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Predicting microRNA precursors with a generalized Gaussian components based density estimation algorithm

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Abstract

Background: MicroRNAs (miRNAs) are short non-coding RNA molecules, which play an important role in post-transcriptional regulation of gene expression. There have been many efforts to discover miRNA precursors (pre-miRNAs) over the years. Recently, *ab initio* approaches have attracted more attention because they do not depend on homology information and provide broader applications than comparative approaches. Kernel based classifiers such as support vector machine (SVM) are extensively adopted in these *ab initio* approaches due to the prediction performance they achieved. On the other hand, logic based classifiers such as decision tree, of which the constructed model is interpretable, have attracted less attention.

Results: This article reports the design of a predictor of pre-miRNAs with a novel kernel based classifier named the generalized Gaussian density estimator (G²DE) based classifier. The G²DE is a kernel based algorithm designed to provide interpretability by utilizing a few but representative kernels for constructing the classification model. The performance of the proposed predictor has been evaluated with 692 human pre-miRNAs and has been compared with two kernel based and two logic based classifiers. The experimental results show that the proposed predictor is capable of achieving prediction performance comparable to those delivered by the prevailing kernel based classification algorithms, while providing the user with an overall picture of the distribution of the data set.

Conclusion: Software predictors that identify pre-miRNAs in genomic sequences have been exploited by biologists to facilitate molecular biology research in recent years. The G²DE employed

in this study can deliver prediction accuracy comparable with the state-of-the-art kernel based machine learning algorithms. Furthermore, biologists can obtain valuable insights about the different characteristics of the sequences of pre-miRNAs with the models generated by the G^2DE based predictor.

Background

MicroRNAs (miRNAs) are short RNAs (~20-22 nt) that direct post-transcriptional regulation of target genes by arresting the translation of mRNAs or by inducing their cleavage [1]. Since the initial discovery of miRNAs in *Caenorhabditis elegans*, RNA molecules are regarded as not only a carrier of gene information, but also a mediator of gene expression [2,3]. Currently, 9539 experimentally verified miRNAs have been collected in the miRBase database [4].

Experimental miRNA identification is accomplished by directional cloning of endogenous small RNAs [5]. Considering both the time and cost of experimental methods, many computational approaches have been proposed [6]. The mature miRNAs is cleaved from a 70-120 nt precursor (pre-miRNA) with a stable hairpin structure. Identifying this specific structure has become an important step in analyzing miRNAs [1]. The earliest computational approaches for discovering pre-miRNAs are mainly based on comparative techniques and can only discover pre-miRNAs that are closely homologous to known miRNAs [7-10]. Alternatively, scientists have resorted to *ab initio* approaches to discover pre-miRNAs based on the characteristics of their secondary structures [11-15]. The *ab initio* approaches based predictors are more generally applicable than those that are based on homology searches, since the *ab initio* approaches do not rely on the existence of homologues. As a result, design of the *ab initio* approaches based predictors has attracted more attention in recent years.

The basis of the *ab initio* approaches is to design a coding scheme that maps the sequence and structure characteristics of pre-miRNAs into distinguishable patterns of feature vectors. Then, a supervised learning algorithm, also commonly referred to as data classification, is invoked to discover pre-miRNAs in the query RNA sequence based on the associated feature vectors. In recent years, the design of the coding scheme for characterizing pre-miRNAs has been extensively studied and several different schemes, including base pairing propensity [16], folding energy [17], base pair distance [18], and degree of compactness [19], have been proposed. On the other hand, most people working on this subject have employed the existing kernel based data classification algorithms such as the hidden Markov model (HMM) [20,21], the support vector machine

(SVM) [22,23], and the kernel density estimator [15] to build the predictors due to the superior prediction performance delivered by these algorithms [24]. Nevertheless, conventional logic based data classification algorithms such as decision trees [25,26] and decision rules [27,28] continue to play a major role in some applications due to the *interpretability* of the logic rules identified by these algorithms. Such a summarized view of the characteristics and distribution of the data set further provides valuable insights about the relations among different features and is highly desirable for in-depth analysis of pre-miRNAs.

Aiming to provide the desirable functionalities of both the kernel based and the logic based data classification algorithms, the study presented in this article has exploited the generalized Gaussian density estimator (G^2DE) that we have recently proposed [29]. The G^2DE identifies a small number of generalized Gaussian components to model the distribution of the data set in the vector space. As a result, the user can examine the parameter values associated with these of Gaussian components to obtain an overview picture of the distribution of the data set. Furthermore, through in-depth analysis of the parameter values, the user can obtain valuable insights about the relations among different features.

