

# Predicting eukaryotic signal peptides using hidden Markov models

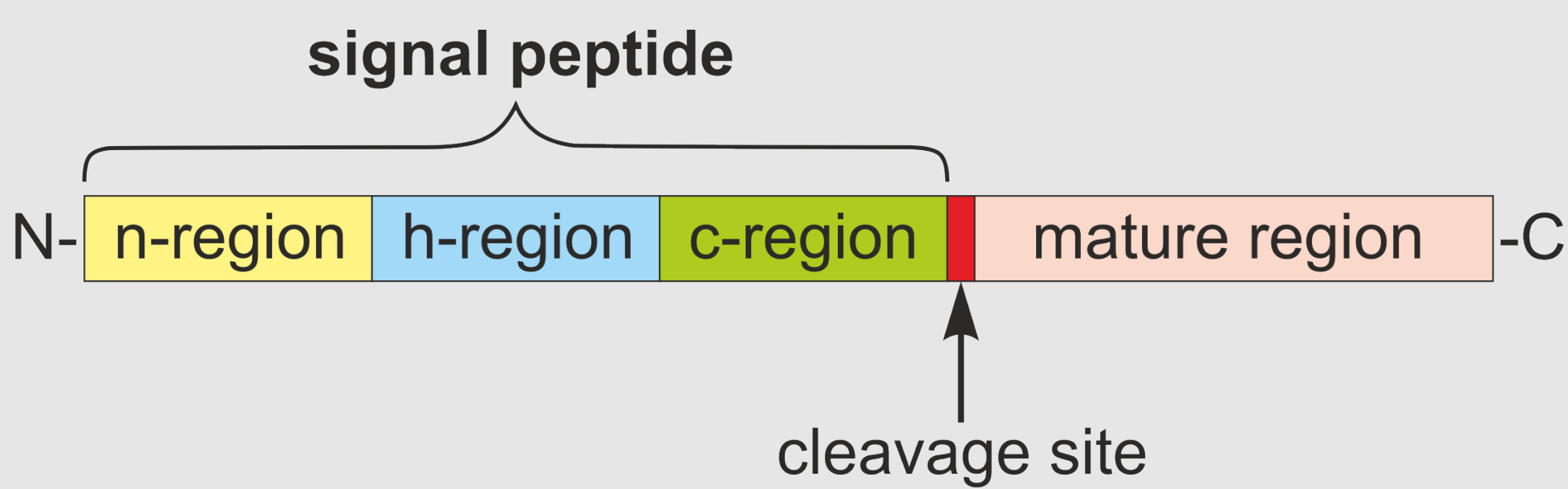
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## Introduction

- Secretory signal peptides:
- are short (20-30 residues) N-terminal amino acid sequences,
  - direct a protein to the endomembrane system and next to the extracellular localization,
  - possess three distinct domains with variable length and characteristic amino acid composition (Hegde and Bernstein, 2006).
  - are universal enough to direct properly proteins in different secretory systems; artificially introduced bacterial signal peptides can guide proteins in mammals (Nagano and Masuda, 2014) and plants (Moeller et al., 2009),
  - tag among others hormones, immune system proteins, structural proteins, and metabolic enzymes.

## Training of AmyloGram



- n-region: mostly basic residues (Nielsen and Krogh, 1998),
- h-region: strongly hydrophobic residues (Nielsen and Krogh, 1998),
- c-region: a few polar, uncharged residues (Jain et al., 1994).

## Hidden semi-Markov model (HSMM) of a signal peptide

- Assumptions of the model:
- the observable distribution of amino acids arises due to being in a certain region (state),
  - a duration of the state (the length of given region) is modeled by a probability distribution (other than geometric distribution as in typical hidden Markov models).

## Training of the signalHsmm algorithm

- Removal of atypical (non-standard amino acids, more than one cleavage site) or poorly annotated records from data set of proteins with signal peptide from UniProtKB 2014 07 (after purification data set contains 3816 eukaryotic proteins with experimentally confirmed signal peptides and 9795 without signal peptide),
- determination of n-, h-, c-regions by the heuristic algorithm,
- reduction of dimensionality by aggregating amino acids to several physicochemical groups,
- calculation of the amino acid group frequency in each region and the average length of the region,
- training of two HSMM models for proteins with and without signal peptide.

