IJICTR
International Journal of Information & Communication Technology Research

Volume 14- Number 3 – 2022 (48 -56)

The Efficient Alignment of Long DNA Sequences Using Divide and Conquer Approach

Mahmoud Naghibzadeh, Samira Babaei
Computer Engineering Department
Ferdowsi University of Mashhad
Mashhad, Iran
naghibzadeh@um.ac.ir, samira.babaei@mail.um.ac.ir

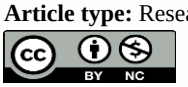
Behshid Behkmal*, Mojtaba Hatami
Computer Engineering Department
Ferdowsi University of Mashhad
Mashhad, Iran
behkamal@um.ac.ir, hatami.mojtaba@mail.um.ac.ir

Received: 12 March 2022 – Revised: 2 May 2022 - Accepted: 18 June 2022

Abstract—**discovering mutations in DNA sequences is the most common approach to diagnosing many genome-related diseases. The optimal alignment of DNA sequences is a reliable approach to discover mutations in one sequence in comparison to the reference sequence. Needleman-Wunsch is the most applicable software for optimal alignment of the sequences and Smith-Waterman is the most applicable one for local optimal alignment of sequences. Their performances are excellent with short sequences, but as the sequences become long their performance degeneration grows exponentially to the point that it is practically impossible to align the sequences such as compete human DNAs. Therefore, many researches are done or being conducted to find ways of performing the alignment with tolerable time and memory consumptions. One such effort is breaking the sequences into same number of parts and align corresponding parts together to produce the overall alignment. With this, there are three achievements simultaneously: run time reduction, main memory utilization reduction, and the possibility to better utilize multiprocessors, multicores and General-Purpose Graphic Processing Units (GPGPUs). In this research, the method for breaking long sequences into smaller parts is based on the divide and conquer approach. The breaking points are selected along the longest common subsequence of the current sequences. The method is evaluated to be very efficient with respect to both time and main memory utilization which are the two confining factors.**

Keywords: DNA sequence alignment; divide and conquer approach; longest common subsequence; big genome data, desease diagnosis.

Article type: Research Article



Publisher: ICT Research Institute

[DOI: 10.52547/itrc.14.3.48]

I. INTRODUCTION

Genomic sequence analysis gained extra momentum since the beginning of human genome project [1]. Nowadays, with the advances in the genome sequencing technologies enormous amount of genomic data is continuously being produced which makes it an explosive big data area. There are variety of purposes such as revealing the relations between genes or proteins, understanding their homology and functionality, deciphering sequences to disclose biological aspects, diagnosing diseases, and producing

* Corresponding Author

drugs, in the analysis of this big data. Many diseases are caused by variations in the genome sequences such as many kinds of cancers [2] and variety of disorders related to nervous system's degeneration such as Huntington's disease [3]. Even a repeat polymorphism in one of the genes, i.e., IL-lra, is shown to be associated with an increased possibility of osteoporotic fractures [4]. Comparing a subject sequence against a reference one to find the differences is commonly used in this analysis. The scientific meaning of comparison here is optimal alignment of sequences and revealing their differences. It is worth mentioning that optimal alignment is not the only method of finding find differences in genomic sequences towards diagnosing some genome-related diseases, and if the goal is to diagnose a particular disease, searching for specific patterns that have been proven to lead to that type of disease is another mechanism that has many applications [5] [6]. However, alignment has a much wider application and detects all kinds of variation in the subject sequence.

Alignment of two sequences is the arranging of these sequences such that similarities and differences are shown in the best possible way while the order of elements of each sequence is preserved. Often, a scoring function is proposed and the quantitative optimal alignment of the two sequences becomes equivalent to minimizing or maximizing this function, depending on the problem being solved and the formulation of the scoring function. Considering the countless number of ways that two sequences can be aligned, when gaps are allowed, the Dynamic Programming (DP) approach is an extremely innovative way of optimally aligning sequences. DP is a Mathematics-Computer optimization problem. A problem has to have the principle of optimality [7] property to be considered for being solved using the DP approach. This principal of optimality states that an optimal principle holds the property that whatever the initial state and initial decision are, the decisions that is followed must create an optimal solution starting from the state resulting from the first decision. The novelty of the DP idea is in systematic solution of all possible sizes of problems starting from the smallest size and going toward the largest, which is actually the given problem to be solved. It stores the solution results of smaller problems and in solving a bigger problem it can use the solution results of any of the previously solved sub-problems in an optimized manner. The optimal sequence alignment has the principle of optimality and it is successfully solved by the DP approach. The two most important approaches for sequence alignments are the Needleman-Wunsch algorithm and the Smith-Waterman algorithm.

