Efficient k-mer Indexing with Application to Mapping-free SNP Genotyping

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Abstract: Advances in sequencing technologies and computational methods have enabled rapid and accurate identification of genetic variants. Accurate genotype calls and allele frequency estimations are crucial for population genomics analyses. One of the most demanding step in the genotyping pipeline is mapping reads to the human reference genome. Recently mapping-free methods, like Lava and VarGeno, have been proposed for the genotyping problem. They are reported to perform 30 times faster than a standard alignment-based genotyping pipeline while achieving comparable accuracy. Moreover, these methods are able to include known genomic variants in the reference making read mapping, and genotyping variant-aware. However, in order to run they require a large k-mers database, of about 60GB, to be loaded in memory. In this paper we study the problem of genotyping using new efficient data structures based on k-mers set compression, and we present a fast mapping-free genotyping tool, named GenoLight. GenoLight reports accuracy results similar to the standard pipeline, but it is up to 8 times faster. Also, GenoLight uses between 5 to 10 times less memory than the other mapping-free tools, and it can be run on a laptop. Availability: https://github.com/CominLab/GenoLight.

1 INTRODUCTION

The discovery and characterization of sequence variations in human populations is crucial in genetic studies. A prime challenge is to efficiently analyze the variations of a freshly sequenced individual with respect to a reference genome and the available genomic variation data. Single nucleotide polymorphism (SNP) genotyping has been widely used in human disease-related research such as genome-wide association studies (Stranger et al., 2011) and a recent study on rare-disease diagnosis (NEJ, 2021).

The approaches to SNP genotyping can be roughly divided into three categories: microarray methods, sequencing alignment-based methods, and alignment-free methods. The first approach uses SNP arrays (Pastinen and et al., 2000). SNP arrays are fast and inexpensive; however, they can only hold a limited number of probes: the state-of-the-art Affymetrix genome-wide SNP array 6.0 has only 906 000 SNP probes, compared to 31 million known common SNPs in dbSNP (build 150).

The second approach is based on high-throughput whole-genome sequencing and read alignment. In most NGS-based genotyping pipelines, the first step after sequencing a genome is to map each read to the reference (Li and Durbin, 2010; Langmead and Salzberg, 2012). Standard tools for genotyping (e.g. Samtools mpileup (Li et al., 2009) and GATK HaplotypeCaller (McKenna and et al., 2010)) require this mapping information for every read before being able to call variants. Yet, despite recent advances in speed (Marco-Sola et al., 2012; Siragusa et al., 2013; Yorukoglu et al., 2016; Zaharia et al., 2011), mapping still remains a computationally expensive step. Furthermore, genotyping pipelines also include variant calling steps, significantly increasing the total runtime.

The third approach is based on high-throughput whole genome sequencing followed by an alignment-free sequence comparison (Zielezinski et al., 2017). Alignment-free methods have been used to save compute time and memory by avoiding the cost of full-scale alignment (Vinga, 2014). The reliance on a single reference human genome could introduce substantial biases in downstream analyses (Brandt et al., 2015; Günther and Nettelblad, 2019; Salavati et al., 2019). Furthermore, including known variants in the reference makes read mapping, variant calling, and genotyping variant-aware. Alignment-free approaches have been applied to SNP genotyping (Shajii et al., 2016; Sun and Medvedev, 2019). They

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introduced two SNP genotyping tools named LAVA and VarGeno, respectively, which build an index from known SNPs (e.g., dbSNP) and then use approximate k-mer matching to genotype the donor from sequencing data. LAVA and VarGeno have been reported to perform 4 to 30 times faster than a standard alignment-based genotyping pipeline while achieving comparable accuracy. Recently, another alignment-free genotyping tool, called MALVA (Denti et al., 2019), has been able to handle indels. Remarkably, alignment-free methods provide in some cases even better results than the most widely adopted SNPs discovery pipeline (Shajii et al., 2016; Sun and Medvedev, 2019). In addition, these tools provide a faster alternative to read mapping, and with the increasing of sequencing capabilities, they are a promising direction of investigation. However, they require a large amount of memory for indexing k-mers, about 60GB, and all tools can be run only on a large server with adequate RAM. In this work, we introduce GenoLight that will address this problem with new efficient data structures based on k-mer set compression.

