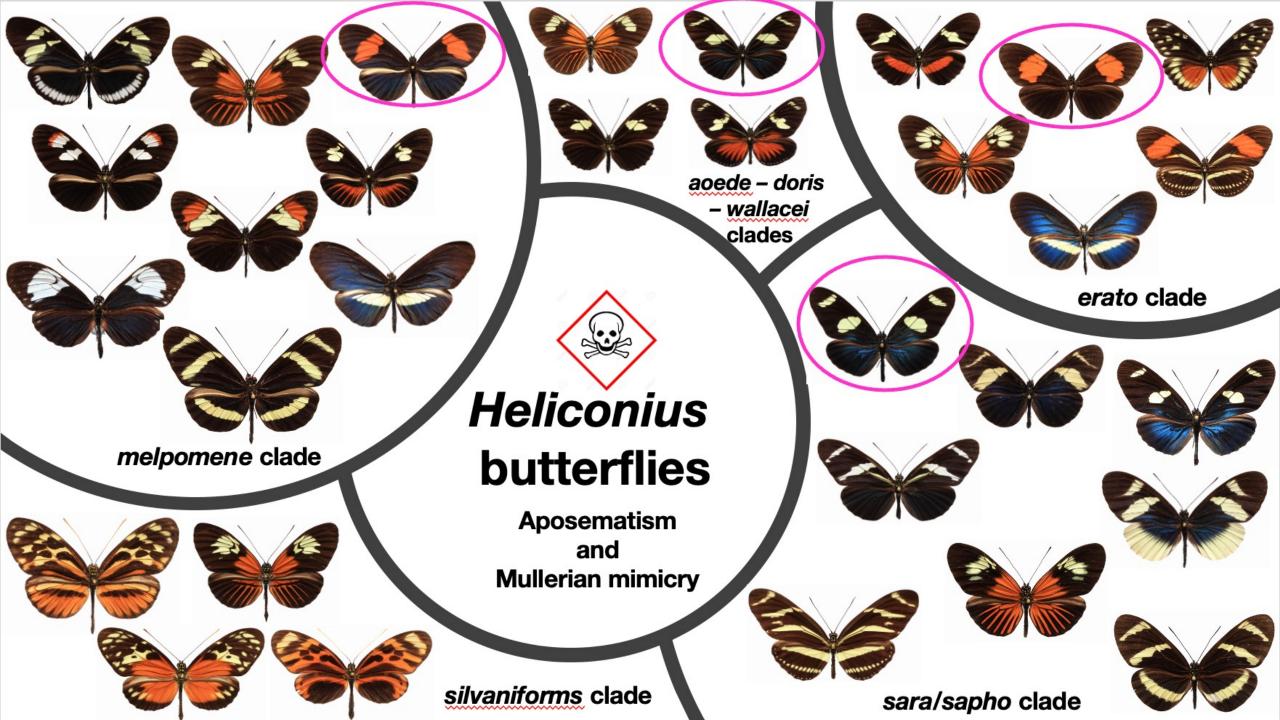
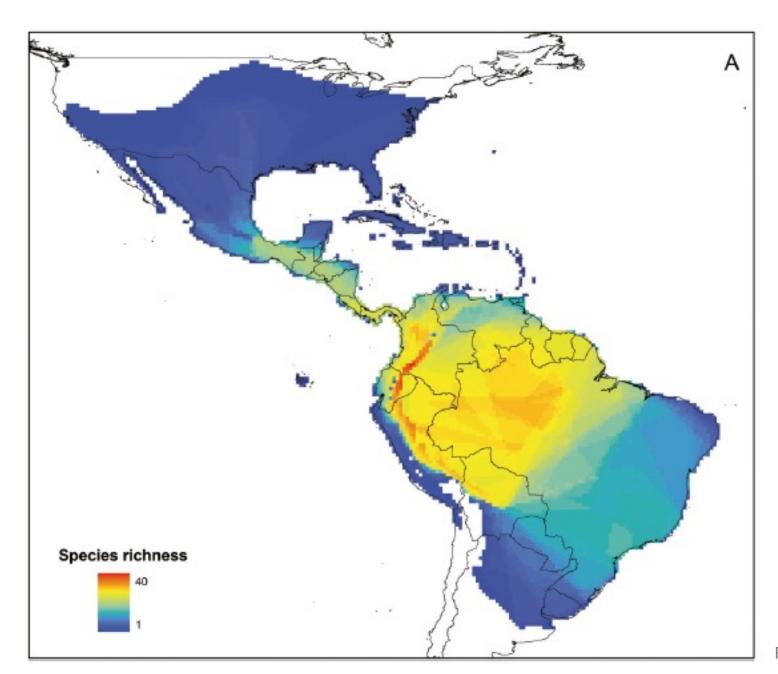