Results and discussion

This section first describes the overall scheme of using G^2DE to analyze pre-miRNAs. Each step of the analysis procedure is further elaborated in the Methods section. Next, the prediction performance of the employed classification algorithm is evaluated and compared with four classification algorithms. A demonstrative analysis is also presented to investigate the interpretability of the employed classification algorithm.

Using G^2DE to analyze pre-miRNAs

This work uses only sequence information to identify pre-miRNAs from pseudo hairpins, which are RNA sequences with similar stem-loop features to pre-miRNAs but containing no mature miRNAs. In this method, each RNA sequence is represented as a feature vector. The characteristics used to generate the feature vector, including sequence composition, folding energy and stem-loop shape, have been shown to be useful for predicting pre-miRNAs in previous studies [17,18,30].

The main task carried out during the learning process of this method is to construct two mixture models of generalized Gaussian components for summarizing pre-miRNAs and pseudo hairpins in the vector space. We model this learning process as a large-parameter-optimization problem (LPOP) and employ an efficient optimization algorithm, Ranking-based Adaptive Mutation Evolutionary (RAME) [31], to decide the parameters associated with each generalized Gaussian component. Finally, the models learnt through the LPOP process are used to predict whether a query RNA sequence is a pre-miRNA. Furthermore, the constructed model of G²DE comprises a small number of generalized Gaussian components and is capable of detecting the sub-clusters or sub-classes of the data set. This study utilizes this feature of G²DE to develop a two-stage analysis where the first stage uses G²DE to partition the data set while the second stage uses G²DE to investigate each of the partitioned subsets.

Prediction performance

The present approach is evaluated using two datasets, HU920 and HU424, combined with four feature sets. See the Methods section for details of the datasets and the feature sets. The prediction performance is compared to two kernel based classifiers, SVM and RVKDE, and two logic based classifiers, C4.5 and RIPPER. The parameters for each classifier are determined by maximizing the prediction accuracy of ten-fold cross-validation on the HU920. A prediction is performed by using the HU920 dataset to predict the HU464 dataset with the selected parameters.

The employed G²DE classifier is compared with two kernel based classifiers, SVM and the relaxed variable kernel density estimator (RVKDE) [32]. The SVM is a commonly used classifier because of its prevailing success in diverse bioinformatics problems [14,33,34], while the RVKDE has been shown to have advantages for predicting species-specific pre-miRNAs [15]. Two well-

known logic based classifiers, C4.5 [35] and RIPPER [36], are also included as representatives of logic based classification algorithms.

As shown in Table 1, the prediction accuracies of one-stage G²DE are 80.39%, 92.03%, 91.60% and 78.66% with different feature sets. The two-stage G²DE further improve the prediction accuracies and achieves the best average accuracy of 86.58%. Though the number of kernels increases from one-stage to two-stage G²DE, it is still much less than the other kernel based classifiers. Table 1 also reveals that the kernel based classifiers generally outperform the logic based classifiers. As a result, the G²DE is capable of delivering satisfactory performance using a smaller number of kernel functions than the other systems.

In addition to compare the alternative classification algorithms, the prediction performance of the proposed method is also compared to two existing pre-miRNA identification packages, miPred [14] and miR-KDE [15]. A number (*nf*) of features from the four feature sets are selected with Wilcoxon rank sum test [37] are utilized as the feature set of G²DE. In current implementation, *nf* is set to seven because that the feature set yielding the best performance in Table 1 contains seven features. In this experiment, a prediction is performed by using the HU920 dataset to predict the HU464 dataset with the parameters maximizing the prediction accuracy of ten-fold cross-validation on the HU920 dataset. The five indices for binary classification (Table 2) used in miPred and miR-KDE are adopted. Table 3 shows the experimental results. G²DE achieves comparable performance with those delivered by miPred and miR-KDE. A notable difference between G²DE and miPred and miR-KDE is that G²DE utilizes much less kernels. G²DE-2 yields the best %ACC, %SE, %Fm and %MCC, which are 93.32%, 90.09%, 93.10% and 87.16%, respectively. Although the number of kernels in G²DE-2 is five times larger than that in G²DE, it is more acceptable to perform further analyses than that in miPred and miR-KDE.

