Conserved Linkage Groups and Rearrangements in Drosophila

Michael U. Mollenhauer, McNair Scholar, Virginia State University

Stephen W. Schaeffer, Ph.D.
Associate Professor of Biology
Biology Department
Penn State University

Abstract

Drosophila is an excellent model to investigate gene organization and the process of genomic rearrangement because it has a small genome size and genes tend to be conserved in the chromosomal location among species. Gene order differences were inferred in a comparison of the completed genomes of *Drosophila virilis* and *D. mojavensis* using *D. melanogaster* to determine derived rearrangements on the two lineages. The six chromosomes in the two species had 444 conserved gene order breakpoints that resulted from 264 inversions that estimate that each breakpoint was used 1.189 times. These data suggest that genomic rearrangements reuse breakpoints at a low to modest level.

Introduction

Sankaranarayanan (1979) examined the results of 1 million conceptions and examined the health of all these embryos. Fifteen percent of the embryos are spontaneously aborted due to a variety of genetic causes. Alterations in gene order account for 5,250 of the 150,000 spontaneous abortions observed in humans. Comparisons of gene order among different species may provide valuable insights into how genome rearrangements occur and potential mechanisms for their control.

Comparisons of complete genomes are elucidating the mechanisms that rearrange gene order among species (Coghlan *et al.* 2002; Coghlan and Wolfe, 2005; Richards *et al.* 2005). Genes within a genome are organized into units called chromosomes. The organization of genes on chromosomes can vary among species and can also differ in the order of genes. Synteny is defined as two or more genes that are found on the same chromosome in two or more species (Ehrlich *et al.* 1997). Conserved linkage groups have two or more syntenic genes that are in the same order in two or more species (Ehrlich et al 1997). Chromosomal inversions provide a mechanism that can alter the arrangement of genes on a chromosome. Inversions occur when the chromosome is broken at two locations on the chromosome and are rejoined in the opposite orientation, which reverses the order of genes (Figure 1) .

The genomes of *Drosophila* consist of six chromosomal arms that have been conserved over evolutionary time (Figure 2) (Muller, 1940). These genes within these six chromosomal arms are largely syntenic among species, however, gene order has been extensively shuffled (Ranz *et al.* 2001; Richards *et al.* 2005). The complete genomes of 12 species of Drosophila provide a model system for the study of the processes that alter gene order on the six chromosomes of the fly genomes. *Drosophila* is an excellent model to investigate gene organization and the process of genomic rearrangement because it has a small genome size (~200 Megabases) and genes tend to be conserved within chromosomal arms among diverse species. *Drosophila* are easily cultured in the laboratory, they have a short (10-14 day) generation time, and there are many phenotypic changes that can be observed. Another advantage of using *Drosophila* is that it only has six chromosomes and 90% of the gene rearrangements occur within a chromosomal arm. Also, there are many biochemical and genetic techniques available to manipulate the genome to create changes, i.e. the ability to make transgenic animals.

This study examined the differences in gene order between *Drosophila virilis* and *Drosophila mojavensis* using *Drosophila melanogaster* as an outgroup to understand the mechanism of genome rearrangement, the most abundant being intrachromosomal inversions.

Materials and Methods

Genome Sequences and Assemblies. Agencourt Bioscience Corporation generated the *D. virilis* and *D. mojavensis* genomes with the whole genome shotgun method (Myers *et al.* 2000). A total of 3.6 million sequence reads were generated for *D. virilis* and 3.1 million reads for *D. mojavensis*. The sequence reads for the two species were assembled into sequence scaffolds using the ARACHNE assembler from the Whitehead/MIT Genome Center.

Orthologous Gene Calls. The 13,363 genes of *D.* melanogaster from release 3.0 were assigned a unique gene identifier beginning with the first gene on Muller A and proceeding sequential across the six chromosomes to Muller F (Figure 2). Each *D. melanogaster* gene sequence was used to identify orthologous genes in the assembled scaffolds with BLAST searches either BLASTN, which matches nucleotide sequences between the two genomes or TBLASTX, which matches the translated nucleotide sequences against the translated target species genome (Altshul *et al.* 1997). An E-value of 1 x 10^{-5} was used as the cutoff for the BLAST screen of the BLASTN and TBLASTX analyses. The best BLAST hit for each gene was concatenated into a Microsoft Excel spreadsheet for each chromosome/element. The sequence scaffolds were then assigned to 1 of the 6 chromosomes or Muller's elements based on majority rule in comparison to the *D.melanogaster* chromosomal assignment. The scaffolds within a chromosomal arm were ordered based on physical and genetic mapping data (*D. virilis* by Bryant McAllister at the University of Iowa; *D. mojavensis* by Luciana Matzkin at the University of Arizona).

Identification of Conserved Linkage Groups. Conserved linkage groups are a set of genes that are in the same order in two or more species. Genes found in only one species were removed from the analysis. These genes were not detected either because the species did not have the gene or the whole shotgun did not provide a sequence for the gene. The residual list of genes was assigned new identification numbers that reflect the gene order in the respective species. This list was used to identify conserved linkage groups and breakpoints. A conserved linkage group was composed of gene identifiers that were sequential. A break in gene identifier number that was greater than 2 units was identified as a conserved linkage breakpoint. A conserved linkage group with just a single gene could be due to a transposition event where a segment of DNA moved to a new location or could be a single gene at the boundary of two inversion events. A gene was considered a transposition if its two adjacent neighbors are each other's closest neighbor. All transposed genes were removed from the analysis of conserved linkage groups.

Linkage Chain Analysis. The *D. virilis* gene order was used as the standard for each chromosome and was compared to the gene order of *D. mojavensis*. Gene order was compared between the two species noting conserved linkage groups, where the gene order was the same between the two species, and conserved linkage breakpoints, where the gene order changed in *D.*

mojavensis relative to *D. virilis*. Each breakpoint was given a unique identifier such as BP_006_007_C, which indicates that the breakpoint is between conserved linkage groups 6 and 7 on Muller element C. The conserved linkage breakpoints can be linked together by following the gene order of genes at the boundaries. For instance, in Figure3B, the next gene in the sequence of BP_006_007 is predicted to be 672. This leads one to BP_005_006, which completes the linkage chain because the boundary genes at both breakpoints are the next genes in the sequence for original breakpoint. When matching breakpoint boundaries, be aware when the same breakpoint halves can occur in the same chain; they cannot be linked to one another (see Figure 3A).

The chain analysis was performed manually using an MS Excel spreadsheet or with the aid of Fortran program (S. W. Schaeffer, Penn State University). Linkage chains were represented by the set of breakpoints that form a complete chain. Each breakpoint showed the first and last gene identifier for the conserved linkage groups before and after the breakpoint. The linkage chain was completed when the gene order from one breakpoint to another comes back to the initial breakpoint in the chain (Figure 4).

The numerical paths of the conserved linkage groups were then checked for possible analysis problems. The gene orders could have been shuffled artificially either because the two or more genes had transcripts that overlapped or because related duplicate genes matched the same region of DNA. In both of these cases, the order of gene identifier numbers could have been artificially rearranged. In these cases, the artificial gene orders were corrected so that gene order was the same in the two species. Some single gene conserved linkage groups resulted from legitimate rearrangement events, but the next gene expected after the boundary was not obvious. In many cases, this was resolved by examining the neighbor for the opposite boundary. Thirdly, gene sections were separated because the grouping criteria was too strict and were rectified by modifying the criteria to the necessary value greater then 5 and finally, genes sections were grouped together because the grouping criteria was not strict enough and were rectified by modifying the criteria to <2.

Inference of Ancestral Breakpoint Arrangements. For each chain, the *D. mojavensis* gene identifiers were replaced with the corresponding gene identifiers of *D. melanogaster*. The ancestral state of the breakpoints was determined by comparing the gene order in *D. melanogaster* to that of *D. virilis* and *D. mojavensis*. Because the original identifiers were designated in *D. melanogaster*, sequential numbers at conserved linkage group boundaries were assumed to represent the ancestral state. If the ancestral states were determined for n-1 breakpoints in a linkage chain with n breakpoints, then the ancestral state for all breakpoints was known (Figure 5). If the sequential genes spanned the same breakpoint in the *D. virilis* sequence, then an inversion was assumed to occur in *D. mojavensis*, otherwise the inversion occurred along the *D. virilis*. This information was then used to construct the gene order of the common ancestor and assembled the gene breakpoints according to their identified ancestral connections (Figure 6). In cases where all ancestral states could not be inferred for all breakpoints in the chain, we assumed that (n-1)/2 inversions occurred on each lineage to allow us to estimate the total number of inversions on the two lineages.

Statistical Analysis. The numbers of inversions that occurred on each species lineage were tallied for each Muller element. The distribution of inversion events among the chromosomal arms and species were tested with either a Chi-square test of heterogeneity or goodness-of-fit (Sokal and Rohlf, 1981). The number of conserved linkage groups, numbers of breakpoints (n_{bp}), and numbers of inversions (n_{inv}) were estimated for comparison of the *D.*

virilis and *D. mojavensis* genomes. The reusage of breakpoints can be evaluated by estimating a reusage statistic (r) (Sankoff and Trihn 2005), $r=(2n_{\mathrm{inv}})/n_{\mathrm{bp}}$. The departure of the observed rearrangement data from random expectations was evaluated by randomly shuffling x genes of a particular chromosome with n_{inv} inversions and estimating the frequency of times the randomly shuffled genes matched the observed data. This would produce a number between 1 and 2. A value of 1 indicates that breakpoints were used an average of 1 time and a value of 2 indicates that breakpoints were used an average of 2 times.

Results

Genome Inversion Distribution. The results of the chain analyses for the six chromosomal arms are found in Appendices A-F. The distribution of inversions among the five major chromosomes is not significantly different between the two species (X^2=0.913, df=4, P>0.05) for the set of breakpoints where ancestral states could be inferred. If we assume that all data continues this trend, then the rate of inversion is 5.28 inversions per million years. This is half the rate observed between *D. pseudoobscura* and *D. melanogaster* (12.34/my) suggesting either an accelerated rate with the *Sophophoran* subgenus or a decelerated rate in the *Drosophila* subgenus. Whether rearrangement rates are accelerated or decelerated will require other pairwise comparisons to make a concrete inference.

Lineage Specific Inversions. Once the chromosomal ancestral states were determined based on the gene order of *D. melanogaster*, the inversion events were placed on either the *D. virilis* or D. mojavensis lineage. *D. virilis* had a total of 79 inversions while *D. mojavensis* had 91 although this difference is not significantly different from the assumption of equal number of inversions on each lineage (Goodness of Fit Test X^2=0.847, df=1, *P*=0.357) (Figure 7). The inversion rate on the different Muller's elements was not the same (Goodness of Fit Test X2=31.823, df=4, P<0.001), however, the rate differences were similar in the two species (Heterogeneity Test X^2=0.913, df=4, *P*=0.923).

Breakpoint Reusage. The breakpoint reusage within the six chromosomes varied between 1.1 and 1.6 times. The statistical analysis of the reusage shows that breakpoints are being reused more than expected given the number of genes and the number of inversions on each chromosome. Some linkage chains include too many breakpoints given a random usage model.

Final Data Result. The six chromosomes in the two species had 444 total conserved gene order breakpoints that resulted from 264 inversions. From this it was estimated that each breakpoint was used 1.189 times. This suggested that genomic rearrangements reuse breakpoint at a low to modest level. This also illustrates that the genetic rearrangements present in the gene order of each species is not random.

Discussion

Rate of chromosomal inversions in *D. mojavensis* and *D. virilis*. The inversion rate was equal on the two species lineages across all chromosomes. This suggests that there is no bias in the process that generates inversions in the two different species. There was a bias in the rate of inversions among the different chromosomes. This bias may emerge either because the nucleotide sequences that are responsible for the rearrangements may differ in their frequency on the different Muller's elements. Alternatively, the genes combinations required for proper protein function and regulation may differ in their sensitivity to chromosomal rearrangements. For instance, Muller's D may be less sensitive to chromosomal rearrangement than Muller's C.

The linkage chain analysis rejects the hypothesis that rearrangements are randomly introduced. The hypothesis of random usage of breakpoints was rejected. This suggests that some breakpoints are used more than expected, while some regions of the chromosomes are not broken by rearrangement breaks. These results may mean that some sequences on chromosomes are more susceptible to double-stranded breaks, while other regions do not have rearrangement breakpoints either because sequences in these regions are not susceptible to double-stranded breaks or breakpoints are introduced, but individuals with these breaks are selected against. One possibility was suggested by Stolc *et al.* (2004) who found that genes with similar expression patterns tend to be clustered in the genome. Breakpoints within these regions may be selected against because these events would break up coordinately expressed genes.

Uses of ancestral origins of genomic inversions. The research indicates that although changes within chromosomes cannot yet be predicted, their origins can be derived from the current arrangement of genes on each chromosomes. Knowing the ancestral state of a chromosome allows researchers to begin tracing each step of a mutation, identifying the sequence of events that gave rise to the current gene arrangement, inferring how each stage affected the species population and distribution, and then approximate the rate of genome rearrangement. The more frequently a conserved linkage group is involved in rearrangement events, the higher the chance of a genetic modification such as a transposition, duplication, or a deletion may cause mutations in the organism. Identifying and understanding the degree, frequency, and effects of chromosomal rearrangement is a valuable tool in the ongoing search for treatments for individuals with genetic conditions. It can also aid in predicting possible evolutionary tracks a specific species may take. Once we understand what causes these gene rearrangements, cures for human genetic disorders may be more easily achieved that will ensure a more promising outlook for future generations.

Breakpoint reusage. It was found that the reusage statistic for Muller Element F was the highest with a reusage of 1.6. This does not take into consideration that it has the least number of genes among the 6 chromosomes and as such any chromosomal inversions on that chromosome would show as significant. It is noteworthy that the rate of reusage does not correlate to the size of the other five chromosomes. Muller Element E, the largest of the chromosomes, has a moderate reusage of 1.208. This is nearly the same as that of Element A, which is the second smallest chromosome (Table 1).

Problems/difficulties encountered. The most difficult issue was making consistent, logical decisions in the assignment of conserved linkage groups. In the chain analysis, it was frequently necessary to reevaluate breakpoints to resolve large gaps between sequential breakpoint neighbors or numerous breaks in a segment of genes that were not necessary. Another challenge was the identification of transpositions. As the research progressed more criteria to make those distinctions were used and additional transpositions were identified. This required re-evaluation of the linkage chains in some of the chromosomes once new transpositions were removed. Another issue was ensuring that a breakpoint that appeared twice in the same chain was not linked to itself in the ancestral inference. Such a mistake would create a circle between the breakpoint ends that did not connect to the reconstructed assembly.

Additional research. The mechanisms that drive genetic rearrangement within the chromosomes of *Drosophila* are still unclear. This leaves several questions that still need to be investigation. For example, what are the factors that initiate a mutation? Chromosomal inversions are not the only type of mutation that occurs within the genome. In other species there are types of rearrangements such as pericentric inversion and inter-chromosomal translocations that play major roles in diversifying the particular species. It is not clear what type of rearrangement is used more than others? Are the forces that cause inversions the same as those that cause transpositions? Can these rearrangements be manipulated to reverse their effects? Deliberate changes in gene order have been achieved in several cases using techniques such as exposure to ultraviolet radiation to create abnormalities. We have yet to understand what is necessary, if possible, to reverse the effects of such treatments on chromosomes.

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Table 1. Distribution of Inversions and Breakpoints among the six Muller Elements based on the comparison of gene order in *D. virilis* and *D. mojavensis*. Breakpoint re-usage for each chromosomal element for the comparison is also shown.

Chromosome	Genes	Inversions	Breakpoints (95% CI)	*r* (95% CI)
Muller A	1,909	48	80 (91- 97)	1.20 (1.00-1.06)
Muller B	2,170	36	62 (69- 73)	1.16 (1.00-1.05)
Muller C	2,455	45	73 (87- 91)	1.23 (1.00-1.05)
Muller D	2,616	73	128 (139-146)	1.14 (1.01-1.06)
Muller E	3,155	58	96 (112-117)	1.21 (1.00-1.05)
Muller F	68	4	5 (7- 9)	1.60 (1.00-1.33)
Total	12,373	264	444 (not done)	1.19 (not done)

r, breakpoint re-usage statistic. 95% CI, 95% confidence interval of the simulations with the given number of inversions and genes on the chromosomal arm.

Figure Legends

Figure 1. Mechanism of chromosomal inversion showing the introduction of two breakpoints and the reversal of gene order.

Figure 2. Phylogeny of five *Drosophila* species. The Drosophila genome is divided into six chromosomal arms or Muller's elements that are conserved among different species. The organization of the chromosomes, however, changes through the fission and fusion of these arms. The gene identifiers of *D. melanogaster* are indicated on its Muller elements.

Figure 3 . Chain analyses A. Incorrect linkage. B. Correct linkage

Figure 4 . Example of a complete linkage chain. Linkage chain analysis moves from breakpoint to breakpoint (red arrows) identifying the nearest neighbor of the gene at the breakpoint (colored boxes) until the breakpoints form an unbroken chain.

Figure 5. Ancestral state inference for conserved linkage breakpoints. The colored boxes indicate the inferred adjacent neighbors in the common ancestor of *D. virilis* and *D. mojavensis*.

Figure 6. Inference of lineage specific inversions. A. If the gene order at the breakpoint is conserved between *D. melanogaster* and *D. mojavensis* (colored boxes), then an inversion occurred on the *D. virilis* lineage (red arrow). B. If the gene order at the breakpoint is conserved between *D. melanogaster* and *D. virilis* (colored boxes), then an inversion occurred on the *D. mojavensis* lineage (red arrow).

Figure 7. Distribution of chromosomal inversions on the *D. virilis* and D. mojavensis lineages where the ancestral states could be unambiguously determined for linkage chains.

