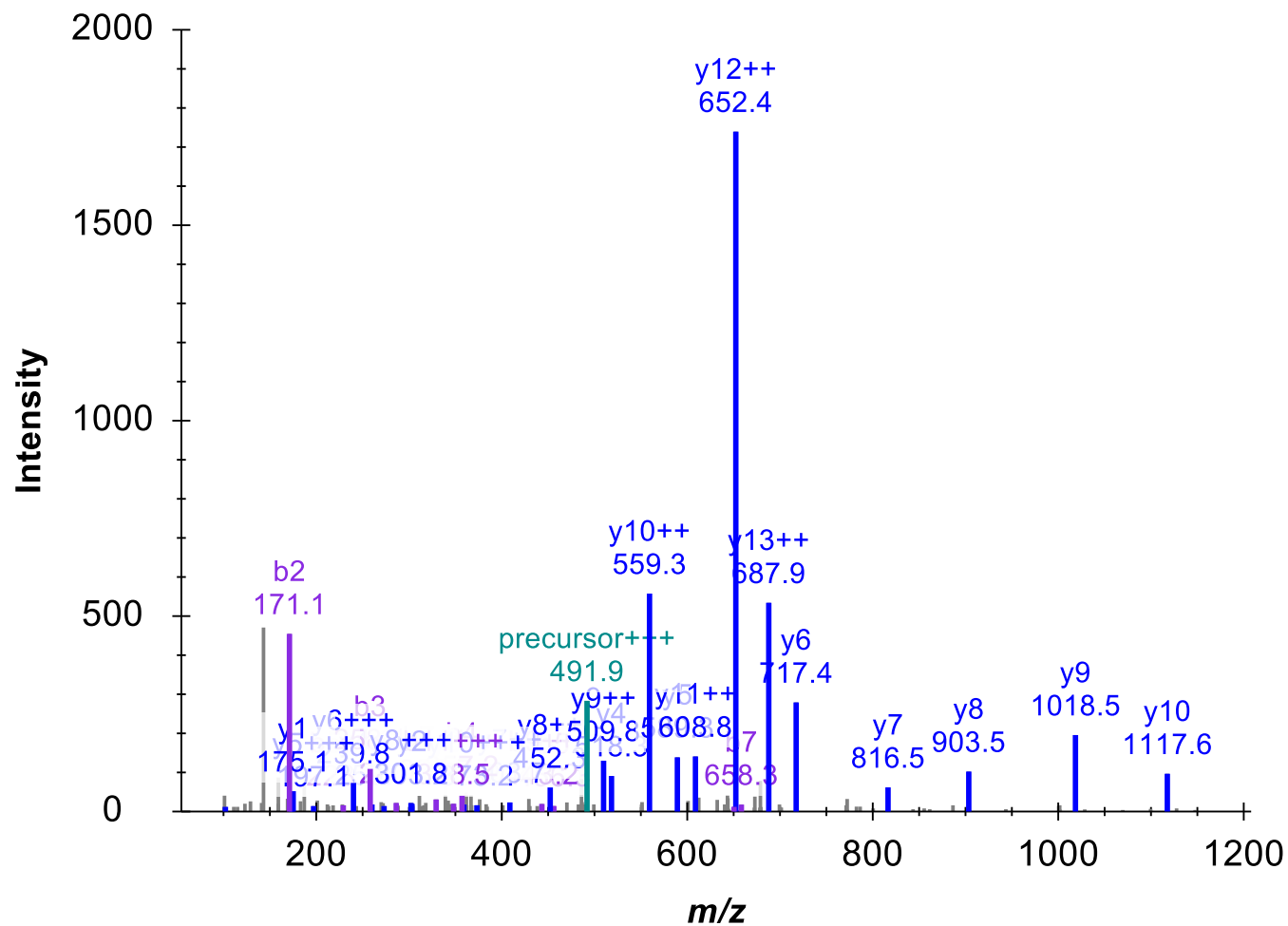
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Fig 2L

