

APL Scripts used in Liao, et al., Modularity and selection of nectar traits in the evolution of the selfing syndrome in *Ipomoea lacunosa* (Convolvulaceae)

Variable Definitions

ΔMASTERDATA AND ΔMASTERDATA1

COL1 TRAIT CODE
COL2 TOTAL RELATIVE HOMOZYGOUS EFFECT (RHE) (TOTAL
INCLUDES NEGATIVE RHE'S IN TOTAL)
COL3 CI LOWER BOUND (CM)
COL4 CI UPPER BOUND (CM)
COL5 IGNORE
COL6 IGNORE
COL7 IGNORE
COL8 ADDITIVE EFFECT?
COL9 RELATIVE HOMOZYGOUS EFFECT (CC-LL)
COL10 NUMBER OF LINES WITH CC GENOTYPE
COL11 NUMBER OF LINES WITH LL GENOTYPE
COL12 UNIQUE ID FOR QTL

ΔALLDATA1 AND ΔALLDATA2 FOR ALL QTLS AND GWS QTLS RESPECTI
VELY

COL1 GENETIC CORRELATION (VARCOMP)
COL2 PREDICTED GENETIC CORRELATION
COL3 AVERAGE TOTAL RHE
COL4 BIAS (= PREDICTED - OBSERVED GENETIC CORRELATIO
NS)
COL5 QTL OVERLAP
COL6 CLASS (1 IF NONSEED, 0 IF SEED, IF BETWEEN-MO
DULE TRAIT PAIR, 0 IF AT LEAST 1 TRAIT IS SEED TRAIT)

I. Permutation Test for Significance of Modules

A. Program SHUFFLECORRS

INPUT IS EITHER Δ OVERLAPCORR1 OR Δ OVERLAPCORR2, WHICH
CORRESPOND TO GENETIC CORRELATIONS CALCULATED FROM RIL
MEANS OR VARIANCE COMPONENTS

COLUMNS OF Δ OVERLAPCORR1

COL1 TRAIT NUMBER FOR TRAIT 1 OF PAIR (CORRESPONDS TO TRAIT
IN Δ TRAITNAMES)
COL2 TRAIT NUMBER FOR TRAIT 2 OF PAIR
COL3 PAIRWISE TRAIT OVERLAP (FROM PROGRAM
CALCQTLOVERLAP)
COL4 PAIRWISE TRAIT GENETIC CORRELATIONS
COL5 1 IF WITHIN-MODULE PAIR, 0 IF BETWEEN-MODULE PAIR

Code:

SHUFFLECORRS

⌘ THIS PROGRAM SHUFFLES CORRELATIONS ASSIGNED TO DIFFERENT MOD
ULES AND DETERMINES PROBABILITY
⌘ THAT DIFFERENCE BETWEEN WITHIN- AND BETWEEN-MODULE AVERAGE
S IS GREATER THAN THAT OBSERVED.

OBSCORRS \leftarrow Δ OVERLAPCORR2[; 4] ⌘ Δ OVERLAPCORR1 HAS GENETI
C CORRELATIONS CALCULATED BY RIL MEANS

OBSTYPE \leftarrow Δ OVERLAPCORR2[; 5] ⌘ Δ OVERLAPCORR2 HAS GENETI
C CORRELATIONS CALCULATED BY VARIANCE COMPONENTS

IND1 \leftarrow (OBSTYPE=1)/ ρ OBSTYPE ⌘ INDEX FOR WITHIN-MODULE CORRE
LATIONS

IND0 \leftarrow (OBSTYPE=0)/ ρ OBSTYPE ⌘ INDEX FOR BETWEEN-MODULE CORR
ELATIONS

NUM1 \leftarrow ρ IND1

NUM0 \leftarrow ρ IND0

OBSAVE1 \leftarrow (+/OBSCORRS[IND1]) \div ρ IND1

OBSAVE0 \leftarrow (+/OBSCORRS[IND0]) \div ρ IND0

OBSDIFF \leftarrow OBSAVE1-OBSAVE0

␣TCLF

'OBSERVED MEAN WITHIN-MODULE CORRELATION: ', OBSAVE1

'OBSERVED MEAN BETWEEN-MODULE CORRELATION: ', OBSAVE0

␣TCLF

'OBSERVED DIFFERENCE: ', OBSDIFF

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SHUFFDIFFS←0ρ0

I←0
RETI:I←I+1
RAND←78?78
NEWTYPE←OBSTYPE[RAND]

IND1←(NEWTYPE=1)/1ρOBSTYPE      A INDEX FOR WITHIN-MODULE CORRE
    LATIONS
IND0←(NEWTYPE=0)/1ρOBSTYPE

AVE1←(+/OBSCORRS[IND1])÷ρIND1
AVE0←(+/OBSCORRS[IND0])÷ρIND0

SHUFFDIFF←AVE1-AVE0

SHUFFDIFFS←SHUFFDIFFS,SHUFFDIFF
→(I<1000)/RETI

PROP←(+/SHUFFDIFFS≥OBSDIFF)÷1000
□TCLF
'PROPORTION OF SHUFFLED DIFFERENCES GREATER THAN OBSERVED: ',P
ROP
□TCLF
'PROGRAM SHUFFLECORRS FINISHED.  VECTOR OF SHUFFLED DIFFERENCE
S IN VARIABLE ''SHUFFDIFFS''.'
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II. QTL overlap permutation test

A. Program CALCMODOVERLAP

PROGRAM CALCMODOVERLAP CALCULATES QTL OVERLAP FOR A MODULE AND PERFORMS RANDOM ASSIGNMENT TEST TO DETERMINE WHETHER AVERAGE QTL OVERLAP FOR A MODULE IS GREATER THAN EXPECTED

INPUT VARIABLES: Δ MODULE1A, Δ MODULE2A, Δ MODULE3A FOR LESS-CONSERVED SET OF QTLS (“ALL QTLs”),
 Δ MODULE1B, Δ MODULE2B, Δ MODULE3B FOR CONSERVED SET OF QTLS (“GWS QTLs”)

