Computational methods in bioinformatics

Lecture 2

Dotplots

A pictorial representation of the similarity between two sequences.

Compare a sequence with itself:
- Repeats
- Palindromic sequences

Compare two sequences:
- Any path from upper left to lower right represents an alignment.
- Horizontal or vertical moves correspond to gaps in one of the sequences.
- Path with highest score corresponds to an optimal alignment.

Measures of sequence similarity

Hamming distance:
- Number of positions with mismatching characters.
- Defined for two strings of equal length.

```
agtc
cgta
```

Levenshtein distance:
- Minimum number of edit operations (delete, insert, change a single character) needed to change one sequence into another.

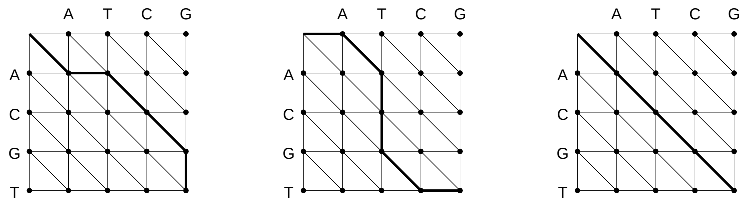
```
agtcc
cgctca
```

Dotplots

	A	G	C	T	A	G	G	A
C			●					
A	●				●			●
C			●					
T				●				
A	●				●			●
G		●				●	●	
G		●				●	●	
C			●					

	A	G	C	T	A	G	C	G	A
A	●				○				○
G		●				○		○	
C			●				○		
A	○				○				○
T				●					
A	○				●				○
G		○				●		○	
G		○				○		●	
A	○				○				●

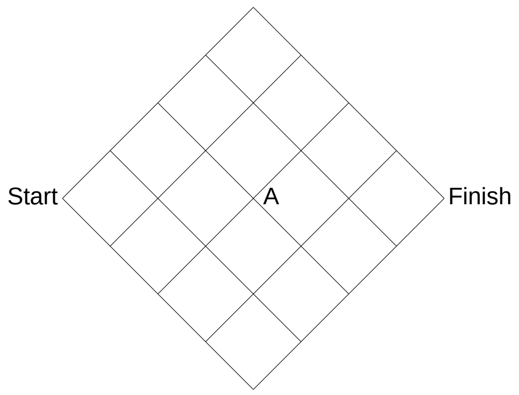
Each path represents an alignment



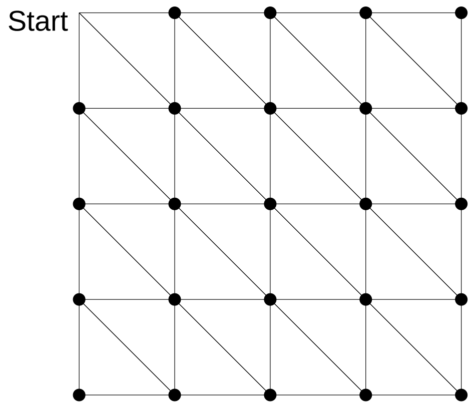
```
A-CGT        -ACGT-        ACGT
|  ||                      |
ATCG-        AT--CG        ATCG
```

- Vertical steps add a gap to the horizontal sequence
- Horizontal steps add a gap to the vertical sequence

Do we have to enumerate all paths?



How many paths?



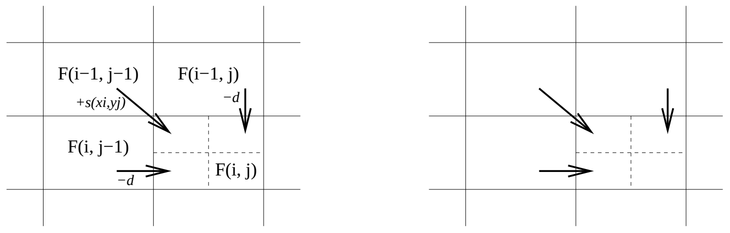
Pairwise global alignment (Needleman-Wunsch algorithm)

Rigorous algorithms use dynamic programming to find an optimal alignment.

- match score
- mismatch score
- gap penalty

$$F(i, j) = \max \begin{cases} F(i-1, j-1) + s(x_i, y_j) \\ F(i-1, j) - d \\ F(i, j-1) - d \end{cases}$$

Dynamic programming



Percent identity

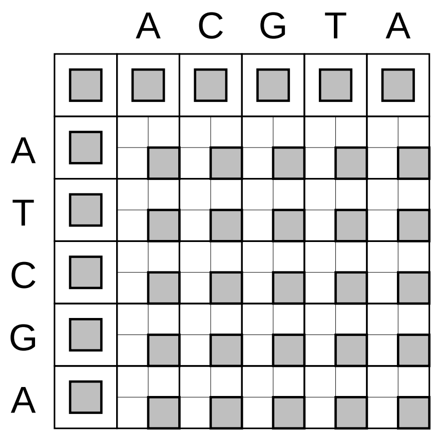
Having obtained an alignment, it is common to quantify the similarity between a pair of sequences by stating the percent identity.

```
-ACGATAG-CGAAACCAAAA
 ||| ||| |||    |
CAGC-TAGCCGATGTC----
```

Count the number of alignment positions with matching characters and divide by ... *what*?