# MURC Muscle-related coiled-coil protein Cavin-4

A2AMM0

VASVVDSVQASQKR [39, 52]

m/z 491.94 3+

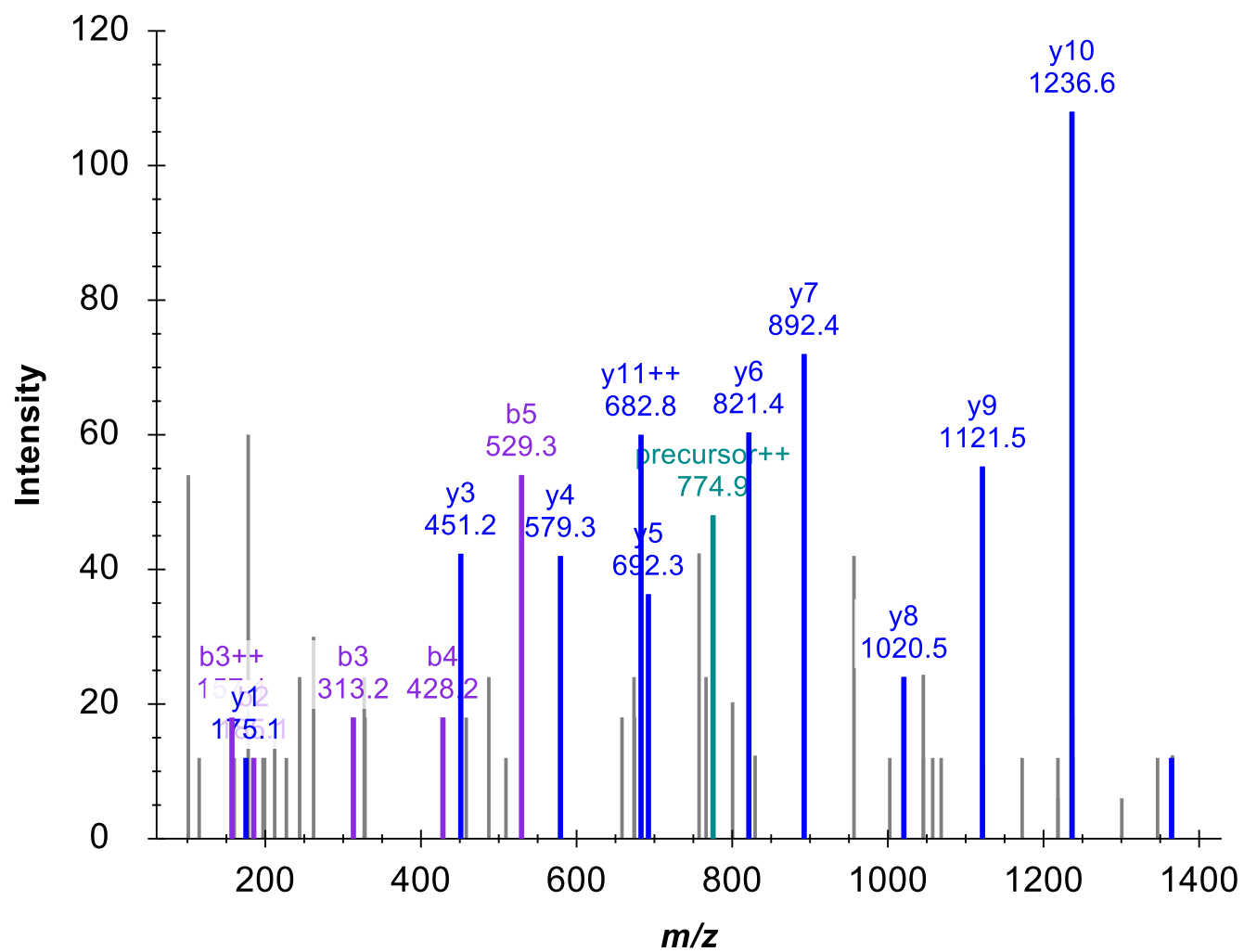


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Fig 2M

# SYNCI Syncoilin

Q9EPM5 ALQDTQAEIQEMR [348, 360]

774.87<sup>2+</sup>



# Figure S3