2 PRELIMINARIES

In this section, we report the basic concepts about the functionality of VarGeno (Sun and Medvedev, 2019) and the notation used inside this document to explain GenoLight more clearly. We describe VarGeno because it is the fastest tool and also because the other methods follow a similar paradigm. Given a database of biallelic SNPs l_S encoded as VCF (Variant Calling Format) file, a database of reads from the genome that we want to genotype, and a reference genome G_R, VarGeno produces a VCF file containing the most probable genotype for each variant inside l_S. SNPs not contained in l_S are not genotyped.

The VarGeno pipeline consists of two phases. During the first phase, VarGeno maps each read r inside the database on G_R, in an alignment-free fashion, and then in the genotyping phase it uses the alignment information to detect the SNPs. To understand how the mapping is performed, we need to introduce some definitions and concepts. We denote by G_S the genome obtained by G_R replacing, at each locus containing an SNP $\in l_S$, the alternative variant.

Conceptually, in this phase VarGeno extracts all non-overlapping k-mer, with k= 32, from a read r and searches for them in G_R and G_S. The position supported by a k-mer is the difference between the position in which the k-mer occurs inside the genome (G_R and/or G_S) and the position in which it appears inside the read. The position supported by the largest number of k-mers and that respect two constraints that we will define later in this section is called the target position (t_p) and represents the initial position where the read is mapped to G_R or G_S. We report a simple example in Figure 1, in which the read r is split into three non-overlapping k-mers, that are searched into the two genomes. All three k-mers are in agreement and support the target position 10.

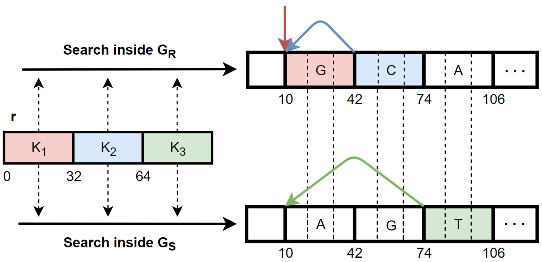
To search k-mers in G_R and G_S efficiently, VarGeno builds two dictionaries D_{ref} and D_{snp} using Bloom filers. D_{ref} stores the binary encoding of each overlapping k-mer, with $k = 32$, from G_R and the relative initials positions. K-mers that occur inside G_R with a frequency greater than 10 are discarded because they likely lead to an incorrect calculation of t_p.

The same process could be used to build D_{snp} from G_S. However, the only k-mers not present in D_{ref} would be the 32 consecutive k-mers for each SNP $\in l_S$ that contain the alternative variant. So, VarGeno stores only such k-mers inside D_{snp} in a binary encoding with the relative initials positions.

Using a specific search algorithm that involves the use of D_{ref} and D_{snp}, VarGeno is able to efficiently obtain all the initial positions of a k-mer in G_R and G_S and thus establishes the target position of a read.

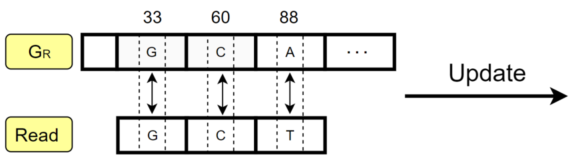
The presence of sequencing errors inside the reads can lead to an incorrect calculation of t_p. To solve this problem, given a k-mer K, LAVA (Shajii et al., 2016) calculates the initial position within G_{ref} and G_{snp} not only by searching for K but also of all k-mers belonging to his Hamming neighbourhood, denoted by $N(K)$. This set is composed of all k-mers having, at most, a Hamming distance equal to 1 from K. We observe that $|N(k)| = 3k + 1$. In this way, k-mers that would be present in G_R or in G_S, but are affected by a single sequencing error contribute to the correct calculation of the target position. To reduce the search space and thus increase the temporal performance, VarGeno uses quality scores to choose which k-mer $\in N(K)$ to search within D_{ref} and D_{snp} (besides obviously K). In particular, it searches for the k-mers $K_n \in N(K)$ whose character $K_n[i]$, $0 \leq i \leq 31$ that differentiates them from K has a quality score value $Q[i]$ higher than a certain threshold. We can now define the constraint that the k-mer that supports position t_p must satisfy. Let t_p be the position supported by the largest number of k-mers, to be the target position, these two conditions must hold:

1. At least one of the k-mers extracted from r supports t_p;
2. Let K_x and K_y be two k-mers extracted from r, $x \neq y$. At least one k-mer belonging to the set $N(K_x)$



K-MER	Start position inside read	Start position in G_R	Start position in G_S	Support position
K_1	0	10	-	10
K_2	32	42	-	10
K_3	64	-	74	10

Figure 1: An example of target position calculation.



Pileup tabel

Position SNP	C_α	C_β
...	...	...
33	+1	
60	+1	
88		+1
...	...	...

Figure 2: Pileup table update.

and one k-mer belonging to the set $N(K_y)$ must support t_p;

If more than one position has the same maximum number of supporter k-mers and respects these two constraints, the current read is considered not mapped. In general, if a read is not mapped, Vargeno tries to map its reverse complement of r, performing the same steps described.

The size of non-overlapping k-mers extracted from reads, and consequently the size of k-mers inside the dictionary, is fixed to 32 for the following reasons. For the human genome, 85% of 32-mer have unit frequency: this is fundamental for how t_p is calculated. The second reason is that the binary representation of the k-mer can be stored exactly in a 64-bit variable without wasting space in a 64-bit architecture.

Once the target position has been identified, VarGeno checks if there are SNPs $\in l_S$ in the range $[tp, tp+|r|]$ within l_s, where $|r|$ is the length of the read. If so, it proceeds with updating a data structure called pileup table. For each SNP, there are two counters C_α and C_β. The first is increased by one unit if a read contains the reference variant, and the second is increased by one unit if the read contains the alternative variant. An example is shown in Figure 2.

Once all the dataset reads are processed, using the data contained in the pileup table, VarGeno proceeds with the genotyping phase, using the Bayesian statistical framework (Sun and Medvedev, 2019).

3 METHODS

The basic idea of LAVA and VarGeno is to build two dictionaries of k-mers, one with all k-mers from the reference genome and another with the k-mers covering known SNPs. In LAVA, these dictionaries are implemented with a hash table where all k-mers are explicitly stored; instead, VarGeno uses a Bloom filter. Both these data structures need to be loaded into memory for efficient queries, and they require about 60-63 GB of RAM. However, the size of these dictionaries can be reduced because most of the information carried by a k-mer is redundant. Given two overlapping k-mers, it is possible to reassemble them into a single (k+1)-mer, thus reducing the storage requirement by k-1 bases. In GenoLight we further exploit this observation: a set of k-mers, associated with known SNPs, is assembled into a string, or set of strings, that contains all of them. This procedure allows us to store the whole dictionary in linear form, reducing the memory requirements from O(kn) to O(n) without losing information.

This problem is closely related to the representation and compression of k-mer sets, which has attracted the attention of many researchers recently (Břinda et al., 2021; Rahman and Medvedev, 2020). K-mer sets are widely used in many bioinformatics applications (Storato and Comin, 2021; Qian et al., 2018; Marchiori and Comin, 2017; Qian and Comin, 2019; Andreace et al., 2021a; Andreace et al., 2021b).