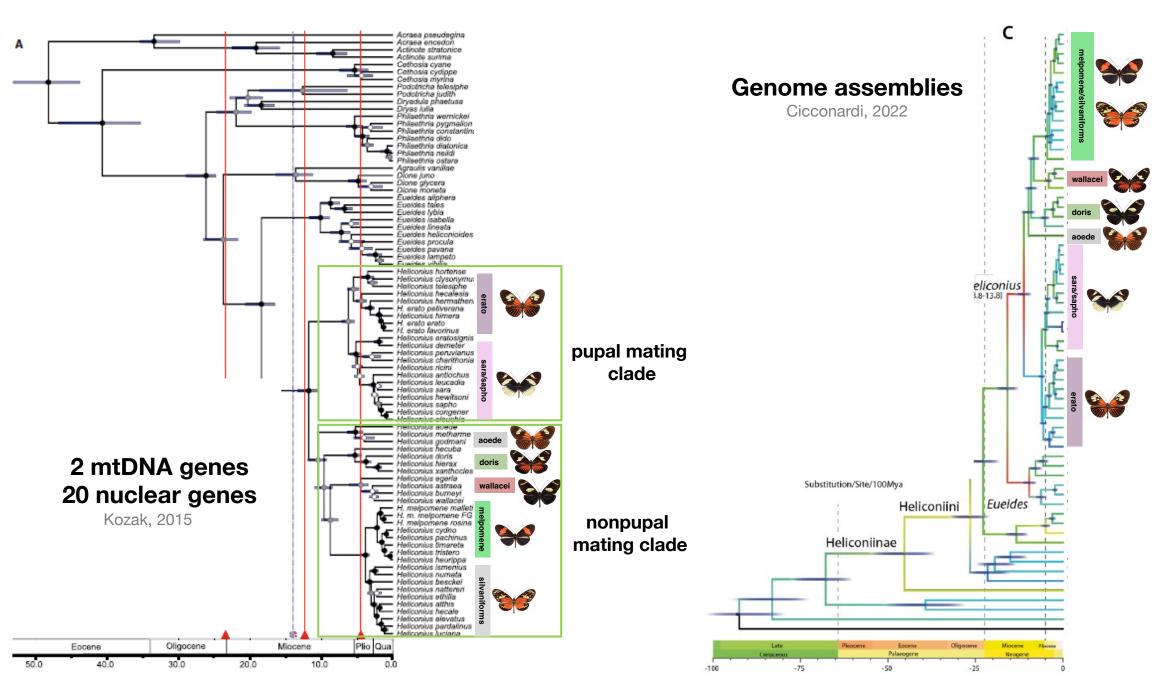
Heliconius butterflies Biodiversity genomics course Tena-Ecuador 2024