Needleman-Wunsch algorithm [8] is often used to compare biological sequences with the goal of computing the similarity score of the sequences. It does so by arranging the sequences in such a way that the scoring function is maximized (in some cases the goal is to minimize the overall penalty). In this paper we are interested in global alignments (not necessarily global optimal) of pairs of sequences of DNA, RNA, or other similar genomic sequences composed of the four nucleotides A(adenine), T(thymine), G(guanine), and C(cytosine). The optimization problem is organized as filling an m by n matrix S of scores, where m is the length of one of the sequences and n is the length of the other. $S_{i,j}$ is the maximum score of aligning the i first characters of the first sequence and the j first characters of the second one. This is the clue to the dynamic program nature of breaking the optimization of a large problem into optimization of smaller problems. At the end, the bottom right corner of that matrix, i.e., $S_{m,n}$, gives the overall score of the alignment. To find the actual alignment, a technique for going from the bottom right corner to the top left corner of the matrix has to be followed [9]. For long sequences, the time complexity of Needleman-Wunsch algorithm which is O(mn) prolongs its execution time, hence continuous efforts are made to make it more practical [10] [11]. For example, for two human genomes of size 3.2 Giga nucleotides each, and considering at least 10 operations to fill each cell of the 3.2Giga by 3.2Giga matrix, the number of operations needed to fill the whole matrix is 1020. Using a single processor computer with the power of 10 million instructions per second, it takes approximately 317,000 years to complete the matrix. The amount of memory required by the Needleman-Wunsch aligner is very high, similar to the amount of runtime required. Calculating it is a simple task. A 3.2 billion by 3.2 billion matrix with each element being four bytes, i.e., one word, is required. It is practically impossible to get this amount of main memory. Using external memory instead, will not work because it further greatly increases the runtime. These are the reasons that lead researchers to break long strings and align the corresponding sections separately. Our effort in this research is in this direction.

Smith-Waterman algorithm [12] is another variant of global sequence alignment. Although the alignment is global, for the following reasons, it can only produce local optimal solutions. An important aspect of this alignment is that if the scoring value of a cell of the scoring matrix is calculated to be negative, its value is set to zero. This means, in such situations, the partial alignment score of sequences up to this point should not affect the alignment score of the rest of the sequences. The net consequence of this assumption is that Smith-Waterman algorithm will better demonstrate the score of local alignments. As a matter of fact, it better shows the local alignment scores and hence it is often used for finding similar regions of sequences. Smith-Waterman algorithm has the same problem as Needleman-Wunsch algorithm has, i.e., its time complexity is high and hence it's high execution time for aligning long sequences is high. In addition, its space complexity is also high such that some later researches have concentrated on lowering its space complexity only [13] [14]. This method has the same high runtime and very high memory requirements of the Needleman-Wunsch method. The novelty of the present study is the use of Longest Common Subsequence (LCM) to break large sequences into shorter ones and then align the corresponding short strings.

One way to reduce the alignment execution time is to break the two sequences into many pairs of smaller sequences, when, possible, and align each pair separately. For example, suppose the length of each of the original two sequences is 100,000 base pairs, i.e., characters. Using the Needleman-Wunsch alignment

[DOI: 10.52547/itrc.14.3.48]

algorithm, the number of operations would be proportional to 10^{10}. Let's assume we can break the two sequences into 10 pairs of sequences such that the length of each of the resulting new sequences is approximately 10,000 base pairs, i.e., bps for short. The number of operations for aligning all these 10 pairs is proportional to $10 * 10^8 = 10^9$ which would need 10 times less execution time compared to the case when the original sequences are aligned.

Examining the required main memory for this small example will also show that by breaking large sequences the need for main memory is also greatly reduced. If the sequences are not broken and they aligned at once, the amount of main memory required is equal to $4 * 10^5 * 10^5 \cong 40$ gigabytes. But if they are broken, the amount of main memory required is equal to 4*10^4*10^4≅0.4 gigabytea. Note that, in neither of cases it is assumed that the alignment is performed in parallel.

On the global alignment of related long genome sequences, many attempts have been made to break pairs of such long sequences into many smaller pairs [10] [15]. The current research is in the same direction to higher the efficiency and the quality of globally aligning two related genomic sequences. It uses the Longest Common Subsequence (LCS) technology to find anchor points, Divide and Conquer (DaC) approach to recursively break pairs of longer sequences, and Needleman-Wunsch algorithm to align every corresponding short pair of sequences.

Finding the LCS of two genome sequences has many applications such as phylogenetic construction and analysis, quick search in genome sequences big data, and identification of motifs. The natural approach to solve LCS of a pair of sequences is to formulate it as a dynamic programming problem similar to Needleman-Wunsch algorithm. However, there are quite newer methods with lower time complexities [16].

In this paper, a new divide and conquer approach to long genome sequence alignment is proposed. The division is along the LCS of the two sequences which is located approximately in the middle of the current two sequences. Only a section in the middle of the two sequences are selected in which the LCS is sought. If the discovered LCS is not long enough the sections length is enlarged until a reasonable length LCS is obtained. The novelties of this approach are highlighted in the following.