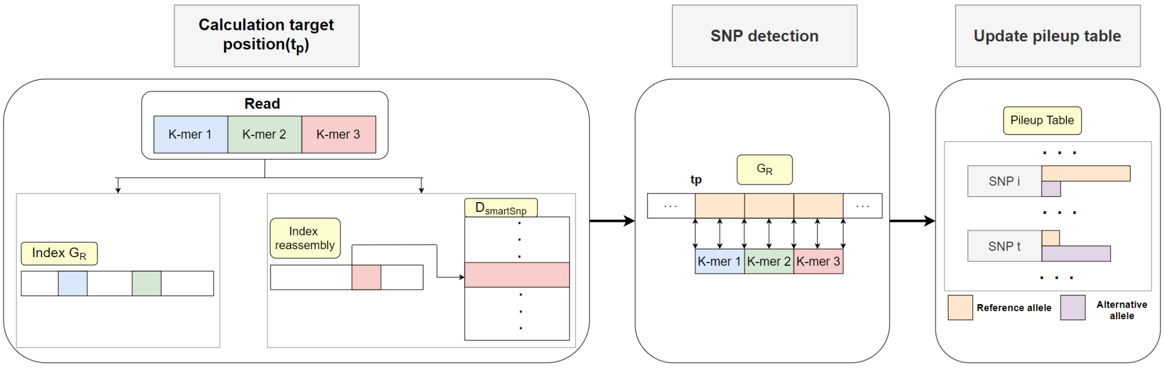


Figure 3: An overview of the genotyping pipeline of GenoLight.

In fact, a k-mers set can be represented by its De Bruijn graph. Instead of explicitly storing the graph, it can be compressed by assembling the graph and computing a string that traverses all nodes in the graph only once. However, this problem is far from trivial, like traditional assembly, and the algorithms proposed in (Břinda et al., 2021; Rahman and Medvedev, 2020) will not produce a single string that traverses all nodes. Instead, the results of this assembly are a set of strings, called unitigs, that describe and cover the De Bruijn graph. These techniques have been successfully applied to quality value compression (Shibuya and Comin, 2019a; Shibuya and Comin, 2019b).

In this work, we use these memory efficient data structures to calculate the target position of a read. The aim of this project is to develop a tool that can be run on a personal computer without the need of a server with a large amount of RAM. Moreover, we implement an efficient search algorithm with time performance similar to that of other alignment-free methods.

In order to use the efficient representation of k-mer set (Břinda et al., 2021), the dictionary D_{snp} needs to be replaced by two new data structures, the *RI* reassembly index and a new dictionary named $D_{SmartSnp}$. Recall that D_{snp} contains all k-mers extracted from G_S that cover an SNP. The binary encoding of these 32-mer isn't stored directly inside $D_{SmartSnp}$. Instead, these k-mers are compressed into a string called the reassembly index, indicated with *RI*, in which each k-mer extracted from G_S is present with unit frequency. In order to create the reassembly index *RI* we use the Prophasm k-mer set assembler (Břinda et al., 2021). Given a dataset of k-mers, Prophasm creates a string that contains all input k-mers exactly once. However, this compressed representation has two characteristics that we need to consider. First, depending on the input k-mers set, it may not be possible to assemble all of them into a single string. Instead, Prophasm produces a set of strings that assemble and cover all input k-mers. The second issue is that a k-mer can be represented in the reassembly index in the forward or reverse complement orientation. Unfortunately, the orientation cannot be determined a priori. Moreover, if in the input k-mer set two k-mers that are one the reverse complement of the other, these two k-mers will be represented only once in the reassembly index and not as two separated k-mers. In summary, a set of k-mers can be compressed using Prophasm into a reassembly index, with a substantial saving in terms of space. However, some important information might be lost during this process, and for this reason we introduce a new dictionary named $D_{SmartSnp}$.

In GenoLight, to compute the target position of a k-mer and the associated SNP position, we need to store for each k-mer in the reassembly index: the original position in the reference; its direction, forward or reverse; and the associated SNP. Instead of storing the k-mers in binary format as in D_{snp}, every k-mer in *RI* can be identified by its position in the reassembly index. The new dictionary $D_{SmartSnp}$ contains a set of records, one for each position in the reassembly index *RI*. In a record, the most important information is the original position of the k-mer inside G_S. While the binary encoding of a 32-length k-mer requires 64 bits to be stored, the integer representing the k-mer's position can be stored using 32 bits. This reduces the size of $D_{SmartSnp}$ compared to D_{snp} by 32 bits for each record. The second information in the record is the SNP position with respect to the k-mer. To correctly calculate the target position, it is essential to keep track of the k-mer orientation inside *RI* with respect to G_S. This information is also stored in the k-mer record inside $D_{SmartSnp}$. If the k-mer appears both as a forward and reverse complement in G_s, then in the reassembly index it will appear only once, in one of the two directions. In this case, the information on the k-mer that is present inside the *RI* is stored in $D_{SmartSnp}$, while the information on other k-mer will be stored in a small auxiliary data structure.

