signalHsmm: prediction of malarial signal peptides

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Signal peptides

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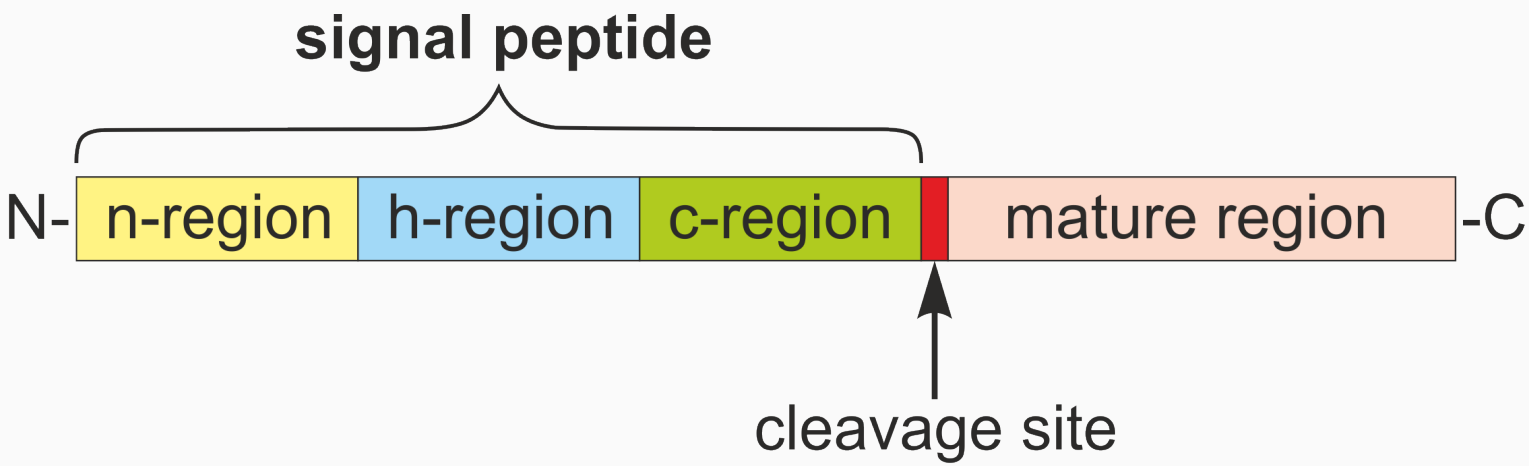
- tag hormons, immune system proteins, structural proteins, and metabolic enzymes.

Architecture



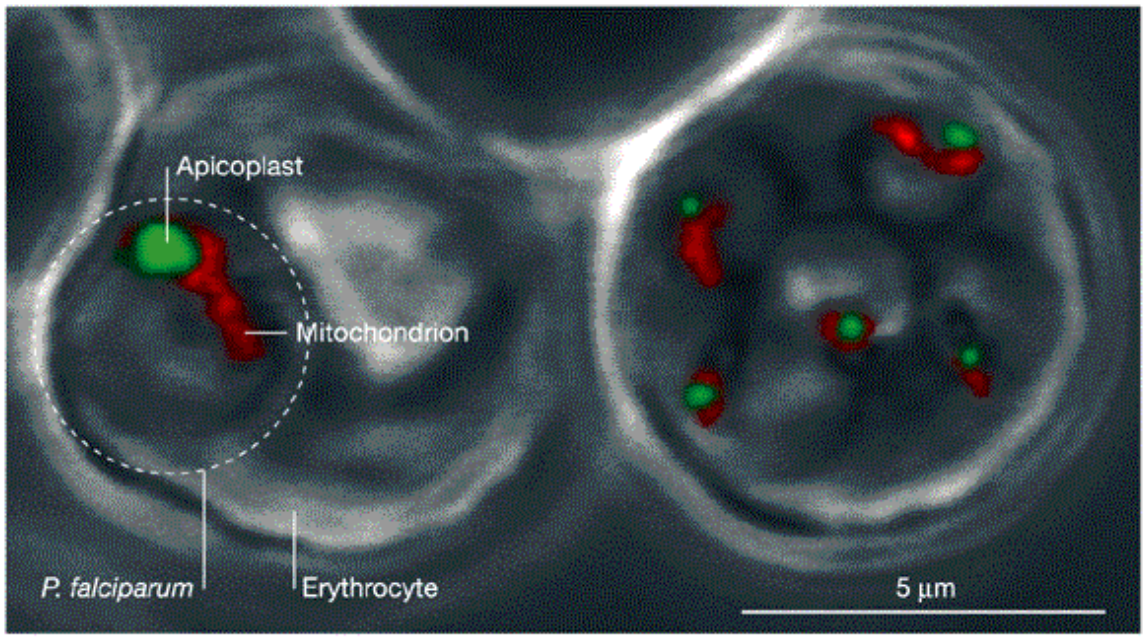
Signal peptides possess three distinct domains with variable length and characteristic amino acid composition (Hegde and Bernstein, 2006):

