Extracting and Evaluating Features from RNA Virus Sequence to Predict Host Species Susceptibility Using Deep Learning

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by

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  - **Computational techniques** could be used to **narrow down** possible hosts

Motivation (contd)

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Kevin Sutanto.
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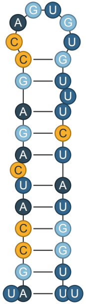
Master of Computer Science, University of Ottawa, School of Electrical Engineering and Computer Science,
2021.

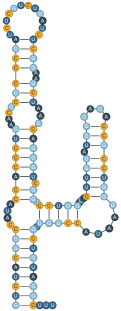
Related Work

- **Deep learning** has been used to identify:
  - **Viruses** from alignment-free metagenomic data [9]
  - **Interactions** between viral and host **proteins** [10]
  - **Hosts** for sequences of “influenza A”, “rabies lyssavirus” and “rotavirus A” [11]
- Data utilized in **prior host identification studies**:
  - **Sequences** of the viruses themselves [11, 12]
  - **Encoded viral proteins** [13]
  - **K-mers** [14, 15]

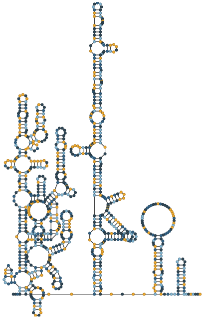
RNA Secondary Structure



piRNA
30 nt, piR-40447



5S Ribosomal RNA
121 nt, CRW V00589



16S Ribosomal RNA
954 nt, CRW J01415

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- **Deep learning**

Methods Overview

- **Features:**
  - **K-mers**
  - **Skip-mers** [21]
  - **Secondary structure fingerprints** [22]
- **Deep learning**
- **Dataset** and filtering

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    - *Match 2 skip 1* (e.g. AC*GT*) with *length of 6, 7, and 9*.

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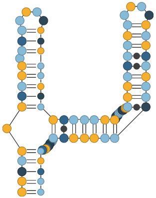
- **RNAMotif** [23] was used to **find and match** secondary structures
- **Circumvent issues** associated with the prediction of **RNA secondary structure**

Related Work Using Secondary Structure

⇒



⇒ **MoSS** ⇒ **feature vector**

Fiannaca, A., Rosa, M. L., Paglia, L. L., Rizzo, R. & Urso, A. nRC: non-coding RNA Classifier based on structural features. *BioData Mining* 10, (2017)

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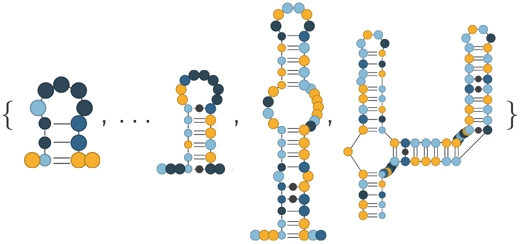
RNA Secondary Structures Fingerprints



$\Rightarrow$ **RNAMotif** $\Rightarrow$ **feature vector**

Deep Learning

- **For each feature set**, 3 different network architectures:
  - 2 consecutive relu-activated dense layers + a softmax-activated dense layer (total depth = 3);
  - 3 consecutive relu-activated dense layers + a softmax-activated dense layer (total depth = 4); and
  - 4 consecutive relu-activated dense layers + a softmax-activated dense layer (total depth = 5).
- The **best performance** among the 3 = performance of the **feature set**.
- **Width of each layer = number of values in the feature set being used**
  - e.g. 256 for 4-mer

Deep Learning

- **10-fold validation** [24] was used
  - **Each fold:** 90% training, 10% evaluation data
  - Splitting into folds takes **class balance** into account
- **Adam** [25] optimizer, **sparse categorical crossentropy** loss
- **300 epochs** for training
- Starting **learning rate** = 0.001, **decay** by 50% every 100 epochs

Dataset

- **RNA virus sequences** and **their host species**
- From **NCBI Virus** [26] as of September 12, 2020
- **Filtering** – the following were **excluded**:
  - Entries with **partial** sequences only
  - Entries which sequence length **exceeds** 40,000
  - Sequences with **unknown nucleotides and/or host species**
  - Hosts with $<$ 100 entries
- **47,266 entries**

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    - Further investigated in a subsequent study [27].

Thank **you**!