- It is much faster than either of Needleman-Wunsch and Smith-Waterman algorithms.
- It is faster than the state-of-the-art anchor-based methods which also use some kind of division. At the same time the space requirement of the method is extremely low.
- It has the potential to be implemented in parallel in three different levels, division of the long sequences, alignment of all short pairs of sequences, and utilization of General-Purpose Graphic Processing Units (GPGPU) within each alignment of short sequences.

In this study, for the first time, longest common subsequence method is used to break long genomic sequences in order to divide them into corresponding shorter sequences and then aligning these the corresponding shorter sequences using Needleman-Wunsch approach. It has been shown that this is not only possible but also efficient with respect to time and main memory utilization. The LCS subsequence is removed from both sequences and they are considered to be aligned, and hence exempted from further processing, which further saves both execution time and memory space.

The structure of the rest of the paper is as follows. In Section 2 a short review of related work is presented. Section 3 is for clarification of the problem being solved. Section 4 details the implementation of the proposed method's solution approach. Section 5 is the evaluation section and finally a short conclusion and future work is documented in Section 6.

II. Related Work

Sequence alignment is the canonical point of most tools of DNA and other genome sequences alignments. Human genome sequencing and analysis [1] formally started in 1990 and with it, computational methods, especially alignment, became an important part of any genome analysis activity. Undoubtedly, with the invention of its dynamic programming implementation and also after that, there has been great progress in making the alignment algorithms efficient and up to date. It is worth mentioning that although we have focused on genome sequence alignment, alignment is widely used in other domains such as protein sequences alignment, protein networks alignment [17] and all kinds of text alignment [18].

Needleman-Wunsch algorithm is the basic method for global sequence alignments [8]. Smith-Waterman algorithm is a variant of Needleman-Wunsch algorithm which is also a global aligner but it is tailored to find local similar regions of sequences being aligned [12]. The problem with these algorithms is their high time complexity which is *O(mn)*, or $O(n^2)$ where the length of the two sequences are the same and it is equal to n. In addition, the space complexity of these algorithms is also *O(mn)* which can be problematic for large sequences. In such cases the solution would be to have part of the scoring matrix in the secondary storage which will further worsen the execution time requirement. Many improvements are reported which we will concentrate on the most recent ones, here.

BLAST is a heuristic algorithm developed to search a short sequence in a large volume of data. Based on a hashing mechanism and a local alignment method, it is capable of finding sequences in the database that are similar to the search sequence with some degree of similarity [19]. BLAST is not originally developed for alignment of genomic sequences and it has the potential to be used in any kind of text data with any kind of alphabet. Later, specific program versions were developed for this purpose. BLASTZ and LASTZ are recent versions of the program that are widely used for local optimal alignment of genomic and DNA data. It is

[DOI: 10.52547/itrc.14.3.48]

much faster than Smith-Waterman method especially for long sequences [20].

Leimeister et al. [10] proposed a new anchor point finding method called filtered spaced word match. Anchor points are short subsequences in the two sequences which will be matched in the final alignment of these sequences. Subsequences between consecutive anchor points of the two sequences are aligned using known alignment algorithms. They claim that their superiority is in finding better anchor points. However, their comparison with that of Mugsy pipeline [21] did not lead to similar quality for closely related sequences but they claim it is superior in alignment of distal sequences. Neither time nor space complexity of the method is reported. Our guess is that its time complexity would be in the level of that of Needleman-Wunsch but there may be improvements is its space complexity.

Another recent development in the field of long genome sequence alignment is MUMmer4 [11] which is the fourth generation of MUMmer. It is based on a 48-bit suffix array data structure. It is capable of using multicores of the host computer however, this is applicable for the case of aligning many sequences to the reference genome; e.g., aligning many short reads to the human reference genome. In such cases, it can handle very large input size up to 141 Tera bps. Although the most important aspect of an algorithmic computational method is its time and space complexity, these are not reported in the paper.

The research reported by Sun et al. [13] is an effort towards space requirement reduction of Smith-Waterman. similar to that of Smith-Waterman, its input is a pair of sequences and it performs the optimal local alignment of the sequences. It is capable of aligning long sequences up to 100 million bps. It claims that the space requirement is tremendously reduced but the order of reduction is not reported. It also claims its time complexity is the same as that of Smith-Waterman. However, because of extra computations required to reduce space, one would expect its time requirement would be higher than that of Smith-Waterman.

A recent method called GSAlign is the last method studied here. It is specifically designed for semi-optimal alignment of long Genome and DNA sequences [22]. Fundamentally, it is composed of three phases: seed identification and pairing of the two sequences' seeds, similar region identification by chaining seed pairs, and finally the local aligning of regions. To produce the overall alignment, local alignment of regions is joined together. It is capable to implement the alignment phase of the process in parallel using a multithreading technique. The authors claim that GSAlign is the fastest semi optimal aligner of long sequences. They also claim that the developed program produces perfect or nearly perfect precision and recalls on the identification of sequence variations in the dataset.

