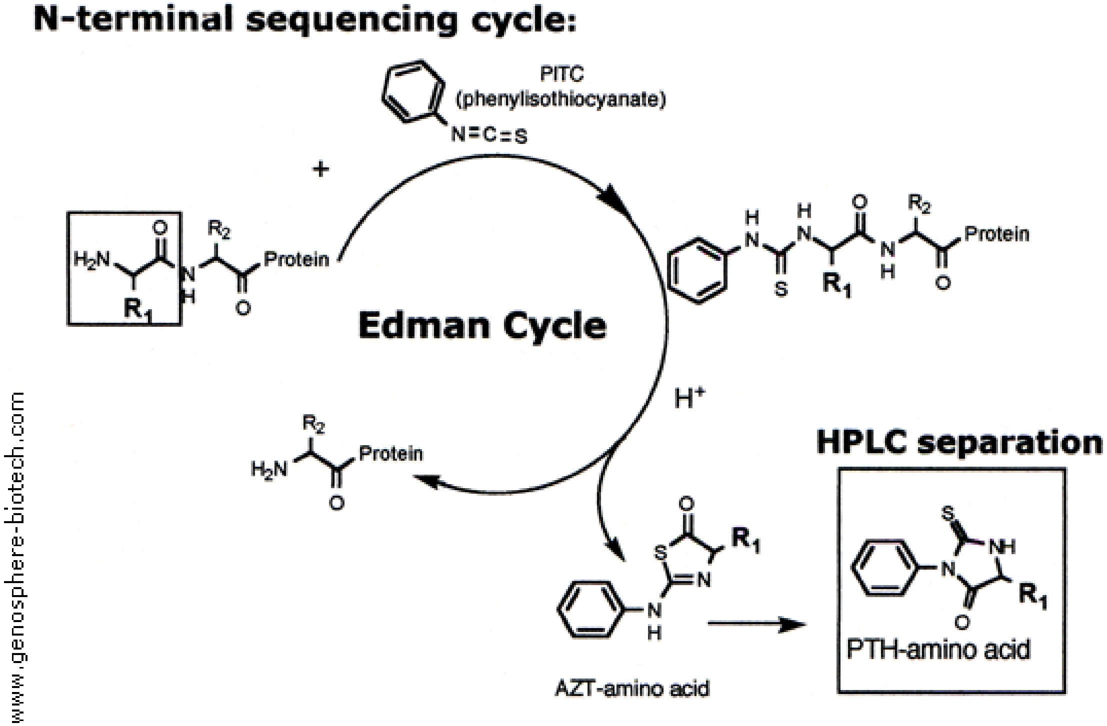
# Sequences

## Breaking things apart

# The old way...



- Starts at N-
- Goes for 10-20
- Stops...

Protein and peptide sequence step by step

# Ion fragmentation

- MSMS ion search (protein ID)
- MSMS sequence confirmation
- MSMS sequence determination

MSMS

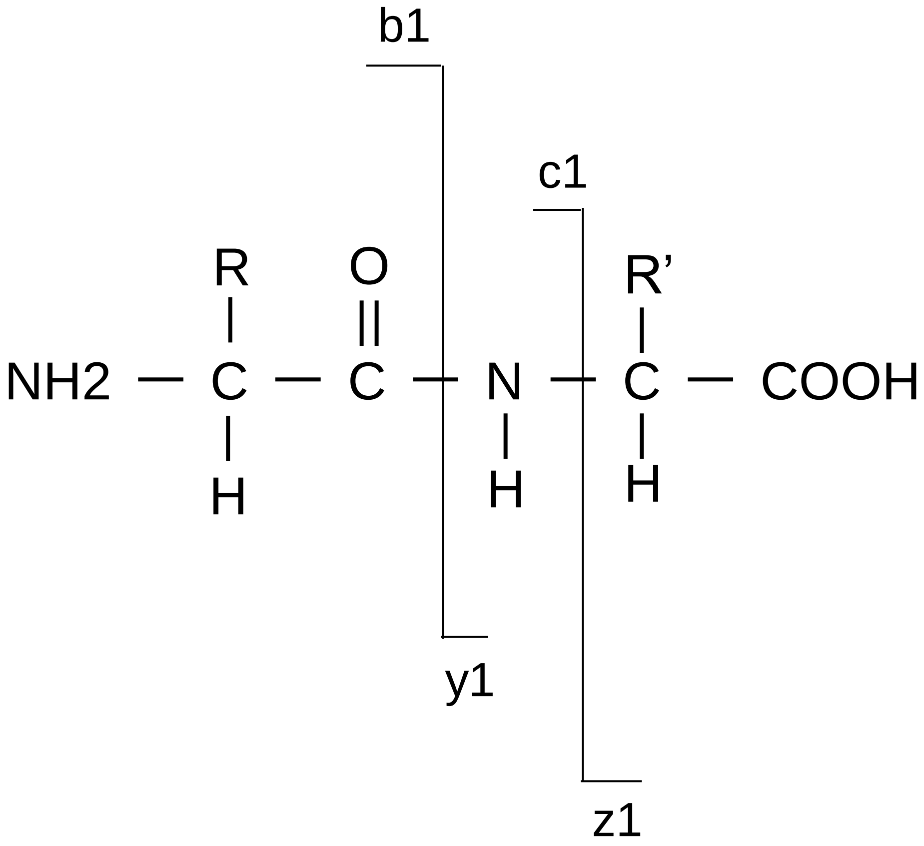
# FragWays

- CAD
- ETD
- SORI-CAD
- IRMPD
- UVPD
- ECD
- IRMPD-ECD

All others...