Table 1: Prediction accuracies achieved by SVM, RVKDE, G²DE, C4.5 and RIPPER

Feature set	Kernel based classifiers				Logic based classifiers	
	SVM	RVKDE	G ² DE	G ² DE-2	C4.5	RIPPER
1	80.17%	77.59%	80.39%	80.60%	77.80%	76.72%
2	93.32%	92.46%	92.03%	93.10%	90.95%	90.52%
3	91.60%	91.16%	91.60%	92.46%	91.16%	91.38%
4	78.66%	79.53%	78.66%	80.17%	77.37%	76.72%
Average	85.94%	85.18%	85.67%	86.58%	84.32%	83.84%
#kernels	361	920	6	36	10	9

The best performance among each feature set is highlighted with bold font. The G²DE-2 indicates the two-stage G²DE, which uses the first stage G²DE to cluster samples and then uses the second stage G²DE to classify each cluster. The #kernels indicate number of kernels in average, where the numbers of logic based classifiers indicate the number of rules they deliver.

Table 2: Evaluation measures employed in this study

Measure	Abbreviation	Equation
Sensitivity (recall)	%SE	$TP/(TP+FN)$
Specificity	%SP	$TN/(TN+FP)$
Accuracy	%ACC	$(TP+TN)/(TP+TN+FP+FN)$
F-measure	%Fm	$2TP/(2TP+FP+FN)$
Matthews' correlation coefficient	%MCC	$(TP \times TN - FP \times FN) / \sqrt{(TP+FP) \times (TN+FN) \times (TP+FN) \times (TN+FP)}$

The definition of the abbreviations used: TP is the number of real pre-miRNAs detected; FN is the number of real pre-miRNAs missed; TN is the number of pseudo hairpins correctly classified; and FP is the number of pseudo hairpins incorrectly classified as pre-miRNA.

Table 3: Comparison of G²DE and two existing pre-miRNA identification packages

Method	#kernels	%SE	%SP	%ACC	%Fm	%MCC
miPred	280	88.80%	96.55%	92.67%	92.38%	85.60%
miR-KDE	920	89.22%	96.12%	92.67%	92.41%	85.55%
G ² DE	6	87.07%	97.84%	92.46%	92.03%	85.41%
G ² DE-2	36	90.09%	96.55%	93.32%	93.10%	87.16%

The best performance among each evaluation index is highlighted with bold font. The G²DE-2 indicates the two-stage G²DE, which uses the first stage G²DE to cluster samples and then uses the second stage G²DE to classify each cluster.

Interpretability of G²DE

Though the two-stage G²DE achieves the best performance in Table 1, the small differences to other classifiers suggests that pre-miRNA prediction algorithms have reached the maximum with current feature sets. Hence, how to interpret the learnt model of machine learning techniques for users is crucial in pre-miRNA prediction. In this subsection, the second feature set is used as an example to explain how to interpret the models generated by the G²DE based predictor. Figure 1 shows the parameters associated with the three Gaussian components used to summarize the pre-miRNAs in the HU920 dataset. To analyze these parameters, we compare them to the Pearson product-moment correlation coefficients (PMCC) [38] of the pre-miRNAs in the HU920 dataset (Figure 2). Parameters in the models generated by G²DE that differ more from the corresponding elements obtained by PMCC are more of our interest. For instance, in Figure 2, the correlation between the first feature (*mfe2*) and the fifth feature (*dQ*) of this feature set is 0.36. See the 'Feature set' subsection for detailed explanations of these features. On the other hand, the correlations between the two features in the three Gaussian components (shown in Figure 1) are 0.12, 0.08 and 0.02, all of which are relatively lower than 0.36. As PMCC summarizes the distribution of all pre-miRNAs, this analysis suggests that the HU920 dataset is composed of multiple clusters of samples, where the relation between *mfe2* and *dQ* varies in different clusters and causes the inconsistency of correlations.

To verify the above analysis, this study depicts the HU920 dataset with their *mfe2* and *dQ* values (Figure 3).