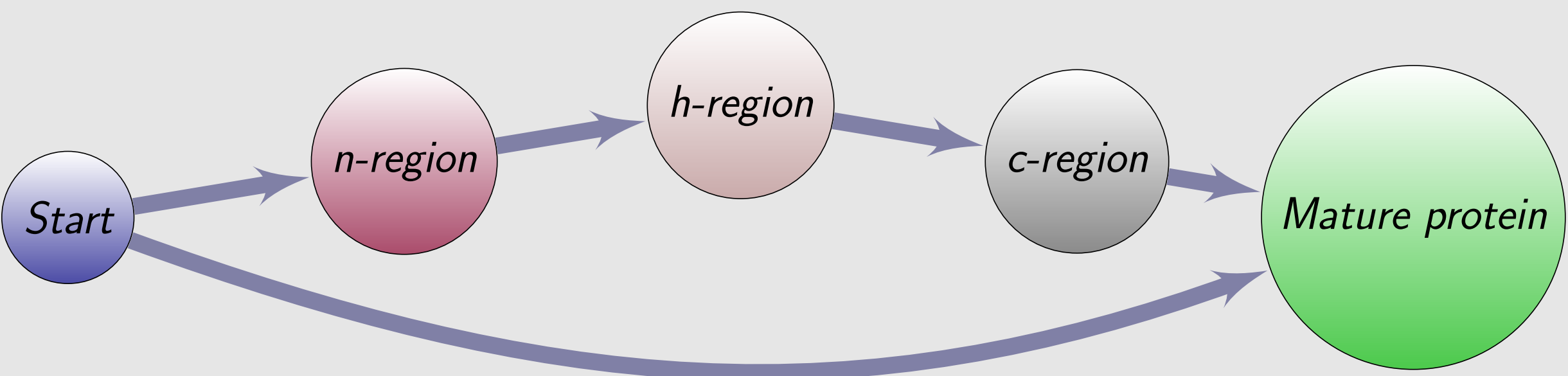
## Classification of amino acids used by signalHsmm

The encoding of amino acids with the best sensitivity.

Group	Amino acids
1	D, E, H, K, N, Q, R
2	G, P, S, T, Y
3	F, I, L, M, V, W
4	A, C

## Signal peptide prediction

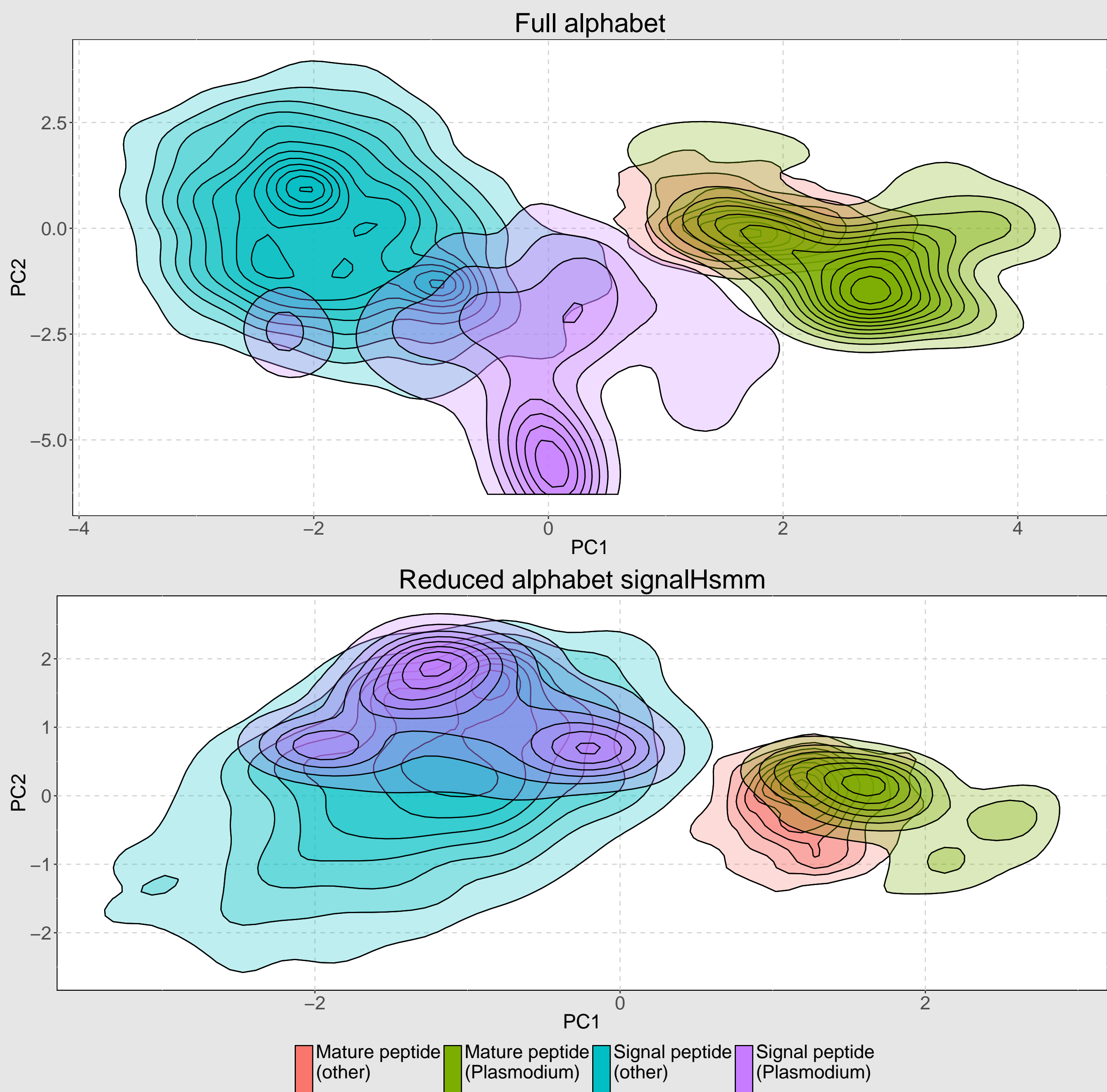
During the test phase, each protein is fitted to two HSMMs representing respectively proteins with and without signal peptides. The probabilities of both fits and predicted cleavage site constitute the software output.