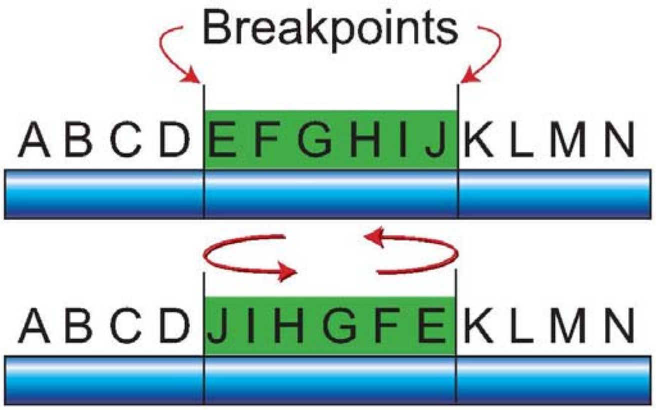

Figure 1

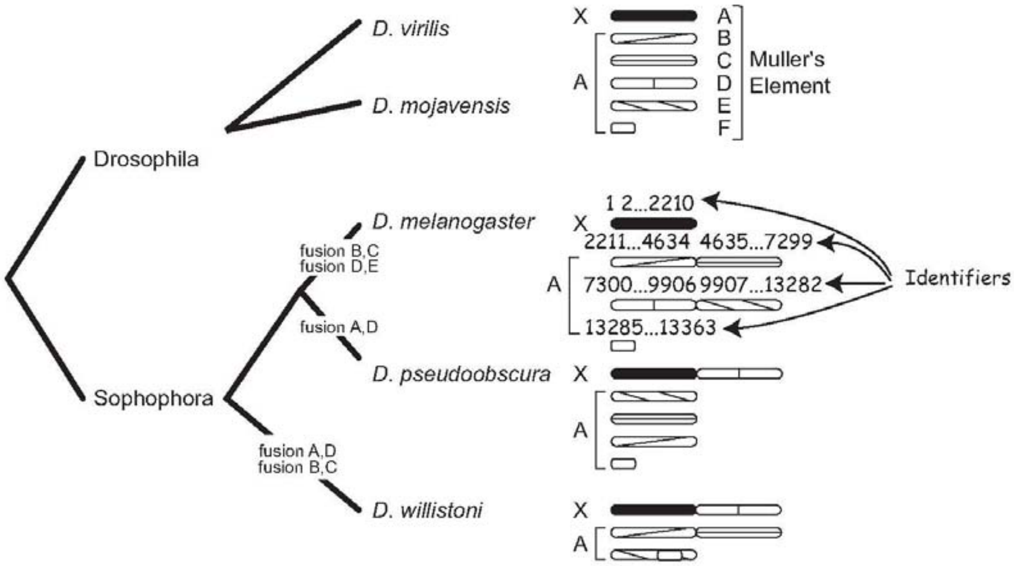

Figure 2

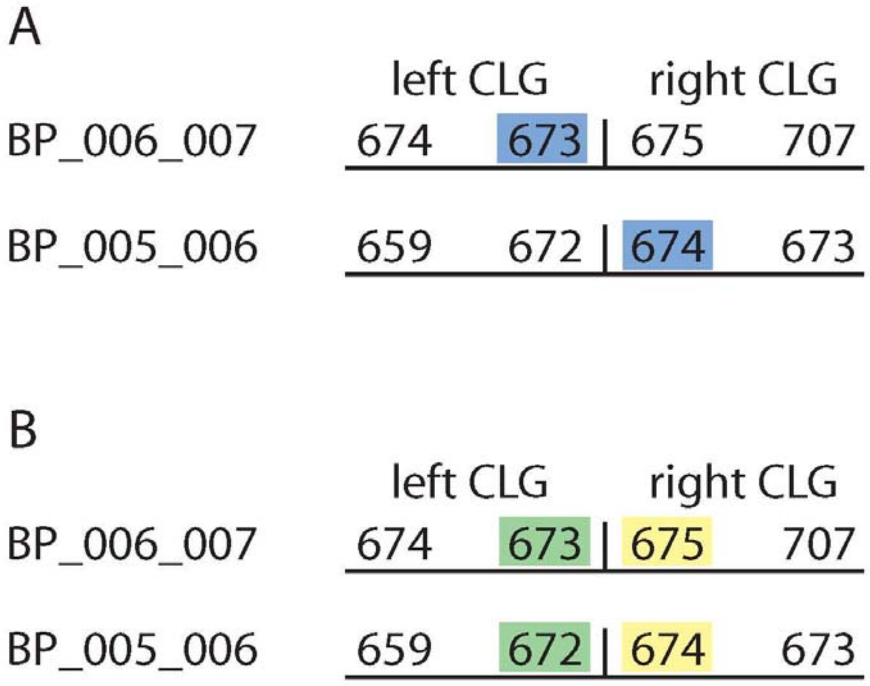

Figure 3

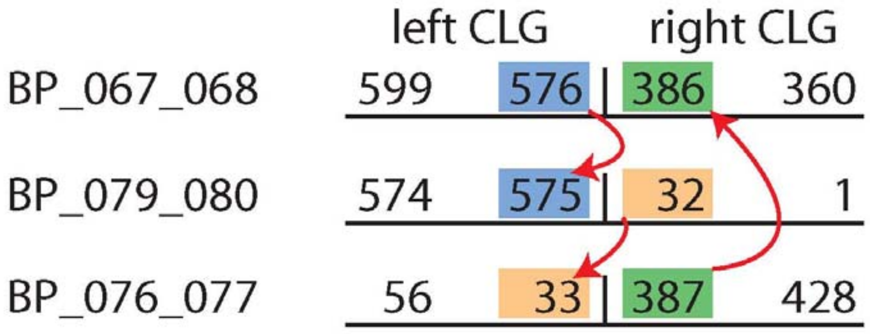

Figure 4

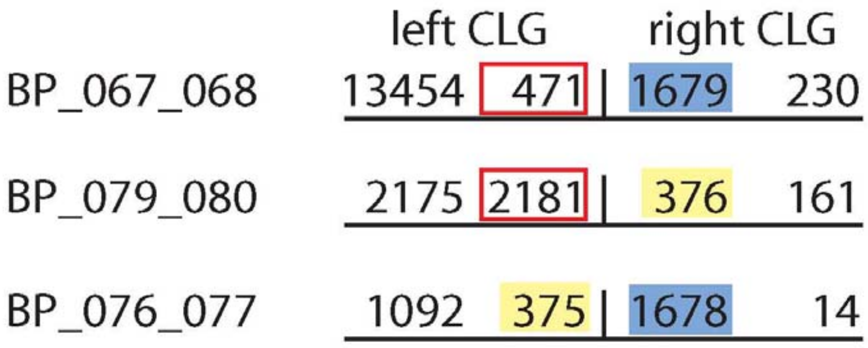

Figure 5

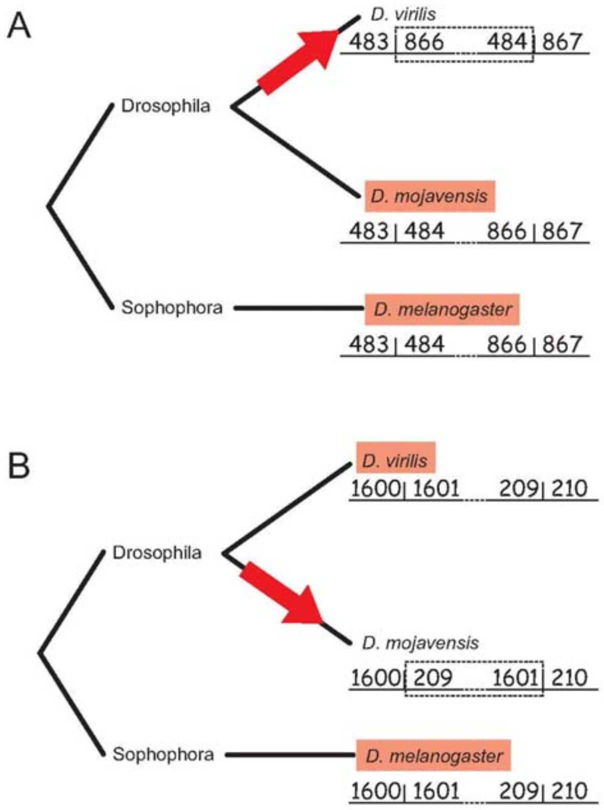

Figure 6

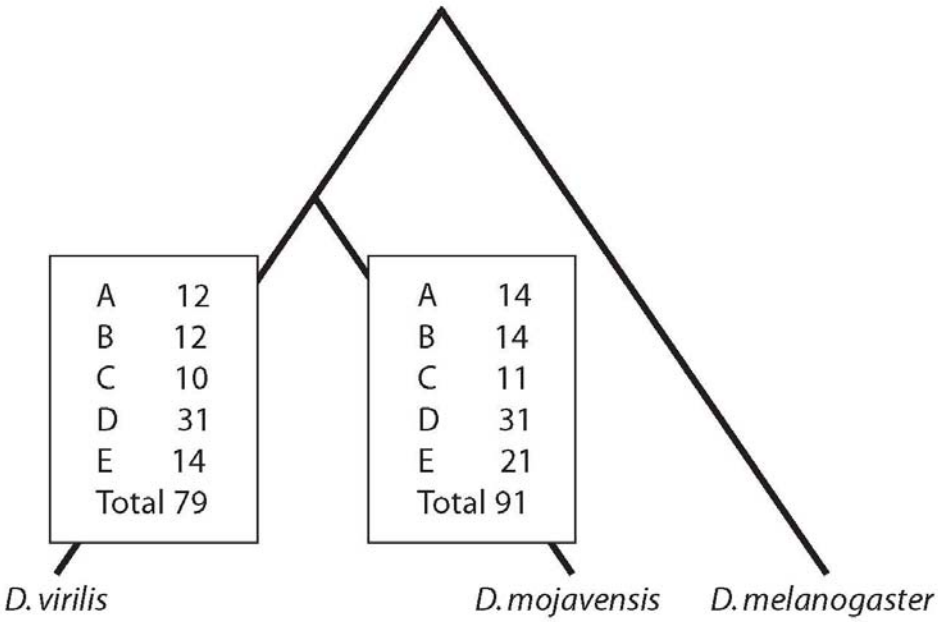
A 12
B 12
C 10
D 31
E 14
Total 79
A 14
B 14
C 11
D 31
E 21
Total 91
D. virilis
D. mojavensis
D. melanogaster

Figure 7

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                                    Chain_Anal_ID_a_Dmoj
Appendix A. Linkage Chain Analysis for the Muller A Element Comparison of D. virilis and D.
mojavensis
 Breakpoints Remaining=  77
 Chain  1            left CLG          right CLG
   BP_001_002           0      0 |  1429   1426
                     ---------------------------
   BP_021_022        1709   1532 |  2206    932
                     ---------------------------
   BP_080_081         376    161 | 13467  13467
                     ---------------------------

 Breakpoints Remaining=  75     (D.moj lineage inversion)(1)
 Chain  2            left CLG          right CLG
   BP_004_005        1969     54 |   500   1932        A by inference
                     ---------------------------
   BP_077_078        1678     14 |    11   2174        A
                     ---------------------------

 Breakpoints Remaining=  73     (D.moj lineage inversion)(1)
 Chain  3            left CLG          right CLG
   BP_006_007        1415   1414 |  1413   1661        A
                     ---------------------------
   BP_005_006         500   1932 |  1415   1414        A by inference
                     ---------------------------

 Breakpoints Remaining=  70     (D.vir lineage inversion)(2)
 Chain  4            left CLG          right CLG
   BP_008_009         293    952 |  2200   2192
                     ---------------------------
   BP_023_024        1202   1965 |  1157   1158
                     ---------------------------
   BP_024_025        1157   1158 |  9599    958
                     ---------------------------

 Breakpoints Remaining=  68     (D.vir lineage inversion)(1)
 Chain  5            left CLG          right CLG
   BP_011_012         516    125 |   118    517
                     ---------------------------
   BP_038_039         897    127 |   119   1032
                     ---------------------------

 Breakpoints Remaining=  65     (D.vir lineage inversion)(2)
 Chain  6            left CLG          right CLG
   BP_013_014        2106    992 |  1784   1701        A by inference
                     ---------------------------
   BP_040_041         199     28 |    27    609        A
                     ---------------------------
   BP_002_003        1429   1426 |  1424    956        A
                     ---------------------------

 Breakpoints Remaining=  56     (D.vir lineage inversion)(8)
 Chain  7            left CLG          right CLG
   BP_062_063        1155   1155 |   652    557
                     ---------------------------
   BP_044_045         492    696 |  1029    200
                     ---------------------------
   BP_046_047        1031   1031 |   697    174
                     ---------------------------
   BP_041_042          27    609 |  1562    699
                     ---------------------------
   BP_016_017        1708   1379 |   928   1726
                     ---------------------------
   BP_056_057        2046    745 |  1521   1519
                     ---------------------------
   BP_035_036        1660   1386 |  1389   1912        A

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 54     54     54     54     54

   8471   8469   8470   8468   8467   8466   8465   8461   8460   8459   8457   8456   8452   8451   8450
8448   8447   8446   8445   8130
     54     54     54     54     54     54     54     55     55     55     55     55     55     55     55
 55     55     55     55     55

   8131   8129   8128   8040   8041   8043   8042   8044   8045   8047   8046   8048   8049   8050   8051
8052   8053   8054   8058   8061
     55     55     55     55     55     55     55     55     55     55     55     55     55     55     55
 55     55     55     55     55

   8062   8063   8064   8065   8066   8069   8068   8070   8071   8073   8074   8400   8401   8402   8403
8404   8405   7625   7624   7623
     55     55     55     55     55     55     55     55     55     55     55     56     56     56     57
 57     57     57     57     57

   7621   7620   7619   7618   7617   7836   7786   8584   8586   8587   8588   7391   7392   9308   9309
7931   7932   7933   7934   7936
     57     57     57     57     57     57     57     57     57     57     57     57     57     57     57
 57     58     58     59     59

   7937   7938   7939   7940   7941   7942   7943   7944   7945   7946   7948   7949   7950   7951   7953
7954   7955   7956   7957   7958
     59     59     59     59     59     59     59     59     59     59     59     59     59     59     59
 59     59     59     59     59

   7959   7964   7960   7961   9273   9274   7764   7579   7577   7578   7580   7581   7582   7584   7586
7588   7589   7590   7685   7576
     59     60     60     61     61     61     61     61     61     61     61     61     61     61     61
 61     61     61     61     61

   7575   7574   7573   7572   7571   7570   7569   7568   7567   7566   7565   7564   7563   7560   7559
7561   7562   7558   7557   7556
     61     61     61     61     61     61     61     61     61     61     61     61     61     62     62
 62     62     63     63     63

   7555   7553   7551   7550   7554   7552   7548   7546   7545   7544   7543   7542   8229   8228   8227
8226   8225   8224   9819   9818
     63     63     63     64     64     64     65     65     65     65     65     65     65     65     65
 65     65     65     66     66

   9817   9816   9815   9814   9813   9812   8838   8839   8840   8841   8842   8843   8846   8847   8848
8849   8853   9465   8855   8856
     66     66     66     66     66     66     66     66     66     66     66     66     66     66     66
 66     66     66     66     66

   8857   8858   8859   8860   8707   9468   9469   9470   9471   9472   9473   9474   9476   9478   9479
9480   9481   9290   9495   9496
     66     66     66     66     66     66     66     66     66     66     66     66     66     66     66
 66     66     66     67     68

   9497   9498   9501   9500   9499   9502   9503   9505   9506   9507   9509   9508   9510   9511   9513
9512   9514   9515   9516   9517
     68     69     69     69     69     69     69     69     69     69     69     69     69     69     69
 69     69     69     69     69

   9518   9519   9520   9521   9522   9523   9524   9525   9526   9527   9464   9463   9462   9460   9459
9458   9457   9456   9454   9453
     69     69     69     69     69     69     69     69     69     69     69     69     69     69     69
 69     69     69     69     69

   9452   9451   9450   9448   9447   9446   9444   9442   9441   9440   9439   9438   9437   9436   9435
9434   9433   9432   9431   9609
     69     69     69     69     69     69     69     69     69     69     69     69     69     69     69
```

```
                                    Chain_Anal_ID_a_Dmoj
                    ---------------------------
  BP_052_053          771   1505 |  578   1105
                    ---------------------------
  BP_051_052         2189   2186 |  771   1505
                    ---------------------------

Breakpoints Remaining=  54     (D.vir lineage inversion)(1)
Chain  8            left CLG          right CLG
  BP_057_058         1521   1519 |  491    485
                    ---------------------------
  BP_043_044         1183   2045 |  492    696
                    ---------------------------

Breakpoints Remaining=  52     (D.moj lineage inversion)(1)
Chain  9            left CLG          right CLG
  BP_073_074         2006    735 |  736    737        A
                    ---------------------------
  BP_074_075          736    737 |  741   1093        A by inference
                    ---------------------------

Breakpoints Remaining=  50     (D.moj lineage inversion)(1)
Chain 10            left CLG          right CLG
  BP_047_048          697    174 |  175    415        A
                    ---------------------------
  BP_068_069         1679    230 |   46    110        A by inference
                    ---------------------------

Breakpoints Remaining=  48     (D.vir lineage inversion)(1)
Chain 11            left CLG          right CLG
  BP_007_008         1413   1661 |  293    952
                    ---------------------------
  BP_034_035          299    294 | 1660   1386
                    ---------------------------

Breakpoints Remaining=  46     (D.moj lineage inversion)(1)
Chain 12            left CLG          right CLG
  BP_014_015         1784   1701 | 1702   1707        A
                    ---------------------------
  BP_015_016         1702   1707 | 1708   1379        A
                    ---------------------------

Breakpoints Remaining=  44     (D.moj lineage inversion)(1)
Chain 13            left CLG          right CLG
  BP_054_055          221   1559 | 1558   1555        A
                    ---------------------------
  BP_065_066         1259  13449 | 1076  13455        A by inference
                    ---------------------------

Breakpoints Remaining=  39     (D.vir lineage inversion)(4)
Chain 14            left CLG          right CLG
  BP_017_018          928   1726 | 1730    849
                    ---------------------------
  BP_027_028         1641   1971 | 1724    844
                    ---------------------------
  BP_003_004         1424    956 | 1969     54
                    ---------------------------
  BP_025_026         9599    958 |  368    301
                    ---------------------------
  BP_063_064          652    557 |  558   2185        A
                    ---------------------------

Breakpoints Remaining=  37     (D.vir lineage inversion)(1)
Chain 15            left CLG          right CLG
  BP_039_040          119   1032 |  199     28
                    ---------------------------
```

```
  BP_045_046          1029    200 |  1031   1031
                     ---------------------------

Breakpoints Remaining=  34     (D.vir lineage inversion)(2)
Chain 16             left CLG          right CLG
  BP_067_068         13454    471 |  1679    230
                     ---------------------------
  BP_079_080          2175   2181 |   376    161
                     ---------------------------
  BP_076_077          1092    375 |  1678     14
                     ---------------------------

Breakpoints Remaining=  32     (D.moj lineage inversion)(1)
Chain 17             left CLG          right CLG
  BP_028_029          1724    844 |   843   1448        A
                     ---------------------------
  BP_018_019          1730    849 |   848    840        A
                     ---------------------------

Breakpoints Remaining=  30     (d.vir lineage inversion)(1)
Chain 18             left CLG          right CLG
  BP_070_071          1548   1547 |  1550    281
                     ---------------------------
  BP_069_070            46    110 |  1548   1547
                     ---------------------------

Breakpoints Remaining=  28     (D.vir lineage inversion)(1)
Chain 19             left CLG          right CLG
  BP_010_011          2193   2103 |   516    125
                     ---------------------------
  BP_012_013           118    517 |  2106    992
                     ---------------------------