Availability

Dataset with the **secondary structure fingerprints** is available at:

- https://www.eecs.uottawa.ca/~turcotte/icbbt2021

Acknowledgments

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- **Indigenous Affirmation** of the Universisity of Ottawa
  - We pay respect to the **Algonquin people**, who are the traditional guardians of this land. We acknowledge their longstanding relationship with this territory, which remains unceded. We pay respect to all **Indigenous people** in this region, from all nations across Canada, who call Ottawa home.
  - We acknowledge the **traditional knowledge keepers**, both young and old.
  - And we honour their **courageous leaders**: past, present, and future.

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Appendix: All the Results (1/4)

K-mer	"Skip-mer" [21]			Secondary Structure Fingerprints	10-Fold Cross Validation Averaged Accuracy		
	Length	Match	Skip		3-Layers Model	4-Layers Model	5-Layers Model
4-mer		-		-	62.48% $\pm$ 0.51%	**64.86%** $\pm$ 0.76%	62.09% $\pm$ 0.77%
5-mer		-		-	**77.29%** $\pm$ 0.22%	75.24% $\pm$ 0.53%	74.31% $\pm$ 0.46%
6-mer		-		-	**84.56%** $\pm$ 0.28%	83.55% $\pm$ 0.48%	83.55% $\pm$ 0.57%
-	6	2	1	-	61.74% $\pm$ 0.31%	**61.85%** $\pm$ 0.94%	59.45% $\pm$ 1.0%
-	7	1	1	-	**55.89%** $\pm$ 0.34%	54.38% $\pm$ 0.99%	48.39% $\pm$ 1.86%
-	7	2	1	-	**77.32%** $\pm$ 0.5%	75.76% $\pm$ 0.8%	71.74% $\pm$ 1.83%
-	9	1	1	-	**75.16%** $\pm$ 0.41%	73.23% $\pm$ 0.46%	65.53% $\pm$ 4.57%
-	9	2	1	-	**84.92%** $\pm$ 0.25%	84.0% $\pm$ 0.36%	82.2% $\pm$ 1.13%
-	11	1	1	-	**84.08%** $\pm$ 0.21%	81.78% $\pm$ 0.98%	81.15% $\pm$ 0.88%
-		-		min. free energy	35.91% $\pm$ 0.42%	**36.75%** $\pm$ 0.76%	35.94% $\pm$ 0.51%
-		-		min., avg. free energy	50.65% $\pm$ 0.57%	52.04% $\pm$ 0.86%	**52.6%** $\pm$ 0.65%
-		-		min., avg., max. free energy	57.37% $\pm$ 0.52%	59.39% $\pm$ 0.58%	**59.42%** $\pm$ 0.76%
4-mer	6	2	1	-	71.57% $\pm$ 0.4%	**71.69%** $\pm$ 0.41%	71.15% $\pm$ 0.49%
4-mer	7	1	1	-	70.52% $\pm$ 0.39%	**71.91%** $\pm$ 0.38%	69.63% $\pm$ 1.01%
5-mer	7	2	1	-	**82.14%** $\pm$ 0.29%	82.08% $\pm$ 0.46%	80.1% $\pm$ 0.65%
5-mer	9	1	1	-	**81.77%** $\pm$ 0.47%	81.13% $\pm$ 0.39%	80.39% $\pm$ 0.69%
6-mer	9	2	1	-	**86.89%** $\pm$ 0.28%	86.09% $\pm$ 0.21%	84.68% $\pm$ 0.83%
6-mer	11	1	1	-	**86.7%** $\pm$ 0.38%	86.17% $\pm$ 0.61%	84.73% $\pm$ 1.58%

Appendix: All the Results (2/4)