FTICR-MS
Fourier transform ion-cyclotron ressonance
Mass spectrometry

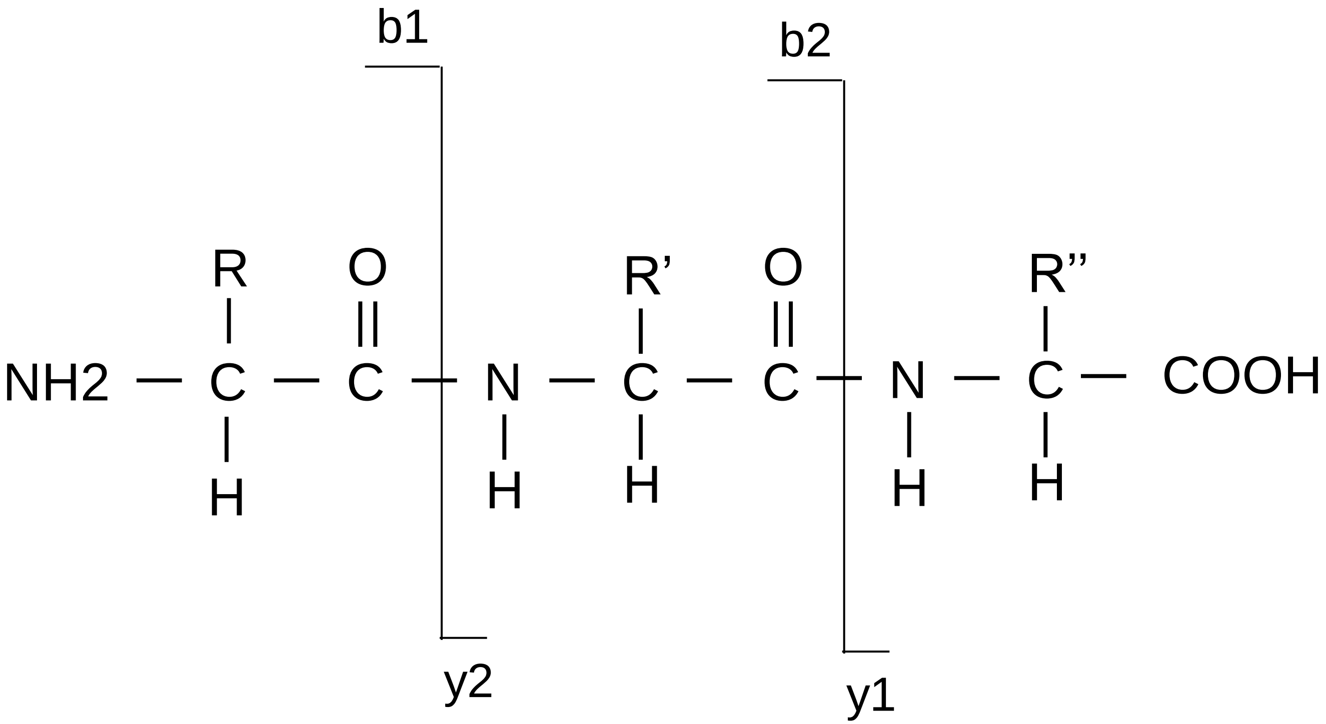
# a e i o u...

