Annotating N termini for the Human Proteome Project:

Nα termini and N-acetylation status differentiate stable cleaved protein species from degradation remnants in the human erythrocyte proteome.

**Supplementary Figure S1.** Glycolysis pathway. Proteins identified in human erythrocytes are mapped onto pathway as derived from KEGG and highlighted in red.
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Supplementary Figure S2. Regulation of the actin cytoskeleton. Proteins identified in human erythrocytes are mapped onto pathway as derived from KEGG and highlighted in red.
Supplementary Figure S3. Ubiquitin mediated proteolysis. Proteins identified in human erythrocytes are mapped onto the pathway as derived from KEGG and highlighted in red.
Supplementary Figure S4. The proteasome. Proteins identified in human erythrocytes are mapped onto the complex as derived from KEGG and highlighted in red.
Supplementary Figure S5. Comparison between erythrocyte proteome studies. Overlap between proteins identified in three independent high throughput studies of the erythrocyte proteome. Pasini et al. (9) used shotgun analyses; Roux-Dalvai et al. used the ProteoMiner approach (13). All reported protein identifiers were mapped to their respective gene name for maximum consistency.
Supplementary Figure S6. Evidence for missing protein cancer/testis antigen 75 (Q6PK30). Representative spectra out of a total of 17 spectra supporting the peptide sequence ECP5LSALR.
Supplementary Figure S7. Evidence for missing protein intercellular adhesion molecule 4 (Q14773). Representative spectra out of a total of ten spectra supporting peptide sequences LNLDGLVVR and VIYSESRL.
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Supplementary Figure S8. Evidence for missing protein leucine-rich repeat transmembrane neuronal protein 1 (Q86UE6). Representative spectra out of a total of 87 spectra supporting peptide sequence LSLRYNSLSE.
Supplementary Figures S1-S13

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Supplementary Figure S9. Evidence for missing protein PR domain zinc finger protein 15 (P57071):
Representative spectra out of a total of 35 spectra supporting peptide sequence AARPPGSPPR
Nα termini and N-acetylation status differentiate stable cleaved protein species from degradation remnants in the human erythrocyte proteome.

Supplementary Figure S10. Evidence for missing protein protein FAM43A (Q8N2R8). Spectra supporting peptide sequence LEQELQEEEE.
Supplementary Figures S1-S13

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Supplementary Figure S11. Evidence for missing protein ‘uncharacterized protein C9orf84’ (Q5VXU9).
Representative spectrum for peptide MDSDGEKHLIKIL.
Supplemental Figure S12. Sequence determinants of cotranslational Met processing and N-terminal acetylation in human erythrocytes. Sequence logos representing A) 111 N termini with intact Met at position 1 of the gene-encoded protein sequence ($-.\text{Met}1$); B) 29 N termini with intact Met, mapping to positions >2 of the gene-encoded protein sequence, suggesting alternative translation start sites (X.Met); C) 304 N termini starting at position 2 after co-translational Met processing (Met1.X); D) 22 N termini with intact Met, mapping to positions >2 of the gene-encoded protein sequence, suggesting Met processing at alternative translation start sites (Met.X); E) 126 acetylated N termini starting with Met (combined canonical and alternative start sites, $-.\text{acMet}1$); F) 14 free N termini starting with Met (combined canonical and alternative start sites, $-.\text{freeMet}1$); G) 289 N termini acetylated after Met processing (combined canonical and alternative start sites, Met.acX); H) 36 free Met-processed N termini (combined canonical and alternative start sites, Met.freeX).