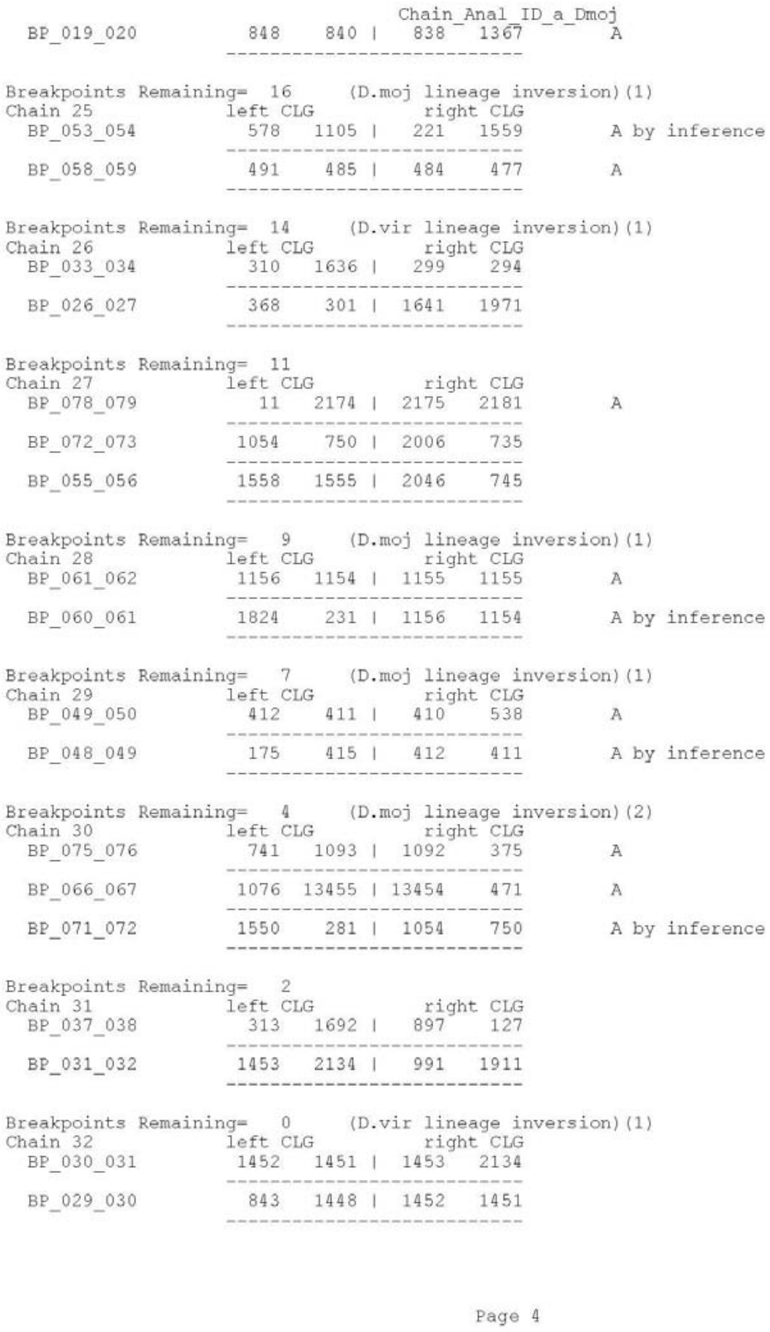
Breakpoints Remaining=  26     (D.vir lineage inversion)(1)
Chain 20             left CLG          right CLG
  BP_059_060           484    477 |  1824    231
                     ---------------------------
  BP_042_043          1562    699 |  1183   2045
                     ---------------------------

Breakpoints Remaining=  24     (D.moj lineage inversion)(1)
Chain 21             left CLG          right CLG
  BP_020_021           838   1367 |  1709   1532        A by inference
                     ---------------------------
  BP_009_010          2200   2192 |  2193   2103        A
                     ---------------------------

Breakpoints Remaining=  22     (D.vir lineage inversion)(1)
Chain 22             left CLG          right CLG
  BP_036_037          1389   1912 |   313   1692
                     ---------------------------
  BP_032_033           991   1911 |   310   1636
                     ---------------------------

Breakpoints Remaining=  20     (D.vir lineage inversion)(1)
Chain 23             left CLG          right CLG
  BP_050_051           410    538 |  2189   2186
                     ---------------------------
  BP_064_065           558   2185 |  1259  13449
                     ---------------------------

Breakpoints Remaining=  18     (D.moj lineage inversion)(1)
Chain 24             left CLG          right CLG
  BP_022_023          2206    932 |  1202   1965        A by inference
                     ---------------------------
```



```
                                    Chain_Anal_ID_a_Dmoj
  BP_019_020            848    840 |  838   1367          A
                     ---------------------------

Breakpoints Remaining=  16     (D.moj lineage inversion)(1)
Chain 25             left CLG           right CLG
  BP_053_054            578   1105 |  221   1559          A by inference
                     ---------------------------
  BP_058_059            491    485 |  484    477          A
                     ---------------------------

Breakpoints Remaining=  14     (D.vir lineage inversion)(1)
Chain 26             left CLG           right CLG
  BP_033_034            310   1636 |  299    294
                     ---------------------------
  BP_026_027            368    301 | 1641   1971
                     ---------------------------

Breakpoints Remaining=  11
Chain 27             left CLG           right CLG
  BP_078_079             11   2174 | 2175   2181          A
                     ---------------------------
  BP_072_073           1054    750 | 2006    735
                     ---------------------------
  BP_055_056           1558   1555 | 2046    745
                     ---------------------------

Breakpoints Remaining=   9     (D.moj lineage inversion)(1)
Chain 28             left CLG           right CLG
  BP_061_062           1156   1154 | 1155   1155          A
                     ---------------------------
  BP_060_061           1824    231 | 1156   1154          A by inference
                     ---------------------------

Breakpoints Remaining=   7     (D.moj lineage inversion)(1)
Chain 29             left CLG           right CLG
  BP_049_050            412    411 |  410    538          A
                     ---------------------------
  BP_048_049            175    415 |  412    411          A by inference
                     ---------------------------

Breakpoints Remaining=   4     (D.moj lineage inversion)(2)
Chain 30             left CLG           right CLG
  BP_075_076            741   1093 | 1092    375          A
                     ---------------------------
  BP_066_067           1076  13455 | 13454   471          A
                     ---------------------------
  BP_071_072           1550    281 | 1054    750          A by inference
                     ---------------------------

Breakpoints Remaining=   2
Chain 31             left CLG           right CLG
  BP_037_038            313   1692 |  897    127
                     ---------------------------
  BP_031_032           1453   2134 |  991   1911
                     ---------------------------

Breakpoints Remaining=   0     (D.vir lineage inversion)(1)
Chain 32             left CLG           right CLG
  BP_030_031           1452   1451 | 1453   2134
                     ---------------------------
  BP_029_030            843   1448 | 1452   1451
                     ---------------------------


                                         Page 4
```

```
                                           Chain_Anal_ID_a_Dmoj
Chain Analysis Summary Data
---------------------------

Chains with  2 Breakpoints=  24
Chains with  3 Breakpoints=   6
Chains with  4 Breakpoints=   0
Chains with  5 Breakpoints=   1
Chains with  6 Breakpoints=   0
Chains with  7 Breakpoints=   0
Chains with  8 Breakpoints=   0
Chains with  9 Breakpoints=   1
Chains with 10 Breakpoints=   0
Chains with 11 Breakpoints=   0
Chains with 12 Breakpoints=   0

Number of Inversions=  48
Number of Breakpoints=  80
Reusage Index=     1.200


Conserved Linkage Group Intervals
---------------------------------

CLG  No.Genes  Left_IDX  Right_IDX  Left_Gene  Right_Gene
  1         1         1          1          0           0
  2         4         2          5       1429        1426
  3         5         6         10       1424         956
  4        11        11         21       1969          54
  5        14        22         35        500        1932
  6         2        36         37       1415        1414
  7        33        38         70       1413        1661
  8        25        71         95        293         952
  9         6        96        101       2200        2192
 10        42       102        143       2193        2103
 11        31       144        174        516         125
 12        25       175        199        118         517
 13        58       200        257       2106         992
 14        24       258        281       1784        1701
 15         5       282        286       1702        1707
 16         6       287        292       1708        1379
 17        11       293        303        928        1726
 18         7       304        310       1730         849
 19        14       311        324        848         840
 20         4       325        328        838        1367
 21         8       329        336       1709        1532
 22       103       337        439       2206         932
 23         9       440        448       1202        1965
 24         2       449        450       1157        1158
 25        26       451        476       9599         958
 26        15       477        491        368         301
 27        17       492        508       1641        1971
 28        10       509        518       1724         844
 29       106       519        624        843        1448
 30         2       625        626       1452        1451
 31        13       627        639       1453        2134
 32        14       640        653        991        1911
 33        12       654        665        310        1636
 34         6       666        671        299         294
 35        10       672        681       1660        1386
 36        29       682        710       1389        1912
 37        21       711        731        313        1692
 38        45       732        776        897         127
 39        94       777        870        119        1032
 40        14       871        884        199          28
 41         5       885        889         27         609

                                               Page 5
```

```
                                    Chain_Anal_ID_a_Dmoj
  42        13       890        902       1562         699
  43        42       903        944       1183        2045
  44        10       945        954        492         696
  45        56       955       1010       1029         200
  46         1      1011       1011       1031        1031
  47         5      1012       1016        697         174
  48        18      1017       1034        175         415
  49         2      1035       1036        412         411
  50        22      1037       1058        410         538
  51         3      1059       1061       2189        2186
  52        76      1062       1137        771        1505
  53        14      1138       1151        578        1105
  54        31      1152       1182        221        1559
  55         4      1183       1186       1558        1555
  56        41      1187       1227       2046         745
  57         3      1228       1230       1521        1519
  58         6      1231       1236        491         485
  59         7      1237       1243        484         477
  60        37      1244       1280       1824         231
  61         2      1281       1282       1156        1154
  62         1      1283       1283       1155        1155
  63        30      1284       1313        652         557
  64        32      1314       1345        558        2185
  65        55      1346       1400       1259       13449
  66        21      1401       1421       1076       13455
  67        24      1422       1445      13454         471
  68        27      1446       1472       1679         230
  69        39      1473       1511         46         110
  70         2      1512       1513       1548        1547
  71        14      1514       1527       1550         281
  72        31      1528       1558       1054         750
  73        19      1559       1577       2006         735
  74         2      1578       1579        736         737
  75        53      1580       1632        741        1093
  76        24      1633       1656       1092         375
  77        42      1657       1698       1678          14
  78        28      1699       1726         11        2174
  79         2      1727       1728       2175        2181
  80        32      1729       1760        376         161
  81         1      1761       1761      13467       13467


 Gene List and Conserved Linkage Group Calls
 -------------------------------------------
      0  1429  1428  1427  1426  1424  1423  1422   955   956  1969  1768  1673  1672   172
171   170   169    56    55
      1     2     2     2     2     3     3     3     3     3     4     4     4     4     4
  4     4     4     4     4

     54   500    51    50    49    48  1924  1925  1926  1927  1928  1929  1930  1931  1932
1415  1414  1413  1412  1410
      4     5     5     5     5     5     5     5     5     5     5     5     5     5     5
  6     6     7     7     7

   1411  1408  1406  1404  1403  1400  1398  1397  1396  1395  1394  1393  1392  1391  1390
1640  1639   187   186   185
      7     7     7     7     7     7     7     7     7     7     7     7     7     7     7
  7     7     7     7     7

    184   183   182   181  1531  1665  1664  1663  1662  1661   293   292   531   807   808
809   810  2165  2166  2167
      7     7     7     7     7     7     7     7     7     7     8     8     8     8     8
  8     8     8     8     8

   1141  1142  1143  1144  1145  1146  1147   946   947   948  1420  1421   954   953   952
```

```
2200  2201   754  2188  2190
     8     8     8     8     8     8     8     8     8     8     8     8     8     8     8
  9     9     9     9     9

   2192  2193  2194  2195  2196  2197  2198   951  1360  1949   884   883   882   880   879
878   877  1966  1968  1846
     9    10    10    10    10    10    10    10    10    10    10    10    10    10    10
 10    10    10    10    10

   1848  1850  1852  1854  1859  1861  1863  1864  1865  1866  1007  1008  1009  2096  2095
2094  2097  2099  2098  2100
    10    10    10    10    10    10    10    10    10    10    10    10    10    10    10
 10    10    10    10    10

   2101  2104  2103   516   136   135   134  1006  1600  1602  1601  1607  1572   283   710
712   713   714   716   717
    10    10    10    11    11    11    11    11    11    11    11    11    11    11    11
 11    11    11    11    11

    721   722   724   725   465   464   463   458   454   453   452   451   449   125   118
116   117   115   114  1542
    11    11    11    11    11    11    11    11    11    11    11    11    11    11    12
 12    12    12    12    12

   1541  1540  1539  1538  1535  1534  2209  2210   505   506 13461 13460 13462 13464   521
520   519   518   517  2106
    12    12    12    12    12    12    12    12    12    12    12    12    12    12    12
 12    12    12    12    13

   2107  2108  2109  2111  2114  2120  2123  2124  2126  2128  2129  2130  2132   901   900
1385  1384  1383   304   305
    13    13    13    13    13    13    13    13    13    13    13    13    13    13    13
 13    13    13    13    13

    303   302   906   908   907   909   910   913   911   912  1934  1923  1922  1933    47
2043  2044  2042   852   853
    13    13    13    13    13    13    13    13    13    13    13    13    13    13    13
 13    13    13    13    13

    856   855  1898  1899  1900  1901  1364  1363  1361  1362  1123  1124   847   846   997
993   992  1784  1783  1781
    13    13    13    13    13    13    13    13    13    13    13    13    13    13    13
 13    13    14    14    14

   1780  1779  1778  1777   936   937   939   938   941   942  1776  1775  1774  1773  1772
1770  1771  1769  1699  1700
    14    14    14    14    14    14    14    14    14    14    14    14    14    14    14
 14    14    14    14    14

   1701  1702  1703  1704  1705  1707  1708  1192  1193  1382  1381  1379   928   929   927
924   926   923   922  1716
    14    15    15    15    15    15    16    16    16    16    16    16    17    17    17
 17    17    17    17    17

   1717  1718  1726  1730  1729  1728  1727  1732   850   849   848   360   361   362  8636
363   364   367  1722  1721
    17    17    17    18    18    18    18    18    18    18    19    19    19    19    19
 19    19    19    19    19

   1720  1719  1733   840   838   839  1342  1367  1709  1710  1713  1711  1671  1670  1669
1532  2206  2205  2204  2203
    19    19    19    19    20    20    20    20    21    21    21    21    21    21    21
 21    22    22    22    22

   2202  1597  1049  1048  1047  1042  1041  1040  1039   915   916   917   918   919   920
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921  1714  1715  1659  1658
     22    22    22    22    22    22    22    22    22    22    22    22    22    22    22
 22    22    22    22    22

   1657  1656  1654  1653  1652  1651  1650  1649  1648  1647  1955  1954  1953  2140  2143
2141  2144  2147  2149  2150
     22    22    22    22    22    22    22    22    22    22    22    22    22    22    22
 22    22    22    22    22

   2146  2154  2159  2160  2161  2162  2163  2164   815   816   817   818  1375  1377  1376
1378   930   931   353   352
     22    22    22    22    22    22    22    22    22    22    22    22    22    22    22
 22    22    22    22    22

    351   347   346   344  1685  1000  1005   778   779   780   782   784   785   786   781
789   791   783   787   788
     22    22    22    22    22    22    22    22    22    22    22    22    22    22    22
 22    22    22    22    22

    790   792   793   795   797   798   801   800  1159  1161   288   287   286   285   943
944   945   934   932  1202
     22    22    22    22    22    22    22    22    22    22    22    22    22    22    22
 22    22    22    22    23

   1203  1204  1205  1207  1206  1209  1210  1965  1157  1158  9599  1341  1366   986   985
984   983   982   979   978
     23    23    23    23    23    23    23    23    24    24    25    25    25    25    25
 25    25    25    25    25

    977   975   974   973   972   971   970   968   966   965   964   962   961   960   959
958   368   371  1148   819
     25    25    25    25    25    25    25    25    25    25    25    25    25    25    25
 25    26    26    26    26

    820  1697  1694  1696  1695  1698  1693   888   889   890   301  1641  1642  1643  1644
1645  1646  1956  8805  1957
     26    26    26    26    26    26    26    26    26    26    26    27    27    27    27
 27    27    27    27    27

   1958  1959  1960  1961  1973  1972  1970  1971  1724  1725  1723  1691  1690  1688  1687
1686   845   844   843   842
     27    27    27    27    27    27    27    27    28    28    28    28    28    28    28
 28    28    28    29    29

   1734  1736  1737  1739  1738  1740  1741  1387   885   886   887  1743  1744   949   950
 34    35    36  2065  2064
     29    29    29    29    29    29    29    29    29    29    29    29    29    29    29
 29    29    29    29    29

   2063  2058  2062  2061  2060  2055  2054  2052  2050  2051  2048  2047   757   758   756
755  1605  1583  1566    58
     29    29    29    29    29    29    29    29    29    29    29    29    29    29    29
 29    29    29    29    29

     57   695  1058  1060  1061  1059  1062  1063  1064  1065  1066  1163  1162  1164     5
  7     8     9    10   507
     29    29    29    29    29    29    29    29    29    29    29    29    29    29    29
 29    29    29    29    29

    504   503   501    32    33  1346  1344  1349  1348  1350  1351  1353  1355  1356  1357
1358   834   833   832   831
     29    29    29    29    29    29    29    29    29    29    29    29    29    29    29
 29    29    29    29    29

    830   829   827   828   826   823   824   825   821  1106  1107  1109  1108  1114  1115
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```
1117  1118  1119  1121  1374
     29     29     29     29     29     29     29     29     29     29     29     29     29     29     29
 29     29     29     29     29

   1444   1445   1446   1448   1452   1451   1453   1454   1950   1951   1576   9266   1582   1578   2137
2138  2136  2135  2134   991
     29     29     29     29     30     30     31     31     31     31     31     31     31     31     31
 31     31     31     31     32

    990    989    988    987   1902   1903   1904   1905   1908   1906   1907   1909   1911    310    309
308   307   306  1194  1195
     32     32     32     32     32     32     32     32     32     32     32     32     32     33     33
 33     33     33     33     33

   1196   1197   1638   1637   1636    299    298    297    296    295    294   1660   1212   1213   1214
1215  1216   851   899  1388
     33     33     33     33     33     34     34     34     34     34     34     35     35     35     35
 35     35     35     35     35

   1386   1389   1635   1634   1633   1631   1632   1630   1629   1627   1628   1626   1625   1624   1623
1622  1621  1620  1619  1616
     35     36     36     36     36     36     36     36     36     36     36     36     36     36     36
 36     36     36     36     36

   1016   1575   1580   1588   1590   1574   1913   1915   1914   1912    313    316    317    318    319
321   322   325   326   327
     36     36     36     36     36     36     36     36     36     36     37     37     37     37     37
 37     37     37     37     37

    329    328    330    331    332    333    334    336    343    335   1692    897   1962   1963   1964
1211  1935  1936  1937  1944
     37     37     37     37     37     37     37     37     37     37     37     38     38     38     38
 38     38     38     38     38

   1946   1947   1948    905    904    903    902   1787   1788   2169   2168   1140   1139   1138   1137
1136  1133  1785  1132  1131
     38     38     38     38     38     38     38     38     38     38     38     38     38     38     38
 38     38     38     38     38

   1130   1129   1128   1127   1126    359   2180   2179    355    354    133    132    131    130    128
127   119   120   121   101
     38     38     38     38     38     38     38     38     38     38     38     38     38     38     38
 38     39     39     39     39

    100     99     91   8302   8301   8739     90     89     88     87     86     84     79     78     77
 75    74    73    72    71
     39     39     39     39     39     39     39     39     39     39     39     39     39     39     39
 39     39     39     39     39

     70     68     67     62     61   1868   1869   1870   1872   1874   1875   1876   1877   1878   1879
1880  1881  1882  1883  1884
     39     39     39     39     39     39     39     39     39     39     39     39     39     39     39
 39     39     39     39     39

   1885   1888   1889   1890   1891   1895   1896   1897    857    858    859    860    861    863    864
865   866   867   868   869
     39     39     39     39     39     39     39     39     39     39     39     39     39     39     39
 39     39     39     39     39

    873    874    872   1767   1765   1764   1763   1762   1755   1756   1754   1750   1749   1748   1746
1419  1417  1418   896   895
     39     39     39     39     39     39     39     39     39     39     39     39     39     39     39
 39     39     39     39     39

