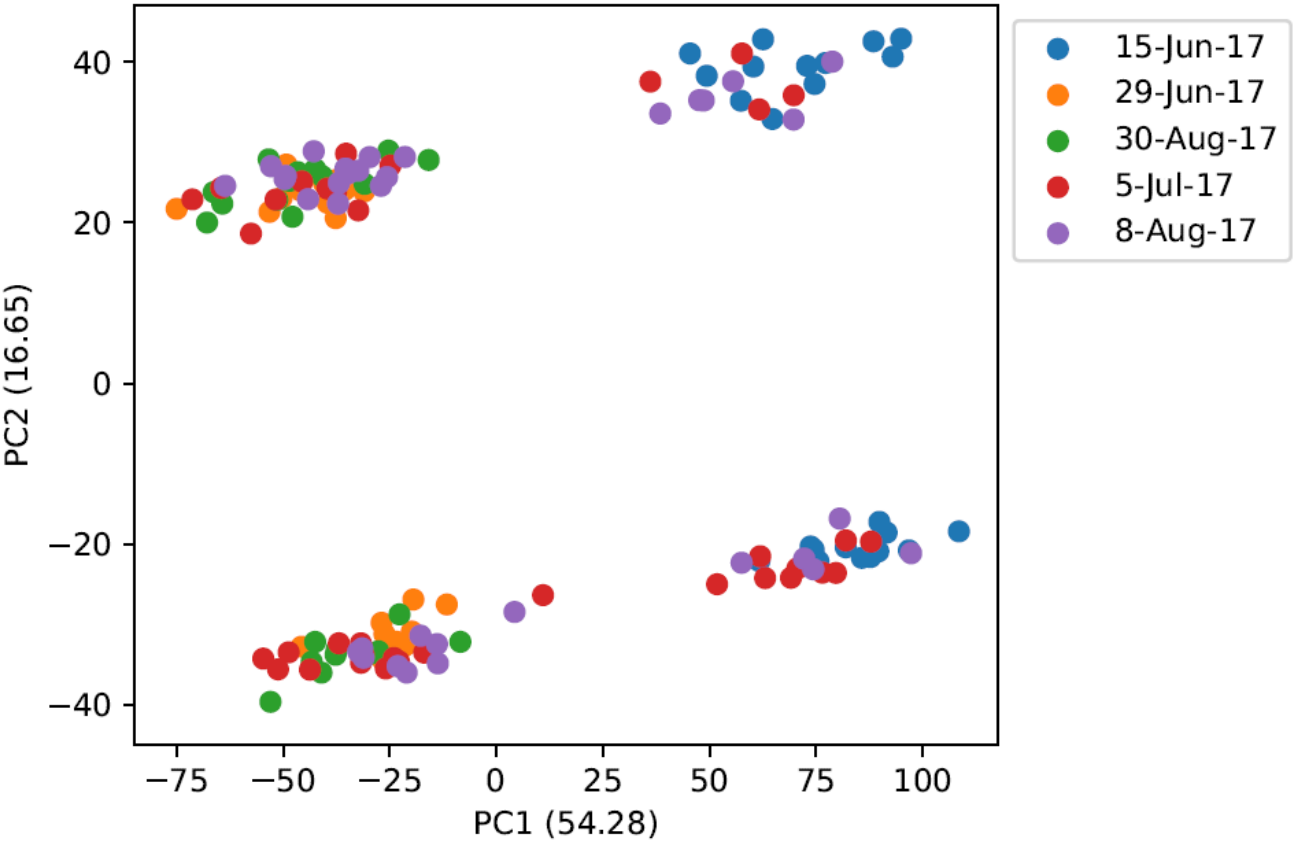
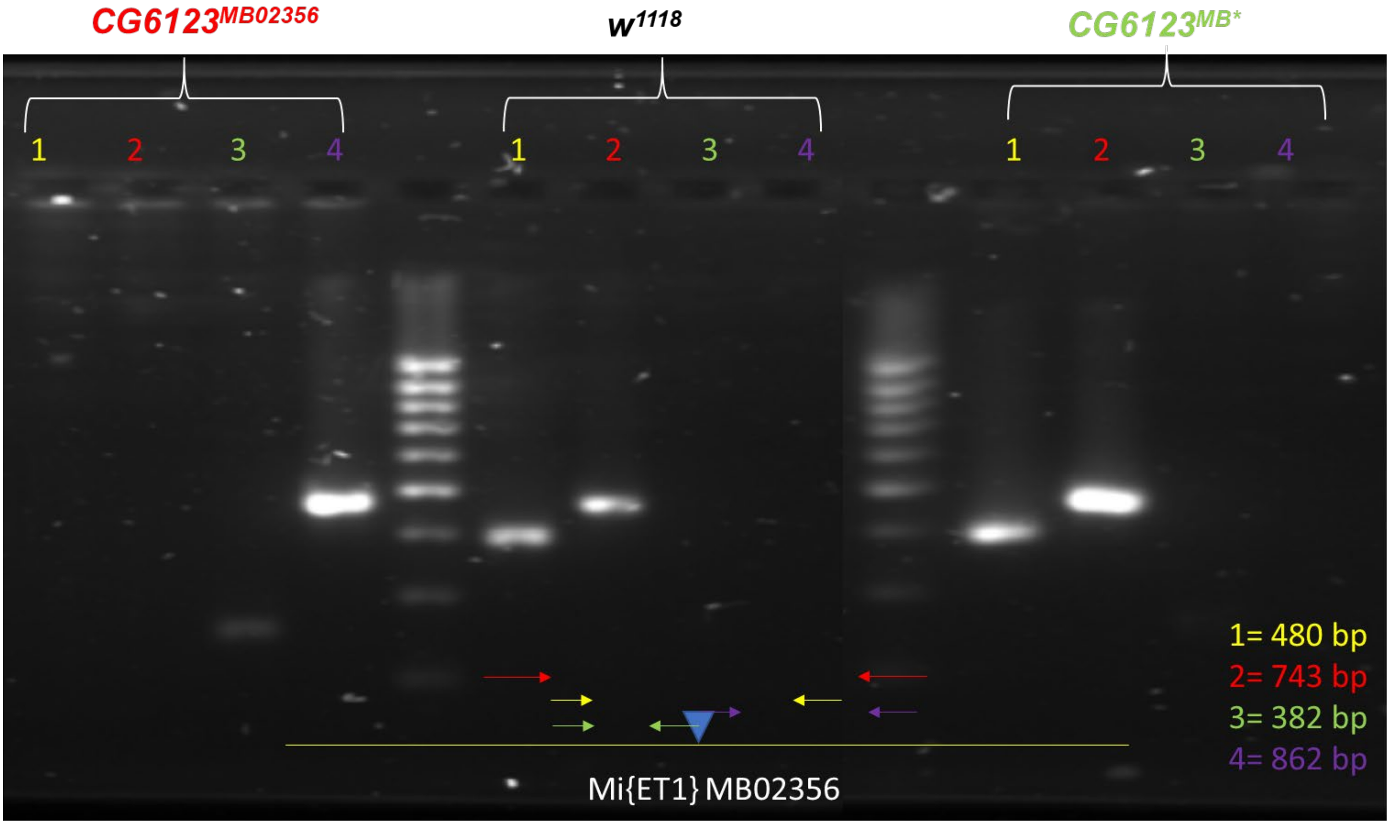
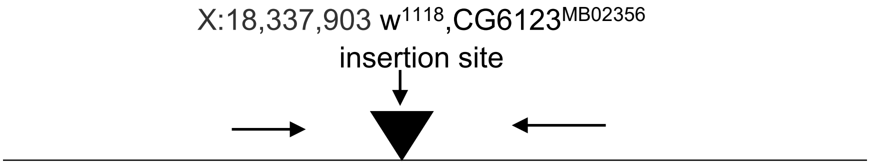


