Genetic space exploration: Overview of low dimension projections of alignment-free characterisation methods applied to SARS-CoV-2

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Introduction

The ongoing epidemic has showed us the necessity to understand better the dynamics of viral evolution to identify variants of concern and variants of interest (VoC/VoI). The common methods of phylogenetical analysis are computationally expensive and struggle to incorporate new data, rely on multi-aligned sequences, performing a pair-wise comparison among all the sampled genomes. Alignment-free methods offer an alternative, and among them those based on word-statistics methods offer an individual representation as a single point in a $\mathbb{R}^n$ space for each genetic code, which allows for easier incorporation of new information. This project analyses approximately 250k SARS-CoV-2 sequences extracted from the GISAID database, characterising them with the aforementioned featuring methods to apply the dimensionality reduction (DR) algorithm PaCMAP[8], to visualise the structures formed within the different genetic spaces. This gives multiple perspectives to understand better the virus ecology. Furthermore, the use of clustering methods, like HDBSCAN[3] and CLASSIX[1] , shows to be a valuable tool to confirm the appearance of clusters formed by new VoC/VoI.

Methods

- Download of around 250k genetic sequences from the GISAID database procuring an appropriate representation of VoC/VoI and even distribution through time;
- Classification of sequences using Pangolin, labelling them through the Scorpio reported lineages;
- Extraction of natural vectors features (NVf). They differ in the details, but can be interpreted as an array of summary statistics of an element $\epsilon \in E$ in a sequence S of length n, generalised as:

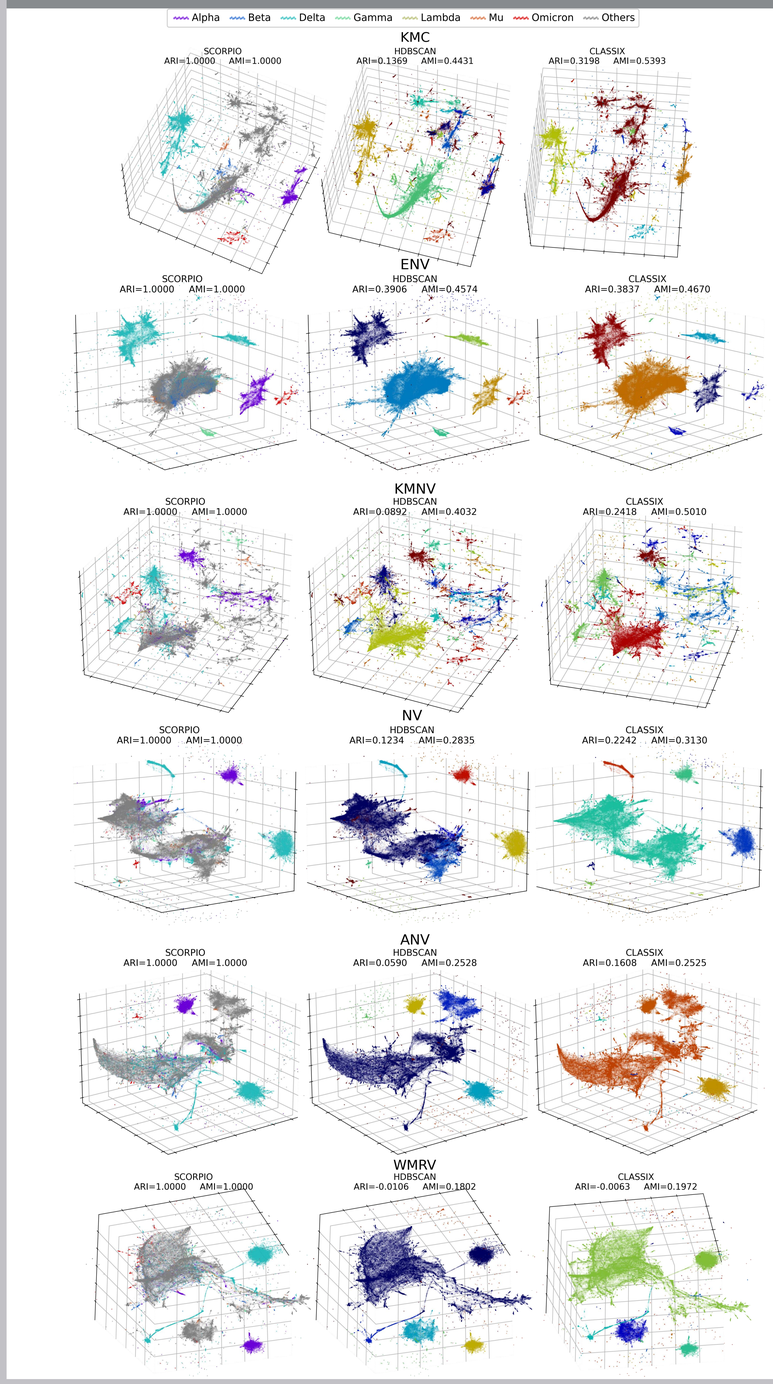
$$NVf(S) = \left(n_{\epsilon_1}, \mu_{\epsilon_1}, D_2^{\epsilon_1}, \dots n_{\epsilon_n}, \mu_{\epsilon_n}, D_2^{\epsilon_n}\right)$$