    894    893    892    891    914   1038   1036   1034   1033   1032    199    198    197    196    194
```

```
1050  1051  1052    53    52
      39    39    39    39    39    39    39    39    39    39    40    40    40    40    40
 40    40    40    40    40

      31    30    29    28    27   612   611   610   609  1562  1563 13445 13446   709   708
707   706   705   704   703
      40    40    40    40    41    41    41    41    41    42    42    42    42    42    42
 42    42    42    42    42

     700   699  1183  1184   561   562   563   564   565   568   570   569   571   572   573
574   576   575  1683  1675
      42    42    43    43    43    43    43    43    43    43    43    43    43    43    43
 43    43    43    43    43

    1808  1172  1173  1174  1175  1176  1178  1179   222   542   545   546   544   543   547
1339  1338  1337  1334  1333
      43    43    43    43    43    43    43    43    43    43    43    43    43    43    43
 43    43    43    43    43

    2039  2040  2041  2045   492   493   494   495   496   498   499    40    39   696  1029
1028  1027  1026  1023  1021
      43    43    43    43    44    44    44    44    44    44    44    44    44    44    45
 45    45    45    45    45

    1017  1020  1018  1015  1014  1013  1010   428   427   624   623   622   621   619   618
617   616   615   614   225
      45    45    45    45    45    45    45    45    45    45    45    45    45    45    45
 45    45    45    45    45

     432   433   769   266   267  1840  1839  1838  1805  1518  1517  1516  1515  1513  1512
1511  1510  1509   743   448
      45    45    45    45    45    45    45    45    45    45    45    45    45    45    45
 45    45    45    45    45

     447   446   445   206   205   204   203   202   201   200  1031   697   698   429   173
174   175   176   224   223
      45    45    45    45    45    45    45    45    45    45    46    47    47    47    47
 47    48    48    48    48

    1071  1070  1069  1068  1067  1190  1191  1455  1456  1457   418   417   416   415   412
411   410   409   406   405
      48    48    48    48    48    48    48    48    48    48    48    48    48    48    49
 49    50    50    50    50

     403   402   401   400   399   398   397   396   395   393   392   391   390   389  1674
541   540   538  2189  2187
      50    50    50    50    50    50    50    50    50    50    50    50    50    50    50
 50    50    50    51    51

    2186   771   671   672 13450     1     2     3     4   651   650   648   647   646   645
644   643   642   641   640
      51    52    52    52    52    52    52    52    52    52    52    52    52    52    52
 52    52    52    52    52

     639   638   620   637   635   634  1561   178   177   208   209   255   254   253   252
251   250   249   248   247
      52    52    52    52    52    52    52    52    52    52    52    52    52    52    52
 52    52    52    52    52

     246   245   244   243  1994  1995   239   238   237   235   236  1321  1523  1522  1524
1526  1527  1528   626   625
      52    52    52    52    52    52    52    52    52    52    52    52    52    52    52
 52    52    52    52    52

     426   425   423   422   420   106   107   108   511   510   508   514   512   515   276
```

```
 613  1505   578   579   277
     52     52     52     52     52     52     52     52     52     52     52     52     52     52     52
  52    52    53    53    53

    278   1473   1217    216    215   1100   1101   1102   1103   1104   1105    221    220    219    218
 217  1269  1270  1271  1272
     53     53     53     53     53     53     53     53     53     53     53     54     54     54     54
  54    54    54    54    54

   1275   1277   1279   1280   1281   1282   1284   1285   1283   1286   1288   1290   1291   1292   1293
1294  1295  1296  1297  1298
     54     54     54     54     54     54     54     54     54     54     54     54     54     54     54
  54    54    54    54    54

   1560   1559   1558   1557   1556   1555   2046    759    760    761    767    762    764    765   1506
 214   213   211   212   662
     54     54     55     55     55     55     56     56     56     56     56     56     56     56     56
  56    56    56    56    56

    661    660    658    657    656    180    179    663    664    666    665    667    669    670   1239
1237  1235  1236  1233  1228
     56     56     56     56     56     56     56     56     56     56     56     56     56     56     56
  56    56    56    56    56

    768   1218    766   1507   1508    744    745   1521   1520   1519    491    490    489    488    486
 485   484   483   481   480
     56     56     56     56     56     56     56     57     57     57     58     58     58     58     58
  58    59    59    59    59

    479    478    477   1824   1825   1826   1828   1829   1831   1832   1833   1834   1835   1837   1804
1802  1801  1803  1798  1800
     59     59     59     60     60     60     60     60     60     60     60     60     60     60     60
  60    60    60    60    60

   1326   1325   1324   1323    259    260    261    262    263    264   1268   1266   1265   1264   1263
1315   680   679  1320   231
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
  60    60    60    60    60

   1156   1154   1155    652    653   1465   1462   1463   1461   1460   1464   1469   1471   1467   1468
1470  1466  1472  1474  1476
     61     61     62     63     63     63     63     63     63     63     63     63     63     63     63
  63    63    63    63    63

   1477   1479   1480   1186   1187   1188   1189    777    776    551    552    553    557    558    191
 190   188   189  1199  1200
     63     63     63     63     63     63     63     63     63     63     63     63     63     64     64
  64    64    64    64    64

   1201   1181   1182    275    274    273    272    271    270    269    268   1842    803    804    806
 532   533   534   535   536
     64     64     64     64     64     64     64     64     64     64     64     64     64     64     64
  64    64    64    64    64

    537    467    466    726   2185   1259   1258   1257   1256   1255   1254   1253   8784   1252   1251
1250  1249  1248  1247  1246
     64     64     64     64     64     65     65     65     65     65     65     65     65     65     65
  65    65    65    65    65

   1244   1242   1241   1240    594    593    592    589    591    590    588   1303   1305   1306   1307
1308  1309  1312  1313   580
     65     65     65     65     65     65     65     65     65     65     65     65     65     65     65
  65    65    65    65    65

    581    582    583    584    586    585    587   1302   1301   1300   1299    633    632    630    629
```

```
628   627 13447 13448 13449
    65    65    65    65    65    65    65    65    65    65    65    65    65    65    65
 65    65    65    65    65

  1076  1075  1074  1073   289   290   291   530   529   528   526   525   524   523   522
13465 13466 13458 13457 13456
    66    66    66    66    66    66    66    66    66    66    66    66    66    66    66
 66    66    66    66    66

 13455 13454 13453 13452 13451   678   677    41    42    43   693   689   690   691   687
686   685   684   681   683
    66    67    67    67    67    67    67    67    67    67    67    67    67    67    67
 67    67    67    67    67

   682   475   474   473   471  1679  1055  1057  1056    37  2066  2067  2068  2069  2071
2070  2072  2073  2074  2075
    67    67    67    67    67    68    68    68    68    68    68    68    68    68    68
 68    68    68    68    68

  2076  2077  2079  2078  2084  2085  1793  1794  1795  1796   229   230    46   431   430
608   607  1332  1331  1330
    68    68    68    68    68    68    68    68    68    68    68    68    69    69    69
 69    69    69    69    69

  1329  1327  2038  2037  2036  2035  2034  2033  2032   241   242  1993  1992  1990  1989
1988  1987  1986  1985  1984
    69    69    69    69    69    69    69    69    69    69    69    69    69    69    69
 69    69    69    69    69

  1982  1981  1977  1979  1980  1975  1610  1568  1615  1613   110  1548  1547  1550  1551
1552  1553  1554   439   440
    69    69    69    69    69    69    69    69    69    69    69    70    70    71    71
 71    71    71    71    71

   441   442   443   444   279   280   281  1054   192   193  1262  1316   437   438   559
560  1482  1483  1484  1485
    71    71    71    71    71    71    71    72    72    72    72    72    72    72    72
 72    72    72    72    72

  1486  1487  1489  1488  1490  1491  1492  1493  1494  1495  1496  1500  1502   388   753
752   751   750  2006  1809
    72    72    72    72    72    72    72    72    72    72    72    72    72    72    72
 72    72    72    73    73

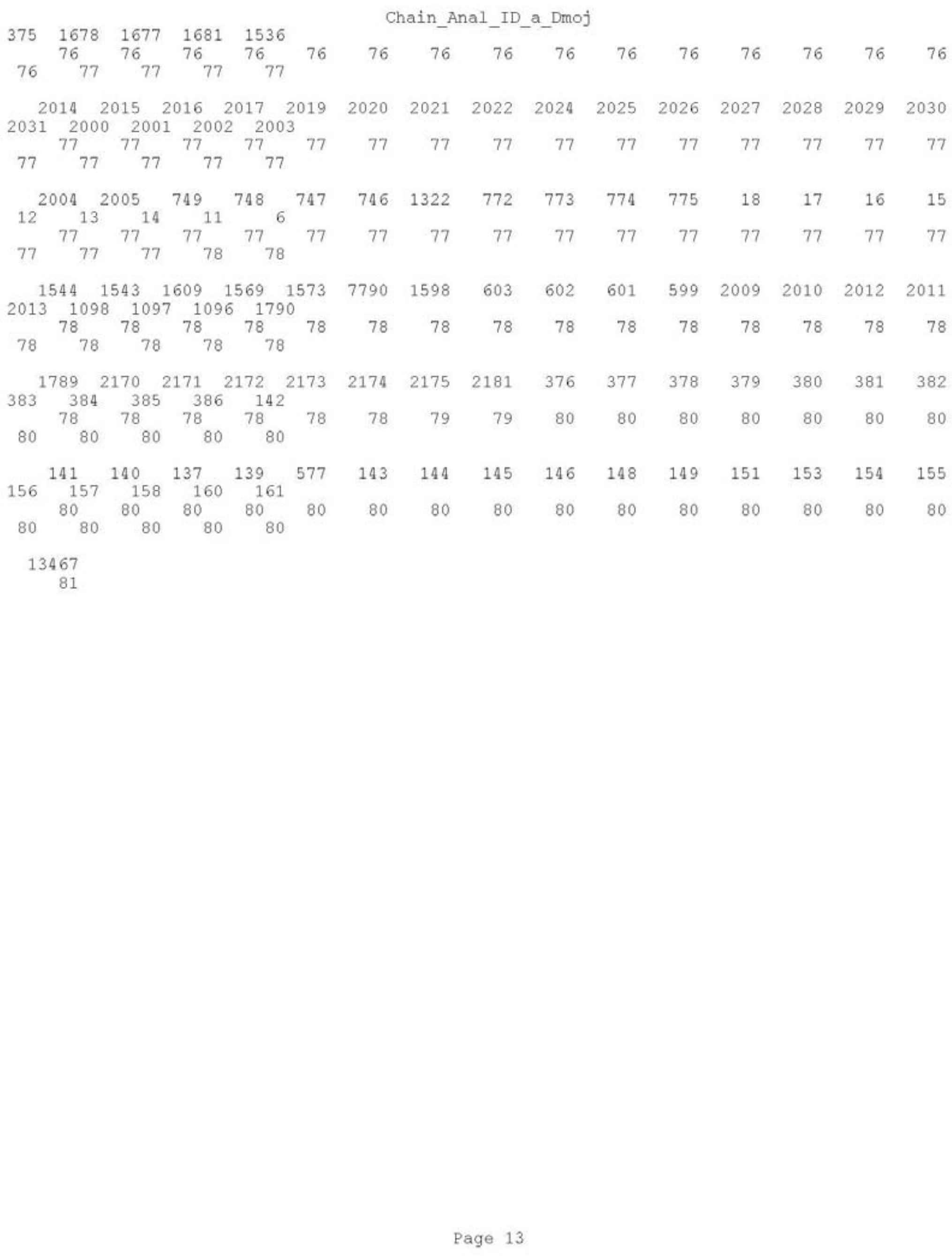
  1810  1811  1812  1813  1814  1815  1816   654   470   727   728   729   730   731   732
733   735   736   737   741
    73    73    73    73    73    73    73    73    73    73    73    73    73    73    73
 73    73    74    74    75

   738   739   124   123   122   102   103   104   469   468  1260  1261   604   606  1165
1166  1167  1168  1169  1170
    75    75    75    75    75    75    75    75    75    75    75    75    75    75    75
 75    75    75    75    75

  1171  2007  2008   598   597   596   595   434   436    26    25    24    22    20    21
 19    23  1820  1822  1823
    75    75    75    75    75    75    75    75    75    75    75    75    75    75    75
 75    75    75    75    75

  2093  2092  2091  2090  2089  2088  2087  2086  1792  1791  1094  1093  1092  1091  1090
1089  1088  1087  1086  1085
    75    75    75    75    75    75    75    75    75    75    75    75    76    76    76
 76    76    76    76    76

  1084  1083  1082  1081  1080  1077    44  1152  1151  1150   373   369   372  1149   374
```



```
                                   Chain_Anal_ID_a_Dmoj
375  1678  1677  1681  1536
     76    76    76    76    76    76    76    76    76    76    76    76    76    76    76
 76    77    77    77    77

   2014  2015  2016  2017  2019  2020  2021  2022  2024  2025  2026  2027  2028  2029  2030
2031  2000  2001  2002  2003
     77    77    77    77    77    77    77    77    77    77    77    77    77    77    77
 77    77    77    77    77

   2004  2005   749   748   747   746  1322   772   773   774   775    18    17    16    15
 12    13    14    11     6
     77    77    77    77    77    77    77    77    77    77    77    77    77    77    77
 77    77    77    78    78

   1544  1543  1609  1569  1573  7790  1598   603   602   601   599  2009  2010  2012  2011
2013  1098  1097  1096  1790
     78    78    78    78    78    78    78    78    78    78    78    78    78    78    78
 78    78    78    78    78

   1789  2170  2171  2172  2173  2174  2175  2181   376   377   378   379   380   381   382
383   384   385   386   142
     78    78    78    78    78    78    79    79    80    80    80    80    80    80    80
 80    80    80    80    80

    141   140   137   139   577   143   144   145   146   148   149   151   153   154   155
156   157   158   160   161
     80    80    80    80    80    80    80    80    80    80    80    80    80    80    80
 80    80    80    80    80

  13467
     81
```

```
                                     Chain_Anal_ID_b_Dmoj
Appendix B. Linkage Chain Analysis for the Muller B Element Comparison of D. virilis and D.
mojavensis
 Breakpoints Remaining=  54
 Chain  1            left CLG          right CLG
   BP_045_046         4610   4610 |  3518   3457
                     ---------------------------
   BP_001_002         2211   2211 |  3479   3469
                     ---------------------------
   BP_010_011         3843   3842 |  3841   2883        A
                     ---------------------------
   BP_016_017         3646   3882 |  3477   3647
                     ---------------------------
   BP_002_003         3479   3469 |  3881   4398
                     ---------------------------
   BP_041_042         3472   3511 |  3463   3524
                     ---------------------------
   BP_046_047         3518   3457 |  2834   3276
                     ---------------------------
   BP_043_044         3499   3499 |  3521   3522
                     ---------------------------

 Breakpoints Remaining=  52     (D.moj lineage inversion)(1)
 Chain  2            left CLG          right CLG
   BP_056_057         4280   4283 |  4284   4285        A
                     ---------------------------
   BP_057_058         4284   4285 |  2264   2722        A by inference
                     ---------------------------

 Breakpoints Remaining=  50     (D.moj lineage inversion)(1)
 Chain  3            left CLG          right CLG
   BP_035_036         2794   2475 |  2476   2582        A
                     ---------------------------
   BP_055_056         4263   4279 |  4280   4283        A
                     ---------------------------

 Breakpoints Remaining=  48     (D.vir lineage inversion)(1)
 Chain  4            left CLG          right CLG
   BP_011_012         3841   2883 |  2549   4459
                     ---------------------------
   BP_020_021         3653   2879 |  3034   3240
                     ---------------------------

 Breakpoints Remaining=  46     (D.moj lineage inversion)(1)
 Chain  5            left CLG          right CLG
   BP_061_062         4195   4186 |  4197   4197        A by inference
                     ---------------------------
   BP_060_061         4555   4194 |  4195   4186        A
                     ---------------------------

 Breakpoints Remaining=  44     (D.moj lineage inversion)(1)
 Chain  6            left CLG          right CLG
   BP_003_004         3881   4398 |  4399   4400        A
                     ---------------------------
   BP_004_005         4399   4400 |  4397   3910        A by inference
                     ---------------------------

 Breakpoints Remaining=  42     (D.moj lineage inversion)(1)
 Chain  7            left CLG          right CLG
   BP_034_035         2585   3171 |  2794   2475        A by inference
                     ---------------------------
   BP_022_023         2983   3288 |  3287   3823        A
                     ---------------------------

 Breakpoints Remaining=  40     (D.vir lineage inversion)(1)
 Chain  8            left CLG          right CLG
```

```
  BP_025_026         3010   2813 |  3157   2814
                    ---------------------------
  BP_026_027         3157   2814 |  4147   3731
                    ---------------------------

Breakpoints Remaining=  38     (D.vir lineage inversion)(1)
Chain  9            left CLG          right CLG
  BP_048_049         3400   3410 |  3412   3411
                    ---------------------------
  BP_049_050         3412   3411 |  3413   3575
                    ---------------------------

Breakpoints Remaining=  36     (D.vir lineage inversion)(1)
Chain 10            left CLG          right CLG
  BP_027_028         4147   3731 |  3574   4173
                    ---------------------------
  BP_050_051         3413   3575 |  3732   3736
                    ---------------------------

Breakpoints Remaining=  34     (D.vir lineage inversion)(1)
Chain 11            left CLG          right CLG
  BP_018_019         4633   2905 |  2907   2906
                    ---------------------------
  BP_019_020         2907   2906 |  3653   2879
                    ---------------------------

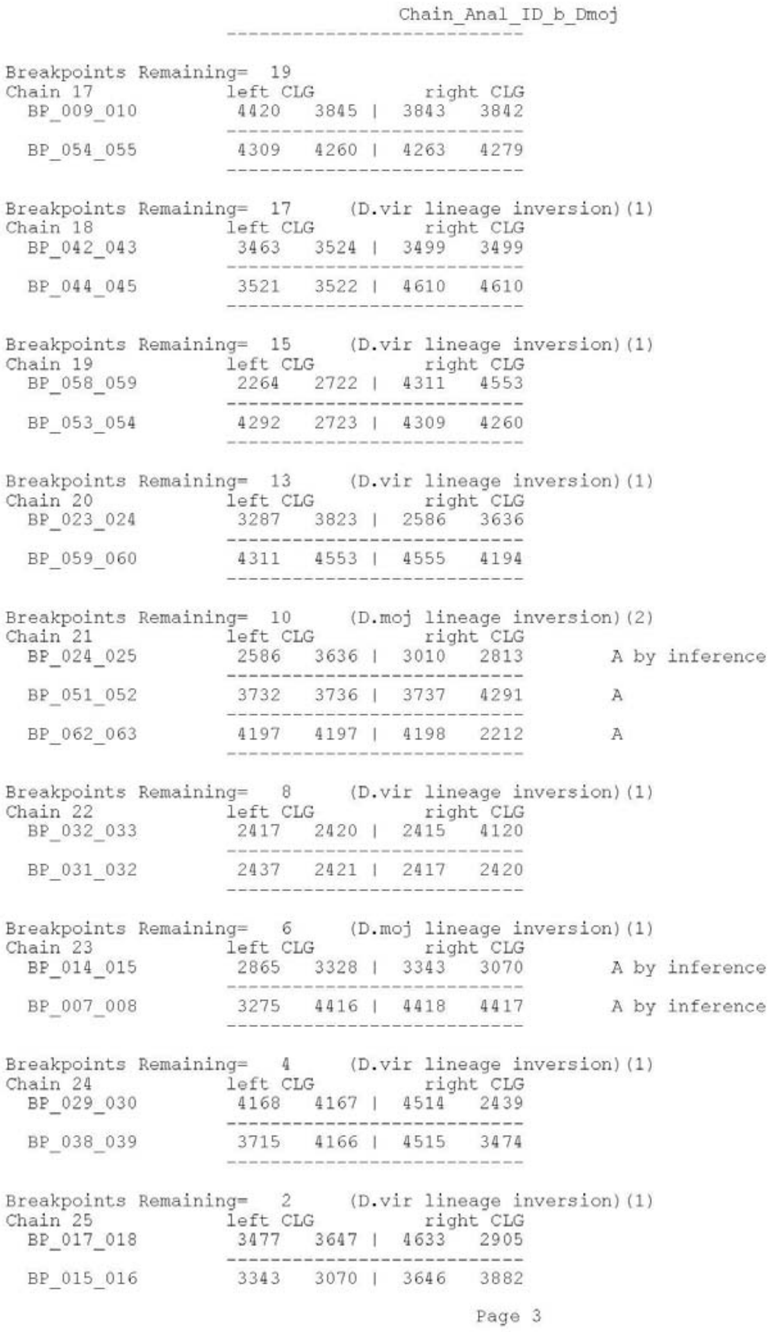
Breakpoints Remaining=  31     (D.moj lineage inversion)(2)
Chain 12            left CLG          right CLG
  BP_005_006         4397   3910 |  3911   3241        A
                    ---------------------------
  BP_030_031         4514   2439 |  2437   2421        A by inference
                    ---------------------------
  BP_012_013         2549   4459 |  4460   3005        A
                    ---------------------------