- 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).

Malarial signal peptides

Apicoplast

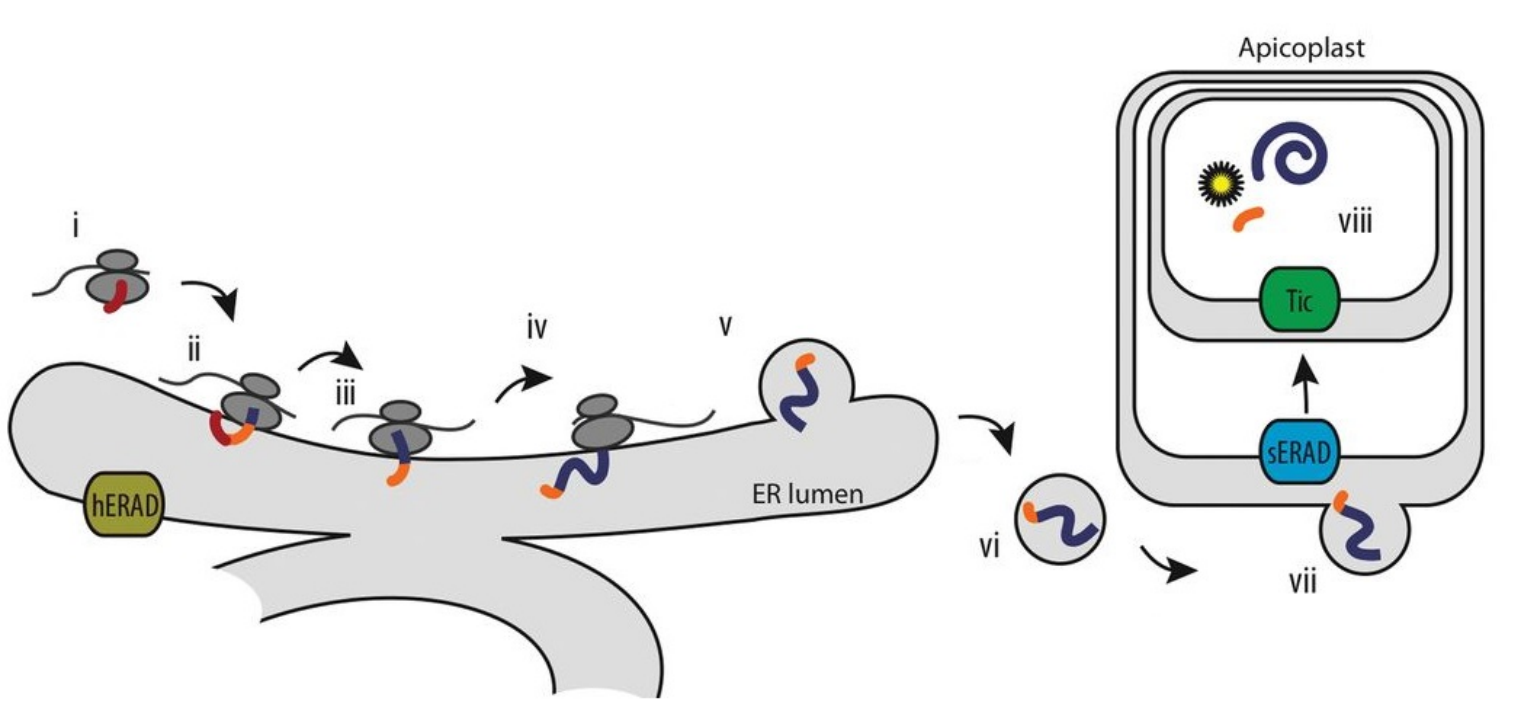
Four membrane-bounded plastid of Plasmodium sp. responsible for several biochemical pathways including the biosynthesis of fatty acids, isoprenoids and haem.