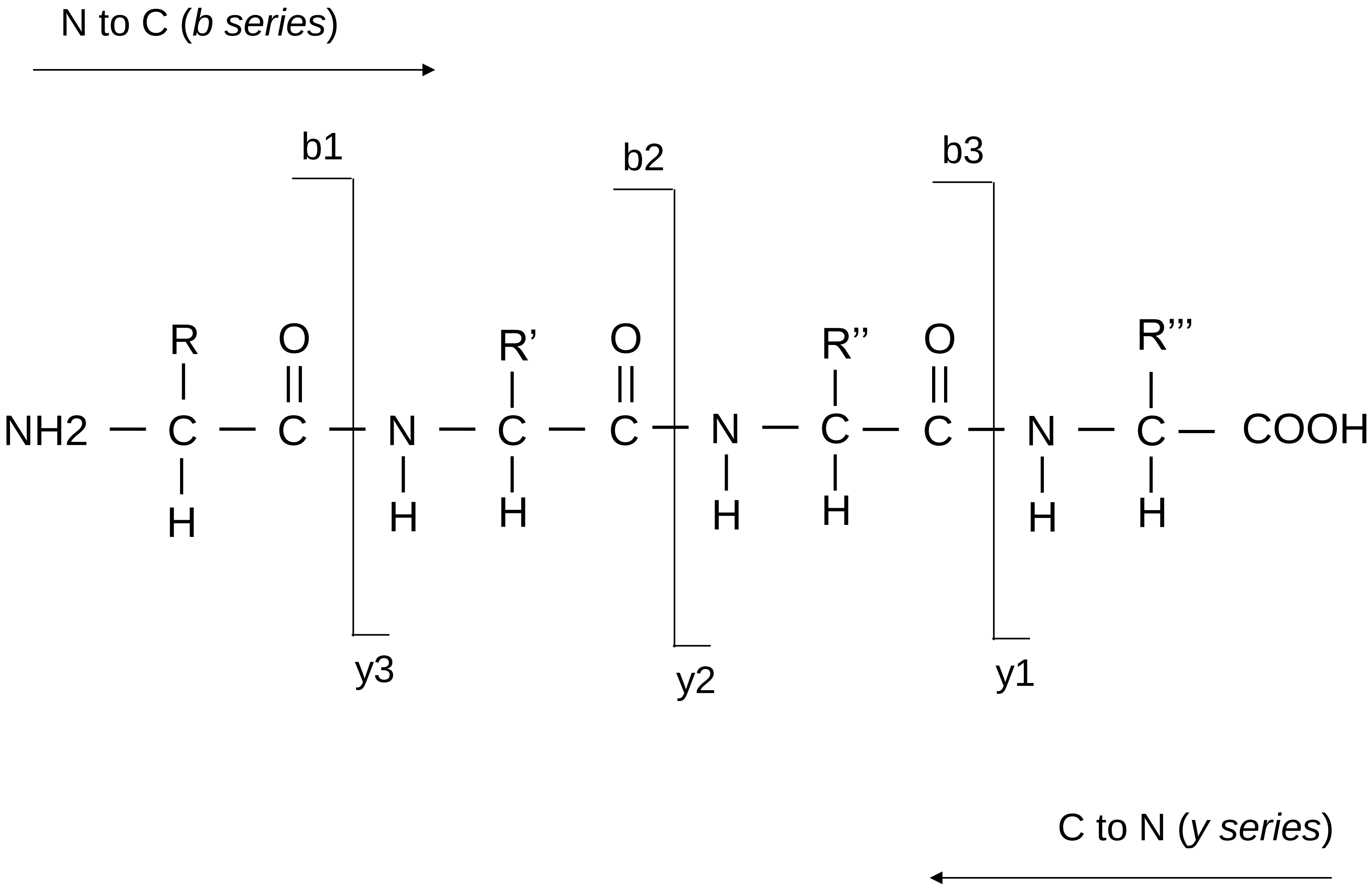


Roepstorff & Folhman (1984)

http://www.matrixscience.com/help/fragmentation_help.htm

# a b c … x y z




N to C (*b series*)

C to N (*y series*)

| **Aminoacid** | Single letter code | **b** | **y** |
|---|---|---|---|
| Alanine | A | 71.037110 | 89.047670 |
| Asparagine | N | 114.042930 | 132.053490 |
| Aspartate | D | 115.026940 | 133.037500 |
| Arginine | R | 156.101110 | 174.111670 |
| Cysteine | C | 103.009190 | 121.019750 |
| Glutamine | Q | 128.058580 | 146.069140 |
| Glutamate | E | 129.042590 | 147.053150 |
| Glycine | G | 57.021460 | 75.032020 |
| Histidine | H | 137.058910 | 155.069470 |
| Isoleucine | I | 113.084060 | 131.094620 |
| Leucine | L | 113.084060 | 131.094620 |
| Lysine | K | 128.094960 | 146.105520 |
| Methionine | M | 131.040490 | 149.051050 |
| Phenylalanine | F | 147.068410 | 165.078970 |
| Proline | P | 97.052760 | 115.063320 |
| Serine | S | 87.032030 | 105.042590 |
| Threonine | T | 101.047680 | 119.058240 |
| Tryptophan | W | 186.079310 | 204.089870 |
| Tyrosine | Y | 163.063330 | 181.073890 |
| Valine | V | 99.068410 | 117.078970 |

# Bottom-up & Top-down

- From peptides to protein
  - Proteolysis
  - Separation and analysis at peptide level

- From protein to aminoacids
  - MSMS of intact protein
  - Separation at protein level

# Bottom up vs Top-down

- Peptide Level
- Well established
- Universal
- Anybody/anywhere
- Incomplete information

- Protein Level
- Novel
- Specialized
- MS dependent
- Complete information
- PTM Analysis
- Structural Proteomics

TOP DOWN PROTEOMICS

http://topdownproteomics.org/