With the exception of Needleman-Wunsch method which is an optimal method for aligning genomic sequences, all the other methods we have introduced in this article are methods that do not guarantee to be optimal. We showed that the Needleman-Wunsch method cannot be used for large sequences. Therefore, efforts are being made to produce methods which require less running time and less memory consumption, and at the same time aligns the sequences more accurately. In the evaluation section, we will show that the proposed method is superior to the state-of-the art methods.

III. Problem Definition

Given two genomic sequences S1 and S2 composed of nucleotides A, C, G, and T are given. The objective is to globally align the two sequences in such a way that differences and similarities of the sequences are clearly recognizable. A scoring function will be defined and the alignment score is supposed to be optimal. However, for very long sequences this may not be possible, hence the aligner has to produce an alignment with and acceptable score. The length of the sequences is assumed to be very large to the point that their optimal alignment using Needleman-Wusch is practically impossible. The length of the sequences could even reach the length of human genome which is around 3.2 billion base pairs. Furthermore, it is assumed that the two sequences are very similar, for example one sequence is the reference genome of a healthy person and the other one is the genome of a person being studied to find its differences with that of the healthy person. It is expected that the method will use the optimal aligner in the cases where the input sequences are short or as a result of breaking them the parts which are to be aligned are short.

The approach is to divide the long input sequences into a number of corresponding sub-sequences and to align these sequences optimally. Finding the right breakpoints is a fundamental issue, and various methods to find the breaking point have been developed by previous researchers. It is important to note that the lengths of the corresponding sub-sequences are not necessarily equal. In this study, the longest common sub-sequence method [15] is used to break a given pair of long sequences into two pairs of sub-sequences and each pair is aligned separately. The process of breaking follows the divide and conquer approach, and continues the division until the pairs to be aligned are small enough.

IV. LCSDAC Implementation

In this section details of the implementation of the proposed Longest Common Subsequence Divide and Conquer (LCSDaC) alignment of two sequences is explained. The system will be able to align similar sequences of any sizes up to the length of a human genome, i,e., 3.2 Giga bps. If both sequences are short, i.e., less than 100 base pairs is assumed here, the Needleman-Wunsch will directly be utilized. Otherwise, the divide and conquer process will systematically break them into many pairs of short sequences. Figure 1 illustrates how this breaking process works. From the middle of each of the sequences a subsequence of length equal to the minimum of 1000 and one third of the current sequence's length, i.e., *Minimum (1000, n_1/3)*, is distinguished which become the input to the LCS procedure. This procedure will find their longest common subsequence. If this subsequence is long

[DOI: 10.52547/itrc.14.3.48]

enough the division is successful, otherwise the lengths of the distinguished subsequences are doubled and the LCS is called again. There is a maximum which is set to 3 for this step and in the worst case the longest common subsequence obtained in the third iteration is accepted as the breaking point. Figure 1 also shows the maximum number of times the LCS is called in each level of the tree.

Figure 2 has illustrated this procedure on a miniature pair of sequences. It is assumed that the first iteration of the procedure gives an acceptable result. In this example, the length of each of the sequences is 50. Although the length of sequences should be more than 100 to perform the division, for this example the division is applied. The LCS procedure will find the longest common subsequence to be GGAGCATGAGCTGG. It is located in Locations 17 to 30 of the first sequence and 19 to 32 of the second sequence. These places are assumed to be aligned in the final alignment of the two sequences and hence they are exempted from further processing. The algorithm remembers this alignment and includes it in the final alignment. In the second level of the tree of Figure 1 we will have to deal with two pairs of sequences to be processed. The first pair is in locations 1 to 16 of the first sequence and Locations 1 to 18 of the second sequence. The second pair is in locations 31 to 50 of the first sequence and Locations 33 to 50 of the second sequence. The division is not continued because the sequences are short. For these two pairs Needleman-Wunsch is directly applied.

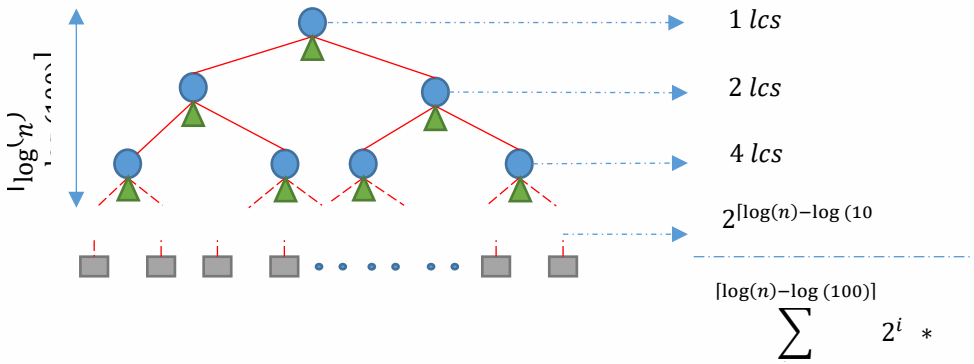


Fig. 1. The division process of LCSDaC showing the number of LCS calls.

```
1  2  3  4  5  6  7   8  9  10 11 12 13 14 15 16 17 18 19 20  21 22 23 24 25 26 27 28 29 30 31 32 33  34 35 36 37 38 39 40 41  42 43 44 45 46 47 48 49 50
CCTTTATCTAATCTTTGGAGCATGAGCTGGCATAGTTGGAACCGCCCTCA
CCTTTATGTAATCTTTGTGGAGCATGAGCTGGGAGTTGGACACGCCCTCA
```

Fig. 2. The effect of applying LCS on two sequences.