INPUT IS AN N X 6 MATRIX OF QTL POSITIONS FOR ALL TRAITS IN A MODULE
COL1: TRAIT NUMBER (1- X)
COL2: LINKAGE GROUP
COL3: LOWER BOUND OF 1.5 LOD CONF INTERVAL (IN CM)
COL4: UPPER BOUND OF 1.5 LOD CONF INTERVAL (IN CM)
COL5: LOWER BOUND OF 1.5 LOD CI (IN BP)
COL6: UPPER BOUND OF 1.5 LOD CI (IN BP)

OUTPUT IS 1000 AVERAGE OVERLAPS FOR RANDOM ASSIGNMENT OF QTLS TO CHROMOSOME POSITIONS
PROPORTION OF AVERAGES GREATER THAN OBSERVED AVERAGE OVERLAP IS ALSO OUTPUT

PROGRAM RANDOMIZEPOSITIONS CREATES NEW Δ MODULEXA WITH POSITIONS ON CHROMOSOMES RANDOMIZED FOR EACH QTL IN Δ MODULEXA
POSITIONS RANDOMIZED USING QTL 1.5 LOD CI IN BP
(CALLED BY CALCMODOVERLAP)

NOTE: THIS PROGRAM USES VARIABLE Δ GENEDENSITY, WHICH HAS THREE COLUMNS:

COL1 LINKAGE GROUP
COL2 NUMBER OF 50KB WINDOW
COL3 NUMBER OF GENES IN WINDOW

ADDITIONAL INPUT VARIABLE IS Δ MASTERDATA OR Δ MASTERDATA1:

COL1 TRAIT CODE
COL2 TOTAL RELATIVE HOMOZYGOUS EFFECT (RHE)
(TOTAL INCLUDES NEGATIVE RHE'S IN TOTAL)
COL3 CI LOWER BOUND (CM)
COL4 CI UPPER BOUND (CM)
COL5 IGNORE

COL6 IGNORE
 COL7 IGNORE
 COL8 ADDITIVE EFFECT
 COL9 RELATIVE HOMOZYGOUS EFFECT (CC-LL)
 COL10 NUMBER OF LINES WITH CC GENOTYPE
 COL11 NUMBER OF LINES WITH LL GENOTYPE
 COL12 UNIQUE ID FOR QTL

CALLS PROGRAMS:

CONSTRUCTMODEOVERLAP
 RANDOMIZEPOSITIONS

Code:

```
CALCMODEOVERLAP X
# THIS PROGRAM CALCULATES QTL OVERLAPS WITHIN A MODULE FOR JO
  ANNA'S DATA AND PERFORMS BOOTSTRAP ON THEM.

□TCLF
'ENTER MODULE NUMBER'
MOD←□
DELETE←0 # 7 MEANS DELETE NOTHING
TRAITQTL←ΔMASTERDATA[;2 3 4]
TRAITCODES←ΔMASTERDATA[;1]
IND←(¬TRAITCODES∈(2.4,2.5))/1ρTRAITCODES
TRAITQTL←TRAITQTL[IND;]
TRAITCODES←TRAITCODES[IND]

# GWINDEX←(ΔGENOMEWIDESIGNIFICANT)/1ρΔGENOMEWIDESIGNIFICANT
# INDEX OF QTL'S THAT ARE SIGNIFICANT GENOME WIDE
# TRAITQTL←TRAITQTL[GWINDEX;]
# TRAITCODES←TRAITCODES[GWINDEX]

# EXECUTE ABOVE THREE STATEMENTS IF WANT TO DO ANALYSIS USING
  JUST GENOMEWIDE QTLS

CONSTRUCTMODEOVERLAP X

OBSAVEOVERLAP←+//+ΔOVERLAPMATALL÷(DIM×(DIM-1))
OBSOEVERLAPMATALL←ΔOVERLAPMATALL

□TCLF
'STARTING RANDMIZATION TEST'
MODAVEOVERLAP←0ρ0

LL←0
RETLL:LL←LL+1
```

RANDOMIZEPOSITIONS

```
CONSTRUCTMODEOVERLAP NEWTRAITQTL  
AVEOVERLAP<+//+/ $\Delta$ OVERLAPMATALL $\div$ (DIM $\times$ (DIM-1))  
MODAVEOVERLAP<MODAVEOVERLAP,AVEOVERLAP
```

\rightarrow (LL<1000)/RETLL

```
 $\square$ TCLF  
'MODULE',(MOD),' RESULTS'  
 $\square$ TCLF  
'OBSERVED AVERAGE OVERLAP = ',OBSAVEOVERLAP  
 $\square$ TCLF  
PROP<(+/ $\Delta$ MODAVEOVERLAP $\geq$ OBSAVEOVERLAP) $\div$ 1000  
'PROPORTION OF RANDOMIZED TRIALS WITH AVERAGE OVERLAP GREATER  
THAN OBSERVED = ',PROP  
 $\square$ TCLF  
  
 $\square$ TCLF  
'PROGRAM CALCMODEOVERLAP FINISHED. DELETE = ',DELETE  
' OUTPUT IN VARIABLES 'MODAVEOVERLAP', 'OBSSEOVERLAP'.'
```

B. Program CONSTRUCTMODEOVERLAP

PROGRAM CONSTRUCTMODEOVERLAP MAKES MATRIX OF OVERLAP
PROPORTIONS FOR TRAITS WITHIN A MODULE (CALLED BY
CALCMODEOVERLAP)

