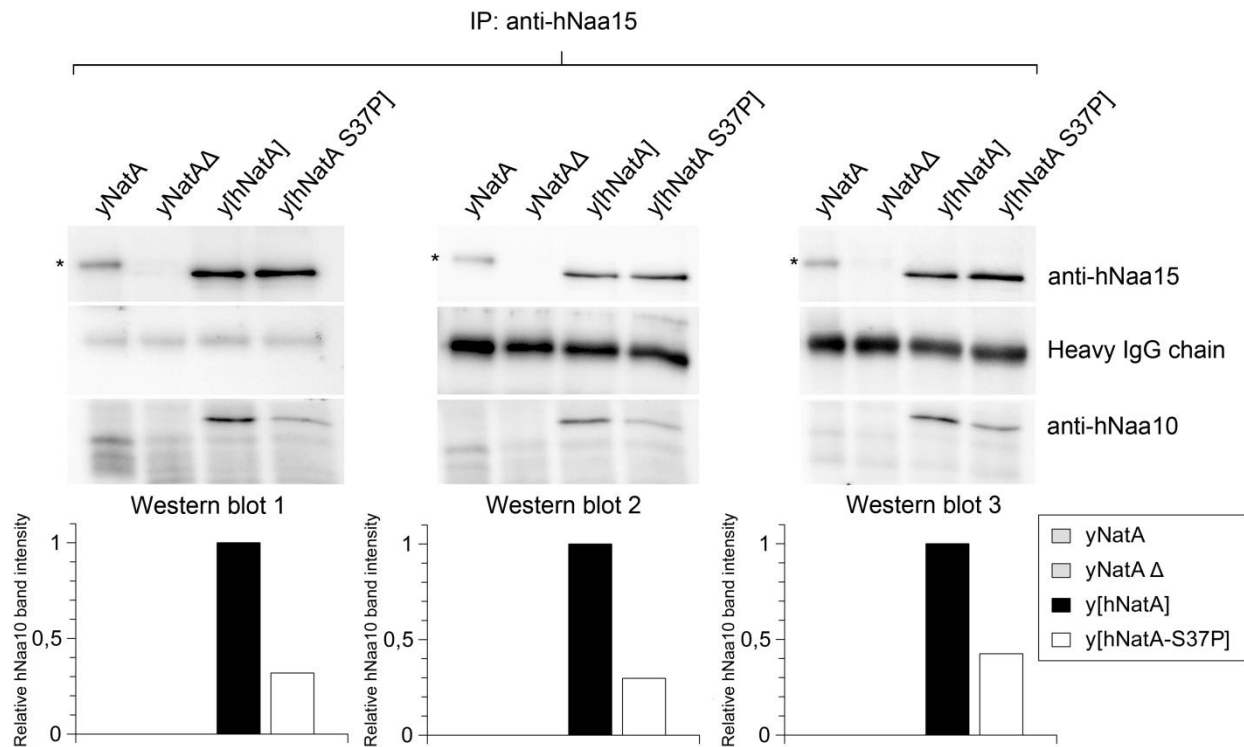


## Supplementary Data: Supplementary Figure 1 and Supplementary Table 1

### Supplementary Figure S1



**Figure S1| Quantification of hNaa10 WT and S37P levels in the anti-hNaa15 immunoprecipitated samples.** hNatA was immunoprecipitated from yeast cell lysates with anti-hNaa15 antibodies. To determine the amount of hNaa10 present in each sample, the immunoprecipitated hNatA complexes were analyzed by SDS-PAGE and Western blotting with anti-hNaa10 and anti-hNaa15. Heavy IgG chains serve as a loading control. Asterisk indicates an unspecific band. The relative band intensity is visualized in bars below each blot. The immunoprecipitated hNatA used in the *in vitro* acetyltransferase assay was analyzed on three separate gels, and the amount of hNaa10 S37P relative to hNaa10 WT was determined by averaging the measured band intensity from the three Western blots.

**Table S1| List of 1,608 unique N-terminal peptides identified in the yeast proteomes.** List of 1,608 yeast N-termini identified in the proteomes of the yNatA, yNatA- $\Delta$ , y[hNatA] and/or the y[hNatA S37P] yeast strain(s). Nat-type specificity, amino acid residue preceding the identified N-terminus (if any), compliance with the rules of N-terminal acetylation and iMet processing (1), identified peptide sequence identified, UniProt database primary accession number, start and end positions, database annotation of start position, NatA substrate (i.e. yNatA, hNatA and/or hNatA S37P substrate), number of identified spectra (per setup/ in total), N-terminal modification status/states confirmed by MS/MS (per setup/in total), % of Nt-acetylation (per setup), Uniprot name, protein description, gene symbols and, whenever a peptide matched to multiple members of a protein family (redundancy), isoforms are given for all uniquely identified yeast N-termini. N-termini are grouped according to their Swiss-Prot database start position and compliance with the rules of N-terminal acetylation and iMet processing and ranked alphabetically according to their protein description and start position.

## References

1. Helsens, K., Van Damme, P., Degroeve, S., Martens, L., Arnesen, T., Vandekerckhove, J., and Gevaert, K. (2011) Bioinformatics analysis of a *Saccharomyces cerevisiae* N-terminal proteome provides evidence of alternative translation initiation and post-translational N-terminal acetylation. *J Proteome Res* 10, 3578-3589