The overall pseudocode of the algorithm of the method presented here is shown in Algorithm 1.

Algorithm 1. The pseudocode for LCSDaC approach

```
1-  Input Sequences S1 and S2
2-  Global SA1, SA2   // the two sequences after alignment
3-  Recursive Procedure LCSDaC (S1, S2)
4-  {
5-          if (S1 < 100 OR S2 <100){
6-             SA (S1, S2) // Short Align and place results in SA1, SA2
7-                    Return
8-          }
9-          SELECT (s1, S1)        //s1 is selected from the middle of S1
10-         SELECT (s2, S2)
11-         LCS (s1, s2)      //find the LCS of s1 and s2
12-         S1=left section of S1 up to LCS start
13-         S2=left section of S2 up to LCS start
14-         LCSDaC (S1, S2)   //recursive call for new sequences S1 and S2
15-          Append (LCS)   //append LCS to current end of SA1, SA2
16-         S1=right section of S1 from end of LCS
17-         S2=right section of S2 from end of LCS
18-         LCSDaC (S1, S2)
19-  }
```


[DOI: 10.52547/itrc.14.3.48]

In Line 2, two new sequences which are the final results of the alignment are obtained. Their length is not necessarily equal to the length of the original sequences and may be a bit longer. The actual processing section, Lines 3 to 19, is declared as a recursive procedure. In this procedure, whenever one of the two inputs is short, i.e., less than 100 base pairs, Needleman-Wunsch is called to align the two sequences (Lines 5 to 8). The aligned results would be place in output sequences SA1 and SA2. In Lines 9 and 10, the SELECT procedure selects a subsequence from the middle of each of the sequences and then the longest common subsequence of the selected sequences are computed (Line 11). A similar procedure has to be applied to either sides of the anchor segments with the LCS being placed in the middle (Lines 12 to 18).

V. EVALUATION

For the alignment of long sequences, which is the interest of this research, both time and space complexities are two major limitations. Otherwise, Needleman-Wunsch produces the optimal alignment. Accuracy is often scarified to be able to obtain a approximate solution in tolerable amount of time and with the available storage capacity. We proceed first with the most important property of the algorithm which is time complexity.

A. Time complexity of Algorithm 1

Finding the time complexity of Algorithm 1 is highly dependent on the time needed to find the LCM of pairs of sequences. In [16]. It is calculated that if the dominant point approach is used the time needed to find the LCM of a pair of strings is proportional to $k^*|\sum|$, where $|\sum|$ represents the cardinality of the alphabet which strings are made of. On the other hand, k is the length of the longest string of the pair of strings. Note that, the sequences considered here consist of 4 characters A, T, G, and C. Therefore, it is safe to say calculation of LCM of a pair of strings is proportional to k. On the other hand, the number of times we can divide an integer n by 2 before the final result becomes less than 100 is $\log_2(n) - \log_2(100)$ or approximately $\log_2(n)$ - 6. This is the depth of the tree of Figure 1. Therefore, the number of times the LCS procedure is called is shown by Formula (1).

$$Number\ of\ LCS\ calls = \sum_{i=0}^{[log(n)-log(100)]} 2^i \quad (1)$$

Assuming the maximum length of the pair of sequences which is sent to the LCS procedure to be k_1, the number of operations needed to run the whole algorithm would satisfy Formula (2). In this formula *O(SA)* represents the time complexity of the Short Align (SA) procedure.

$$T(n) \le C_1 * k_1 \sum_{i=0}^{[log(n)-log(100)]} 2^i + 2^{log(n)+1}\, O(SA) \quad (2)$$

Or,

$$T(n) \le C_1 * k_1\, n + n * O(SA). \quad (3)$$

Recall that even if the length of the original sequences to be aligned is as large as a whole genome or any other longer length, in the algorithm, alignment for only short sequences with length less than 100 bps is called. Let's assume that the number of operations needed to do this alignment is *k*. It is usually very small compared to the length of the input sequences and it is bounded by a constant. However, it is not too small to satisfy the definition of the big O notation of the time complexity to consider it a constant. On the average, for alignment of short sequences there would be 50*50=2500 cells to fill. As this is represented by *k*, the time complexity of the method would be *O(kn)*. Of course, there is always an option to set the length of short sequences to be less than 100 bps.