Breakpoints Remaining=  28     (D.vir lineage inversion)(2)
Chain 13            left CLG          right CLG
  BP_013_014         4460   3005 |  2865   3328
                    ---------------------------
  BP_008_009         4418   4417 |  4420   3845        A
                    ---------------------------
  BP_033_034         2415   4120 |  2585   3171
                    ---------------------------

Breakpoints Remaining=  26     (D.moj lineage inversion)(1)
Chain 14            left CLG          right CLG
  BP_039_040         4515   3474 |  3489   3473        A by inference
                    ---------------------------
  BP_040_041         3489   3473 |  3472   3511        A
                    ---------------------------

Breakpoints Remaining=  24     (D.moj lineage inversion)(1)
Chain 15            left CLG          right CLG
  BP_037_038         3711   3714 |  3715   4166        A
                    ---------------------------
  BP_036_037         2476   2582 |  3711   3714        A by inference
                    ---------------------------

Breakpoints Remaining=  21     (D.vir lineage inversion)(2)
Chain 16            left CLG          right CLG
  BP_021_022         3034   3240 |  2983   3288
                    ---------------------------
  BP_006_007         3911   3241 |  3275   4416
                    ---------------------------
  BP_047_048         2834   3276 |  3400   3410
```



```
                              Chain_Anal_ID_b_Dmoj
                    ---------------------------

Breakpoints Remaining=  19
Chain 17             left CLG          right CLG
  BP_009_010          4420   3845 |  3843   3842
                     ---------------------------
  BP_054_055          4309   4260 |  4263   4279
                     ---------------------------

Breakpoints Remaining=  17     (D.vir lineage inversion)(1)
Chain 18             left CLG          right CLG
  BP_042_043          3463   3524 |  3499   3499
                     ---------------------------
  BP_044_045          3521   3522 |  4610   4610
                     ---------------------------

Breakpoints Remaining=  15     (D.vir lineage inversion)(1)
Chain 19             left CLG          right CLG
  BP_058_059          2264   2722 |  4311   4553
                     ---------------------------
  BP_053_054          4292   2723 |  4309   4260
                     ---------------------------

Breakpoints Remaining=  13     (D.vir lineage inversion)(1)
Chain 20             left CLG          right CLG
  BP_023_024          3287   3823 |  2586   3636
                     ---------------------------
  BP_059_060          4311   4553 |  4555   4194
                     ---------------------------

Breakpoints Remaining=  10     (D.moj lineage inversion)(2)
Chain 21             left CLG          right CLG
  BP_024_025          2586   3636 |  3010   2813        A by inference
                     ---------------------------
  BP_051_052          3732   3736 |  3737   4291        A
                     ---------------------------
  BP_062_063          4197   4197 |  4198   2212        A
                     ---------------------------

Breakpoints Remaining=   8     (D.vir lineage inversion)(1)
Chain 22             left CLG          right CLG
  BP_032_033          2417   2420 |  2415   4120
                     ---------------------------
  BP_031_032          2437   2421 |  2417   2420
                     ---------------------------

Breakpoints Remaining=   6     (D.moj lineage inversion)(1)
Chain 23             left CLG          right CLG
  BP_014_015          2865   3328 |  3343   3070        A by inference
                     ---------------------------
  BP_007_008          3275   4416 |  4418   4417        A by inference
                     ---------------------------

Breakpoints Remaining=   4     (D.vir lineage inversion)(1)
Chain 24             left CLG          right CLG
  BP_029_030          4168   4167 |  4514   2439
                     ---------------------------
  BP_038_039          3715   4166 |  4515   3474
                     ---------------------------

Breakpoints Remaining=   2     (D.vir lineage inversion)(1)
Chain 25             left CLG          right CLG
  BP_017_018          3477   3647 |  4633   2905
                     ---------------------------
  BP_015_016          3343   3070 |  3646   3882

                                      Page 3
```

```
                                    Chain_Anal_ID_b_Dmoj
                    ---------------------------

Breakpoints Remaining=   0      (D.moj lineage inversion)(1)
Chain 26            left CLG         right CLG
  BP_028_029         3574   4173 |  4168   4167        A by inference
                    ---------------------------
  BP_052_053         3737   4291 |  4292   2723        A
                    ---------------------------


Chain Analysis Summary Data
---------------------------

Chains with  2 Breakpoints=  21
Chains with  3 Breakpoints=   4
Chains with  4 Breakpoints=   0
Chains with  5 Breakpoints=   0
Chains with  6 Breakpoints=   0
Chains with  7 Breakpoints=   0
Chains with  8 Breakpoints=   1
Chains with  9 Breakpoints=   0
Chains with 10 Breakpoints=   0
Chains with 11 Breakpoints=   0
Chains with 12 Breakpoints=   0

Number of Inversions=  36
Number of Breakpoints=  62
Reusage Index=     1.161


Conserved Linkage Group Intervals
---------------------------------

CLG  No.Genes  Left_IDX  Right_IDX  Left_Gene  Right_Gene
  1         1         1          1       2211        2211
  2         3         2          4       3479        3469
  3        15         5         19       3881        4398
  4         2        20         21       4399        4400
  5        21        22         42       4397        3910
  6        82        43        124       3911        3241
  7        43       125        167       3275        4416
  8         2       168        169       4418        4417
  9        70       170        239       4420        3845
 10         2       240        241       3843        3842
 11        75       242        316       3841        2883
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 Gene List and Conserved Linkage Group Calls
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  3521  3522  4610  3518  3496  3457  2834  2835  2836  2837  2838  2839  2840  2842  2598
2597  2596  3282  3281  3280
    44    44    45    46    46    46    47    47    47    47    47    47    47    47    47
 47    47    47    47    47

  3278  3277  3276  3400  3401  3402  3403  3404  3405  3406  3407  3408  3409  3410  3412
3411  3413  3414  3415  3416
    47    47    47    48    48    48    48    48    48    48    48    48    48    48    49
 49    50    50    50    50