Ralph et al. (2004)

Bi-partite transit signal

The **signal peptide** is required for targeting the apicoplast protein to the endomembrane system, whereas the **transit peptide** is required to traffic the protein to the apicoplast.

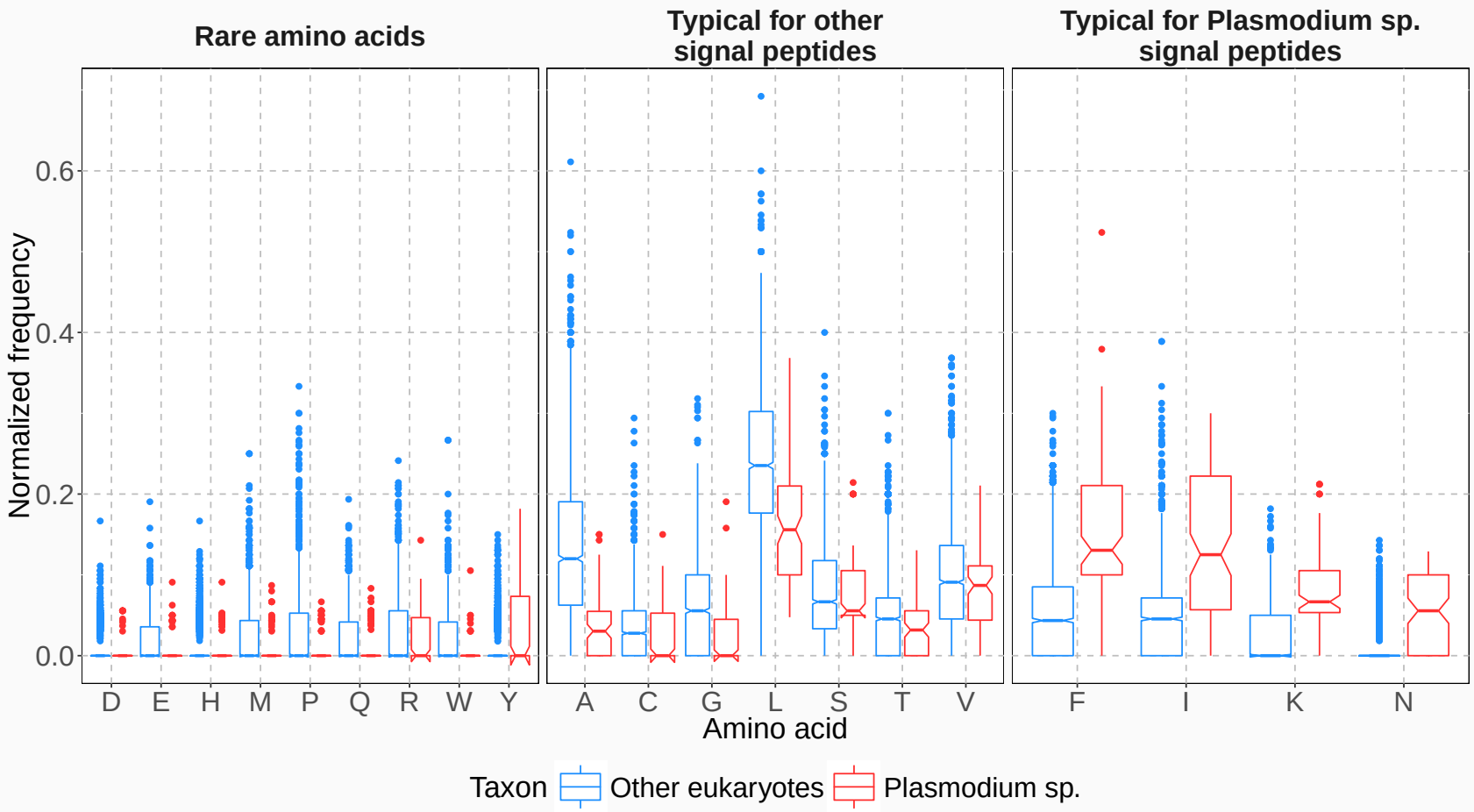


Kalanon and McFadden (2010)