PROGRAM CALCMODEOVERLAP2 DOES SAME THING BUT AVERAGES OVER ALL
MODULES

INPUTS ARE Δ MODULE1 - Δ MODULE5

CALLED FROM: CALCMODEOVERLAP

Output: Δ OVERLAPMATALL

Code:

```
CONSTRUCTMODEOVERLAP Y  
# THIS PROGRAM CONSTRUCTS QTL OVERLAP MATRIX FOR ALL QTLs  
# ENTRY I,J = 0 IF NO OVERLAP, =1 IF OVERLAP  
QTLNUMS<106TRAITCODES  
  
TRAITQTL<Y  
TRAITCODES<Y[;1]  
  
TRSH<TRAITCODES  
TRSH2<-1 $\Phi$ TRAITCODES
```

```

UNIQUE←(TRSH≠TRSH2)/TRAITCODES
NUNIQUE←ρUNIQUE
DIM←ρUNIQUE

SIZE←ρTRAITCODES      A TOTAL NUMBER OF QTLS
NUMMAT1←NUMMAT2←MAT←(DIM,DIM)ρ0      A INITIALIZE OVERLAP MATRIX
ΔOVERLAPMATALL←(DIM,DIM)ρ0

LINES2←0 8ρ0

I←0
RETI:I←I+1      A QTL1 LOOP

J←I
RETJ:J←J+1      A QTL2 LOOP
→(J>SIZE)/DOWN
LINEI←TRAITQTL[I;]  A GET QTL DATA FOR QTL1  (CHROMOSOME, CI L
OW, CI HIGH)
LINEJ←TRAITQTL[J;]  A GET QTL DATA FOR QTL2

TEST1←LINEI[1]=LINEJ[1]  A 1 IF BOTH QTLS FOR SAME TRAIT
→(TEST1=1)/RETJ

TEST1←LINEI[2]=LINEJ[2]      A QTL1 AND QTL2 ON SAME CHROMOSOME?
→(TEST1=0)/RETJ      A IF NOT, SKIP AND LEAVE ZERO IN M
ATRIX

TEST←((LINEI[6]=LINEJ[5])∨(LINEJ[6]=LINEI[5]))
→(TEST=1)/RETJ
CRIT1←(LINEI[5]≥LINEJ[5]) ∧ (LINEI[5]≤LINEJ[6])      A THESE TW
O LINES TEST WHETHER THERE IS OVERLAP
CRIT2←(LINEI[6]≥LINEJ[5]) ∧ (LINEI[6]≤LINEJ[6])
CRIT3←(LINEJ[5]≥LINEI[5]) ∧ (LINEJ[5]≤LINEI[6])
CRIT4←(LINEJ[6]≥LINEI[5]) ∧ (LINEJ[6]≤LINEI[6])

TEST2←CRIT1∨CRIT2∨CRIT3∨CRIT4

→(TEST2=0)/RETJ

LINES2←LINES2,[1](LINEI[14],LINEJ[14])

CODE1←TRAITCODES[I]
CODE2←TRAITCODES[J]
INDEX1←(UNIQUE=CODE1)/1ρUNIQUE
INDEX2←(UNIQUE=CODE2)/1ρUNIQUE

NUM1←+/(TRAITCODES=CODE1)
NUM2←+/(TRAITCODES=CODE2
      A TEST2 =1 IF THERE IS OVERLAP

```

```

  (TEST2=1) / 'MAT[INDEX1;INDEX2]←MAT[INDEX2;INDEX1]←MAT[INDEX2;I
INDEX1]+1'          A IF THERE IS OVERLAP, ENTER 1 IN
    MAT FOR ENTRIES I,J AND J,I
  (TEST2=1) / 'NUMMAT1[INDEX1;INDEX2]←NUMMAT1[INDEX2;INDEX1]←NUM1
  ,
  (TEST2=1) / 'NUMMAT2[INDEX1;INDEX2]←NUMMAT2[INDEX2;INDEX1]←NUM2
  ,
→RETJ

DOWN:→(I<(SIZE-1))/RETI

SIZE2←1↑ρMAT

K←0
RETK:K←K+1

L←K
RETL:L←L+1

→(MAT[K;L]=0)/DOWN2

AVE←(MAT[K;L])÷(NUMMAT1[K;L]+NUMMAT2[K;L]-MAT[K;L])      A THIS
    IS THE JACCARD INDEX
ΔOVERLAPMATALL[K;L]←ΔOVERLAPMATALL[L;K]←AVE

DOWN2:→(L<SIZE2)/RETL
→(K<(SIZE2-1))/RETK

→0

□TCLF
'PROGRAM CONSTRUCTOVERLAP FINISHED. TRAITCODE ',(⌘DELETE),'. M
ATRIX IN VARIABLE 'MAT''.'

```

C. Program RANDOMIZEPOSITIONS

PROGRAM RANDOMIZEPOSITIONS CREATES NEW ΔMODULEXA WITH POSITIONS
ON CHROMOSOMES RANDOMIZED FOR EACH QTL IN ΔMODULEXA
POSITIONS RANDOMIZED USING QTL 1.5 LOD CI IN BP
(CALLED BY CALCMODOVERLAP)

NOTE: THIS PROGRAM USES VARIABLES ΔCHROMDENSITIES AND
ΔCUMDENSITY, PRODUCED BY PROGRAM CALCGENDENSITIES

CALLED FROM: CALCMODOVERLAP
Output: NEWTRAITQTL

Code:

```
RANDOMIZEPOSITIONS
  A THIS PROGRAM RANDOMIZES THE POSITIONS OF THE QTLS IN A GIVEN
  MODULE

NEWTRAITQTL←0 600

SIZE←1↑ρTRAITQTL
MAXII←SIZE
II←0
RETI:II←II+1
LINE←TRAITQTL[II;]      A CHOOSE QTL II
NEWLINE←LINE[1]
MAXDEN←ΔCHROMDENSITIES[15]      AΔCHROMDENSITIES IS VECTOR
    OF CUMULATIVE CHROMOSOME SIZES TOTAL GENES
RAND←?MAXDEN
LINKGRP←1+(+/RAND≥ΔCHROMDENSITIES )
  ⚡(RAND=MAXDEN)/'LINKGRP←15'

NEWLINE←NEWLINE, LINKGRP

QTLsize←LINE[6]-LINE[5]      A SIZE OF QTL 1.5 LOD INTERVAL IN BP
QTLHALFSIZE←QTLsize÷2

IND←(ΔCUMDENSITY[;1]=LINKGRP)/1↑ρΔCUMDENSITY
PART←ΔCUMDENSITY[IND;]
MAX←↑/PART[;3]
RAND2←?MAX
WINDOW←1+(+/PART[;3]≤RAND2)
BASEPOS←(WINDOW-1)×50000
RAND3←?50000
POSITION←BASEPOS+RAND3
  A POSITION←?(↑ΔLGSIZESBP[LINKGRP])      A CHOSE POSITION OF QTL
  ON NEW LG

LOWER←POSITION-QTLHALFSIZE
UPPER←POSITION+QTLHALFSIZE

  ⚡(LOWER<0)/'POSITION←POSITION-LOWER'
  ⚡(UPPER>ΔLGSIZESBP[LINKGRP])/'DIFF←UPPER-ΔLGSIZESBP[LINKGRP] ⚡
  POSITION←POSITION-DIFF'
LOWER←POSITION-QTLHALFSIZE
UPPER←POSITION+QTLHALFSIZE

  ⚡(LOWER<0)/'LOWER←0'

NEWLINE←NEWLINE, LINE[3 4], LOWER, UPPER
NEWTRAITQTL←NEWTRAITQTL, [1]NEWLINE
→(II<MAXII)/RETI
```