  3364  3363  3362  3361  3359  3007  2828  3008  3009  3011  3012  3013  3059  3060  3061
3062  3063  3064  3015  3014
    50    50    50    50    50    50    50    50    50    50    50    50    50    50    50
```

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 50    50    50    50    50

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3045  3044  3043  3042  3041
     50    50    50    50    50    50    50    50    50    50    50    50    50    50    50
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   3040  3038  3037  3035  3036  2686  2683  2682  2681  2680  2679  2677  2675  2674  2673
2672  2670  2671  2668  2669
     50    50    50    50    50    50    50    50    50    50    50    50    50    50    50
 50    50    50    50    50

   2666  2665  2664  2663  2662  2661  2660  2659  2658  2657  2656  2655  2654  2652  2651
2650  2649  2648  2647  2646
     50    50    50    50    50    50    50    50    50    50    50    50    50    50    50
 50    50    50    50    50

   3116  3115  3114  3113  3112  3111  3110  3109  3108  3107  3106  3104  3103  3102  3101
3100  3099  3098  3096  3097
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   3095  3092  3091  3090  3089  3088  3087  3086  3085  3083  2778  2779  2780  2781  3602
3600  3599  3598  3597  3596
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   3595  3601  3594  3591  3590  3589  3588  3587  3586  3585  3584  3583  3581  3579  3578
3577  3576  3575  3732  3733
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3219  3220  3221  3222  3223
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   3224  3225  3226  3227  3228  3229  3230  3231  3232  3233  3234  4331  4329  4328  4327
4326  4325  4324  4323  4322
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3182  3183  3184  3185  3187
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 53    53    53    53    53

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4215  4216  4217  4220  4219
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 53    53    53    53    53

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4243  4245  4244  4246  3749
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 53    53    53    53    53

   3750  3754  3751  3756  3757  3758  3759  3760  4490  4488  4489  4487  3782  4486  4484
4483  4482  4481  4480  4478
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 53    53    53    53    53

   2736  2735  2734  2733  2732  2730  2729  2728  2727  2726  2725  2724  2723  4309  4310
4307  4306  4308  4305  4304
     53    53    53    53    53    53    53    53    53    53    53    53    53    54    54
```

```
 54    54    54    54    54

   4303  4300  4299  4302  4248  4250  4249  4251  4253  4254  4255  4256  4258  4259  4260
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   4269  4270  4271  4272  4273  4274  4275  4276  4278  4279  4280  4282  4281  4283  4284
4285  2264  2265  2266  2267
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   2268  2269  2270  2271  2272  2273  2274  2275  2276  2277  2278  2279  2280  2281  2282
2283  2284  2285  2286  2287
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2312  2313  2314  2315  2316
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   2317  2318  2319  2320  2321  2322  2323  2324  2325  2326  2327  2328  2329  2330  2331
2334  2335  3131  3132  3136
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   3135  3134  3133  3137  3141  3139  3138  3140  4560  4558  4561  4562  4563  4564  4565
4566  4567  4568  2863  2862
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   2861  2860  2859  2858  2857  2856  2855  2853  2854  2851  2850  2849  2848  2846  2847
2845  2602  2603  2604  2605
     58    58    58    58    58    58    58    58    58    58    58    58    58    58    58
 58    58    58    58    58

   2356  2355  2354  2353  2352  2351  2350  2348  2347  2346  2345  2344  2343  2342  2341
2340  2339  2337  2716  2717
     58    58    58    58    58    58    58    58    58    58    58    58    58    58    58
 58    58    58    58    58

   2719  2720  2721  2722  4311  2691  2690  2689  2688  2687  2773  2771  2770  2769  2768
2767  2766  2765  2764  2763
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 59    59    59    59    59

   2762  2454  4551  4552  4553  4555  4556  4557  4559  3143  3142  3144  3145  3147  3148
3793  3792  3790  3788  3786
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 60    60    60    60    60

   3787  3784  3785  3783  3781  3779  3780  3778  3774  3771  3773  3768  3767  3766  3765
3764  3761  3547  3548  2774
     60    60    60    60    60    60    60    60    60    60    60    60    60    60    60
 60    60    60    60    60

   2775  2776  3082  3080  3079  3078  3077  3075  4099  4100  3612  3610  3609  2448  3608
3607  3605  3604  3603  2785
     60    60    60    60    60    60    60    60    60    60    60    60    60    60    60
 60    60    60    60    60

   2786  2791  2544  4176  4179  4181  4182  4188  4193  4194  4195  4186  4197  4198  4199
4200  4201  4203  4202  4204
     60    60    60    60    60    60    60    60    60    60    61    61    62    63    63
```

```
 63    63    63    63    63

   4205  2758  2759  3125  3126  3127  3129  3128  3130  2714  2711  2710  2709  2708  2232
2231  2230  2228  2229  2227
     63    63    63    63    63    63    63    63    63    63    63    63    63    63    63
 63    63    63    63    63

   2226  2224  2225  2222  2221  2214  2215  2216  2217  2218  2219  2212     0     0     0
  0     0     0     0     0
     63    63    63    63    63    63    63    63    63    63    63    63     0     0     0
  0     0     0     0     0
```

```
                                   Chain_Anal_ID_c_Dmoj
Appendix C. Linkage Chain Analysis for the Muller C Element Comparison of D. virilis and D.
mojavensis
 Breakpoints Remaining=  70     (D.vir lineage inversion)(2)
 Chain  1            left CLG         right CLG
   BP_001_002         5972   7034 |  6247   4668
                     ---------------------------
   BP_003_004         4913   5233 |  4842   5905
                     ---------------------------
   BP_005_006         4918   4919 |  5109   5108
                     ---------------------------

 Breakpoints Remaining=  68     (D.vir lineage inversion)(2)
 Chain  2            left CLG         right CLG
   BP_004_005         4842   5905 |  4918   4919
                     ---------------------------
   BP_002_003         6247   4668 |  4913   5233
                     ---------------------------

 Breakpoints Remaining=  66     (D.moj lineage inversion)(1)
 Chain  3            left CLG         right CLG
   BP_006_007         5109   5108 |  5107   4800        A
                     ---------------------------
   BP_064_065         4884   5964 |  5232   5089        A by inference
                     ---------------------------

 Breakpoints Remaining=  64     (D.vir lineage inversion)(1)
 Chain  4            left CLG         right CLG
   BP_008_009         5090   6338 |  6341   6340
                     ---------------------------
   BP_009_010         6341   6340 |  6343   5494
                     ---------------------------

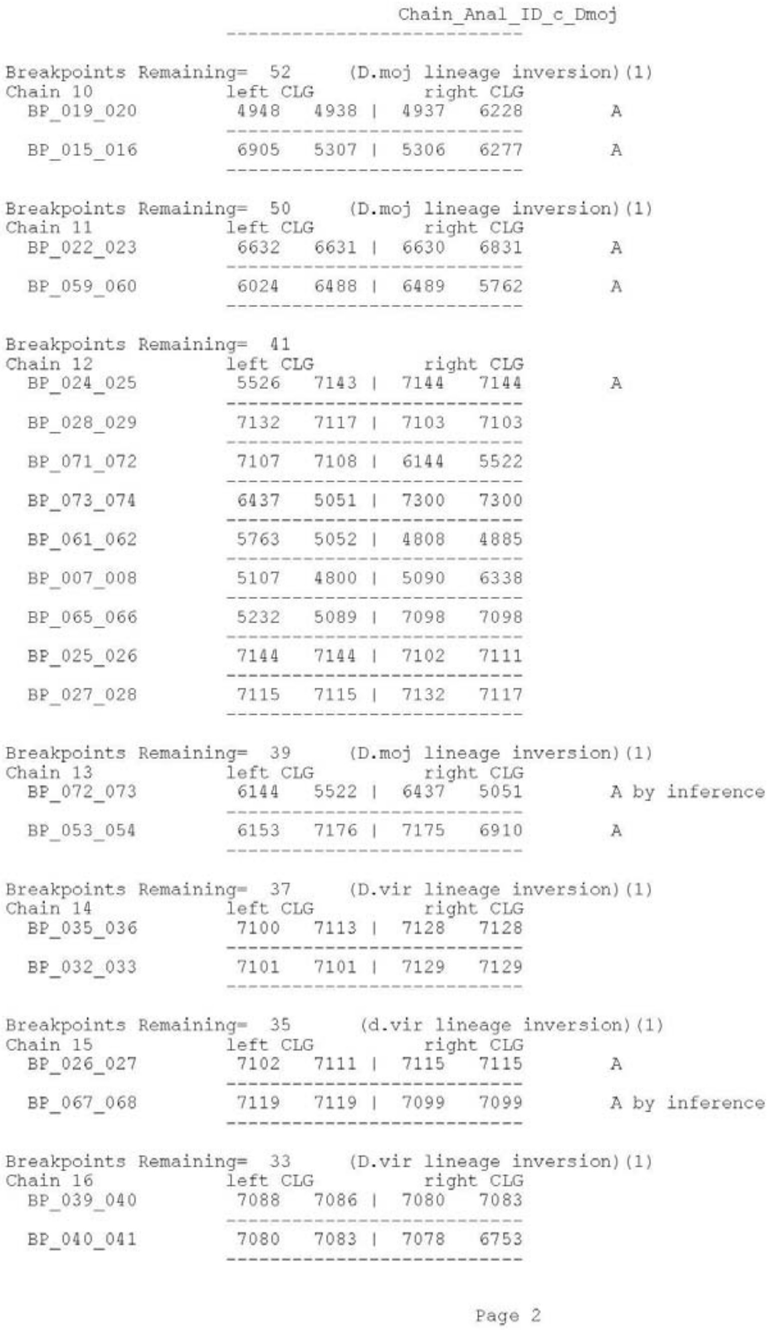
 Breakpoints Remaining=  62     (D.vir lineage inversion)(1)
 Chain  5            left CLG         right CLG
   BP_010_011         6343   5494 |  6829   6633
                     ---------------------------
   BP_012_013         6415   5495 |  6828   4849
                     ---------------------------

 Breakpoints Remaining=  60     (D.vir lineage inversion)(1)
 Chain  6            left CLG         right CLG
   BP_011_012         6829   6633 |  6415   5495
                     ---------------------------
   BP_021_022         5796   6414 |  6632   6631
                     ---------------------------

 Breakpoints Remaining=  58     (D.moj lineage inversion)(1)
 Chain  7            left CLG         right CLG
   BP_014_015         4848   6906 |  6905   5307        A
                     ---------------------------
   BP_013_014         6828   4849 |  4848   6906        A
                     ---------------------------

 Breakpoints Remaining=  56     (D.moj lineage inversion)(1)
 Chain  8            left CLG         right CLG
   BP_016_017         5306   6277 |  6278   6279        A
                     ---------------------------
   BP_017_018         6278   6279 |  6274   4949        A by inference
                     ---------------------------

 Breakpoints Remaining=  54     (D.moj lineage inversion)(1)
 Chain  9            left CLG         right CLG
   BP_018_019         6274   4949 |  4948   4938        A
                     ---------------------------
   BP_020_021         4937   6228 |  5796   6414        A by inference
```



```
                         Chain_Anal_ID_c_Dmoj
                   ---------------------------

Breakpoints Remaining=  52     (D.moj lineage inversion)(1)
Chain 10            left CLG          right CLG
  BP_019_020         4948   4938 |  4937   6228        A
                   ---------------------------
  BP_015_016         6905   5307 |  5306   6277        A
                   ---------------------------

Breakpoints Remaining=  50     (D.moj lineage inversion)(1)
Chain 11            left CLG          right CLG
  BP_022_023         6632   6631 |  6630   6831        A
                   ---------------------------
  BP_059_060         6024   6488 |  6489   5762        A
                   ---------------------------

Breakpoints Remaining=  41
Chain 12            left CLG          right CLG
  BP_024_025         5526   7143 |  7144   7144        A
                   ---------------------------
  BP_028_029         7132   7117 |  7103   7103
                   ---------------------------
  BP_071_072         7107   7108 |  6144   5522
                   ---------------------------
  BP_073_074         6437   5051 |  7300   7300
                   ---------------------------
  BP_061_062         5763   5052 |  4808   4885
                   ---------------------------
  BP_007_008         5107   4800 |  5090   6338
                   ---------------------------
  BP_065_066         5232   5089 |  7098   7098
                   ---------------------------
  BP_025_026         7144   7144 |  7102   7111
                   ---------------------------
  BP_027_028         7115   7115 |  7132   7117
                   ---------------------------

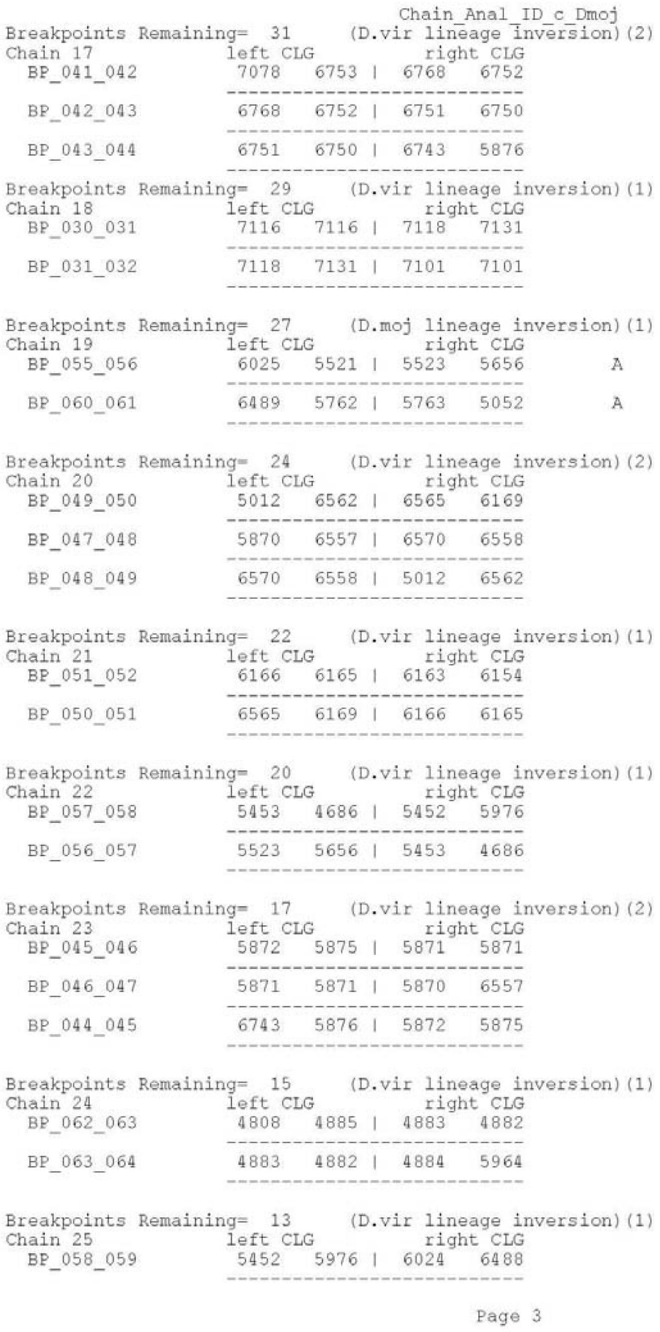
Breakpoints Remaining=  39     (D.moj lineage inversion)(1)
Chain 13            left CLG          right CLG
  BP_072_073         6144   5522 |  6437   5051        A by inference
                   ---------------------------
  BP_053_054         6153   7176 |  7175   6910        A
                   ---------------------------

Breakpoints Remaining=  37     (D.vir lineage inversion)(1)
Chain 14            left CLG          right CLG
  BP_035_036         7100   7113 |  7128   7128
                   ---------------------------
  BP_032_033         7101   7101 |  7129   7129
                   ---------------------------

Breakpoints Remaining=  35      (d.vir lineage inversion)(1)
Chain 15            left CLG          right CLG
  BP_026_027         7102   7111 |  7115   7115        A
                   ---------------------------
  BP_067_068         7119   7119 |  7099   7099        A by inference
                   ---------------------------

Breakpoints Remaining=  33     (D.vir lineage inversion)(1)
Chain 16            left CLG          right CLG
  BP_039_040         7088   7086 |  7080   7083
                   ---------------------------
  BP_040_041         7080   7083 |  7078   6753
                   ---------------------------

                              Page 2
```



```
                                   Chain_Anal_ID_c_Dmoj
Breakpoints Remaining=  31     (D.vir lineage inversion)(2)
Chain 17            left CLG          right CLG
  BP_041_042         7078   6753 |  6768   6752
                    ---------------------------
  BP_042_043         6768   6752 |  6751   6750
                    ---------------------------
  BP_043_044         6751   6750 |  6743   5876
                    ---------------------------
Breakpoints Remaining=  29     (D.vir lineage inversion)(1)
Chain 18            left CLG          right CLG
  BP_030_031         7116   7116 |  7118   7131
                    ---------------------------
  BP_031_032         7118   7131 |  7101   7101
                    ---------------------------

Breakpoints Remaining=  27     (D.moj lineage inversion)(1)
Chain 19            left CLG          right CLG
  BP_055_056         6025   5521 |  5523   5656        A
                    ---------------------------
  BP_060_061         6489   5762 |  5763   5052        A
                    ---------------------------

Breakpoints Remaining=  24     (D.vir lineage inversion)(2)
Chain 20            left CLG          right CLG
  BP_049_050         5012   6562 |  6565   6169
                    ---------------------------
  BP_047_048         5870   6557 |  6570   6558
                    ---------------------------
  BP_048_049         6570   6558 |  5012   6562
                    ---------------------------

Breakpoints Remaining=  22     (D.vir lineage inversion)(1)
Chain 21            left CLG          right CLG
  BP_051_052         6166   6165 |  6163   6154
                    ---------------------------
  BP_050_051         6565   6169 |  6166   6165
                    ---------------------------

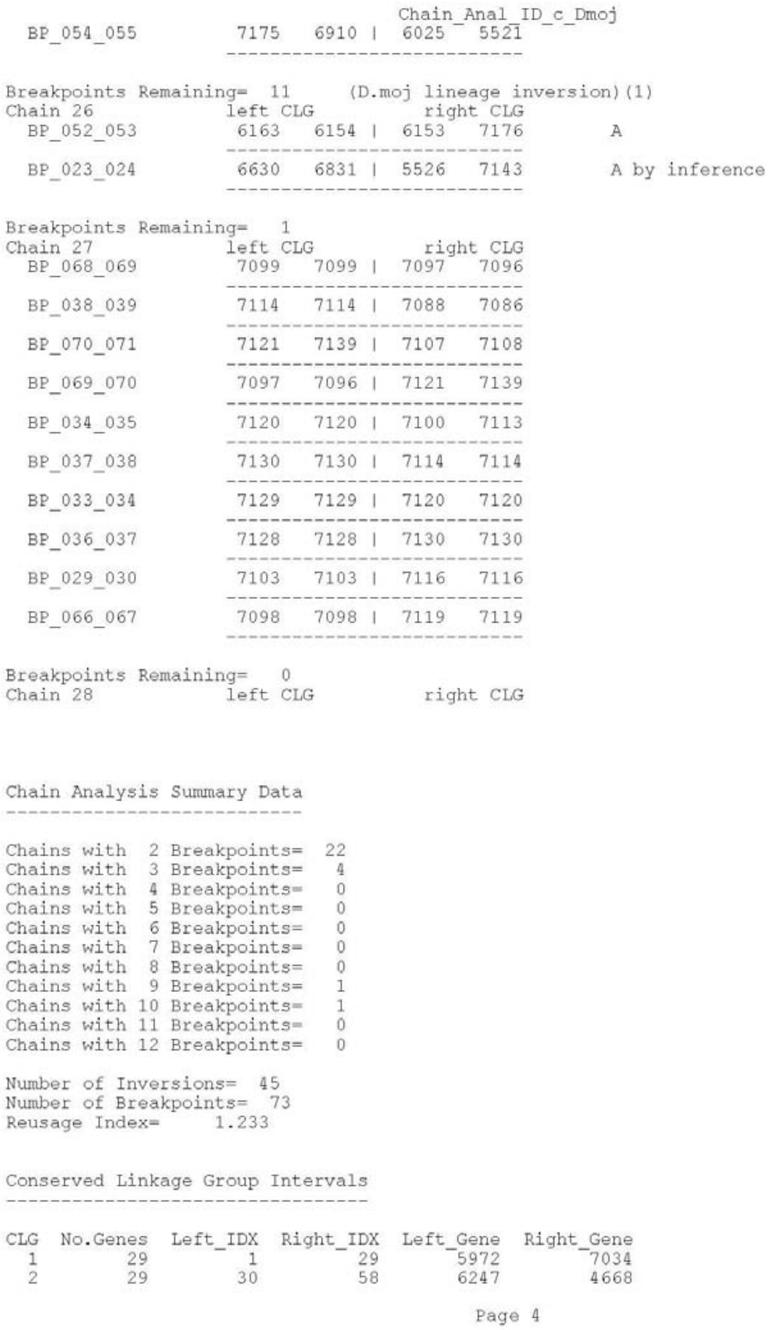
Breakpoints Remaining=  20     (D.vir lineage inversion)(1)
Chain 22            left CLG          right CLG
  BP_057_058         5453   4686 |  5452   5976
                    ---------------------------
  BP_056_057         5523   5656 |  5453   4686
                    ---------------------------

Breakpoints Remaining=  17     (D.vir lineage inversion)(2)
Chain 23            left CLG          right CLG
  BP_045_046         5872   5875 |  5871   5871
                    ---------------------------
  BP_046_047         5871   5871 |  5870   6557
                    ---------------------------
  BP_044_045         6743   5876 |  5872   5875
                    ---------------------------

Breakpoints Remaining=  15     (D.vir lineage inversion)(1)
Chain 24            left CLG          right CLG
  BP_062_063         4808   4885 |  4883   4882
                    ---------------------------
  BP_063_064         4883   4882 |  4884   5964
                    ---------------------------

Breakpoints Remaining=  13     (D.vir lineage inversion)(1)
Chain 25            left CLG          right CLG
  BP_058_059         5452   5976 |  6024   6488
                    ---------------------------

                                          Page 3
```



```
                                    Chain_Anal_ID_c_Dmoj
  BP_054_055          7175   6910 |  6025   5521
                    ---------------------------

Breakpoints Remaining=  11     (D.moj lineage inversion)(1)
Chain 26            left CLG          right CLG
  BP_052_053          6163   6154 |  6153   7176        A
                    ---------------------------
  BP_023_024          6630   6831 |  5526   7143        A by inference
                    ---------------------------

Breakpoints Remaining=   1
Chain 27            left CLG          right CLG
  BP_068_069          7099   7099 |  7097   7096
                    ---------------------------
  BP_038_039          7114   7114 |  7088   7086
                    ---------------------------
  BP_070_071          7121   7139 |  7107   7108
                    ---------------------------
  BP_069_070          7097   7096 |  7121   7139
                    ---------------------------
  BP_034_035          7120   7120 |  7100   7113
                    ---------------------------
  BP_037_038          7130   7130 |  7114   7114
                    ---------------------------
  BP_033_034          7129   7129 |  7120   7120
                    ---------------------------
  BP_036_037          7128   7128 |  7130   7130
                    ---------------------------
  BP_029_030          7103   7103 |  7116   7116
                    ---------------------------
  BP_066_067          7098   7098 |  7119   7119
                    ---------------------------

Breakpoints Remaining=   0
Chain 28            left CLG          right CLG



Chain Analysis Summary Data
---------------------------

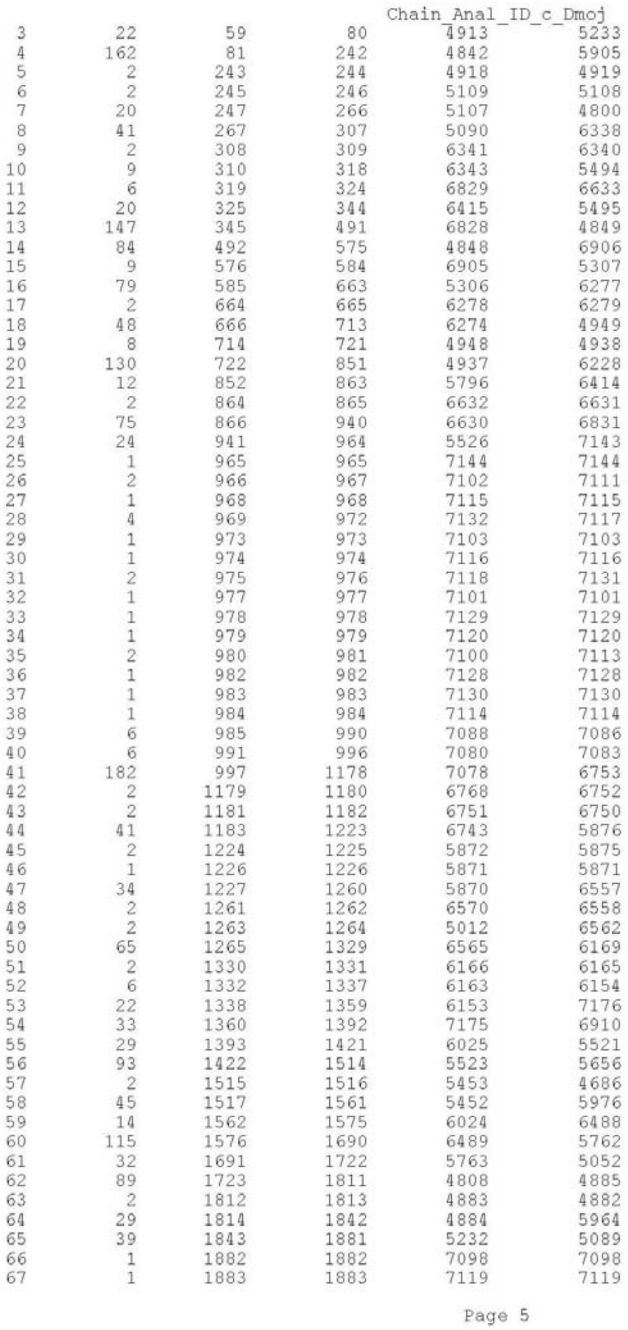

Chains with  2 Breakpoints=  22
Chains with  3 Breakpoints=   4
Chains with  4 Breakpoints=   0
Chains with  5 Breakpoints=   0
Chains with  6 Breakpoints=   0
Chains with  7 Breakpoints=   0
Chains with  8 Breakpoints=   0
Chains with  9 Breakpoints=   1
Chains with 10 Breakpoints=   1
Chains with 11 Breakpoints=   0
Chains with 12 Breakpoints=   0

Number of Inversions=  45
Number of Breakpoints=  73
Reusage Index=     1.233


Conserved Linkage Group Intervals
---------------------------------


CLG  No.Genes  Left_IDX  Right_IDX  Left_Gene  Right_Gene
  1        29         1         29       5972        7034
  2        29        30         58       6247        4668

                                          Page 4
```



```
                                          Chain_Anal_ID_c_Dmoj
 3      22       59       80       4913       5233
 4     162       81      242       4842       5905
 5       2      243      244       4918       4919
 6       2      245      246       5109       5108
 7      20      247      266       5107       4800
 8      41      267      307       5090       6338
 9       2      308      309       6341       6340
10       9      310      318       6343       5494
11       6      319      324       6829       6633
12      20      325      344       6415       5495
13     147      345      491       6828       4849
14      84      492      575       4848       6906
15       9      576      584       6905       5307
16      79      585      663       5306       6277
17       2      664      665       6278       6279
18      48      666      713       6274       4949
19       8      714      721       4948       4938
20     130      722      851       4937       6228
21      12      852      863       5796       6414
22       2      864      865       6632       6631
23      75      866      940       6630       6831
24      24      941      964       5526       7143
25       1      965      965       7144       7144
26       2      966      967       7102       7111
27       1      968      968       7115       7115
28       4      969      972       7132       7117
29       1      973      973       7103       7103
30       1      974      974       7116       7116
31       2      975      976       7118       7131
32       1      977      977       7101       7101
33       1      978      978       7129       7129
34       1      979      979       7120       7120
35       2      980      981       7100       7113
36       1      982      982       7128       7128
37       1      983      983       7130       7130
38       1      984      984       7114       7114
39       6      985      990       7088       7086
40       6      991      996       7080       7083
41     182      997     1178       7078       6753
42       2     1179     1180       6768       6752
43       2     1181     1182       6751       6750
44      41     1183     1223       6743       5876
45       2     1224     1225       5872       5875
46       1     1226     1226       5871       5871
47      34     1227     1260       5870       6557
48       2     1261     1262       6570       6558
49       2     1263     1264       5012       6562
50      65     1265     1329       6565       6169
51       2     1330     1331       6166       6165
52       6     1332     1337       6163       6154
53      22     1338     1359       6153       7176
54      33     1360     1392       7175       6910
55      29     1393     1421       6025       5521
56      93     1422     1514       5523       5656
57       2     1515     1516       5453       4686
58      45     1517     1561       5452       5976
59      14     1562     1575       6024       6488
60     115     1576     1690       6489       5762
61      32     1691     1722       5763       5052
62      89     1723     1811       4808       4885
63       2     1812     1813       4883       4882
64      29     1814     1842       4884       5964
65      39     1843     1881       5232       5089
66       1     1882     1882       7098       7098
67       1     1883     1883       7119       7119

                                      Page 5
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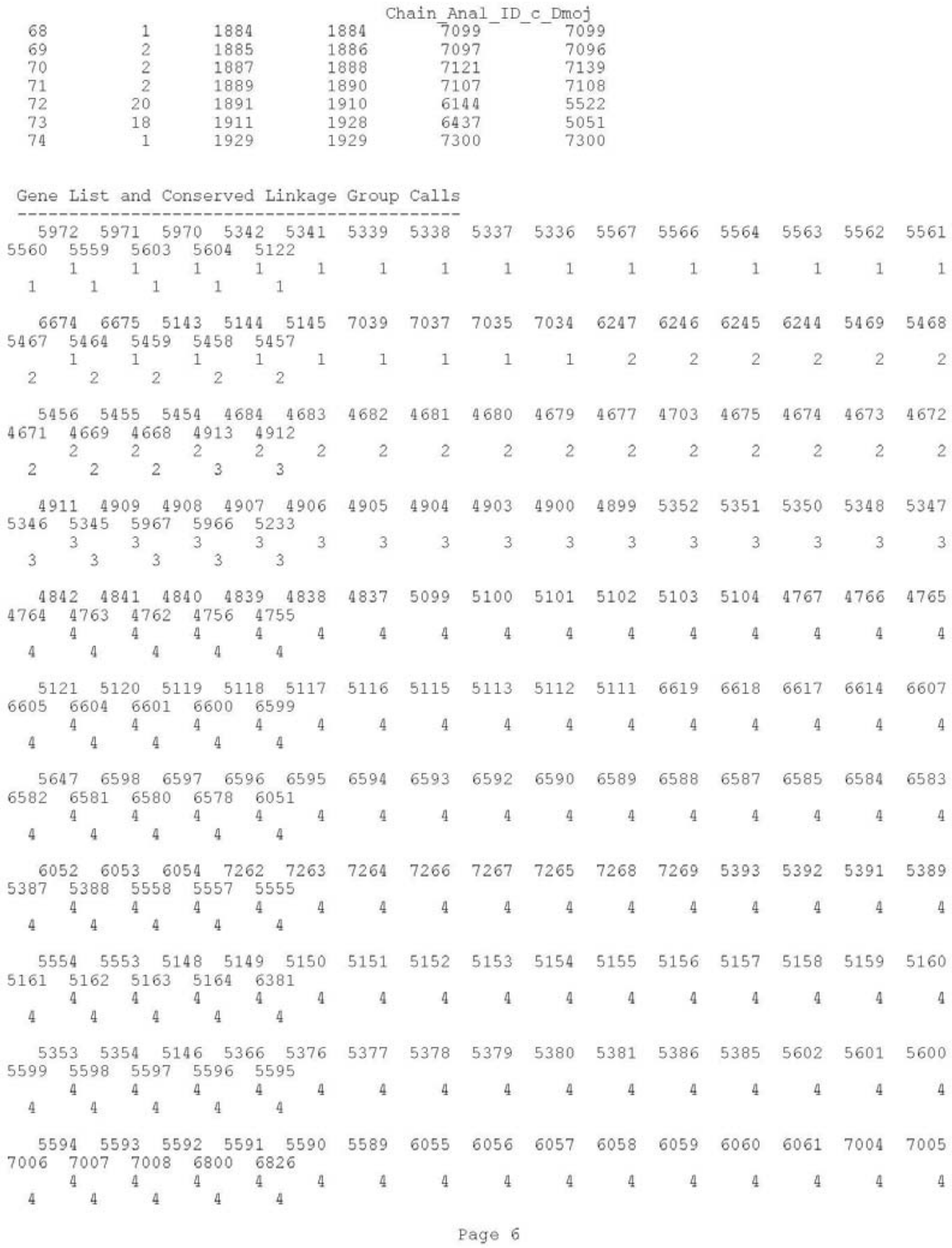