Apicoplast

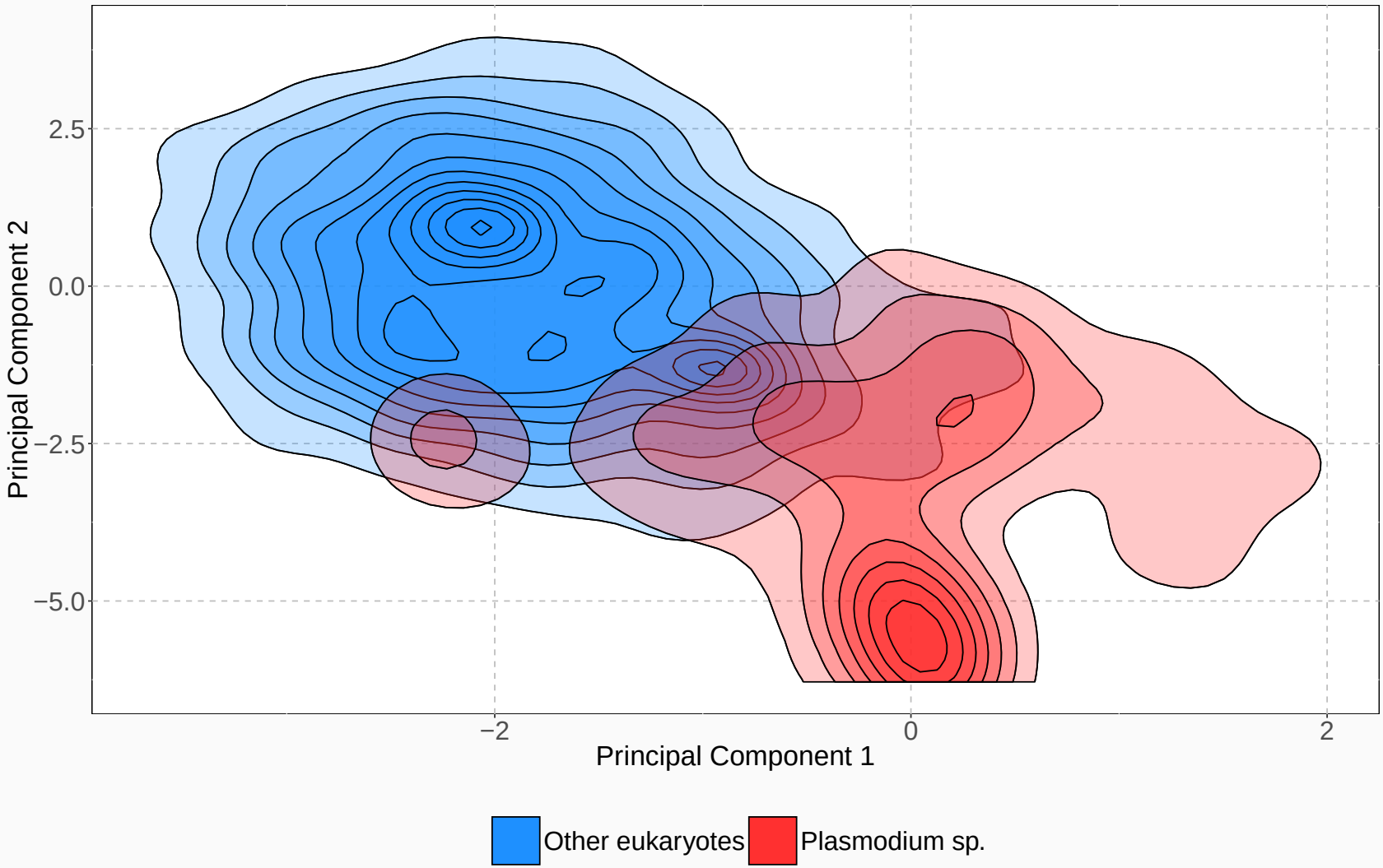
The absence of a metabolic counterpart in human host make the apicoplast proteins promising targets for anti-malarial drug development.