D. Program CALCGENEDENSITIES

INPUT VARIABLE Δ GENEDENSITY HAS THREE COLUMNS:

COL1 LINKAGE GROUP
COL2 NUMBER OF 50KB WINDOW
COL3 NUMBER OF GENES IN WINDOW

PRODUCES TWO VARIABLES:

Δ CHROMDENSITIES: VECTOR WITH 15 ELEMENTS, EACH CORRESPONDING TO TOTAL NUMBER OF GENES ON CHROMOSOMES 1-15

Δ CUMDENSITY HAS 3 COLUMNS:

COL1 LINKAGE GROUP
COL2 NUMBER OF 50KB WINDOW
COL3 CUMULATIVE NUMBER OF GENES ON CHROMOSOME

Code:

CALCGENEDENSITIES

⌘ THIS PROGRAM INPUTS Δ GENEDENSITY AND CALCULATES CUMULATIVE GENE DENSITIES PER CHROMOSOME

ROWS \leftarrow 1 \uparrow ρ Δ GENEDENSITY

Δ CUMDENSITY \leftarrow 0 3 ρ 0

Δ CHROMDENSITIES \leftarrow 0 ρ 0

CUMDEN \leftarrow 0 ρ 0

I \leftarrow 0

RETI:I \leftarrow I+1

IND \leftarrow (Δ GENEDENSITY[;1]=I)/ \uparrow ROWS

PART \leftarrow Δ GENEDENSITY[IND;]

LINE \leftarrow PART[1;]

PREVCUM \leftarrow LINE[3]

Δ CUMDENSITY \leftarrow Δ CUMDENSITY,[1]LINE

MAXJ \leftarrow 1 \uparrow ρ PART

J \leftarrow 1

RETJ:J \leftarrow J+1

LINE \leftarrow PART[J;]

TEMP \leftarrow LINE[2]

TEST \leftarrow TEMP \neq TEMP

\rightarrow (TEST=1)/DOWN

LINE[3] \leftarrow LINE[3]+PREVCUM

PREVCUM \leftarrow LINE[3]

```

ΔCUMDENSITY←ΔCUMDENSITY,[1]LINE
DOWN:→(J<MAXJ)/RETJ

TEMP←-1↑ΔCHROMDENSITIES
ΔCHROMDENSITIES←ΔCHROMDENSITIES,(TEMP+PREVCUM)

→(I<15)/RETI

□TCLF
'PROGRAM CALCTGENEDENSITIES FINISHED.  OUTPUT IN VARIABLES ''Δ
CUMDENSITY'' AND ''ΔCHROMDENSITIES''.'
```

III. Predicting genetic correlations from QTL overlap

PROGRAM PREDICTCORR DOES THIS. INPUT FROM Δ MASTERDATA OR Δ MASTERDATA1 FOR LESS CONSERVATIVE ("ALL QTLs") AND CONSERVATIVE ("GWS QTLs") QTL SETS

ADDITIONAL INPUT: Δ MIDPOINTS

COL1: QTL1 ID
COL2: QTL2 ID
COL3: LINKAGE GROUP
COL4: MIDPOINT OF OVERLAP INTERVAL (CM)
COL5: NEAREST MARKER POSITION (CM)
COL6: NUMBER OF LINES WITH CC GENOTYPE AT MARKER POSITION
COL7: NUMBER OF LINES WITH LL GENOTYPE AT MARKER POSITION

PROGRAM CALLS CALCCOV2 WHICH CALCULATES COV AND VARIANCES CONTRIBUTED BY QTLs FROM A PAIR OF TRAITS, THEN SUMS TO GET PREDICTED TOTAL GENETIC COVARIANCE AND GENETIC VARIANCES FOR THE PAIR OF TRAITS.

PROGRAM PRODUCES VARIABLE Δ GENCORRS' (ORDER OF TRAITS AS IN Δ TRAITNAMES)

PROGRAM TRIANGLE RUN ON Δ GENCORRS TO OBTAIN LINEAR VECTOR OF CORRELATIONS

THIS VECTOR WAS COMBINED WITH Δ GENCORRS2 TO PRODUCE A MATRIX WITH TWO COLUMNS:

COL1: GENETIC CORRELATION BETWEEN PAIRS OF TRAITS (COVAR- AND VAR-COMP CORRELATIONS)
COL2: PREDICTED GENETIC CORRELATION FROM GENCORRS
COL3: BIAS (COL2 - COL1)
COL4: AVERAGE TOTAL RHE FOR EACH PAIR OF TRAITS
COL5: QTL OVERLAP (JACCARD INDEX) FOR EACH PAIR OF TRAITS

COMPARISON OF PREDICTED WITH OBSERVED CORRELATIONS ANALYZED BY SAS PROGRAM PREDICTCORR IN WORKSPACE 'IRENE'

PROGRAM SHUFFLEPRED TESTS SIGNIFICANCE OF CORRELATIONS FROM PREDICTCORR

INPUTS ARE Δ MASTERDATA OR Δ MASTERDATA1 AND Δ MIDPOINTS OR Δ MIDPOINTS2

PROGRAM PERMUTES TRAITS FOR INDIVIDUAL QTLS

A. Program PREDICTCORR

CALLS: CALCCORR, TRIANGLE, CALCCOV2

Code:

PREDICTCORR

A THIS PROGRAM PREDICTS PAIRWISE TRAIT GENETIC CORRELATIONS FROM QTL OVERLAP AND EFFECT SIZES

MAXJ← ρ UNIQCODES

MAXI←MAXJ-1

MIDPOINTS←0 4 ρ 0 A THIS VARIABLE HOLDS MIDPOINTS OF OVERLAP INTERVALS FOR QTLS THAT OVERLAP

A FIRST TWO COLUMNS ARE QTL UNIQUE ID, THIRD IS LG AND FOURTH IS MIDPOINT

GENCORRS←((ρ UNIQCODES),(ρ UNIQCODES)) ρ 0

AVETRHE←((ρ UNIQCODES),(ρ UNIQCODES)) ρ 0

OVERLAPMAT←((ρ UNIQCODES),(ρ UNIQCODES)) ρ 0

I←0

RETI:I←I+1 A FIRST TRAIT LOOP

J←I

RETJ:J←J+1 A SECOND TRAIT LOOP

TRAITCODEI←UNIQCODES[I]

TRAITCODEJ←UNIQCODES[J]

TOTRHE← Δ TOTRHEA[;1]

INDI←(TRAITCODEI=TOTRHE)/ ι ρ UNIQCODES

INDJ←(TRAITCODEJ=TOTRHE)/ ι ρ UNIQCODES

TOTRHEI← Δ TOTRHEA[INDI;2]

TOTRHEJ← Δ TOTRHEA[INDJ;2]

AVETOTRHE←(TOTRHEI+TOTRHEJ) \div 2

IND1←(TRAITCODES=UNIQCODES[I])/ ι ρ TRAITCODES

PART1←QTLDATA[IND1;]

IND2←(TRAITCODES=UNIQCODES[J])/ ι ρ TRAITCODES

PART2←QTLDATA[IND2;]

OVERLAPS←0 4 ρ 0 A NUMBERS ARE QTLNUMBERS FROM PARTS 1 AND 2 FOR OVERLAPPING QTL

MAXII←1 \uparrow ρ PART1

MAXJJ←1 \uparrow ρ PART2

```

II←0
RETI:II←II+1      a PART1 LOOP

JJ←0
RETJJ:JJ←JJ+1     a PART2 LOOP

LINEII←PART1[II;] a GET QTL DATA FOR QTL1 (CHROMOSOME, CI LOW
, CI HIGH)
LINEJJ←PART2[JJ;] a GET QTL DATA FOR QTL2

TEST1←LINEII[2]=LINEJJ[2] a QTL1 AND QTL2 ON SAME CHROMOSOME?

→(TEST1=0)/DOWN      a IF NOT, SKIP AND LEAVE ZERO IN
MATRIX

TEST←((LINEII[4]=LINEJJ[3])∨(LINEJJ[4]=LINEII[3]))
→(TEST=1)/DOWN      a DO NOT COUNT AS OVERLAP IF CI'S ABOUT
CRIT1←(LINEII[3]≥LINEJJ[3]) ∧ (LINEII[3]≤LINEJJ[4]) a THES
E TWO LINES TEST WHETHER THERE IS OVERLAP
CRIT2←(LINEII[4]≥LINEJJ[3]) ∧ (LINEII[4]≤LINEJJ[4])
CRIT3←(LINEJJ[3]≥LINEII[3]) ∧ (LINEJJ[3]≤LINEII[4])
CRIT4←(LINEJJ[4]≥LINEII[3]) ∧ (LINEJJ[4]≤LINEII[4])

TEST2←CRIT1∨CRIT2∨CRIT3∨CRIT4
→(TEST2=0)/DOWN

OVERLAPS←OVERLAPS,[1](II,JJ,LINEII[12],LINEJJ[12])

TEMP←LINEII[3 4],LINEJJ[3 4] a THIS NEXT SET OF 6 LINE
S CALCULATES MIDPOINTS OF OVERLAP INTERVALS AND STORES IN V
ARIABLE MIDPOINTS
TEMP2←ATEMP
TEMP3←TEMP[TEMP2]
INTERVAL←TEMP3[2 3]
MIDPOINT←(+/INTERVAL)÷2
MIDPOINTS←MIDPOINTS,[1](LINEII[12],LINEJJ[12],LINEII[2],MIDPOI
NT)

DOWN:→(JJ<MAXJJ)/RETJJ

→(II<MAXII)/RETI

a □TCLF
a 'CALCULATING OVERLAPS FINISHED FOR I,J = ',I,J

MAXK←1+pOVERLAPS

a FOLLOWING VARIABLE HAS COLUMNS: COL1 RHE ON TRAITI RHE ON T
RAITJ
RHE←0 4p0

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SIZE←1↑ρOVERLAPS
→(SIZE=0)/DOWN4

K←0
RETK:K←K+1  @ THIS LOOP ADDS RHE FOR OVERLAPPING QTLS

RHEVALS←(PART1[OVERLAPS[K;1];9],PART2[OVERLAPS[K;2];9],OVERLAP
S[K;3 4])

RHE←RHE,[1]RHEVALS

→(K<MAXK)/RETK

DOWN4:MAXL←1↑ρPART1
L←0
RETL:L←L+1

TEST←L∈OVERLAPS[;1]
→(TEST=1)/DOWN2

RHE←RHE,[1](PART1[L;9],0,PART1[L;12],0)

DOWN2:→(L<MAXL)/RETL

MAXLL←1↑ρPART2
LL←0
RETLL:LL←LL+1

TEST←LL∈OVERLAPS[;2]
→(TEST=1)/DOWN3

RHE←RHE,[1](0,PART2[LL;9],0,PART2[LL;12])

DOWN3:→(LL<MAXLL)/RETLL

CALCCOV2

GENCORRS[I;J]←R
AVETRHESES[I;J]←AVETOTRHE

IND1←((RHE[;1]≠0)^(RHE[;2]≠0))/1↑ρRHE
A←ρIND1
B←+/RHE[;1]≠0
C←+/RHE[;2]≠0
OV←A÷(B+C-1)
OVERLAPMAT[I;J]←OV

→(J<MAXJ)/RETJ
→(I<MAXI)/RETI

TRIANGLE  GENCORRS

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⊕(ALLQTLS≠'N')/'DATA←ΔGENCORRS2,78 1ρVEC'
⊕(ALLQTLS='N')/'DATA←ΔGENCORRS2[NONSEEDIND;],45 1ρVEC'
BIAS←DATA[;2]-DATA[;1]
⊕(ALLQTLS≠'N')/'DATA←DATA,78 1ρBIAS'
⊕(ALLQTLS='N')/'DATA←DATA,45 1ρBIAS'
TRIANGLE AVETRHEs
⊕(ALLQTLS≠'N')/'DATA←DATA,78 1ρVEC'
⊕(ALLQTLS='N')/'DATA←DATA,45 1ρVEC'

TRIANGLE OVERLAPMAT
⊕(ALLQTLS≠'N')/'DATA←DATA,78 1ρVEC'
⊕(ALLQTLS='N')/'DATA←DATA,45 1ρVEC'

CALCCORR DATA[;1 2]
OBSCORR1←CORR
CALCCORR DATA[;1 3]
OBSCORR2←CORR
CALCCORR DATA[;3 4]
OBSCORR3←CORR

CALCCORR DATA[;2 5]
OBSCORR4←CORR

□TCLF
'CORRELATION OF GENETIC CORRELATIONS AND PREDICTED CORRELATION
S: ',OBSCORR1
'CORRELATION OF BIAS AND GENETIC CORRELATIONS: ',OBSCORR2
'CORRELATION OF AVERAGE TOTAL RHE AND BIAS: ',OBSCORR3
'CORRELATION OF PREDICTED GENETIC CORRELATIONS AND QTL OVERLAP
: ',OBSCORR4
□TCLF

'PROGRAM PREDICTCORR FINISHED.  OUTPUT IN VARIABLES ''GENCORRS
'' AND ''AVETRHEs''.'