## Validation procedure

- Choose randomly (without replacement) 3816 proteins without signal peptides, reshuffle 3816 proteins with signal peptides.
- Perform 5-fold cross-validation.
- Repeat step 1. and 2. 250 times.

## Classification of amino acids used by signalHsmm



## Benchmark with other predictors of signal peptides

Benchmark data set: 140 eukaryotic proteins with signal peptide and 280 randomly chosen eukaryotic proteins without signal peptide added after 2010.

signalHsmm1987: trained on data set of 496 eukaryotic proteins with signal peptides added before year 1987.

signalHsmm2010: trained on data set of 3676 eukaryotic proteins with signal peptides added before year 2010.

	Sensitivity	Specificity	MCC	AUC
signalP 4.1 (no tm)	0.8235	0.9100	0.6872	0.8667
signalP 4.1 (tm)	0.6471	0.9431	0.6196	0.7951
signalP 3.0 (NN) (Bendtsen et al., 2004)	0.8824	0.9052	0.7220	0.8938
signalP 3.0 (HMM) (Bendtsen et al., 2004)	0.6275	0.9194	0.5553	0.7734
PrediSi	0.3333	<b>0.9573</b>	0.3849	0.6453
Philius	0.6078	0.9336	0.5684	0.7707
Phobius	0.6471	0.9289	0.5895	0.7880
signalHsmm-2010	0.9804	0.8720	0.7409	0.9262
signalHsmm-2010 (hom. 50%)	<b>1.0000</b>	0.8768	<b>0.7621</b>	<b>0.9384</b>
signalHsmm-2010 (raw aa)	0.8431	0.9005	0.6853	0.8718
signalHsmm-1987	0.9216	0.8910	0.7271	0.9063
signalHsmm-1987 (hom. 50%)	0.9412	0.8768	0.7194	0.9090
signalHsmm-1987 (raw aa)	0.7647	0.9052	0.6350	0.8350

## Conclusions

Hidden semi-Markov models can be used to accurately predict the presence of secretory signal peptides effectively extracting information from very data sets. Prediction of cleavage site position still requires refinement.

## Availability and funding

signalHsmm web server:

[www.smorfland.uni.wroc.pl/signalhsmm](http://www.smorfland.uni.wroc.pl/signalhsmm)

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## Bibliography

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