- the length of the shortest sequence?
- the length of the alignment?
- the average length of the sequences?
- the number of non-gap positions?
- the number of equivalenced positions excluding overhangs?

Score matrix



Pairwise local alignment (Smith-Waterman algorithm)

Local similarities may be masked by long unrelated regions.

A minor modification to the global alignment algorithm.

- If the score for a subalignment becomes negative, set the score to zero.

$$F(i, j) = \max \begin{cases} 0 \\ F(i-1, j-1) + s(x_i, y_j) \\ F(i-1, j) - d \\ F(i, j-1) - d \end{cases}$$

- Trace back from the position in the score matrix with the highest value.

- Stop at cell where score is zero.

global_alignment.c

```c
#include <stdio.h>

#define MAX_LENGTH     100

#define MATCH_SCORE      2
#define MISMATCH_SCORE  -1
#define GAP_PENALTY      2

#define STOP             0
#define UP               1
#define LEFT             2
#define DIAG             3


main()
{
    int  i, j;
    int  m, n;
    int  alignmentLength, score, tmp;
    char X[MAX_LENGTH+1] = "ATCGAT";
    char Y[MAX_LENGTH+1] = "ATACGT";

    int  F[MAX_LENGTH+1][MAX_LENGTH+1]; /* score matrix */
    int  trace[MAX_LENGTH+1][MAX_LENGTH+1];
    char alignX[MAX_LENGTH*2];   /* aligned X sequence */
    char alignY[MAX_LENGTH*2];   /* aligned Y sequence */


    /*
     * Find lengths of (null-terminated) strings X and Y
     */
    m = 0;
    n = 0;
    while ( X[m] != 0 ) {
        m++;
    }
    while ( Y[n] != 0 ) {
        n++;
    }


    /*
     * Initialise matrices
     */

    F[0][0] = 0;
    trace[0][0] = STOP;
    for ( i=1 ; i<=m ; i++ ) {
        F[i][0] = F[i-1][0] - GAP_PENALTY;
        trace[i][0] = STOP;
    }
    for ( j=1 ; j<=n ; j++ ) {
        F[0][j] = F[0][j-1] - GAP_PENALTY;
        trace[0][j] = STOP;
    }



    /*
     * Fill matrices
     */

    for ( i=1 ; i<=m ; i++ ) {

        for ( j=1 ; j<=n ; j++ ) {

            if ( X[i-1]==Y[j-1] ) {
                score = F[i-1][j-1] + MATCH_SCORE;
            } else {
                score = F[i-1][j-1] + MISMATCH_SCORE;
            }
            trace[i][j] = DIAG;

            tmp = F[i-1][j] - GAP_PENALTY;
            if ( tmp>score ) {
                score = tmp;
                trace[i][j] = UP;
            }

            tmp = F[i][j-1] - GAP_PENALTY;
            if( tmp>score ) {
                score = tmp;
                trace[i][j] = LEFT;
            }

            F[i][j] = score;
         }
    }


    /*
     * Print score matrix
     */

    printf("Score matrix:\n      ");
    for ( j=0 ; j<n ; ++j ) {
        printf("%5c", Y[j]);
    }
    printf("\n");
    for ( i=0 ; i<=m ; i++ ) {
        if ( i==0 ) {
            printf(" ");
        } else {
            printf("%c", X[i-1]);
        }
        for ( j=0 ; j<=n ; j++ ) {
            printf("%5d", F[i][j]);
        }
        printf("\n");
    }
    printf("\n");



    /*
     * Trace back from the lower-right corner of the matrix
     */

    i = m;
    j = n;
    alignmentLength = 0;

    while ( trace[i][j] != STOP ) {

        switch ( trace[i][j] ) {

            case DIAG:
                alignX[alignmentLength] = X[i-1];
                alignY[alignmentLength] = Y[j-1];
                i--;
                j--;
                alignmentLength++;
                break;

            case LEFT:
                alignX[alignmentLength] = '-';
                alignY[alignmentLength] = Y[j-1];
                j--;
                alignmentLength++;
                break;

            case UP:
                alignX[alignmentLength] = X[i-1];
                alignY[alignmentLength] = '-';
                i--;
                alignmentLength++;
        }
    }


    /*
     * Unaligned beginning
     */

    while ( i>0 ) {
        alignX[alignmentLength] = X[i-1];
        alignY[alignmentLength] = '-';
        i--;
        alignmentLength++;
    }

    while ( j>0 ) {
        alignX[alignmentLength] = '-';
        alignY[alignmentLength] = Y[j-1];
        j--;
        alignmentLength++;
    }


    /*
     * Print alignment
     */

    for ( i=alignmentLength-1 ; i>=0 ; i-- ) {
        printf("%c",alignX[i]);
    }
    printf("\n");
    for ( i=alignmentLength-1 ; i>=0 ; i-- ) {
        printf("%c",alignY[i]);
    }
    printf("\n");

    return(1);
}
```