```

B. Program CALCCORR

Called by PREDICTCORR

Code:

```

CALCCORR  X
⌘ THIS PROGRAM CALCULATES CORRELATION BETWEEN TWO COLUMNS IN X
ROWS←1↑ρX
MEANS←+/[1]X÷ROWS
X1←,X[;1]
X2←,X[;2]
DIFF1←X1-MEANS[1]

```



```

DIFF2←X2-MEANS[2]
COV←+/(DIFF1×DIFF2)÷ROWS
VAR1←+/(DIFF1★2)÷ROWS
VAR2←+/(DIFF2★2)÷ROWS
CORR←COV÷((VAR1×VAR2)★.5)
A  □TCLF
A  'THE CORRELATION IS ',CORR

```

C. Program TRIANGLE

CALLED BY PREDICTCORR

Code:

```

TRIANGLE X
A  THIS PRPGRAM MAKES A VECTOR OUT OF THE UPPER TRIANGLE (NOT I
    NCLUDING DIAGONAL) OF A MATRIX

VEC←0ρ0
MAXI←-1+1↑ρX
I←0
RETI:I←I+1

MAXJ←1↑ρX
J←I
RETJ:J←J+1
VEC←VEC,X[I;J]

→(J<MAXJ)/RETJ

→(I<MAXI)/RETI

```

D. Program CALCCOV2

Called from PREDICTCORR

Code:

```

CALCCOV2
A  THIS PROGRAM CALCULATES GENETIC COVARIANCE AND VARIANCES FO
    R A PAIR OF TRAITS TAKING GENOTYPE FREQS INTO ACCOUNT
A  INPUT IS RHE FROM PROGRAM 'PREDICTCORR'

KKMAX←1↑ρRHE

```

```

PAIRS←ΔMIDPOINTS[;1 2]      A VARIABLE WITH ALL PAIRS OF QTLS
                              THAT OVERLAP; NUMBERS ARE QTL IDS; ΔMIDPOINTS FOR ALL QTL,
                              ΔMIDPOINTS1 FOR GWS QTL

VARCOV←0 3ρ0

KK←0
RETKK:KK←KK+1
LINE←RHE[KK;]
EFFECTS←0.5×LINE[1 2]

TEST←(EFFECTS[1]≠0)^(EFFECTS[2]≠0)      A TEST FOR WHETHER OVERLAPPING QTLS

COV←0

→(TEST=0)/DOWN
A CALCULATE COVARIANCE FOR QTL PAIR
PAIR←2 1ρLINE[3 4]      A THIS IS THE PAIR OF QTL UNIQUE IDS

IND←(,PAIRS^.=PAIR)/11↑ρPAIRS      A ROW INDEX OF QTL PAIR IN ΔMIDPOINTS
TEST←0=ρIND
⊥(TEST=1)/'PAIR←2 1ρLINE[4 3] ♦ IND←(,PAIRS^.=PAIR)/11↑ρPAIRS
'

NUMS←,ΔMIDPOINTS[IND;6 7]
      A VECTOR OF NUMBER OF LINES WITH CC AND WITH LL GENOTYPE

PROPCC←NUMS[1]÷(NUMS[1]+NUMS[2])      A PROPORTION OF LINES WITH CC GENOTYPE

MEAN1←(PROPCC×EFFECTS[1])-(1-PROPCC)×EFFECTS[1]      A MEAN EFFECT FOR QTL1
MEAN2←(PROPCC×EFFECTS[2])-(1-PROPCC)×EFFECTS[2]      A MEAN EFFECT FOR QTL2

PROD1←PROPCC×(EFFECTS[1]-MEAN1)×(EFFECTS[2]-MEAN2)
      A CALCULATE COVARIANCE
PROD2←(1-PROPCC)×((-1×EFFECTS[1]-MEAN1)×((-1×EFFECTS[2]-MEAN2))
COV←PROD1+PROD2

DOWN:

A CALCULATE VARIANCES
VARI←VARJ←0
PAIR2←,PAIR
→(EFFECTS[1]=0)/DOWN2      A SKIP FOR QTL1 IF EFFECT IS 0
A CALCULATE VARIANCE FOR QTL1