Heavy adenine-thymine bias of malarial genomes alters amino acid composition of malarial signal peptides making them hard to predict using software trained on other eukaryotes.



If notches are overlapping, two groups can be considered equal.

PCA of amino acid frequency



Since amino acid composition of signal peptides differ between Plasmodium sp. and other eukaryotes, predictors of signal peptides do not predict malarial signal peptides accurately.

There are no enough malarial signal peptides to train a specialized predictor.

Aim

Can we employ decision rules used for prediction of eukaryotic signal peptides to correctly detect malarial signal peptides?

Even nonbiological sequences can be effective signal peptides provided they fulfill general requirements (Tonkin et al., 2008).

NH_2-SKINNYSLINKYKINKYTHING-COOH - targets apicoplast.

NH_2-ITWILLNEVERTARGETPLASTID-COOH - does not target apicoplast.

Methods

Reduced amino acid alphabets

To date, several reduced amino acid alphabets have been proposed, which have been applied to (among others) protein folding and protein structure prediction.

Novel reduced amino acid alphabets

13 physicochemical properties handpicked from AAIndex database relevant to the regional architecture of signal peptides.

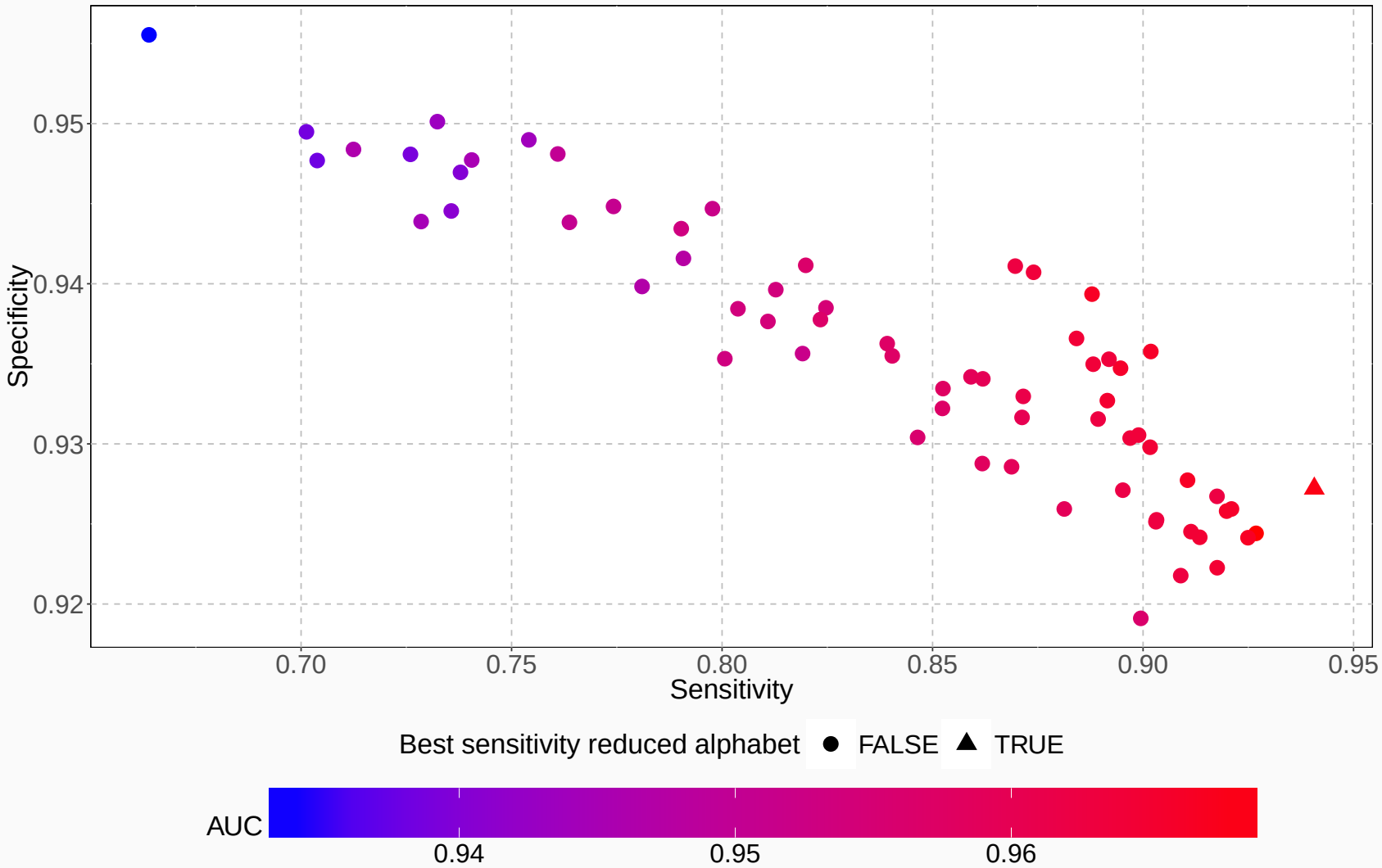
Property name	Amino acid scale
Size	Size
Size	Molecular weight
Size	Residue volume
Size	Bulkiness
Hydrophobicity	Normalized hydrophobicity scales for α-proteins
Hydrophobicity	Consensus normalized hydrophobicity scale
Hydrophobicity	Hydropathy index
Hydrophobicity	Surrounding hydrophobicity in α-helix
Polarity	Polarity
Polarity	Mean polarity
Occurrence in α-helices	Signal sequence helical potential
Occurrence in α-helices	Normalized frequency of N-terminal helix
Occurrence in α-helices	Relative frequency in α-helix