There is a hidden benefit in the proposed method which is the exemption of longest common subsequences of each pair of sequences from any further processing. In the calculation of the time complexity, this is ignored because it does not affect the time complexity itself but reduces the hidden constant of the time complexity, and as a result it has a positive effect in processing time of the algorithm.

B. Space complexity of Algorithm 1

Space complexity of the alignment methods is as important as their time complexities. Some recent methods such as Sun et al. [13] have left the time complexity of the aligner untouched and have concentrated on reducing its space complexity. The actual space complexity of their proposed algorithm is not reported because, it is calculated for the worst case and in the worst case it would not be impressive. However, they claim the main memory of a "normal personal computer" would be able to align sequences as long as 100,000,000 nucleotides.

Here we explicitly express that the space complexity of our algorithm is *O(n)* where *n* is the length of longer input sequence. There are two input sequences of length say *n* and two output sequences of approximate length *n*, too. LCSDaC would need *4n* bytes to keep them all in the main memory. Within this algorithm, each Needleman-Wunsch execution will require at the most 10,000 cells and considering 4 bytes for each cell, adds up to 40,000 bytes. In the non-parallel version of LCSDaC, there would be only one running Needleman-Wunsch at any given time. Other minor memory requirements are ignorable. Therefore, the total memory space needed is expressed by Formula (4).

$$S(n) = 4n + 40000 \quad (4)$$

For sequences larger than 40000 bps, the space requirement would be

$$S(n) \le 5n \quad (5)$$

Therefore, the space complexity is $S(n) \in O(n)$. Even for the case of parallel implementation of the LCSDaC using multicores, the number of multicores is limited and it is small, say 16, which still leaves the space complexity to be *O(n)*. This is another achievement of the current research. Thus, it can be said that the proposed method is much more efficient than both

[DOI: 10.52547/itrc.14.3.48]

Needleman-Wunsch and Smith-Waterman methods with respect to both time and space complexities. Compared to newer algorithms, its superiority is at least in the hidden constants of time and space complexities. In any case, the actual time and space utilization of the proposed algorithm is less than each of the state of the art methods.

The method presented here has a high potential to be implemented in parallel. The simplest section that can be paralleled is the "S*hort align*" section of the algorithm which is responsible for alignment of short sequences. This section corresponds to the lowest level of the tree of Figure 1. Within each *short align* instance one can utilize General Purpose Graphic Processing Unit (GPGPU) to elevate the degree of parallelism. Furthermore, the LCS recognition can become parallel. For example, in Level 2 (third level) of the tree of Figure 1 four LCS instances could run in parallel. In this paper, the whole idea is developed sequentially and the compassions are with sequential competitors. Actually, we intended to put our idea into practice and evaluate the practicality of the method, first. The parallelization of the whole algorithm is left for future work.

For short sequences, there is no need for comparison, and the choice is definitely Needleman-Wunsch [8] with optimal alignment capability. For local optimal alignment, the choice is Smith-Waterman [12] for local optimal alignment. The latter method completely ignores many parts of the pair of sequences meaning. It only looks for those parts the sequences which are somewhat similar to each other. In some applications that parts which should be focused on are actually those which are very different. Those parts could actually be the cause of some diseases. For medium length sequences it depends on the available computer, its number of cores, number of General Purpose Graphic Process Units (GPGPUs), and the capabilities of the employed software program. For large and very large sequences the choice is definitely not Needleman-Wunsch or Smith-Waterman. A practical choice is an efficient heuristic semi-optimal methods. Therefore, two such recent methods are selected for the comparison part: MUMmer [11], and GSAlign [22]. The experiment includes both short genomes and long ones.

BRCA1 is a human gene responsible for suppressing tumors and repairing DNA, ATF6 is a human gene which acts as a transcription factor inside the nucleus, and CFTR gene that is the provider of instructions for making a kind of protein. The approximate sizes of these genes are expressed in thousands (K) of nucleotides in Table 1. These genes are taken from 1000 genomes project dataset [23]. Escherichia coli (E.Coli), Shigella, and Salmonella are three bacteria with the approximate sizes that are expressed in millions (M) of nucleotides in this table. The bacteria sequences are taken from NCBI site [24]. For each gene and bacteria two different variants are selected to be aligned.

The computer used for the experiments is Intel(R) core(TM) i7-353U CPU 2GHz, RAM 6GB, and Linux Ubuntu 18.0 operating system.

Table 1 summarizes the result of comparisons of MUMmer, GSAlign, and LCSDac methods.