```

```

IND←(,PAIR2[1]=QTLDATA[;12])/1↑ρQTLDATA
NCC←QTLDATA[IND;10]
NLL←QTLDATA[IND;11]
PROPCC1←NCC÷(NCC+NLL)

MEAN3←(PROPCC1×EFFECTS[1])-(1-PROPCC1)×EFFECTS[1]
VARI←(PROPCC1×(EFFECTS[1]-MEAN3))★2)+(1-PROPCC1)×((-1×EFFECTS[1]-MEAN3))★2)

DOWN2: →(EFFECTS[2]=0)/DOWN3      ␣ SKIP FOR QTL2 IF EFFECT IS 0

␣ CALCULATE VARIANCE FOR QTL2

IND←(,PAIR2[2]=QTLDATA[;12])/1↑ρQTLDATA
NCC←QTLDATA[IND;10]
NLL←QTLDATA[IND;11]
PROPCC2←NCC÷(NCC+NLL)

MEAN4←(PROPCC2×EFFECTS[2])-(1-PROPCC2)×EFFECTS[2]
VARJ←(PROPCC2×(EFFECTS[2]-MEAN4))★2)+(1-PROPCC2)×((-1×EFFECTS[2]-MEAN4))★2)

DOWN3:

VARCOV←VARCOV,[1](COV,VARI,VARJ)

→(KK<KKMAX)/RETKK

SUM←+/[1]VARCOV
R←SUM[1]÷((SUM[2]×SUM[3])★.5)

```

E. Program SHUFFLEPRED

Calls PREDICTCORR, PREDICTCORR2

Code:

```

SHUFFLEPRED
␣ THIS PROGRAM SHUFFLES QTLS TO CALCULATE PROBABILITY OF CORRELATION BETWEEN GENETIC CORRELATIONS AND PREDICTED CORRELATIONS

␣ CAN RUN THIS FOR COMBINATIONS OF GWS OR ALL QTLS AND ALL TRAITS OR JUST FLOWER AND NECTAR TRAITS
␣ CHANGE TITLE APPROPRIATELY BELOW
␣ IN PREDICTCORR2, INDICATE ALLQTLS = 'Y' OR 'N'      (NO MEANS JUST FLOWER AND NECTAR TRAITS)

```

```

      A IN PREDICTCORR2 USE EITHER ΔMIDPOINTS OR ΔMIDPOINTS1, ΔMASTE
        RDATA OR ΔMASTERDATA1, AND ΔTOTRHE OR ΔTOTRHE1, FOR LESS OR
        MORE CONSERVED QTLS
      A IN PREDICTCORR2 USE EITHER ΔMIDPOINTS OR ΔMIDPOINTS1

ALLQTLS←'N'      A THIS VARIABLE SHOULD REALLY BE NAMED 'ALLTRA
  ITS'
TITLE←'GWS QTLS, FLORAL AND NECTAR TRAITS'

NONSEEDIND←1 2 3 4 5 6 10 11 12 13 14 15 16 17 21 22 23 24 25
26 27 31 32 33 34 35 36 40 41 42 43 44 48 49 50 51 55 56 57 61
62 63 76 77 78
ΔMIDPOINTSΔ←ΔMIDPOINTS1      A USE ΔMIDPOINTS FOR LESS-CONSERVA
  TIVE QTLS, ΔMIDPOINTS1 FOR GWS QTLS
ΔTOTRHEA←ΔTOTRHE1      A USE ΔTOTRHE FOR LESS-CONSERVATIV
  E QTLS, ΔTOTRHE1 FOR GWS QTLS
QTLDATAA←QTLDATA←ΔMASTERDATA1      A USE ΔMASTERDATA FOR LES
  S-CONSERVATIVE QTL SET, ΔMASTERDATA1 FOR CONSERVATIVE DATAS
  ET
TRAITCODESA←TRAITCODES←ΔMASTERDATA1[;1]      A AND USE ΔMA
  STERDATA FOR LESS-CONSERVATIVE QTL SET, ΔMASTERDATA1 FOR CO
  NSERVATIVE DATASET

IND←(~TRAITCODES∈(2.4,2.5))/1ρTRAITCODES      A LIST TRAITS TO EXC
  LUDE NOTE: 2.4 AND 2.5 ARE ALTERNATES FOR 2.3 (NECTARY SIZ
  E)

Δ(ALLQTLS='N')/'IND←(~TRAITCODES∈(3.1,3.2,3.3,2.4,2.5))/1ρTRAI
  TCODES'      A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS

TRAITCODES←TRAITCODES[IND]
QTLDATA←QTLDATA[IND;]
QTLDATAB←QTLDATA

UNIQCODES←1.1,1.2,1.3,1.4,1.5,1.6,1.7,3.1,3.2,3.3,2.1,2.2,2.3

Δ(ALLQTLS='N')/'UNIQCODES←1.1,1.2,1.3,1.4,1.5,1.6,1.7,2.1,2.2,
  2.3'      A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS
Δ(ALLQTLS='N')/'ΔTOTRHEA←ΔTOTRHEA[1 2 3 4 5 6 7 11 12 13;]'
      A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS

SCORRS1←SCORRS2←SCORRS3←SCORRS4←0ρ0
PREDICTCORR

□TCLF
'OBSERVED CORRELATIONS CALCULATED'
□TCLF
'BEGINNING SHUFFLE ANALYSIS'

IJ←0
RETIJ:IJ←IJ+1