Cross-validation

We built 96 reduced amino acid alphabets (each based on one scale per a given property category) of length 4 (four distinct regions: n-, h-, c-region, mature protein).

Alphabets were evaluated in a cross-validation experiment using hidden semi-Markov models trained on eukaryotic sequences.

Results


Specificity
0.95
0.94
0.93
0.92
0.70
0.75
0.80
0.85
0.90
0.95
Sensitivity
Best sensitivity reduced alphabet
FALSE
TRUE
AUC
0.94
0.95
0.96

Group	Amino acids
I	D, E, H, K, N, Q, R
II	G, P, S, T, Y
III	F, I, L, M, V, W
IV	A, C

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I. Charged or uncharged but polar amino acids absent in h-region.

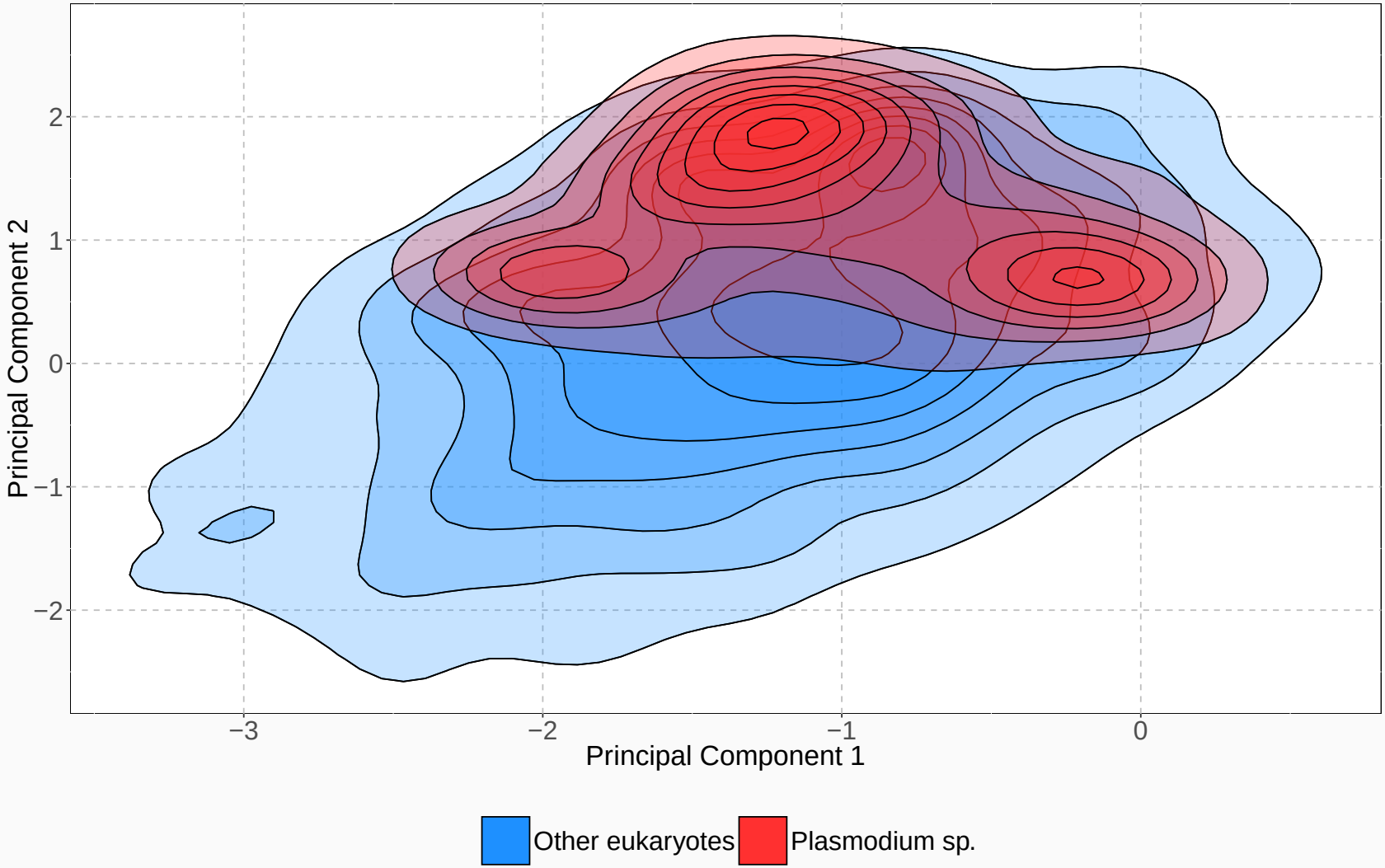
Group	Amino acids
I	D, E, H, K, N, Q, R
II	G, P, S, T, Y
III	F, I, L, M, V, W
IV	A, C

II. Polar and uncharged amino acids common in c-region.

Group	Amino acids
I	D, E, H, K, N, Q, R
II	G, P, S, T, Y
III	F, I, L, M, V, W
IV	A, C