TABLE I. SUMMARY OF THE TIMING COMPARISON RESULTS

Sequence → Method↓	BRCA1 #127K	ATF6 #198K	CFTR #430K	E.coli #5M	Shigella #5M	Sal* #4.9M
MUMmer	5s	5s	9s	328s	350s	246s
GSAlign	2s	3s	4s	125s	240s	160s
LCSDaC	1s	2s	4s	138s	218s	127s

*Salmonella

The overall results of Table 1 for the six tested sequences, show that, on the average, LCSDaC is 2.61 times faster than MUMmer, and 1.29 times faster than GSAlign. For example in comparing MUMmer and LCSDaC the following computation is used.

$$({}^{5}/_{1} + {}^{5}/_{2} + {}^{9}/_{4} + {}^{328}/_{138} + {}^{330}/_{218} + {}^{246}/_{127})/6 = 2.61$$

Another major area of comparison is the accuracy of the methods. It is obvious that Needleman-Wunsch is the most accurate one because it is an optimal aligner. The problem arises when the sequences are long and its time and space requirements using Needleman-Wunsch is absolutely intolerable. Smith-Waterman is not an optimal aligner but, it is a locally optimal one. MUMmer, GSAlign, and LCSDaC fall into this category and none of them could be used as a fully accurate one. In the absence of an optimal alignment the number of exact matches of the two sequences are taken to be a measure for the purpose of correctness evaluations. A higher value of this measure is interpreted as the method being more accurate. Table 2 shows that the method presented in this paper is more accurate than others, in all cases. Therefore, the Relative Accuracies (RA) of other methods are computed in comparison to LCSDaC.

Evaluating their accuracy in terms of score, precision, recall, and F-measure requires extensive experiments on numerous sequences which is left for the future work.

TABLE II. RELATIVE ACCURACY COMPARISON RESULTS

Sequence→ Method↓	BRCA1 #127K	ATF6 #198K	CFTR #430K	E.coli #5M	Shigella #5M	Sal* #4.9M
MUMmer	103521	15250	36624	281466	250537	249410
GSAlign	98531	12895	32358	245326	237856	210856
LCSDaC	123087	19727	42144	295584	295876	275470

* Salmonella

Therefore, the average relative accuracies of other methods are computed in comparison to LCSDaC. Details of calculations for MUMmer is shown in the following.

$$\mathrm{RA}_{\mathrm{MUMmer}} = (103521/123087 + 152501/197277 + 366248/421446 + 2814666/2955849 + 2505372/2958760 + 2494108/2754703)/6 = 0.864$$

Performing a similar calculation for GSAlign evaluates its relative accuracy to be $RA_{GSALign} = 0.6554$.

[DOI: 10.52547/itrc.14.3.48]

Therefore the accuracy of LCSDaC is the highest and that of MUMmer is 86 percent of LCSDaC. With respect to relative accuracy, the GSAlign is the lowest with 66 percent of the LCSDaC.

VI. Conclusion and Future Work

In this paper, we introduced LCSDaC which is a novel long DNA sequence aligner base on divide and conquer approach in which, the division takes place along the longest common subsequence of the middle portions of the current sequences. The time complexity of the method is analyzed and it is shows to be superior to traditional aligners. The space complexity of the algorithm is also calculated to be *O(n)* which is superior to all classic aligners such as Needleman-Wunsch and Smith-Waterman methods. Further, it outperforms all state of the art methods. The sequential version of the presented method is implemented and it is compared against two state of the art heuristic aligners called MUMmer, and GSAlign. It is shown that on the average, the proposed algorithm, LCSDaC, is 2.61 times faster than MUMmer, and 1.29 times faster than GSAlign. For the accuracy we showed that the accuracy of MUMmer is 86 percent of LCSDaC and that of GSAlign is 66 percent of the LCSDaC.

References

[1] H. Chial, "DNA sequencing technologies key to the Human Genome Project," Nat. Educ., vol. 1, no. 1, p. 219, 2008.," *Nature Education*, vol. 1, no. 1, p. 219, 2008.

[2] J. Lever, E. Zhao, J. Grewal, M. Jones and S. Jones, "CancerMine: a literature-mined resource for drivers, oncogenes and tumor suppressors in cancer," *Nature Methods*, vol. 16, pp. 505-507, 2019.

[3] I. Kovtun and C. McMurray, "Features of trinucleotide repeat instability in vivo.," *Cell Research*, vol. 18, pp. 198-213, 2008.

[4] B. Langdahl, E. Løkke, M. Carstens, L. Stenkjaer and E. Eriksen, "Osteoporotic Fractures Are Associated with an 86-Base Pair Repeat PolymCorphism in the Interleukin-1–Receptor Antagonist Gene But Not with olymorphisms in the Interleukin-1b Gene," *Bone Min. Res.*, vol. 15, no. 3, pp. 402-414, 2000.

[5] Y. Deng, S. Kumar and W. Byrne, "Y. Deng, S. Kumar, and W. Byrne, "Segmentation and alignment of parallel text for statistical machine translation," *Nat. Lang. Eng.*, vol. 13, no. 3, pp. 235-260, 2007.

[6] P. Neamatollahi, M. Hadi and M. Naghibzadeh, "Simple and Efficient Pattern Matching Algorithms for Biological Sequences," *IEEE Access*, vol. 8, pp. 23838 - 23846, 2020.

