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Prediction of amyloidogenicity based on the n-gram analysis

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Abstract

Motivation: Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text.

Results: Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text

Availability: Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text

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Supplementary information: Supplementary data are available at *Bioinformatics* online.

1 Introduction

Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text. Figure 1 shows that the above method Text Text Text Text Text Text Text Text Text Text Text Text. (**?**) wants to know about …… text follows.

$$\sum x + y = Z \tag{1}$$

2 Approach

Equation (1) Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text. Figure 2 shows that the above method Text Text Text Text Text Text Text Text Text Text Text Text Text. **?** might want to know about text text text text .....

3 Methods

3.1 Data set

The data used in the study was extracted from AmyLoad data base (Wozniak and Kotulska, 2015). Aside from eight sequences shorter than five residues that were removed from the final data set, we obtained 418 amyloidogenic sequences and 1039 non-amyloidogenic sequences (1457 peptides in total).

Sequences shorter than 6 amino acids and longer than 25 amino acids were removed from data set. The former were too short to be processed in the devised analysis framework and the latter were too diversified and rare, preventing the proper analysis.

The final data set contains 397 amyloidogenic and 1033 non-amyloidogenic sequences (1430 peptides in total).

3.2 Encodings of amino acids

The amyloidogenicity of given peptide may not depend on the exact sequence of amino acids, but on its more general properties. To verify this hypothesis, we created 18 537 reduced amino acid alphabets (encodings) with different lengths (from three to six letters) using Ward's clusterization on the selected physicochemical properties from AAIndex database (Kawashima *et al.*, 2008). We handpicked several measures belonging to more general categories important in process of amyloidogenicity as size, hydrophobicity, solvent surface area, frequency in β-sheets and contactivity. As the rule of thumb, we limited ourselves to properties introduced after 1980 when, thanks to the technological advancements, the measurements were more accurate.

Since highly correlated or, contrarily, uncorrelated measures create very similar encodings, we further reduced the number of properties to 17 by selecting measures with the absolute value of Pearson's correlation coefficient for normalized values larger than 0.95.

3.3 Traning of learners

In addition to the reduced amino acid alphabets created through the clusterization of physicochemical properties, we also examined two reduced alphabets from the literature and amino acid alphabet of normal length.

Category	Property
Contactivity	Average flexibility indices (Bhaskaran and Ponnuswamy, 1988)
Contactivity	14 A contact number (Nishikawa and Ooi, 1986)
Contactivity	Accessible surface area (Radzicka and Wolfenden, 1988)
Contactivity	Buriability (Zhou and Zhou, 2004)
Contactivity	Values of Wc in proteins from class Beta, cutoff 12 A, separation 5 (Wozniak and Kotulska, 2014)
Contactivity	Values of Wc in proteins from class Beta, cutoff 12 A, separation 15 (Wozniak and Kotulska, 2014)
β-frequency	Average relative probability of inner beta-sheet (Kanehisa and Tsong, 1980)
β-frequency	Relative frequency in beta-sheet (Prabhakaran, 1990)
β-frequency	Thermodynamic beta sheet propensity (Kim and Berg, 1993)
Hydrophobicity	Hydrophobicity index (Argos *et al.*, 1982)
Hydrophobicity	Optimal matching hydrophobicity (Sweet and Eisenberg, 1983)
Hydrophobicity	Hydrophobicity-related index (Kidera *et al.*, 1985)
Hydrophobicity	Scaled side chain hydrophobicity values (Black and Mould, 1991)
Polarity	Polarizability parameter (Charton and Charton, 1982)
Polarity	Mean polarity (Radzicka and Wolfenden, 1988)
Size	Average volumes of residues (Pontius *et al.*, 1996)
Stability	Side-chain contribution to protein stability (kJ/mol) (Takano and Yutani, 2001)

Table 1. Physicochemical properties used during creation of reduced amino acid alphabets.

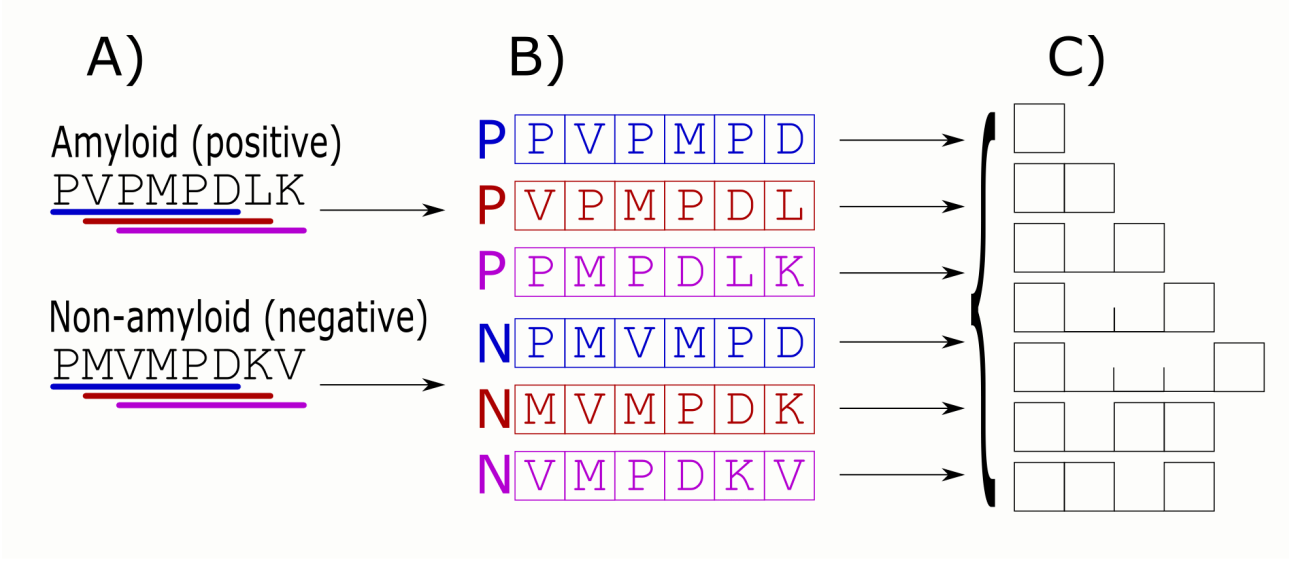

Fig. 1. Caption, caption.