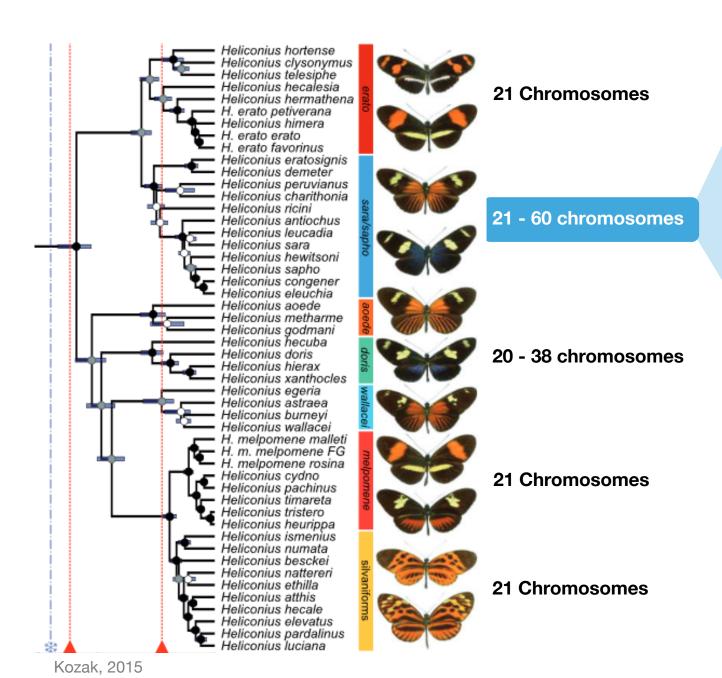
Introduction

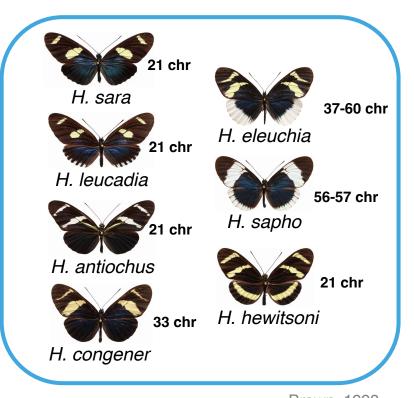


Introduction



Introduction



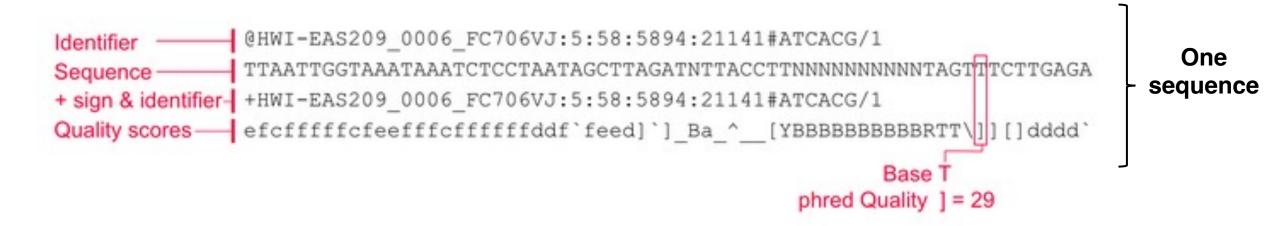


Brown, 1992

Raw sequences files and quality control

Fastq Format

This format is designed to handle base quality metrics output from sequencing machines.



Line 1 begins with the '@' character and is followed by a sequence identifier and an optional description.

Line 2 is the sequence letters.

Line 3 begins with a '+' character; it marks the end of the sequence and is optionally followed by the same sequence identifier again in line 1.

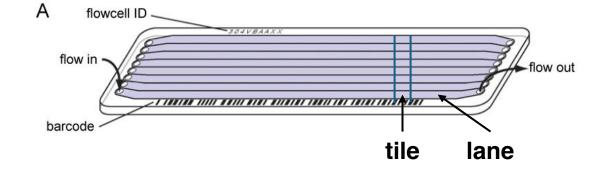
Line 4 encodes the quality values for the sequence in Line 2, and must contain the same number of symbols as letters in the sequence.