```
                                      Chain_Anal_ID_c_Dmoj
 68        1        1884        1884        7099         7099
 69        2        1885        1886        7097         7096
 70        2        1887        1888        7121         7139
 71        2        1889        1890        7107         7108
 72       20        1891        1910        6144         5522
 73       18        1911        1928        6437         5051
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 Gene List and Conserved Linkage Group Calls
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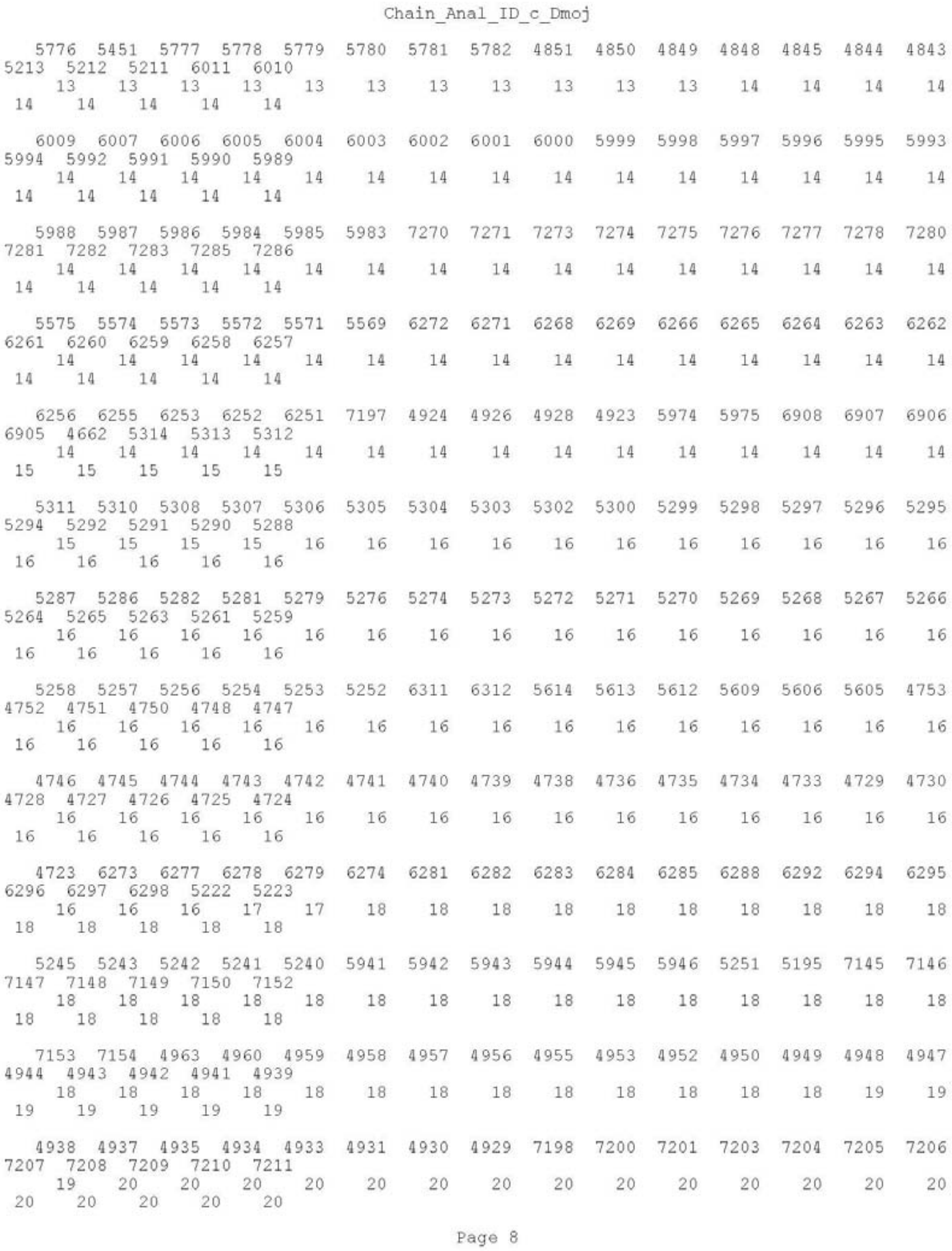

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   7056  7057  7058  7059  7060  5646  5648  5649  5650  5651  5652  5654  5655  5656  5453
4686  5452  5450  5449  5448
     56    56    56    56    56    56    56    56    56    56    56    56    56    56    57
 57    58    58    58    58
```

```
   4717   5440   5441   5442   5445   5443   5444   5446   5447   4716   4715   4714   4713   4710   4709
4706   4705   4704   4701   4699
     58     58     58     58     58     58     58     58     58     58     58     58     58     58     58
 58     58     58     58     58

   4696   4695   4694   4693   4692   6531   4691   4690   4689   6219   6218   6217   6216   6215   6214
6213   6212   6211   5978   5977
     58     58     58     58     58     58     58     58     58     58     58     58     58     58     58
 58     58     58     58     58

   5976   6024   6023   6022   6021   6020   6019   6018   6017   6483   6484   6485   6486   6487   6488
6489   6491   6493   6494   6495
     58     59     59     59     59     59     59     59     59     59     59     59     59     59     59
 60     60     60     60     60

   6496   6497   6498   6500   6501   6502   6503   6505   6506   6507   6508   6512   6513   6514   6515
6517   5512   6520   7192   6522
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
 60     60     60     60     60

   6523   6525   6526   6527   6528   6529   6530   6532   6533   6534   6535   6536   6537   6539   5568
6140   6720   6719   6718   6717
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
 60     60     60     60     60

   6715   6714   6713   6971   6970   6968   6965   6961   6958   6954   6953   6950   6949   6944   6943
6942   6938   6933   6932   6795
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
 60     60     60     60     60

   6796   6458   6456   6088   6089   6090   6091   6092   6093   6094   6095   6096   6098   6097   6099
6103   6106   6107   6108   5588
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
 60     60     60     60     60

   5587   5586   5585   5584   5147   6119   6120   6125   6126   6127   6128   6130   6248   7289   7290
7291   7292   7296   7299   5072
     60     60     60     60     60     60     60     60     60     60     60     60     60     60     60
 60     60     60     60     60

   7298   7297   5696   5757   5755   5758   5759   5760   5761   5762   5763   5764   6725   6726   6727
5686   5687   5690   5691   5693
     60     60     60     60     60     60     60     60     60     60     61     61     61     61     61
 61     61     61     61     61

   6730   6729   6728   5070   5069   5068   5067   5066   5065   5064   5063   5062   5061   5060   5059
5058   5057   5056   5055   5054
     61     61     61     61     61     61     61     61     61     61     61     61     61     61     61
 61     61     61     61     61

   5053   5052   4808   4809   4810   4811   4812   4813   4814   4815   6111   6110   6109   7261   7260
7259   7258   7256   7255   7254
     61     61     62     62     62     62     62     62     62     62     62     62     62     62     62
 62     62     62     62     62

   7253   7252   7251   7250   7249   7248   7247   7245   7244   7243   7242   7241   7240   7239   7238
7237   5227   5225   5142   5139
     62     62     62     62     62     62     62     62     62     62     62     62     62     62     62
 62     62     62     62     62

   5137   5136   5135   4854   7033   7032   7030   7029   7028   7027   6972   6712   6711   6842   6843
6844   6845   6846   6847   6848
     62     62     62     62     62     62     62     62     62     62     62     62     62     62     62
 62     62     62     62     62
```

```
   6849  6851  5947  7236  7235  7234  7233  7232  7231  7230  7229  7227  7226  7225  7223
7222  7221  7219  7218  6383
     62    62    62    62    62    62    62    62    62    62    62    62    62    62    62
 62    62    62    62    62

   4897  4896  4895  4894  4893  4892  5237  4888  4887  4886  4885  4883  4882  4884  7216
7217  5644  5642  5641  5639
     62    62    62    62    62    62    62    62    62    62    62    63    63    64    64
 64    64    64    64    64

   5638  5637  5636  5634  5632  5631  5630  5629  5627  5625  5624  5623  5622  5621  5620
6379  6378  6377  6375  6374
     64    64    64    64    64    64    64    64    64    64    64    64    64    64    64
 64    64    64    64    64

   6372  5964  5232  5230  5229  5228  5948  5949  5950  6313  5095  5096  5098  5662  5663
5664  5665  5666  5667  5669
     64    64    65    65    65    65    65    65    65    65    65    65    65    65    65
 65    65    65    65    65

   5672  5673  5674  5675  5676  5677  5678  5680  5682  5073  5074  5076  5075  5077  5078
5079  5083  5084  5087  5088
     65    65    65    65    65    65    65    65    65    65    65    65    65    65    65
 65    65    65    65    65

   5089  7098  7119  7099  7097  7096  7121  7139  7107  7108  6144  6143  6142  6141  6722
6723  5694  5695  5697  5698
     65    66    67    68    69    69    70    70    71    71    72    72    72    72    72
 72    72    72    72    72

   5699  5701  5702  5703  5704  5705  5706  5708  5710  5522  6437  6438  6439  6440  6442
6443  6444  6445  6446  6448
     72    72    72    72    72    72    72    72    72    72    73    73    73    73    73
 73    73    73    73    73

   6449  6454  6455  5044  5046  5047  5050  5051  7300     0     0     0     0     0     0
  0     0     0     0     0
     73    73    73    73    73    73    73    73    74     0     0     0     0     0     0
  0     0     0     0     0
```

```
Appendix D. Linkage Chain Analysis for the Muller D Element Comparison of D. virilis and D.
mojavensis
 Breakpoints Remaining= 126     (D.moj lineage inversion)(1)
 Chain  1               left CLG          right CLG
   BP_092_093            8377   9871 |  9868   9415        A
                        ---------------------------
   BP_076_077            8868   8864 |  8410   8409        A by inference
                        ---------------------------

 Breakpoints Remaining= 123
 Chain  2               left CLG          right CLG
   BP_117_118            9068   9064 |  9066   9059
                        ---------------------------
   BP_116_117            9347   9072 |  9068   9064
                        ---------------------------
   BP_118_119            9066   9059 |  9057   8832
                        ---------------------------

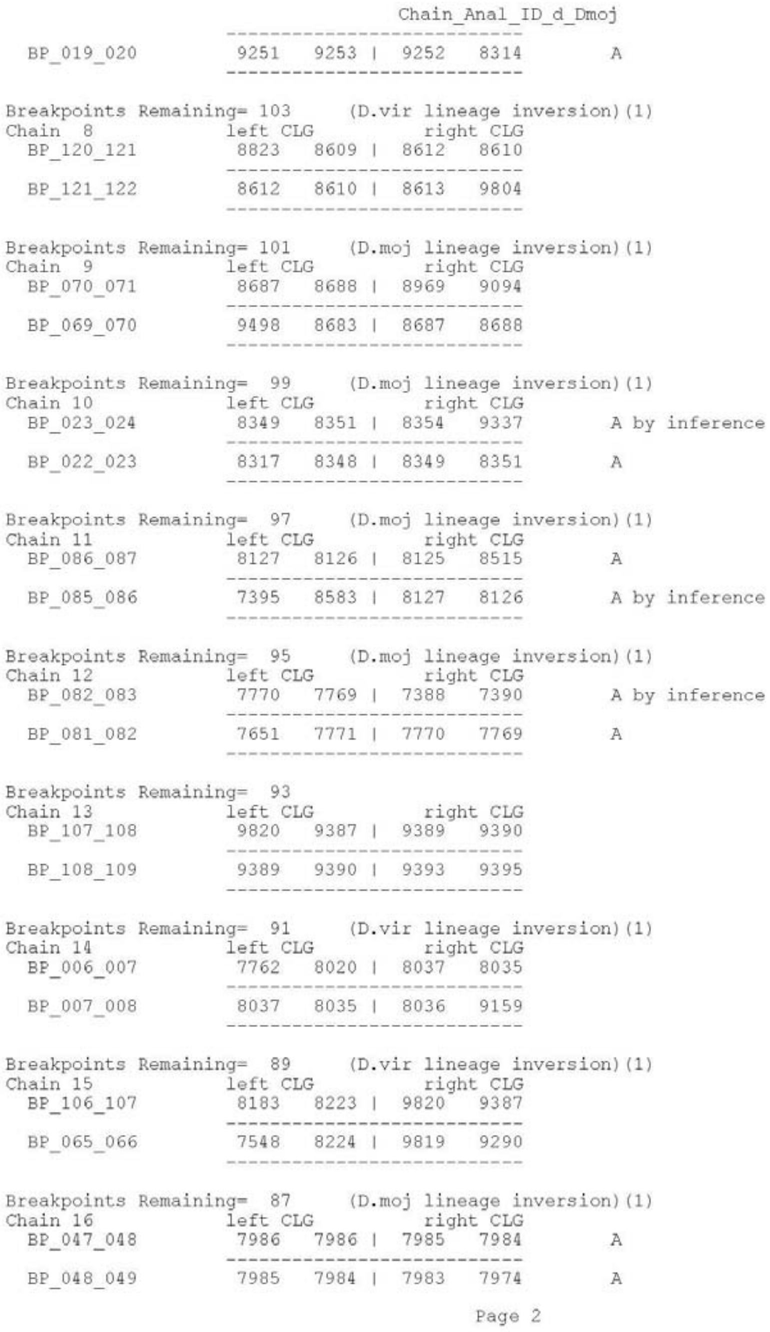
 Breakpoints Remaining= 120     (D.vir lineage inversion)(2)
 Chain  3               left CLG          right CLG
   BP_104_105            8863   9732 |  9733   9736        A
                        ---------------------------
   BP_105_106            9733   9736 |  8183   8223
                        ---------------------------
   BP_041_042            8235   8202 |  9737   9739
                        ---------------------------

 Breakpoints Remaining= 118     (D.vir lineage inversion)(1)
 Chain  4               left CLG          right CLG
   BP_113_114            9353   9354 |  9350   9350
                        ---------------------------
   BP_112_113            9405   9355 |  9353   9354
                        ---------------------------

 Breakpoints Remaining= 115     (D.moj lineage inversion)(2)
 Chain  5               left CLG          right CLG
   BP_066_067            9819   9290 |  9495   9495        A by inference
                        ---------------------------
   BP_067_068            9495   9495 |  9496   9497        A
                        ---------------------------
   BP_068_069            9496   9497 |  9498   8683        A
                        ---------------------------

 Breakpoints Remaining= 107
 Chain  6               left CLG          right CLG
   BP_054_055            8114   8465 |  8461   8074
                        ---------------------------
   BP_010_011            9162   9163 |  8464   8462
                        ---------------------------
   BP_001_002            7308   7308 |  7339   7753
                        ---------------------------
   BP_013_014            7524   7525 |  7522   7738
                        ---------------------------
   BP_119_120            9057   8832 |  8823   8609
                        ---------------------------
   BP_012_013            7982   7982 |  7524   7525
                        ---------------------------
   BP_046_047            7988   7987 |  7986   7986
                        ---------------------------
   BP_011_012            8464   8462 |  7982   7982
                        ---------------------------

 Breakpoints Remaining= 105     (D.moj lineage inversion)(1)
 Chain  7               left CLG          right CLG
   BP_018_019            8182   8188 |  9251   9253        A by inference
```



```
                                    Chain_Anal_ID_d_Dmoj
                         ---------------------------
  BP_019_020              9251   9253 |  9252   8314        A
                         ---------------------------

Breakpoints Remaining= 103     (D.vir lineage inversion)(1)
Chain  8                 left CLG          right CLG
  BP_120_121              8823   8609 |  8612   8610
                         ---------------------------
  BP_121_122              8612   8610 |  8613   9804
                         ---------------------------

Breakpoints Remaining= 101     (D.moj lineage inversion)(1)
Chain  9                 left CLG          right CLG
  BP_070_071              8687   8688 |  8969   9094
                         ---------------------------
  BP_069_070              9498   8683 |  8687   8688
                         ---------------------------

Breakpoints Remaining=  99     (D.moj lineage inversion)(1)
Chain 10                 left CLG          right CLG
  BP_023_024              8349   8351 |  8354   9337        A by inference
                         ---------------------------
  BP_022_023              8317   8348 |  8349   8351        A
                         ---------------------------

Breakpoints Remaining=  97     (D.moj lineage inversion)(1)
Chain 11                 left CLG          right CLG
  BP_086_087              8127   8126 |  8125   8515        A
                         ---------------------------
  BP_085_086              7395   8583 |  8127   8126        A by inference
                         ---------------------------

Breakpoints Remaining=  95     (D.moj lineage inversion)(1)
Chain 12                 left CLG          right CLG
  BP_082_083              7770   7769 |  7388   7390        A by inference
                         ---------------------------
  BP_081_082              7651   7771 |  7770   7769        A
                         ---------------------------

Breakpoints Remaining=  93
Chain 13                 left CLG          right CLG
  BP_107_108              9820   9387 |  9389   9390
                         ---------------------------
  BP_108_109              9389   9390 |  9393   9395
                         ---------------------------

Breakpoints Remaining=  91     (D.vir lineage inversion)(1)
Chain 14                 left CLG          right CLG
  BP_006_007              7762   8020 |  8037   8035
                         ---------------------------
  BP_007_008              8037   8035 |  8036   9159
                         ---------------------------

Breakpoints Remaining=  89     (D.vir lineage inversion)(1)
Chain 15                 left CLG          right CLG
  BP_106_107              8183   8223 |  9820   9387
                         ---------------------------
  BP_065_066              7548   8224 |  9819   9290
                         ---------------------------