The count of (n_ϵ), mean distance of to the origin (μ_ϵ) and variance of said distance (D_2^ϵ), characterise its distribution within S, these three magnitudes can be defined as:

$$n_\epsilon = \sum_{i=0}^{n} w_\epsilon(S_i)\,, \mu_\epsilon = \sum_{i=0}^{n} \frac{i}{n_\epsilon} w_\epsilon(s_i), D_2^\epsilon = \sum_{i=0}^{n} \frac{(i-\mu_\epsilon)^2}{n_\epsilon n} w_\epsilon(s_i)$$

Where $s_i \in S$, and w_ϵ is a weight of 1 if $\epsilon = s_i$. Among the explored NVf we can mention: natural vector (NV)[5], which is a 12-D array, since $\epsilon \in \{A,C,G,T\}$; Accumulated NV (ANV)[4] adds the covariance of the accumulation of nucleotides through S; degenerate bases NV ($WMRV$)[7] maps S by pairs of degenerate bases, namely $\epsilon \in \{W,S,M,K,R,Y\}$; k-mers NV ($KMNV$)[2] describes k-mer distribution; and extended NV (ENV)[6] characterise the intensities of the pixels in a 2-d frequency chaos representation of S, in which $\epsilon \in \{0, \dots 255\}$. Additionally, the method of k-mer counts (KMC) was also included among the NVf. The value k was set to 3 for all k-dependant algorithms;
- DR projection for the different NVf were produced to observe structures in these genetic spaces. From these the PaCMAP[8] method proved to be robust for replication, while reliable on representing global and local structures.
- Two clustering algorithms were applied and evaluated, HDBSCAN[3] and CLASSIX[1], both yielded similar and consistent results according feature projected.

Results



Results (cont.)

- The figure (left) shows the formation of structures and clusters on the genetic spaces that in most of them correspond to VoC/VoI,
- The unsupervised algorithms, HDBSCAN and CLASSIX, are able to detect the formation of these clusters,
- The best NVf, by ARI and AMI metrics, appear to be KMC and ENV, while the worst $WMRV$, results are summarised on right table.

Feature	HDBSCAN		CLASSIX	
	ARI	AMI	ARI	AMI
KMC	0.1369	0.4431	0.3198	0.5393
ENV	0.3906	0.4574	0.3837	0.4670
$KMNV$	0.0892	0.4032	0.2418	0.5010
NV	0.1234	0.2835	0.2242	0.3130
ANV	0.0590	0.2528	0.1608	0.2525
$WMRV$	-0.0106	0.1802	-0.0063	0.1972

Conclusions and Future work

- The low dimension projection of most of NVf seem to rescue important information to identify new variants through clustering methods,
- Monitoring of emergence of clusters could make possible to police the emergence of variants of concern,
- Simple KMC was the one of the bests NVf for clustering formation followed by ENV, modification on this methods could improve their accuracy and sensibility
- The application of different featuring methods, such as: protein NV[9], graphical representation[10], of Fourier power spectrum should be investigated[11],
- It would be also of interest to assess different metrics of distance like: Kull-Leiber, Yaus-Hausdorff[10], among others; to explore possibilities.

References

[1] X. Chen and S. Guettel, 'Fast and explainable clustering based on sorting', 2022, doi: 10.48550/ARXIV.2202.01456.
[2] J. Wen, et al., 'K-mer natural vector and its application to the phylogenetic analysis of genetic sequences', Gene, vol. 546, 2014.
[3] L. McInnes and J. Healy, 'Accelerated hierarchical density based clustering', Nov. 2017.
[4] R. Dong et al., 'A novel approach to clustering genome sequences using inter-nucleotide covariance', Frontiers in Genetics, vol. 10, 2019.
[5] M. Deng, et al., 'A novel method of characterizing genetic sequences: Genome space with biological distance and applications', PLoS ONE, vol. 6, 2011.
[6] S. Pei, et al., 'Fast and accurate genome comparison using genome images: The Extended Natural Vector Method', Molecular Phylogenetics and Evolution, vol. 141, 2019.
[7] Y. Li, et al., 'A novel fast vector method for genetic sequence comparison', Scientific Reports, vol. 7, 2017.
[8] Y. Wang, et al., 'Understanding How Dimension Reduction Tools Work: An Empirical Approach to Deciphering t-SNE, UMAP, TriMap, and PaCMAP for Data Visualization', Journal of Machine Learning Research, vol. 22, 2021.
[9] C. Yu, et al., 'Protein space: A natural method for realizing the nature of protein universe', Journal of Theoretical Biology, vol. 318, 2013
[10] K Tian, et al., 'Two Dimensional Yau-Hausdorff Distance with Applications on Comparison of 417 DNA and Protein Sequences', PLoS ONE, vol. 10, 2015.
[11] C. Yin, et al., 'A measure of DNA sequence similarity by Fourier Transform with applications on hierarchical clustering', Journal of Theoretical Biology, vol. 359, 2014.

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