K-mer	"Skip-mer" [21]			Secondary Structure Fingerprints	10-Fold Cross Validation Averaged Accuracy		
	Length	Match	Skip		3-Layers Model	4-Layers Model	5-Layers Model
4-mer		-		min. free energy	$67.96\% \pm 0.56\%$	$70.97\% \pm 0.54\%$	**72.6%** $\pm 0.63\%$
5-mer		-		min. free energy	$78.93\% \pm 0.24\%$	$80.49\% \pm 0.62\%$	**81.05%** $\pm 0.46\%$
6-mer		-		min. free energy	**84.33%** $\pm 0.51\%$	$84.05\% \pm 0.71\%$	$77.7\% \pm 5.36\%$
4-mer		-		min., avg. free energy	$69.92\% \pm 0.56\%$	$72.28\% \pm 0.52\%$	**75.38%** $\pm 0.6\%$
5-mer		-		min., avg. free energy	$74.93\% \pm 1.66\%$	**81.28%** $\pm 0.43\%$	$81.02\% \pm 0.33\%$
6-mer		-		min., avg. free energy	$83.42\% \pm 0.39\%$	**83.73%** $\pm 0.32\%$	$82.23\% \pm 0.32\%$
4-mer		-		min., avg., max. free energy	$71.14\% \pm 0.49\%$	$74.63\% \pm 0.54\%$	**75.85%** $\pm 0.54\%$
5-mer		-		min., avg., max. free energy	$79.28\% \pm 0.75\%$	$80.74\% \pm 0.52\%$	**81.23%** $\pm 0.75\%$
6-mer		-		min., avg., max. free energy	$83.21\% \pm 0.37\%$	**83.53%** $\pm 0.13\%$	$81.87\% \pm 0.44\%$
-	6	2	1	min. free energy	$66.94\% \pm 0.58\%$	$69.98\% \pm 0.65\%$	**71.02%** $\pm 0.83\%$
-	7	1	1	min. free energy	$66.83\% \pm 0.22\%$	$69.69\% \pm 0.48\%$	**71.23%** $\pm 0.34\%$
-	7	2	1	min. free energy	$78.66\% \pm 0.55\%$	$80.35\% \pm 0.41\%$	**80.72%** $\pm 0.59\%$
-	9	1	1	min. free energy	$77.78\% \pm 0.27\%$	**80.05%** $\pm 0.29\%$	$79.43\% \pm 1.73\%$
-	9	2	1	min. free energy	**84.58%** $\pm 0.34\%$	$79.17\% \pm 3.18\%$	$80.65\% \pm 1.28\%$
-	11	1	1	min. free energy	$83.61\% \pm 0.52\%$	**83.88%** $\pm 0.32\%$	$77.77\% \pm 5.17\%$

Appendix: All the Results (3/4)

K-mer	"Skip-mer" [21]			Secondary Structure Fingerprints	10-Fold Cross Validation Averaged Accuracy		
	Length	Match	Skip		3-Layers Model	4-Layers Model	5-Layers Model
-	6	2	1	min., avg. free energy	69.62% $\pm$ 0.49%	71.83% $\pm$ 0.74%	**74.16%** $\pm$ 0.6%
-	7	1	1	min., avg. free energy	68.11% $\pm$ 0.99%	71.45% $\pm$ 0.73%	**73.93%** $\pm$ 0.59%
-	7	2	1	min., avg. free energy	78.64% $\pm$ 0.35%	79.75% $\pm$ 0.85%	**80.9%** $\pm$ 0.32%
-	9	1	1	min., avg. free energy	78.48% $\pm$ 0.59%	79.58% $\pm$ 0.55%	**81.29%** $\pm$ 0.32%
-	9	2	1	min., avg. free energy	83.2% $\pm$ 0.54%	**83.37%** $\pm$ 0.43%	82.45% $\pm$ 0.57%
-	11	1	1	min., avg. free energy	**82.83%** $\pm$ 0.41%	82.75% $\pm$ 0.44%	82.3% $\pm$ 0.45%
-	6	2	1	min., avg., max. free energy	70.77% $\pm$ 0.42%	74.04% $\pm$ 0.75%	**75.38%** $\pm$ 0.36%
-	7	1	1	min., avg., max. free energy	69.28% $\pm$ 0.75%	74.32% $\pm$ 0.52%	**74.74%** $\pm$ 0.75%
-	7	2	1	min., avg., max. free energy	79.02% $\pm$ 0.58%	80.43% $\pm$ 0.52%	**81.16%** $\pm$ 0.52%
-	9	1	1	min., avg., max. free energy	78.73% $\pm$ 0.45%	80.42% $\pm$ 0.7%	**81.3%** $\pm$ 0.33%
-	9	2	1	min., avg., max. free energy	**83.93%** $\pm$ 0.2%	83.38% $\pm$ 0.42%	82.34% $\pm$ 0.63%
-	11	1	1	min., avg., max. free energy	**83.04%** $\pm$ 0.4%	83.0% $\pm$ 0.18%	82.47% $\pm$ 0.35%

Appendix: All the Results (4/4)