In Figure 3, the red squares and green circles represent the pre-miRNAs and the pseudo hairpins, respectively. The red ellipses, named GGC₁, GGC₂ and GGC₃, are the generalized Gaussian components shown in Figure 1, and the black ellipse is the Gaussian distribution shown in Figure 2. Figure 3 reveals that there are potentially two clusters of pre-miRNAs in this dataset and form a shape of a mirrored 'L' in the feature space of *mfe2* and *dQ*. *mfe2* measures the energy of folding while *dQ* measures the arrangement of base pairing. Table 4 shows the detailed descriptions of these features. Figure 3 suggests that if a RNA sequence has low folding energy, it is probably a pre-miRNA regardless of the arrangement of its base pairing. On the other hand, there is another cluster of pre-miRNAs that have similar folding energies to those of pseudo hairpins. No obvious correlation exists in both clusters of pre-miRNAs. These findings coincide with the analysis based on the models generated by G²DE. In this example, the Gaussian components learnt by G²DE help users to identify features of interest without plotting all pairs of features along with the relations between them.

Another useful analysis provided by the learnt model of the G²DE based predictor is sub-class detection. By defining that a sample *belongs* to the Gaussian component reporting the maximum function value at the location of that sample, the learnt Gaussian components of G²DE suggests that "a sample that belongs to GGC₂ is a pre-miRNA." This statement is similar to the normal decision rule "a sample that has *mfe2* < 0.4 is a pre-miRNA," except that the conditions (belong vs. *mfe2* < 0.4) within the rule inferred by G²DE is non-linear. This non-linearity of a single rule is an important feature of

G²DE to describe more complicated model than traditional logic based classifiers when the number of kernels of G²DE is limited to the number of rules of logic based algorithms. One immediate application of the sub-class detection is to group samples into clusters and then construct a classifier for each of the clusters. The performance improved by applying such two-stage framework has been shown in the previous subsection.

Generalized Gaussian component 1 (GGC₁):

Mean = [0.58, 0.27, 0.82, 0.18, -0.55, -0.93, -0.37];

Std. = [0.78, 0.63, 0.84, 0.67, 0.60, 0.43, 0.81]

$$\text{Corr. matrix} = \begin{bmatrix} 1.00 & 0.15 & -0.66 & 0.56 & 0.12 & 0.05 & 0.41 \\ 0.15 & 1.00 & 0.52 & -0.43 & -0.06 & -0.80 & 0.44 \\ -0.66 & 0.52 & 1.00 & -0.53 & -0.17 & -0.56 & 0.03 \\ 0.56 & -0.43 & -0.53 & 1.00 & 0.00 & 0.42 & 0.42 \\ \blacktriangleright 0.12 & -0.06 & -0.17 & 0.00 & 1.00 & -0.04 & -0.07 \\ 0.05 & -0.80 & -0.56 & 0.42 & -0.04 & 1.00 & -0.29 \\ 0.41 & 0.44 & 0.03 & 0.42 & -0.07 & -0.29 & 1.00 \end{bmatrix}$$

Weight: 0.826

Generalized Gaussian component 2 (GGC₂):

Mean = [0.09, -0.21, 0.73, -0.19, 0.09, -0.70, 0.28];

Std. = [0.14, 0.79, 0.47, 0.19, 0.76, 0.81, 0.50];

$$\text{Corr. matrix} = \begin{bmatrix} 1.00 & 0.04 & -0.43 & -0.12 & 0.08 & -0.34 & 0.42 \\ 0.04 & 1.00 & -0.01 & 0.22 & -0.42 & -0.03 & -0.33 \\ -0.43 & -0.01 & 1.00 & 0.14 & 0.55 & -0.09 & 0.28 \\ -0.12 & 0.22 & 0.14 & 1.00 & 0.33 & 0.29 & -0.10 \\ \blacktriangleright 0.08 & -0.42 & 0.55 & 0.33 & 1.00 & -0.09 & 0.71 \\ -0.34 & -0.03 & -0.09 & 0.29 & -0.09 & 1.00 & -0.69 \\ 0.42 & -0.33 & 0.28 & -0.10 & 0.71 & -0.69 & 1.00 \end{bmatrix}$$

Weight: 0.488

Generalized Gaussian component 3 (GGC₃):

Mean = [-0.04, -0.49, 0.24, 0.36, -0.40, 0.62, 0.73];