[7] R. L. E. Bellman, "History and development of dynamic programming," *IEEE Control Systems Magazine*, pp. 24 - 28, 1984.

[8] S. Needleman and C. Wunsch, "A general method applicable to the search for similarities in the amino acid sequence of two proteins," *Journal of Molecular Biology*, pp. 443-453, 1970.

[9] M. Naghibzadeh, New Generation Computer Algorithms, Available on Amazon, 2021.

[10] C. Leimeister, T. Dencker and B. Morgenstern, "Accurate multiple alignment of distantly related genome sequences using filtered spaced word matches as anchor points," *Bioinformatics*, vol. 35, no. 2, pp. 211-218, 2019.

[11] G. MarcËais, A. L. Delcher, A. M. Phillippy, R. Coston, S. L. Salzberg and A. Zimin, "G. MarcËais, A. L. Delcher, A. M. Phillippy, R. Coston, S. L. Salzberg, and A. Zimin, "MUMmer4: A fast and versatile genome alignment system," *PLOS Comput. Biol.*, 2018.

[12] T. W. M. Smith, "Identification of common molecular subsequences," *Journal of Molecular Biology*, pp. 195-197, 1981.

[13] J. Sun, K. Chen and Z. Hao, "Pairwise alignment for very long nucleic acid sequences," Biochem Biophys Res Commun," vol. 502, no. 3, pp. 313-317, 2018.

[14] M. Zhao, W. Lee, E. P. Garrison and G. T. Marth, "SSW library: an SIMD Smith-Waterman C/C++ library for use in genomic applications," *PLoS One*, vol. 8, no. 12, 2013.

[15] M. Brudno, C. Do, C. G. M. M. F. Kim and et.al., "LAGAN and Multi-LAGAN: Efficient Tools for Large-Scale Multiple Alignment of Genomic DNA," *Genome Res.*, vol. 13, no. 4, pp. 721-731, 2003.

[16] Q. Wang, D. Korkin and Y. Shang, "A Fast Multiple Longest Common Subsequence (MLCS) Algorithm," *IEEE Trans. Knowl. Data Eng.*, vol. 23, no. 3, pp. 321-334, 2011.

[17] A. Mir, M. Naghibzadeh and N. Saadati, "INDEX: Incremental depth extension approach for protein–protein interaction networks alignment," *BioSystems*, vol. 162, pp. 24-34, 2017.

[18] B. Chowdhury and G. Garai, "A review on multiple sequence alignment from the perspective of genetic algorithm," *Genomics*, vol. 109, no. 5-6, pp. 419-431, 2017.

[19] S. F. Altschul, W. Gish, W. Miller, E. W. Myers and D. J. Lipman, "Basic local alignment search tool," *J. Mol. Biol.*, vol. 215, no. 3, pp. 403-410, 1990.

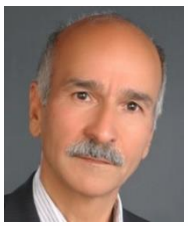
[20] S. Schwartz, W. J. S. A. Kent, Z. Zhang, R. Baertsch and et.al., "Human-mouse alignments with BLASTZ," *Genome Res.*, vol. 13, no. 1, pp. 103-107, 2003.

[21] S. S. S. Angiuoli, "S. V Angiuoli and S. L. Salzberg, "Mugsy: fast multiple alignment of closely related whole genomes," *Bioinformatics*, vol. 27, no. 3, pp. 334-342, 2011.

[22] H. Lin and W. L. Hsu, "GSAlign: an efficient sequence alignment tool for intra-species genomes," *BMC genomics*, vol. 21, no. 1, pp. 1-10, 2020.

[23] IGSR: The International Genome Sample Resource, [Online]. Available: https://www.internationalgenome.org/data/. [Accessed July 2020].

[24] National Center for Biotechnology Information, [Online]. Available: http://www.ncbi.nlm.nih.gov/. [Accessed July 2019].



Mahmoud Naghibzadeh completed his M.Sc. and Ph.D. in Computer Science at the University of Southern California (USC). He has published about 300 scientific research articles, 10 Persian books, and 2 internationally available English text books.

Samira Babaei received her M.Sc. degree in Computer Science from Ferdowsi University of Mashhad in 2021, Iran. Her main research interests include Algorithms and Bioinformatics.

[DOI: 10.52547/itrc.14.3.48]

Behshid Behkamal received her Ph.D. degree in Computer Engineering in 2014. Currently, she is an Assistant Professor at Computer Engineering Department of Ferdowsi University of Mashhad. She works in the area of Data Science, Specifically on Data Quality Assessment and Metrics.

Mojtaba Hatami is a Ph.D. Candidate in Computer Science at Ferdowsi University of Mashhad. His current research interests include the scheduling aspects of Real-Time Systems, Cloud, Multiprocessors, Multicores and IoT. He is also interested in Randomize Algorithms and Complexity Theory.

[DOI: 10.52547/itrc.14.3.48]