The read mapping phase of GenoLight is similar to that of VarGeno. A read is split into non-overlapping k-mers, and these k-mers are searched in the reference sequence G_R and the alternative reference G_S, in order to detect a candidate target position, see Figure 3. However, the representation of these references is different with respect to VarGeno. In VarGeno, both G_R and G_S are represented by Bloom filters. Whereas in GenoLight, we use the original reference sequence for G_R, and the reassembly index *RI* and $D_{SmartSnp}$ for G_S. Both G_R and *RI* contain a set of sequences in which we want to search for k-mers.

In order to allow for fast queries we use a variant of the FM-index (Ferragina and Manzini, 2000), the FDM-Index also used in BWA (Li and Durbin, 2010). GenoLight uses FDM-Index to index G_R and *RI*, and the bound backtracking algorithm described in (Li and Durbin, 2010) to search for k-mers inside it. This algorithm solves the inexact string match problem and is used by the Burrows-Wheeler Aligner (BWA). BWA performs local alignment of reads on a genome according to the seed-and-extend alignment paradigm. To perform the seeding phase efficiently, it indexes the reference genome using FDM-Index and performs searches with at most m mismatches to find areas with high similarity using this algorithm. We adapt this library allowing only for one mismatch and using the additional constraint provided by quality values so that possibly wrong bases are not aligned.

A pseudocode of the read mapping phase is reported above. Each read is decomposed into non-overlapping k-mers. For each k-mer we compute its neighbourhood also using the quality values. Then, these k-mers are searched in the reference genome through its index. If a match is found, these positions are stored in the vector P. To obtain the position p_{G_S} where a k-mer k occurs inside G_S, the alternative reference, GenoLight first identifies the starting position $p_{reassembly}$ of k inside *RI*. Next, it accesses $D_{SmartSnp}$ using $p_{reassembly}$ as key to get p_{G_S}. To efficiently search k in *RI*, *RI* is also indexed using FDM-Index. Once all matching positions have been stored in the vector P, we can compute the target position of the read, as in Figure 1. Finally, we can update the pileup table and compute the genotyping using the Bayesian statistical framework as in (Sun and Medvedev, 2019; Shajii et al., 2016).

Algorithm 1: GenoLight search.

Input:
IndexG: FDM-Index of the reference genome G_R
IndexRI: FDM-Index of the reassembly *RI*
$D_{SmartSnp}$: Dictionary containing k-mer having the alternative allele
r : Read to map into the reference genome
Q[K] : Quality scores associated with a k-mer K extracted from r

Output
t_p: target position of reads

Start
$S \leftarrow ExtractNonOverlapKmer(r)$
$P \leftarrow \emptyset$
for each $K \in \mathcal{S}$ **do**
  $N(K)_f \leftarrow Filter(N(K), Q[K])$
  $P \leftarrow SearchReferenceGenome(N(K)_f, IndexG)$
  for each $k \in \mathcal{N}(K)_f$ **do**
    $p_{reassembly} \leftarrow SearchRI(k, IndexRI)$
    $p_{Gs} \leftarrow D_{SmartSnp}[p_{reassembly}]$
    $P \leftarrow P \bigcup p_{Gs}$
$t_p \leftarrow CalculateTargetPosition(P)$
End

4 RESULTS

In this section, we report the results obtained by the tools GenoLight, VarGeno (Sun and Medvedev, 2019) and Lava (Shajii et al., 2016), as well as the standard alignment-based genotyping pipeline (BWA (Li and Durbin, 2010) as aligner, and BCFtools (Li, 2011) as variant caller). The analysis focuses on two fundamental points, the computation time and memory taken to complete the alignment and genotyping process, and the accuracy of the results obtained by the respective tools.