Std. = [0.66, 0.93, 0.91, 0.49, 0.43, 0.73, 0.33];

$$\text{Corr. matrix} = \begin{bmatrix} 1.00 & 0.62 & -0.76 & 0.81 & 0.02 & -0.16 & -0.75 \\ 0.62 & 1.00 & -0.84 & 0.58 & -0.41 & -0.06 & -0.61 \\ -0.76 & -0.84 & 1.00 & -0.76 & 0.45 & 0.07 & 0.57 \\ 0.81 & 0.58 & -0.76 & 1.00 & -0.27 & 0.15 & -0.56 \\ \blacktriangleright 0.02 & -0.41 & 0.45 & -0.27 & 1.00 & -0.43 & -0.16 \\ -0.16 & -0.06 & 0.07 & 0.15 & -0.43 & 1.00 & 0.54 \\ -0.75 & -0.61 & 0.57 & -0.56 & -0.16 & 0.54 & 1.00 \end{bmatrix}$$

Weight: 0.545

Figure 1
Parameters of the three generalized Gaussian components generated by G²DE. This figure shows the three generalized Gaussian components of G²DE with the pre-miRNAs in the HU920 dataset and the second feature set. The correlation of interest is indicated with an arrow.

Mean = [0.63, 0.18, 0.24, 0.01, -0.71, -0.64, -0.61];

Std. = [0.26, 0.35, 0.31, 0.33, 0.23, 0.27, 0.30]

$$\text{Corr. matrix} = \begin{bmatrix} 1.00 & 0.63 & -0.42 & 0.67 & 0.36 & 0.37 & -0.90 \\ 0.63 & 1.00 & -0.62 & 0.47 & 0.55 & 0.54 & -0.56 \\ -0.42 & -0.62 & 1.00 & -0.59 & -0.48 & -0.45 & 0.20 \\ 0.67 & 0.47 & -0.59 & 1.00 & 0.38 & 0.37 & -0.32 \\ \blacktriangleright 0.36 & 0.55 & -0.48 & 0.38 & 1.00 & 0.99 & -0.28 \\ 0.37 & 0.54 & -0.45 & 0.37 & 0.99 & 1.00 & -0.30 \\ -0.90 & -0.56 & 0.20 & -0.32 & -0.28 & -0.30 & 1.00 \end{bmatrix}$$

Figure 2
Parameters obtained by basic statistics. These parameters are obtained by calculating the mean, standard deviation and Pearson product-moment correlation coefficients with the pre-miRNAs of the HU920 dataset and the second feature set. The correlation of interest is indicated with an arrow.

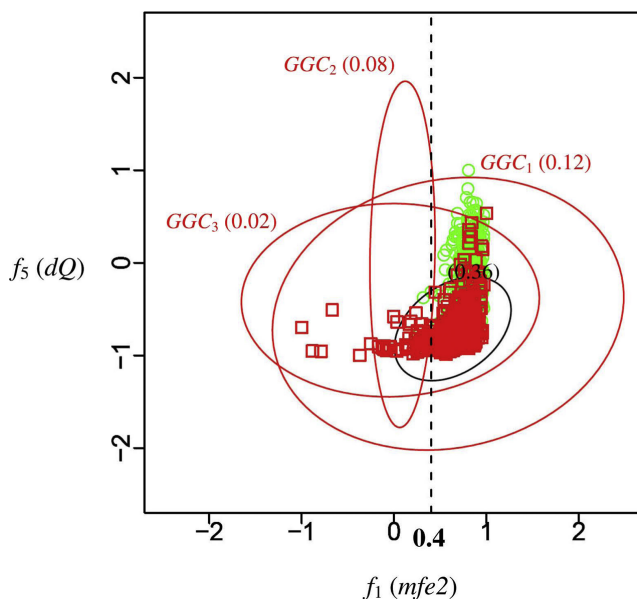


Figure 3
Distribution of the HU920 dataset. The x-axis is the first feature of the second feature set, ratio of MFE to the number of stems; the y-axis is the fifth feature of the second feature set, adjusted Shannon entropy. Red ellipses represent the generalized Gaussian components shown in Figure 1; the black ellipse represents the Gaussian component shown in Figure 2. The red squares and green circles represent the pre-miRNAs and the pseudo hairpins, respectively. Values within the parentheses indicate the correlations between these two features in the corresponding Gaussian components.