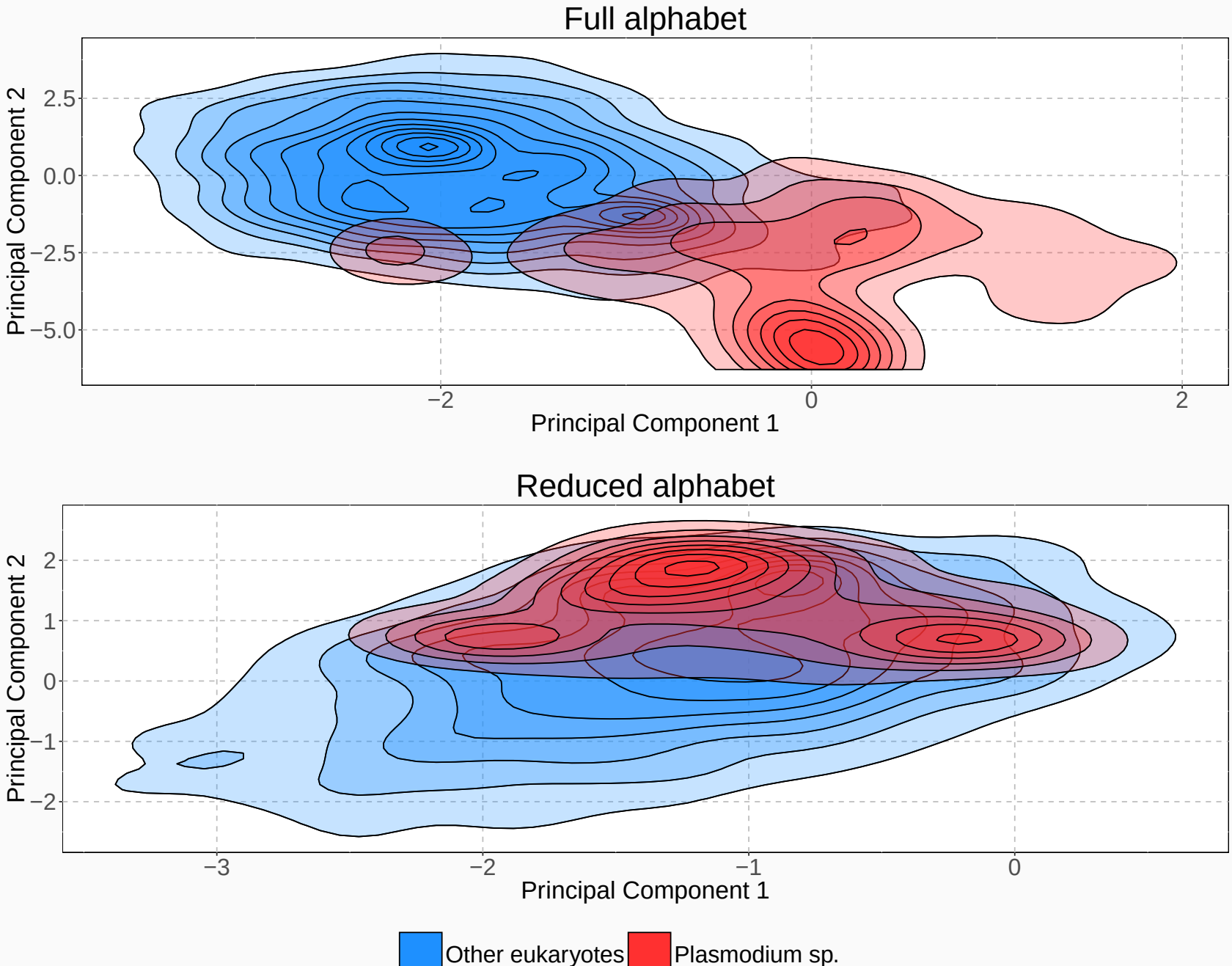
III. Hydrophobic amino acids common in h-region.

PCA of amino acid frequency



Signal peptides, after the reduction of the amino acid alphabet, group together despite their different origins.

PCA of amino acid frequency



Benchmark

Benchmark data set: 51 proteins with signal peptide and 211 proteins without signal peptide from members of *Plasmodiidae*.

Benchmark

	MCC	AUC
signalP 4.1 (no tm) (Petersen et al., 2011)	0.6872	0.8667
signalP 4.1 (tm) (Petersen et al., 2011)	0.6196	0.7951
signalP 3.0 (NN) (Bendtsen et al., 2004)	0.7220	0.8938
signalP 3.0 (HMM) (Bendtsen et al., 2004)	0.5553	0.7734
Phobius (Käll et al., 2004)	0.5895	0.7880
signalHsmm-2010	0.7409	0.9262
signalHsmm-2010 (hom. 50%)	**0.7621**	**0.9384**
signalHsmm-2010 (full alphabet)	0.6853	0.8718

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

Conclusions and perspectives

Eukaryotic signal peptides have very similar amino acid composition in their regions considering only the physicochemical properties of residues.

signalHsmm allows sensitive scanning of malarial proteome for potential drug targets.

Availability

signalHsmm web-server

`http://smorfland.uni.wroc.pl/shiny/signalHsmm`.

signalHsmm R package

`https://CRAN.R-project.org/package=signalHsmm`.

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- Paweł Mackiewicz.
- **biogram** package (`https://cran.r-project.org/package=biogram`):
  - Piotr Sobczyk,
  - Chris Lauber.

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