Breakpoints Remaining=  87     (D.moj lineage inversion)(1)
Chain 16                 left CLG          right CLG
  BP_047_048              7986   7986 |  7985   7984        A
                         ---------------------------
  BP_048_049              7985   7984 |  7983   7974        A

                                           Page 2
```

```
                                    Chain_Anal_ID_d_Dmoj
                    ---------------------------

Breakpoints Remaining=  85     (D.moj lineage inversion)(1)
Chain 17            left CLG          right CLG
  BP_098_099         9614   9617 |  8798   8797       A by inference
                    ---------------------------
  BP_097_098         9673   9615 |  9614   9617       A
                    ---------------------------

Breakpoints Remaining=  83     (D.moj lineage inversion)(1)
Chain 18            left CLG          right CLG
  BP_094_095         9575   8926 |  8927   8928       A
                    ---------------------------
  BP_095_096         8927   8928 |  8929   9582       A
                    ---------------------------

Breakpoints Remaining=  81     (D.vir lineage inversion)(1)
Chain 19            left CLG          right CLG
  BP_102_103         8413   8416 |  8412   8417
                    ---------------------------
  BP_101_102         8411   8415 |  8413   8416
                    ---------------------------

Breakpoints Remaining=  79     (D.moj lineage inversion)(1)
Chain 20            left CLG          right CLG
  BP_035_036         7684   7440 |  7438   7379       A by inference
                    ---------------------------
  BP_099_100         8798   8797 |  8796   8418       A
                    ---------------------------

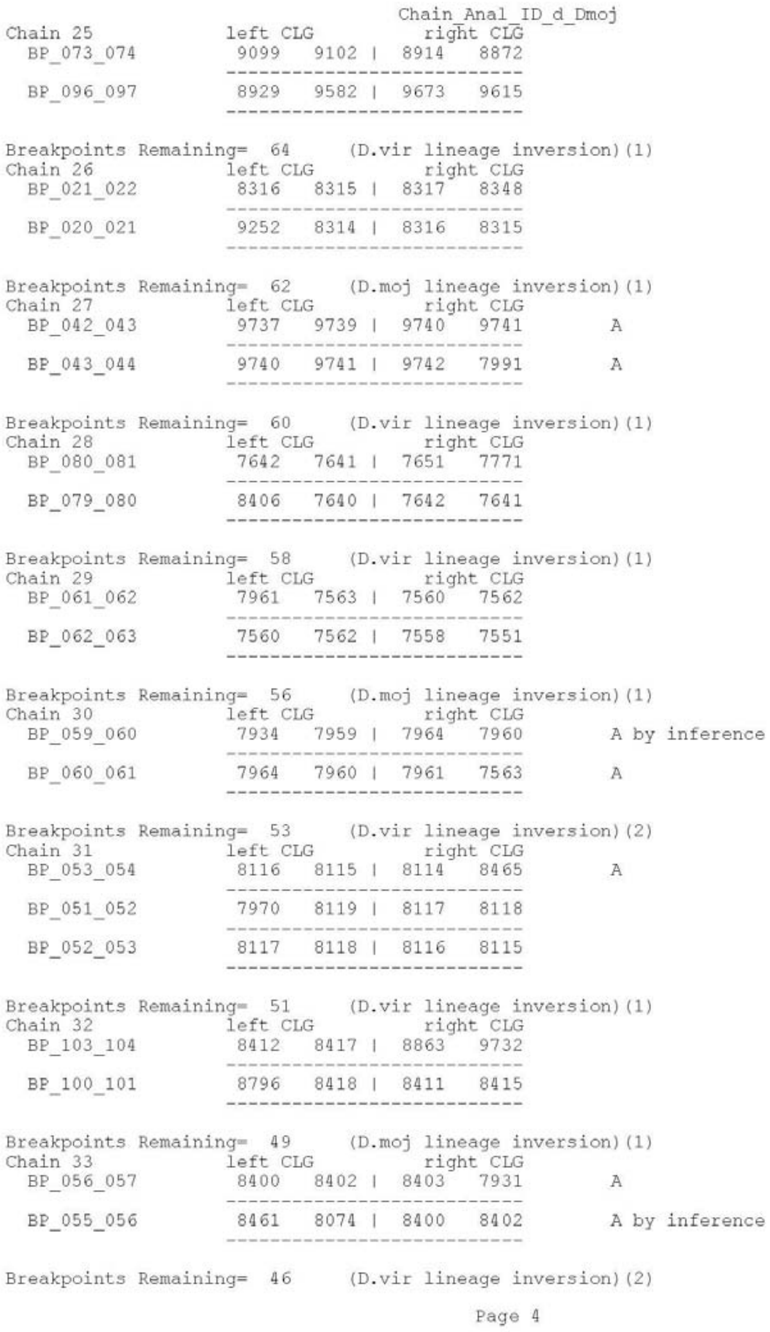
Breakpoints Remaining=  74     (D.vir lineage inversion)(4)
Chain 21            left CLG          right CLG
  BP_032_033         8242   7678 |  7680   7679
                    ---------------------------
  BP_033_034         7680   7679 |  7683   7681
                    ---------------------------
  BP_034_035         7683   7681 |  7684   7440       A
                    ---------------------------
  BP_017_018         8179   8181 |  8182   8188       A
                    ---------------------------
  BP_016_017         7735   8178 |  8179   8181       A
                    ---------------------------

Breakpoints Remaining=  72     (D.moj lineage inversion)(1)
Chain 22            left CLG          right CLG
  BP_088_089         8516   8518 |  8520   8549       A
                    ---------------------------
  BP_087_088         8125   8515 |  8516   8518       A
                    ---------------------------

Breakpoints Remaining=  70     (D.vir lineage inversion)(1)
Chain 23            left CLG          right CLG
  BP_044_045         9742   7991 |  7989   7990
                    ---------------------------
  BP_045_046         7989   7990 |  7988   7987
                    ---------------------------

Breakpoints Remaining=  68     (D.vir lineage inversion)(1)
Chain 24            left CLG          right CLG
  BP_040_041         8762   8762 |  8235   8202
                    ---------------------------
  BP_029_030         9755   8761 |  8236   8239
                    ---------------------------

Breakpoints Remaining=  66     (D.vir lineage inversion)(1)
```



```
                                    Chain_Anal_ID_d_Dmoj
Chain 25                left CLG          right CLG
  BP_073_074             9099   9102 |  8914   8872
                        ---------------------------
  BP_096_097             8929   9582 |  9673   9615
                        ---------------------------

Breakpoints Remaining=  64     (D.vir lineage inversion)(1)
Chain 26                left CLG          right CLG
  BP_021_022             8316   8315 |  8317   8348
                        ---------------------------
  BP_020_021             9252   8314 |  8316   8315
                        ---------------------------

Breakpoints Remaining=  62     (D.moj lineage inversion)(1)
Chain 27                left CLG          right CLG
  BP_042_043             9737   9739 |  9740   9741        A
                        ---------------------------
  BP_043_044             9740   9741 |  9742   7991        A
                        ---------------------------

Breakpoints Remaining=  60     (D.vir lineage inversion)(1)
Chain 28                left CLG          right CLG
  BP_080_081             7642   7641 |  7651   7771
                        ---------------------------
  BP_079_080             8406   7640 |  7642   7641
                        ---------------------------

Breakpoints Remaining=  58     (D.vir lineage inversion)(1)
Chain 29                left CLG          right CLG
  BP_061_062             7961   7563 |  7560   7562
                        ---------------------------
  BP_062_063             7560   7562 |  7558   7551
                        ---------------------------

Breakpoints Remaining=  56     (D.moj lineage inversion)(1)
Chain 30                left CLG          right CLG
  BP_059_060             7934   7959 |  7964   7960        A by inference
                        ---------------------------
  BP_060_061             7964   7960 |  7961   7563        A
                        ---------------------------

Breakpoints Remaining=  53     (D.vir lineage inversion)(2)
Chain 31                left CLG          right CLG
  BP_053_054             8116   8115 |  8114   8465        A
                        ---------------------------
  BP_051_052             7970   8119 |  8117   8118
                        ---------------------------
  BP_052_053             8117   8118 |  8116   8115
                        ---------------------------

Breakpoints Remaining=  51     (D.vir lineage inversion)(1)
Chain 32                left CLG          right CLG
  BP_103_104             8412   8417 |  8863   9732
                        ---------------------------
  BP_100_101             8796   8418 |  8411   8415
                        ---------------------------

Breakpoints Remaining=  49     (D.moj lineage inversion)(1)
Chain 33                left CLG          right CLG
  BP_056_057             8400   8402 |  8403   7931        A
                        ---------------------------
  BP_055_056             8461   8074 |  8400   8402        A by inference
                        ---------------------------

Breakpoints Remaining=  46     (D.vir lineage inversion)(2)
                                          Page 4
```

```
                                Chain_Anal_ID_d_Dmoj
Chain 34             left CLG          right CLG
  BP_111_112          9401   9403 |  9405   9355         A
                     ---------------------------
  BP_109_110          9393   9395 |  9397   9396
                     ---------------------------
  BP_110_111          9397   9396 |  9401   9403
                     ---------------------------

Breakpoints Remaining=  44     (D.moj lineage inversion)(1)
Chain 35             left CLG          right CLG
  BP_083_084          7388   7390 |  7393   7394         A by inference
                     ---------------------------
  BP_084_085          7393   7394 |  7395   8583         A
                     ---------------------------

Breakpoints Remaining=  42     (D.moj lineage inversion)(1)
Chain 36             left CLG          right CLG
  BP_063_064          7558   7551 |  7550   7552         A
                     ---------------------------
  BP_064_065          7550   7552 |  7548   8224         A by inference
                     ---------------------------

Breakpoints Remaining=  39
Chain 37             left CLG          right CLG
  BP_128_129          9413   9409 |  9343   9363
                     ---------------------------
  BP_093_094          9868   9415 |  9575   8926
                     ---------------------------
  BP_127_128          9797   9785 |  9413   9409
                     ---------------------------

Breakpoints Remaining=  37     (D.vir lineage inversion)(2)
Chain 38             left CLG          right CLG
  BP_024_025          8354   9337 |  8366   9336         A by inference
                     ---------------------------
  BP_025_026          8366   9336 |  9333   9335
                     ---------------------------
  BP_026_027          9333   9335 |  9332   8099
                     ---------------------------


Breakpoints Remaining=  35     (D.moj lineage inversion)(1)
Chain 39             left CLG          right CLG
  BP_014_015          7522   7738 |  7737   7736         A
                     ---------------------------
  BP_015_016          7737   7736 |  7735   8178         A
                     ---------------------------

Breakpoints Remaining=  33     (D.vir lineage inversion)(1)
Chain 40             left CLG          right CLG
  BP_003_004          7755   7754 |  7756   7759
                     ---------------------------
  BP_002_003          7339   7753 |  7755   7754
                     ---------------------------

Breakpoints Remaining=  30     (D.vir lineage inversion)(2)
Chain 41             left CLG          right CLG
  BP_090_091          8381   8378 |  8380   8380
                     ---------------------------
  BP_091_092          8380   8380 |  8377   9871
                     ---------------------------
  BP_089_090          8520   8549 |  8381   8378
                     ---------------------------

Breakpoints Remaining=  28     (D.vir lineage inversion)(1)
                                      Page 5
```

```
                                   Chain_Anal_ID_d_Dmoj
Chain 42            left CLG         right CLG
  BP_077_078         8410   8409 |  8407   8408
                    ---------------------------
  BP_078_079         8407   8408 |  8406   7640
                    ---------------------------

Breakpoints Remaining=  26     (D.moj lineage inversion)(1)
Chain 43            left CLG         right CLG
  BP_037_038         7370   7375 |  7374   8766
                    ---------------------------
  BP_036_037         7438   7379 |  7370   7375
                    ---------------------------

Breakpoints Remaining=  24     (D.vir lineage inversion)(1)
Chain 44            left CLG         right CLG
  BP_038_039         7374   8766 |  8763   8764
                    ---------------------------
  BP_039_040         8763   8764 |  8762   8762
                    ---------------------------

Breakpoints Remaining=  22     (D.moj lineage inversion)(1)
Chain 45            left CLG         right CLG
  BP_058_059         7932   7933 |  7934   7959        A
                    ---------------------------
  BP_057_058         8403   7931 |  7932   7933        A
                    ---------------------------

Breakpoints Remaining=  20     (D.moj lineage inversion)(1)
Chain 46            left CLG         right CLG
  BP_074_075         8914   8872 |  8870   8869        A
                    ---------------------------
  BP_075_076         8870   8869 |  8868   8864        A
                    ---------------------------

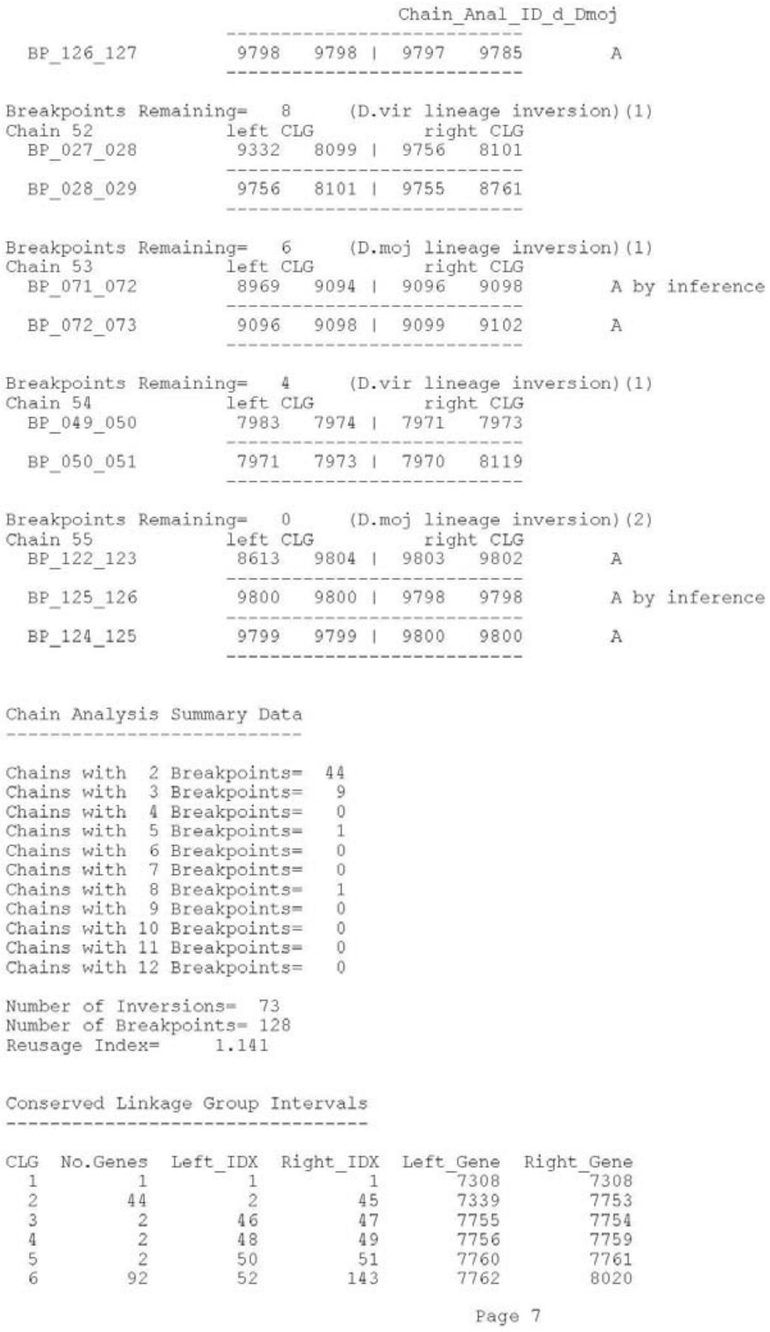
Breakpoints Remaining=  18     (D.moj lineage inversion)(1)
Chain 47            left CLG         right CLG
  BP_008_009         8036   9159 |  9160   9161        A
                    ---------------------------
  BP_009_010         9160   9161 |  9162   9163        A
                    ---------------------------

Breakpoints Remaining=  16     (D.moj lineage inversion)(1)
Chain 48            left CLG         right CLG
  BP_114_115         9350   9350 |  9349   9348        A
                    ---------------------------
  BP_115_116         9349   9348 |  9347   9072        A
                    ---------------------------

Breakpoints Remaining=  14     (D.moj lineage inversion)(1)
Chain 49            left CLG         right CLG
  BP_004_005         7756   7759 |  7760   7761        A
                    ---------------------------
  BP_005_006         7760   7761 |  7762   8020        A
                    ---------------------------

Breakpoints Remaining=  12     (D.moj lineage inversion)(1)
Chain 50            left CLG         right CLG
  BP_031_032         8240   8241 |  8242   7678        A
                    ---------------------------
  BP_030_031         8236   8239 |  8240   8241        A
                    ---------------------------

Breakpoints Remaining=  10     (D.moj lineage inversion)(1)
Chain 51            left CLG         right CLG
  BP_123_124         9803   9802 |  9799   9799        A by inference
```



```
                                Chain_Anal_ID_d_Dmoj
                    ---------------------------
  BP_126_127         9798   9798 |  9797   9785        A
                    ---------------------------

Breakpoints Remaining=   8     (D.vir lineage inversion)(1)
Chain 52            left CLG          right CLG
  BP_027_028         9332   8099 |  9756   8101
                    ---------------------------
  BP_028_029         9756   8101 |  9755   8761
                    ---------------------------

Breakpoints Remaining=   6     (D.moj lineage inversion)(1)
Chain 53            left CLG          right CLG
  BP_071_072         8969   9094 |  9096   9098        A by inference
                    ---------------------------
  BP_072_073         9096   9098 |  9099   9102        A
                    ---------------------------

Breakpoints Remaining=   4     (D.vir lineage inversion)(1)
Chain 54            left CLG          right CLG
  BP_049_050         7983   7974 |  7971   7973
                    ---------------------------
  BP_050_051         7971   7973 |  7970   8119
                    ---------------------------

Breakpoints Remaining=   0     (D.moj lineage inversion)(2)
Chain 55            left CLG          right CLG
  BP_122_123         8613   9804 |  9803   9802        A
                    ---------------------------
  BP_125_126         9800   9800 |  9798   9798        A by inference
                    ---------------------------
  BP_124_125         9799   9799 |  9800   9800        A
                    ---------------------------


Chain Analysis Summary Data
---------------------------

Chains with  2 Breakpoints=  44
Chains with  3 Breakpoints=   9
Chains with  4 Breakpoints=   0
Chains with  5 Breakpoints=   1
Chains with  6 Breakpoints=   0
Chains with  7 Breakpoints=   0
Chains with  8 Breakpoints=   1
Chains with  9 Breakpoints=   0
Chains with 10 Breakpoints=   0
Chains with 11 Breakpoints=   0
Chains with 12 Breakpoints=   0

Number of Inversions=  73
Number of Breakpoints= 128
Reusage Index=     1.141


Conserved Linkage Group Intervals
---------------------------------

CLG  No.Genes  Left_IDX  Right_IDX  Left_Gene  Right_Gene
  1         1         1          1       7308        7308
  2        44         2         45       7339        7753
  3         2        46         47       7755        7754
  4         2        48         49       7756        7759
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  6        92        52        143       7762        8020
                                     Page 7
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63	6	938	943	7558	7551
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65	12	947	958	7548	8224
66	40	959	998	9819	9290
67	1	999	999	9495	9495
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 Gene List and Conserved Linkage Group Calls
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