Supplemental Fig S1. Principal component analysis reveals a bias in the data according to the date of the sequencing run.



Supplemental Fig S2. **Agarose gel showing genotyping strategy used for determining the presence of *Minos* insertion in the *CG6123* genomic region as well as its precise excision in the rescued line. Yellow and red denote the oligos used for PCR amplification flanking the *Minos* insertion whereas green and purple arrows denote the oligos used for checking the presence of the *Minos* insertion in the original *CG6123*$^{\text{MB02356}}$ mutant.**



>gnl|dmel|X type=golden_path_region; loc=X:1..23542271; ID=X; dbxref=GB:AE014298, GB:AE014298, REFSEQ:NC_004354; MD5=f7228bbf9f8fa0af457bc2fb7e396e4d; length=23542271; release=r6.30; species=Dmel;
Length = 23542271

HSP # = 1 , **Score =** 860.835 bits (434) , **Expect =** 0
Identities = 587 / 632 (92.9%) , **Positives =** 587 / 632 (92.9%) , **Gaps =** 11 / 632 (1.7%)
Strand = Plus / Minus

```
Query:   21          ATGTGGTGGAA-CTTTTGCATACTAGGCTCCAAGGGA--GGGGGGCTGATGATGAAAACA    77
                     ||||||||||| ||||||||||||||| ||||||| |  |||||||||||||||||||||
Subject: 18338392    ATGTGGTGGAAACTTTTGCATACTAGGGTCCAAGGAATGGGGGGCTGATGATGAAAACA    18338333

Query:   78          TATCGAATCGAATTTAAAGCTAACAAGGTCCACTCTAAAAGCTGNNNNNNNNNNNNNNNN    137
                     ||||||||||||||||||||||||||||||||||||||||||||
Subject: 18338332    TATCGAATCGAATTTAAAGCTAACAAGGTCCACTCTAAAAGCTGAAAAAAAAAAAAAAAC    18338273

Query:   138         -TACCGATGGGACAAAACGACAAGCCGAAACTTAATTATGCGATTTTATGGGGGTATTTG    196
                      ||||||||| ||||||||||||||||| ||||||||||||||||||||| |||||||||
Subject: 18338272    ATACCGATGGCACAAAACGACAAGCCGAGACTTAATTATGCGATTTTATGTGGGTATTTG    18338213

Query:   197         CACAAAAATAATTGTATTGATTGCCCCCCTCTGCGAGTCTCAAGTTTTCCCTGGAACTTT    256
                     |||| ||||||||||||||||||||| |||||||||||||||||||||| ||||||||||
Subject: 18338212    CACAGAAATAATTGTATTGATTGCCCGCCTCTGCGAGTCTCAAGTTTTCACTGGAACTTT    18338153

Query:   257         CAACTTTGATCAAAAGCAGCTGCGCGATGAAAATCCGCTCCGGCATTAAGGGGCTTTTCC    316
                     |||||||||||||||||||||||||||||| |||||||||||||||||||||||||||||
Subject: 18338152    CAACTTTGATCAAAAGCAGCTGCGCGATGATAATCCGCTCCGGCATTAAGGGGCTTTTCC    18338093

Query:   317         CAGTAAATGTTTAATTAAATAAACAAATCAAATCTGACGCGGCGGAACACCCACATCCGT    376
                     ||||||||||||||||| |||||||||||||||||||||||||||| |||||||||||||
Subject: 18338092    CAGTAAATGTTTAATTAGATAAACAAATCAAATCTGACGCGGCGGAGCACCCACATCCGT    18338033

Query:   377         CGACCCGACGACCCGACGACCCGGAAGCCATGGGGTTCGCATACCCATCCGTATCCGTAT    436
                     |||||||||||||||||||||||||||||||| ||||||||||| |||||||||||||||
Subject: 18338032    CGACCCGACGACCCGACGACCCGGAAGCCATGTGGTTCGCATACGCATCCGTATCCGTAT    18337973

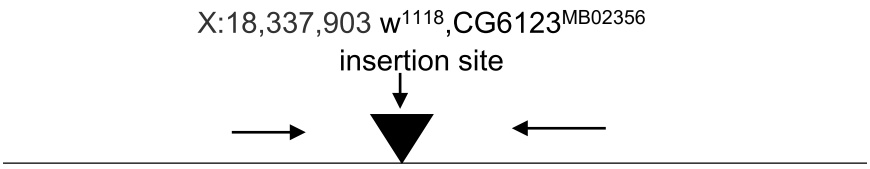
Query:   437         CCGCATCTGTATTCCCATCCGGATCCGTATTCGGTTGCGTACCAGGGGATGGCATCGTAT    496
                     |||||||||||||| ||||||||||||||||||||||||||||||| |||||||||||||
Subject: 18337972    CCGCATCTGTATTCGCATCCGGATCCGTATTCGGTTGCGTACCAGGTGATGGCATCGTAT    18337913

Query:   497         GGATGTACATATGTAAATGCATTAATTGCGGCACCCCCAACTTCGTTAA--TAACAATTA    554
                     ||||||||||||||| ||||||||||||||||||| |||||||||||||   ||||||||
Subject: 18337912    GGATGTACATATGTAGATGCATTAATTGCGGCACCACCAACTTCGTTAAATAAACAATTA    18337853

Query:   555         ATTAATGGCTGTGGGGAGTGACCGAGCATTATGAATGAT-AATCATGCATG-TTCACTGC    612
                     ||||||||||||||||||||||||||||||||||||||| ||||||||||| ||||||||
Subject: 18337852    ATTAATGGCTGTGGGGAGTGACCGAGCATTATGAATGATAAATCATGCATGTTTCACTGC    18337793

Query:   613         TGA-TATTT-CCAATGA-TCTATGTATATTTA    641
                     ||| ||||| ||||||| ||||||||||||||
Subject: 18337792    TGATTATTTCCCAATGATTCTATGTATATTTA    18337761
```

Supplemental Fig S3. Top panel shows the presence of the *Minos* insertion in the *CG6123* genomic region. Black flanking arrows denote the oligos used for Sanger based sequencing. Lower panel shows the snapshot of nucleotide blast of *w*^{1118} control line. The transparent red box denotes the location of the *Minos* insertion in the original mutant of *CG6123*.