K-mer	"Skip-mer" [21]			Secondary Structure Fingerprints	10-Fold Cross Validation Averaged Accuracy		
	Length	Match	Skip		3-Layers Model	4-Layers Model	5-Layers Model
4-mer	6	2	1	min. free energy	74.26% $\pm$ 0.46%	76.11% $\pm$ 0.66%	**78.78%** $\pm$ 0.3%
4-mer	7	1	1	min. free energy	74.22% $\pm$ 0.29%	77.44% $\pm$ 0.83%	**78.54%** $\pm$ 0.47%
5-mer	7	2	1	min. free energy	**83.74%** $\pm$ 0.39%	83.54% $\pm$ 0.39%	83.65% $\pm$ 0.18%
5-mer	9	1	1	min. free energy	82.17% $\pm$ 0.47%	**83.21%** $\pm$ 0.42%	83.07% $\pm$ 0.45%
6-mer	9	2	1	min. free energy	**85.9%** $\pm$ 0.28%	84.37% $\pm$ 0.71%	83.1% $\pm$ 0.73%
6-mer	11	1	1	min. free energy	**85.86%** $\pm$ 0.35%	84.94% $\pm$ 0.34%	82.39% $\pm$ 0.57%
4-mer	6	2	1	min., avg. free energy	75.06% $\pm$ 0.53%	77.33% $\pm$ 0.66%	**78.89%** $\pm$ 0.48%
4-mer	7	1	1	min., avg. free energy	74.78% $\pm$ 0.46%	77.11% $\pm$ 0.39%	**78.47%** $\pm$ 0.48%
5-mer	7	2	1	min., avg. free energy	82.52% $\pm$ 0.38%	**82.77%** $\pm$ 0.41%	82.68% $\pm$ 0.27%
5-mer	9	1	1	min., avg. free energy	81.26% $\pm$ 0.38%	**82.59%** $\pm$ 0.6%	82.37% $\pm$ 0.41%
6-mer	9	2	1	min., avg. free energy	**84.39%** $\pm$ 0.52%	84.2% $\pm$ 0.3%	82.54% $\pm$ 1.01%
6-mer	11	1	1	min., avg. free energy	**84.33%** $\pm$ 0.53%	84.19% $\pm$ 0.65%	82.06% $\pm$ 0.7%
4-mer	6	2	1	min., avg., max. free energy	75.73% $\pm$ 0.57%	79.56% $\pm$ 0.2%	**79.65%** $\pm$ 0.67%
4-mer	7	1	1	min., avg., max. free energy	75.77% $\pm$ 0.61%	77.99% $\pm$ 0.38%	**79.23%** $\pm$ 0.44%
5-mer	7	2	1	min., avg., max. free energy	82.54% $\pm$ 0.39%	**82.92%** $\pm$ 0.28%	82.16% $\pm$ 0.61%
5-mer	9	1	1	min., avg., max. free energy	81.41% $\pm$ 0.34%	**83.13%** $\pm$ 0.17%	81.55% $\pm$ 1.31%
6-mer	9	2	1	min., avg., max. free energy	**84.73%** $\pm$ 0.32%	83.71% $\pm$ 0.16%	82.33% $\pm$ 0.64%
6-mer	11	1	1	min., avg., max. free energy	**84.61%** $\pm$ 0.26%	83.33% $\pm$ 0.68%	80.4% $\pm$ 2.18%

Appendix: Included Hosts

- **47 different host species:**
  - *Allium sativum*, *Anas carolinensis*, *Anas clypeata*, *Anas platyrhynchos*, *Anatidae*, *Apodemus agrarius*, *Aves*, *Bos taurus*, *Canis lupus familiaris*, *Capra hircus*, *Capsicum annuum*, *Columbidae*, *Corvus brachyrhynchos*, *Cricetulus griseus*, *Culex*, *Culex pipiens*, *Culex quinquefasciatus*, *Culicidae*, *Culiseta melanura*, *Cyanocitta cristata*, *Equus caballus*, *Felis catus*, *Gallus gallus*, *Glycine max*, *Homo sapiens*, *Macaca mulatta*, *Malus domestica*, *Meleagris gallopavo*, *Melogale*, *Mus musculus*, *Oryza sativa*, *Ovis aries*, *Procyon lotor*, *Prunus*, *Prunus avium*, *Prunus persica*, *Pyrus communis*, *Rattus norvegicus*, *Rosa sp.*, *Solanum lycopersicum*, *Solanum tuberosum*, *Sus scrofa*, *Sus scrofa domesticus*, *Triticum aestivum*, *Vitis vinifera*, *Vulpes vulpes*, and *Zea mays*