4.1 Experimental Setup

The datasets used in this study are two sets of real reads from the same individual (NA12878) from the 1000 Genomes Project (Project, 2008): SRR622461 with a coverage of 6X (40 GB) and SRR622457 with a coverage of 10X (65 GB). These datasets have been widely used for benchmarking in other studies (Shibuya and Comin, 2019a; Shibuya and Comin, 2019b; Shajii et al., 2016; Sun and Medvedev, 2019; Monsu and Comin, 2021). For validation, we used an up-to-date high-quality genotype annotation generated by the Genome in a Bottle Consortium (Zook et al., 2019). The GIAB gold standard contains validated genotype information for NA12878, from 14 sequencing datasets with five sequencing technologies, seven read mappers, and three different variant callers. The number of SNPs validated for

Table 1: Resources and Datasets used for testing.

DATASET	DESCRIPTION
hg19	Reference genome
dbSNP	Biallelic SNPs dataset (Sherry et al., 2001)
Affymetrix	Affymetrix Genome-Wide Human SNP Array
NA12878	Gold standard from GIAB (Zook et al., 2019)
SRR622461 (Low Coverage)	Dataset of reads with coverage 6X
SRR622457 (High Coverage)	Dataset of reads with coverage 10X

NA12878 in the gold standard is 3.7M. The reference genome used is hg19, also described as Genome Reference Consortium Human Build 37 (GRCh37). For the SNPs dataset, we choose dbSNP (Sherry et al., 2001), which contains human single nucleotide variations, and more specifically 11M SNPs biallelic, and Affymetrix Genome-Wide Human SNP Array 6.0 with about 1M SNPs. A summary of the datasets used for the evaluation is reported in Table 1.

To measure the accuracy, we use RTG Tools and the SNP list which are also genotyped in the GIAB gold standard. All tests were performed on a 14 lame blade cluster DELL PowerEdge M600 where each lame is equipped with two Intel Xeon E5450 at 3.00 GHz, 16GB RAM and two 72GB hard disk in RAID-1 (mirroring).

4.2 Computational Resources: Time and Memory

In this section we test the requirement of computational resources for all tools. Table 2 reports a summary of the results obtained for the two read datasets, with the two SNP databases, for all tools.

We consider the standard alignment-based genotyping pipeline as the reference to compare the three mapping-free algorithms. As expected, the standard pipeline is always more time consuming while requiring only 4 GB of memory. On the low coverage dataset the standard pipeline requires 1929 minutes to align 179 millions reads, that is more than 1 day of computation. On this dataset VarGeno is able to genotype the reads in only 59 and 44 minutes, depending on the SNP databases, remaining the fastest tool, thanks to the Bloom filter, as expected (Sun and Medvedev, 2019). GenoLight is the second fastest tool, it can process the low coverage dataset in 406 and 295 minutes, using the two SNP databases, which results in a speed-up of 4.75x and 6.5x with respect to the standard pipeline. LAVA is the slowest of the three mapping-free methods.

If we consider the high coverage dataset, that contains 287 million reads, the standard pipeline requires 2560 minutes, again more than one day of computation on a server. The time performance of the mapping-free tools is similar to that of the low coverage dataset. VarGeno is the fastest tool with 80 and 55 minutes, GenoLight needs 503 and 296 minutes, and Lava requires 723 and 435 minutes, on the two SNP databases. On the high coverage datasets, the speed-up of GenoLight with respect to the standard pipeline is 5.1x and 8.6x, for the two SNP databases. GenoLight is again the second fastest tool; however, on the high coverage dataset the speed-up w.r.t. standard pipeline increases.

In terms of memory requirement, the standard pipeline is the least expensive, with only 4 GB of RAM needed. For the mapping-free tools, the memory is mainly dominated by the size of the k-mers database and the corresponding data structures, and it varies only slightly with the number of the input reads. On the smallest SNP database, e.g. Affymetrix, GenoLight requires only 6 GB of RAM, whereas Lava requires 57.6 GB and VarGeno about 59 GB. On the largest SNP database, e.g. dbSNP, GenoLight needs 12.5 GB of memory, whereas Lava requires 60 GB and VarGeno 63 GB.