Read header

Colon

@EAS139:136:FC706VJ:2:2104:15343:197393 1:Y:18:ATCACG

| EAS139 | the unique instrument name | |
|---------|--|--|
| 136 | the run id | |
| FC706VJ | the flowcell id | |
| 2 | flowcell lane | |
| 2104 | tile number within the flowcell lane | |
| 15343 | 'x'-coordinate of the cluster within the tile | |
| 197393 | 'y'-coordinate of the cluster within the tile | |
| 1 | the member of a pair, 1 or 2 (paired-end or mate-pair reads only) | |
| Y | Y if the read is filtered, N otherwise | |
| 18 | 0 when none of the control bits are on, otherwise it is an even number | |
| ATCACG | index sequence | |
| | | |



Quality scores

```
@EAS139:136:FC706VJ:2:2104:15343:197393 1:Y:18:ATCACG
CCGTCAATTCATTAGTTTTTAACCTTTGCGGCCGTACTCCCCAGGCGGT
+
AAAAAAAAAAAAAAA:9@::::??@@::FFAAAAAACCAA::::BB@@?A?
```

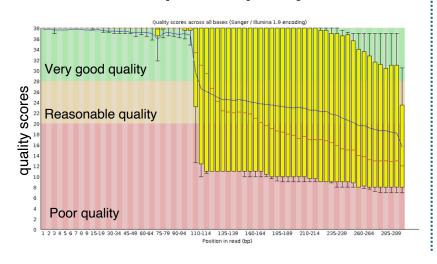
Quality score: ASCII encoding

| 40:0 | 90:Z | 141 : a |
|------|------|----------------|
| 41:A | 91:[| 142:b |
| 42:B | 92:\ | 143:c |
| 43:C | 93:] | 144:d |
| 44:D | 94:^ | 145 : e |
| 45:E | 95:_ | 146:f |
| • | : | : |

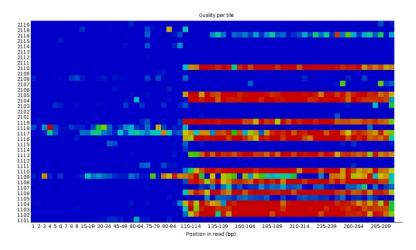
| Quality Score | Probability of incorrect base call | Base call accuracy |
|---------------|------------------------------------|--------------------|
| 10 | I in 10 | 90% |
| 20 | I in 100 | 99% |
| 30 | I in 1000 | 99.9% |
| 40 | I in 10000 | 99.99% |

Assess quality with FastQC → .html ¥

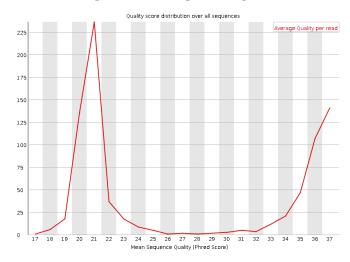
Per base sequence quality



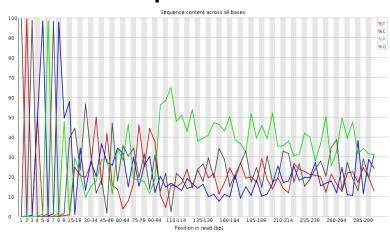
Per tile sequence quality



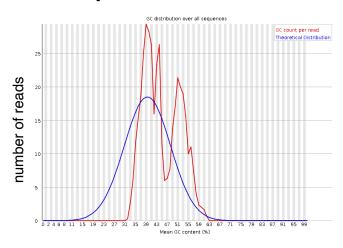
Per sequence quality scores



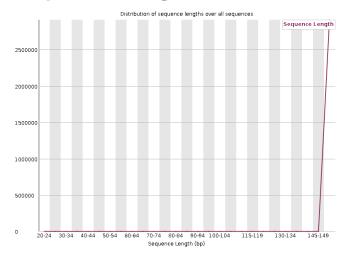
Per base sequence content



Per sequence GC content



Sequence length distribution



Let's have a look at the first few sequences and check the sequencing quality with fastqc