During the training phase, we extracted overlapping hexamers from each sequence. Each hexamer was tagged with the same etiquette (amyloid/nonamyloid) as the original peptide. For example, the sequence of length 6 residues yields only one hexamer and the sequence of 8 residues yields 3 hexamers. Assuming that in longer amyloids only a short part of the sequence is responsible for amyloidogenicity, our method might results in many false positives in the training data set. To evade this problem, we restricted the maximum length of peptides in training data set to fifteen amino acids to easy the extraction of probable hot-spots.

The inquire further the length of amyloidogenicity signal, we trained nine classifiers for each encoding on the sequences of different length. We considered six-residue sequences, shorter of equal to ten residues and shorter or equal to fifteen residues. We specified separately length of peptides for negative and positive training set, obtaining in total nine ways of constructing training set for each reduced amino acids alphabet.

3.4 Cross-validation

The cross-validation was repeated five times for each combination of the encoding as well as the length of sequences in positive data set and negative data set.

Since we are interested if our classiffiers are able to use decision rules extracted from sequences of given length to correctly classify longer or shorter sequences, we calculate performance measures separately for four ranges of lengths of sequences:

- 6;
- 7-10;
- 11-15;
- 16-25.

The number of sequences from the given length range was roughly comparable between folds of cross-validation.

4 Discussion

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5 Conclusion

(Table **??**) Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text. Figure 2 shows that the above method Text Text Text Text Text Text Text Text Text Text Text Text. **?** might want to know about text text text text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text. Figure 2 shows that the above method Text Text Text Text Text Text Text Text Text Text Text. **?** might want to know about text text text text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text Text. Figure 2 shows that the above method Text Text Text Text Text Text Text Text Text Text Text Text.

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2. this is item, use enumerate
3. this is item, use enumerate

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References

Argos, P., Rao, J. K., and Hargrave, P. A. (1982). Structural prediction of membrane-bound proteins. *Eur. J. Biochem.*, **128**(2-3), 565–575.

Bhaskaran, R. and Ponnuswamy, P. (1988). Positional flexibilities of amino acid residues in globular proteins. *International Journal of Peptide and Protein Research*, **32**(4), 241–255.

Black, S. D. and Mould, D. R. (1991). Development of hydrophobicity parameters to analyze proteins which bear post- or cotranslational modifications. *Anal. Biochem.*, **193**(1), 72–82.

Charton, M. and Charton, B. I. (1982). The structural dependence of amino acid hydrophobicity parameters. *Journal of Theoretical Biology*, **99**(4), 629–644.

Kanehisa, M. I. and Tsong, T. Y. (1980). Local hydrophobicity stabilizes secondary structures in proteins. *Biopolymers*, **19**(9), 1617–1628.

Kawashima, S., Pokarowski, P., Pokarowska, M., Kolinski, A., Katayama, T., and Kanehisa, M. (2008). AAindex: amino acid index database, progress report 2008. *Nucl. Acids Res.*, **36**(suppl 1), D202–D205.

Kidera, A., Konishi, Y., Oka, M., Ooi, T., and Scheraga, H. A. (1985). Statistical analysis of the physical properties of the 20 naturally occurring amino acids. *J Protein Chem*, **4**(1), 23–55.

Kim, C. A. and Berg, J. M. (1993). Thermodynamic beta-sheet propensities measured using a zinc-finger host peptide. *Nature*, **362**(6417), 267–270.

Nishikawa, K. and Ooi, T. (1986). Radial locations of amino acid residues in a globular protein: correlation with the sequence. *J. Biochem.*, **100**(4), 1043–1047.

Pontius, J., Richelle, J., and Wodak, S. J. (1996). Deviations from Standard Atomic Volumes as a Quality Measure for Protein Crystal Structures. *Journal of Molecular Biology*, **264**(1), 121–136.

Prabhakaran, M. (1990). The distribution of physical, chemical and conformational properties in signal and nascent peptides. *Biochem. J.*, **269**(3), 691–696.

Radzicka, A. and Wolfenden, R. (1988). Comparing the polarities of the amino acids: side-chain distribution coefficients between the vapor phase, cyclohexane, 1-octanol, and neutral aqueous solution. *Biochemistry*, **27**(5), 1664–1670.

Sweet, R. M. and Eisenberg, D. (1983). Correlation of sequence hydrophobicities measures similarity in three-dimensional protein structure. *J. Mol. Biol.*, **171**(4), 479–488.

Takano, K. and Yutani, K. (2001). A new scale for side-chain contribution to protein stability based on the empirical stability analysis of mutant proteins. *Protein Eng.*, **14**(8), 525–528.

Wozniak, P. P. and Kotulska, M. (2014). Characteristics of protein residue-residue contacts and their application in contact prediction. *J Mol Model*, **20**(11).

Wozniak, P. P. and Kotulska, M. (2015). AmyLoad: website dedicated to amyloidogenic protein fragments. *Bioinformatics*.

Zhou, H. and Zhou, Y. (2004). Quantifying the effect of burial of amino acid residues on protein stability. *Proteins*, **54**(2), 315–322.