In summary, only GenoLight and the standard pipeline can run on a laptop with 16 GB of RAM because the memory requirements of both methods are low. However, GenoLight is much faster than the standard pipeline, up to 8.6x.

4.3 Genotyping Accuracy

In this section we test the genotyping accuracy of all tools on both datasets. The VCF file produced by the standard pipeline and all mapping-free tools are compared with the gold standard (Zook et al., 2019). In Table 3 are shown the results in terms of accuracy, in line with other studies (Shajii et al., 2016; Sun and Medvedev, 2019).

For the Low Coverage dataset, the standard pipeline has an accuracy of 93%. VarGeno and GenoLight have similar performance, varying from 91% to 93.5%, depending on the SNP databases. Lava shows a lower accuracy in some cases. The different performance of Lava can be explained by the fact that it does not use quality value information, unlike the

Table 2: Performance results of the genotyping for all tools on various datasets.

Dataset	SNP db	Algorithm	Time (d:h:m)	RAM (GB)
Low Coverage	None	Standard Pipeline	1:08:09	4
	dbSNP	VarGeno	59	63.2
		Lava	8:26	61
		GenoLight	6:46	12.5
	Affymetrix	VarGeno	44	58.9
		Lava	5:18	57.6
		GenoLight	4:55	6
High Coverage	None	Standard Pipeline	1:18:40	4
	dbSNP	VarGeno	1:20	63.242
		Lava	12:03	60
		GenoLight	8:23	12.5
	Affymetrix	VarGeno	55	59
		Lava	7:15	57.6
		GenoLight	4:56	6

Table 3: Performance results of the genotyping for all tools on various datasets.

Dataset	SNP db	Algorithm	Accuracy
Low Coverage	None	Standard Pipeline	0.930
	dbSNP	VarGeno	0.911
		Lava	0.819
		GenoLight	0.912
	Affymetrix	VarGeno	0.935
		Lava	0.935
		GenoLight	0.934
High Coverage	None	Standard Pipeline	0.969
	dbSNP	VarGeno	0.949
		Lava	0.845
		GenoLight	0.951
	Affymetrix	VarGeno	0.977
		Lava	0.974
		GenoLight	0.976

other two mapping-free methods.

On the High Coverage dataset, thanks to the 10X coverage, all tools reported higher accuracy values. In this case, the standard pipeline has an accuracy of 96.9%. Also, on this dataset the behaviour of all mapping-free tools is similar to the previous case. VarGeno and GenoLight have similar accuracies, in line with the standard pipeline, whereas Lava is less precise.

In summary, among the mapping-free tools, VarGeno and Genolight achieve the best overall performance in terms of accuracy, in line with the standard pipeline. GenoLight reports precision results comparable to VarGeno, but it uses between 5 and 10 times less memory than the other two tools. The standard alignment-based pipeline is extremely slow, and it requires more than a day of computation on a cluster. GenoLight achieves almost the precision levels and memory usage of the standard pipeline while being significantly faster, thus resolving the high memory issue of VarGeno and Lava.

5 CONCLUSIONS

In this paper we presented GenoLight an algorithm to speed up the alignment-based genotyping of reads with application to SNP detection. In the case of SNP detection, a k-mer database can be exploited for efficiently mapping of reads. Popular mapping-free tools, like Lava and VarGeno, require a large amount of memory to store these k-mers databases, more than 60 GB of RAM. In GenoLight we introduce a novel k-mers set compression technique that allows to store the same information in limited space, less than 12.5 GB. We tested different tools in popular benchmarking datasets for SNP genotyping. The results show that GenoLight is able to detect SNP with an accuracy similar to that of the standard pipeline, in line with VarGeno. However, GenoLight is up to 8 times faster than the standard pipeline, while requiring a limited amount of RAM, and it can be executed on a standard laptop, unlike the other mapping-free tools. As a future direction of investigation, it would be interesting to extend GenoLight for the detection of other genetic variations such as insertions and deletions.

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