Table 4: Summary of the adopted feature sets

Feature	Description
Set 1	
AA, AC, ..., UU	Frequencies of 16 dinucleotide pairs
%G+C	Percentage of nitrogenous bases which are either G or C
Set 2	
<i>mfe2</i>	Ratio of <i>dG</i> to the number of stems
<i>mfe1</i>	Ratio of <i>dG</i> to %G+C
<i>dP</i>	Adjusted base pairing propensity. <i>dP</i> is the number of base pairs observed in the secondary structure divided by the sequence length.
<i>dG</i>	Adjusted minimum free energy of folding. <i>dG</i> is the minimum free energy (MFE) divided by the sequence length.
<i>dQ</i>	Adjusted Shannon entropy. <i>dQ</i> measures the entropy of the base pairing probability distribution (BPPD).
<i>dD</i>	Adjusted base pair distance. <i>dD</i> measures the average distance between all base pairs of structures inferred from the sequence.
<i>dF</i>	Compactness of the tree-graph representation of the sequence.
Set 3	
<i>zG, zQ, zD, zP, zF</i>	5 normalized variants of <i>dP, dG, dQ, dD</i> and <i>dF</i>
Set 4	
<i>IH</i>	Hairpin length
<i>IL</i>	Loop length
<i>IC</i>	Consecutive base-pairs
<i>%L</i>	Ratio of loop length to hairpin length

The table shows the order of a feature within the feature set. For example, the fifth feature in the second feature set is *dQ*.

Conclusion

Software predictors that identify pre-miRNAs in genomic sequences have been exploited by biologists to facilitate molecular biology research in recent years. However, design of advanced predictors of pre-miRNAs has focused mostly on coding the distinguishable sequence as well as structure characteristics of miRNAs. The study presented in this article addresses this issue from the aspect of exploiting advanced machine learning algorithms. The G²DE employed in this study has been designed to deliver prediction accuracy comparable with the state-of-the-art kernel based machine learning algorithms, while providing the user with good interpretability. As demonstrated by the experiments reported in this study, the models generated by the G²DE based classifier provide the user with crucial clues about the different characteristics of the sequences of pre-miRNAs.

Methods

Feature set

This work adopts 33 characteristic features which have been shown to be useful for miRNA detection in previous studies [16-19,30,39-41]. To investigate how alternative classifiers perform when using different features, these features are grouped as four different sets according to their biochemical properties. The first feature set includes 17 sequence composition variables, which comprise frequencies of 16 dinucleotide pairs and proportion of G and C in the RNA molecule. The second feature set includes seven folding measures: Minimum Free Energy (MFE) and two of its variants [17,18,39], base pairing propensity [16], Shannon entropy [18], base

pair distance [18,40] and degree of compactness [19,41]. The third feature set uses the Z-score [42] to normalize the features in the second feature set except the two MFE variants. The fourth feature set includes four stem-loop features: hairpin length [15], loop length [15], consecutive base-pairs [15] and the ratio of loop length to hairpin length [15]. Table 4 shows a summary of these features.

Dataset

The process of data preparation is the same as that in the compared pre-miRNA identification packages [14,15] for a fair comparison. 692 human miRNA precursors are collected from the miRBase registry database [43] (release 12.0) as the positive set. For the negative set, 8494 pseudo hairpins collected from the protein-coding regions (CDSs) according to RefSeq [44] and UCSC refGene [45] annotations are analyzed. These RNA sequences are extracted from genomic regions where no experimentally validated splicing event has been reported [12]. The secondary structures of the 8494 RNA sequences are obtained by executing RNAfold [46]. RNA sequences with <18 base pairs on the stem, MFE > -25 kcal/mol and multiple loops are excluded. As a result, 3988 pseudo hairpins, which are similar to genuine pre-miRNAs in terms of length, stem-loop structure, and number of bulges but not have been reported as pre-miRNAs, are used as the negative set.

Based on the positive and negative sets, one training set and one testing set are built to evaluate the pre-miRNA predictors. The training set, HU920, comprises 460

human pre-miRNAs and 460 pseudo hairpins randomly selected from the positive and negative sets, respectively. The HU920 dataset is used for parameter selection and model construction of the pre-miRNA predictors. The testing set, HU464, comprises the remaining 232 human pre-miRNAs and 232 randomly selected pseudo hairpins. Care has been taken to ensure that no pseudo hairpin is included in both datasets. Before performing any experiments on these datasets, the features are rescaled linearly by the svm-scale program [47] to the interval of [-1.0, 1.0].