>gnl|dmel|X type=golden_path_region; loc=X:1..23542271; ID=X; dbxref=GB:AE014298, GB:AE014298, REFSEQ:NC_004354; MD5=f7228bbf9f8fa0af457bc2fb7e396e4d; length=23542271; release=r6.30; species=Dmel;
Length = 23542271

HSP # = 1 , **Score =** 448.505 bits (226) , **Expect =** 1.18976e-124
Identities = 431 / 511 (84.3%) , **Positives =** 431 / 511 (84.3%) , **Gaps =** 4 / 511 (0.8%)
Strand = Plus / Minus

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Query:    23        ATGTGGTGGAA-CTTTTGCATACTAGGCTCCAAGGGA--GGGGGGCTGATGATGAAAACA   79
                    ||||||||||| ||||||||||||||| ||||||| |  |||||||||||||||||||||
Subject: 18338392   ATGTGGTGGAAACTTTTGCATACTAGGGTCCAAGGAATGGGGGGGCTGATGATGAAAACA   18338333

Query:    80        TATCGAATCGAATTTAAAGCTAACAAGGTCCACTCTAAAAGCTGNNNNNNNNNNNNNNNN   139
                    ||||||||||||||||||||||||||||||||||||||||||||
Subject: 18338332   TATCGAATCGAATTTAAAGCTAACAAGGTCCACTCTAAAAGCTGAAAAAAAAAAAAAAAC   18338273

Query:    140       -TACCGATGGGGCAAAACAACAAGCCGAAAATTATTTATGCGATTTTTTGTGGGTATTTG   198
                     |||||||||  |||||| ||||||||| | ||| |||||||||||| ||||||||||||
Subject: 18338272   ATACCGATGGCACAAAACGACAAGCCGAGACTTAATTATGCGATTTTATGTGGGTATTTG   18338213

Query:    199       CCCNNNNNNNNTTGTTTTGATTGNNNNNNNNTGCGAGTCTCAAGTTTTCCCTGGAACTTT   258
                    | |        |||| |||||||        |||||||||||||||||| ||||||||||
Subject: 18338212   CACAGAAATAATTGTATTGATTGCCCGCCTCTGCGAGTCTCAAGTTTTCACTGGAACTTT   18338153

Query:    259       CAACTTTTATCAAAAACAGCTGCGCGATGAAAATNNNNNNNGGCATTAAGGGGGTTTTCC   318
                    ||||||| ||||||| |||||||||||||| |||       |||||||||||| ||||||
Subject: 18338152   CAACTTTGATCAAAAGCAGCTGCGCGATGATAATCCGCTCCGGCATTAAGGGGCTTTTCC   18338093

Query:    319       CCCTAAATGTTTAATTAAATNNNNNNNTCAAATCTGACGCGGCGGAGCACCCCCCTCCGT   378
                    |  |||||||||||||| ||       ||||||||||||||||||||||||| | |||||
Subject: 18338092   CAGTAAATGTTTAATTAGATAAACAAATCAAATCTGACGCGGCGGAGCACCCACATCCGT   18338033

Query:    379       CGACCCCACGACCCCACGACCCGGAAGCCATGGGGTTCGCATACGCATCCGTATCCGTAT   438
                    |||||| ||||||| ||||||||||||||||| |||||||||||||||||||||||||||
Subject: 18338032   CGACCCGACGACCCGACGACCCGGAAGCCATGTGGTTCGCATACGCATCCGTATCCGTAT   18337973

Query:    439       CCCCATCTGTATTCCCATCCGGATCCGTATTCGGTTGGGTACCAGGGGATGGCATCCTAT   498
                    || ||||||||||| |||||||||||||||||||||| |||||||| ||||||||| |||
Subject: 18337972   CCGCATCTGTATTCGCATCCGGATCCGTATTCGGTTGCGTACCAGGTGATGGCATCGTAT   18337913

Query:    499       GGAGGTACATATGTAGATGCATTAATTGCGG    529
                    ||| |||||||||||||||||||||||||||
Subject: 18337912   GGATGTACATATGTAGATGCATTAATTGCGG    18337882
```

Supplemental Fig S4. Top panel shows the presence of the *Minos* insertion in the *CG6123* genomic region. Black flanking arrows denote the oligos used for Sanger based sequencing. Lower panel shows the snapshot of nucleotide blast of rescued *CG6123 Minos* insertion line. The transparent red box denotes the location of the original *Minos* insertion.