Generalized Gaussian density estimator

This work transforms samples into feature vectors and then uses them to construct a generalized Gaussian density estimator (G²DE) [29]. A density estimator is in fact an approximate probability density function. With the G²DE algorithm, one approximate probability density function of the following form is generated for each class of samples:

$$\hat{f}(\mathbf{v}) = \frac{1}{\sum w_i} \sum \frac{1}{(2\pi)^{d/2}} \text{GGC}(w_i, \boldsymbol{\mu}_i, \boldsymbol{\Sigma}_i) \text{ and}$$

$$\text{GGC}(w_i, \boldsymbol{\mu}_i, \boldsymbol{\Sigma}_i) = w_i \frac{1}{|\boldsymbol{\Sigma}_i|^{1/2}} \exp\left(-\frac{1}{2}(\mathbf{v} - \boldsymbol{\mu}_i)^T \boldsymbol{\Sigma}_i^{-1}(\mathbf{v} - \boldsymbol{\mu}_i)\right), \tag{1}$$

where d is the dimension of the vector space and w_i , $\boldsymbol{\mu}_i$, and $\boldsymbol{\Sigma}_i$ are the weight, mean vector, and the covariance matrix of the i -th Gaussian component. Let \hat{f}_j denote the approximate probability density function for the j -th class of samples. Then, a query sample located at \mathbf{v} is predicted to belong to the class of which the corresponding likelihood function defined in the following gives the maximum value:

$$L_h(\mathbf{v}) = \frac{|S_h| \cdot \hat{f}_h(\mathbf{v})}{\sum_j |S_j| \cdot \hat{f}_j(\mathbf{v})},$$

where S_j is the set of class- j training samples and $|S_j|$ is the number of class- j training samples.

In current G²DE implementation, the user can specify the maximum number of generalized Gaussian components that can be incorporated to generate the approximate probability density function for one class of samples. If the user sets this number to k and the total number of features of the data set is d , then the learning algorithm of G²DE needs to figure out the optimal combination of the values of the following $k(d+2)(d+1)/2$ parameters in order to generate one approximate probability density function: k d -dimensional vectors as the means of the generalized Gaussian components; k sets of $d(d+1)/2$ coefficients with

each corresponding to the covariance matrix of one generalized Gaussian components; k weights. The optimal combination of parameter values are figured out using the Ranking-based Adaptive Mutation Evolutionary (RAME) algorithm [31].

In the evolutionary optimization algorithm, the objective function to be maximized is as follows:

$$O(\mathbf{Z}) = \# \text{Correct} + \theta \cdot \sum_j \log(\text{likelihood of class } j) \tag{2}$$

where

- (1) \mathbf{Z} is the vector formed by concatenating all the $k(d+2)(d+1)/2$ parameters associated with the approximate probability density function of one class of samples;
- (2) $\# \text{Correct}$ is the number of correctly classified training samples;
- (3) θ is a user-defined parameter;
- (4) likelihood of class $j = \prod_{\mathbf{s}_i \in \text{class } j} \hat{f}_j(\mathbf{s}_i)$, where \mathbf{s}_i is the i -th training sample of class j .

The objective function adopted in the learning process of G²DE consists of two terms. Both terms have specific mathematical meanings. Maximizing term $\# \text{Correct}$ implies that the number of training samples of which the class can be correctly predicted with the decision model is maximized. Meanwhile, maximizing the second term implies that the mixture models give the maximum likelihood with the training samples.

Two-stage G²DE

The learnt model of G²DE is composed of a small number of Gaussian components. In this study, a sample is defined as *belonging* to the Gaussian component reporting the maximum function value at the location of that sample. The two-stage classification framework is performed by first grouping samples belonging to the same Gaussian component into clusters and then constructing a classifier for each of the clusters. In the first stage, all training samples would be submitted to G²DE for constructing a mixture model of generalized Gaussian components. Suppose that the learnt model contains n_1 generalized Gaussian components, essentially dividing the training dataset into n_1 clusters. Each training sample would then be assigned to the Gaussian component to which it belongs. In the second stage, G²DE is invoked n_1 times to construct n_1 mixture models for each cluster. If each of the learnt models in the second stage contains n_2 generalized Gaussian components, the final classifier will contain $n_1 \times n_2$ generalized Gaussian components.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

Author C.-H. Hsieh performed all calculations and analysis and drafted the manuscript. Author D. T.-H. Chang aided in design of the methodology, interpretation of the data and manuscript preparation. Author C.-H. Hsueh and C.-Y. Wu participated in the data preparation. Author Y.-J. Oyang conceived the design of G²DE classifier. All authors have read and approved this manuscript.

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