```

```

TEST←(IJ÷100)=L(IJ÷100)
⌚(TEST=1)/''REPS COMPLETED '',IJ'
PREDICTCORR2

SCORRS1←SCORRS1,SHUFCORR1
SCORRS2←SCORRS2,SHUFCORR2
SCORRS3←SCORRS3,SHUFCORR3
SCORRS4←SCORRS4,SHUFCORR4

→(IJ<1000)/RETIJ

SUM1←+/SCORRS1≥OBSCORR1
SUM2←+/SCORRS2≥OBSCORR2
SUM3←+/SCORRS3≥OBSCORR3
SUM4←+/SCORRS4≥OBSCORR4
□TCLF
TITLE
□TCLF
'CORRELATION BETWEEN GENETIC CORRELATIONS AND PREDICTED CORREL
ATION'
'    OBSERVED CORRELATION: ',OBSCORR1
'    PROPORTION OF SHUFFLED CORRELATIONS ≥ OBSERVED: ',SUM1÷10
00
'CORRELATION BETWEEN GENETIC CORRELATIONS AND BIAS'
'    OBSERVED CORRELATION: ',OBSCORR2
'    PROPORTION OF SHUFFLED CORRELATIONS ≥ OBSERVED: ',SUM2÷10
00
'CORRELATION BETWEEN BIAS AND AVE TOT RHE: '
'    OBSERVED CORRELATION: ',OBSCORR3
'    PROPORTION OF SHUFFLED CORRELATIONS ≥ OBSERVED: ',SUM3÷10
00
'CORRELATION BETWEEN QTL OVERLAP AND PREDICTED CORRELATION'
'    OBSERVED CORRELATION: ',OBSCORR4
'    PROPORTION OF SHUFFLED CORRELATIONS ≥ OBSERVED: ',SUM4÷10
00

```

F. Program PREDICTCORR2

Called from SHUFFLEPRED

NOTE: This program is essentially the same as PREDICTCORR, slightly modified to be called multiple times by SHUFFLEPRED

Code:

```

PREDICTCORR2
⌘ THIS PROGRAM PREDICTS PAIRWISE TRAIT GENETIC CORRELATIONS FR
  OM QTL OVERLAP AND EFFECT SIZES FOR RANDOM ASSIGNMENT OF TR
  AITS TO QTLS

```

```

QTLDATA←QTLDATAA      A USE ΔMASTERDATA FOR LESS-CONSERVATIVE
    QTL SET, ΔMASTERDATA1 FOR CONSERVATIVE DATASET
TRAITCODES←TRAITCODESA      A AND USE ΔMASTERDATA FOR LESS
    -CONSERVATIVE QTL SET, ΔMASTERDATA1 FOR CONSERVATIVE DATASET
MIDNUMS←ΔMIDPOINTSΔ[;1 2]
IND←(∼TRAITCODES∈(2.4,2.5))/1ρTRAITCODES  A LIST TRAITS TO EXC
    LUDE NOTE: 2.4 AND 2.5 ARE ALTERNATES FOR 2.3 (NECTARY SIZE)

Δ(ALLQTLS='N')/'IND←(∼TRAITCODES∈(3.1,3.2,3.3,2.4,2.5))/1ρTRAI
TCODES'  A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS

TRAITCODES←TRAITCODES[IND]
QTLDATA←QTLDATA[IND;]
CODES←,QTLDATA[;1]
NUM←ρCODES
ORDER←NUM?NUM

QTLDATA[;1]←CODES[ORDER]
TRAITCODES← CODES[ORDER]

UNIQCODES←1.1,1.2,1.3,1.4,1.5,1.6,1.7,3.1,3.2,3.3,2.1,2.2,2.3

Δ(ALLQTLS='N')/'UNIQCODES←1.1,1.2,1.3,1.4,1.5,1.6,1.7,2.1,2.2,
2.3'      A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS
A Δ(ALLQTLS='N')/'ΔTOTRHEA←ΔTOTRHEA[1 2 3 4 5 6 7 11 12 13;]'
    A USE THIS FOR JUST FLOWER AND NECTAR TRAIT ANALYSIS

MAXJ←ρUNIQCODES
MAXI←MAXJ-1

MIDPOINTS←0 4ρ0      A THIS VARIABLE HOLDS MIDPOINTS OF OVERLAP I
    NTERVALS FOR QTLS THAT OVERLAP
    A FIRST TWO COLUMNS ARE QTL UNIQUE ID, THIRD
    IS LG AND FOURTH IS MIDPOINT

GENCORRS←((ρUNIQCODES),(ρUNIQCODES))ρ0
AVETRHEA←((ρUNIQCODES),(ρUNIQCODES))ρ0
OVERLAPMAT←((ρUNIQCODES),(ρUNIQCODES))ρ0

I←0
RETI:I←I+1  A FIRST TRAIT LOOP

J←I
RETJ:J←J+1  A SECOND TRAIT LOOP

TRAITCODEI←UNIQCODES[I]
TRAITCODEJ←UNIQCODES[J]

IND1←(TRAITCODES=UNIQCODES[I])/1ρTRAITCODES

```

```

PART1←QTLDATA[IND1;]
IND2←(TRAITCODES=UNIQCODES[J])/1ρTRAITCODES
PART2←QTLDATA[IND2;]

OVERLAPS←0 4ρ0      A NUMBERS ARE QTLNUMBERS FROM PARTS 1 AND 2 F
OR OVERLAPPING QTL

MAXII←1↑ρPART1
MAXJJ←1↑ρPART2

II←0
RETII:II←II+1      A PART1 LOOP

JJ←0
RETJJ:JJ←JJ+1      A PART2 LOOP

LINEII←PART1[II;] A GET QTL DATA FOR QTL1 (CHROMOSOME, CI LOW
, CI HIGH)
LINEJJ←PART2[JJ;] A GET QTL DATA FOR QTL2

TEST1←LINEII[2]=LINEJJ[2] A QTL1 AND QTL2 ON SAME CHROMOSOME?
→(TEST1=0)/DOWN      A IF NOT, SKIP AND LEAVE ZERO IN MATRIX

TEST←((LINEII[4]=LINEJJ[3])∨(LINEJJ[4]=LINEII[3]))
→(TEST=1)/DOWN      A DO NOT COUNT AS OVERLAP IF CI'S ABUT
CRIT1←(LINEII[3]≥LINEJJ[3]) ∧ (LINEII[3]≤LINEJJ[4])      A THES
E TWO LINES TEST WHETHER THERE IS OVERLAP
CRIT2←(LINEII[4]≥LINEJJ[3]) ∧ (LINEII[4]≤LINEJJ[4])
CRIT3←(LINEJJ[3]≥LINEII[3]) ∧ (LINEJJ[3]≤LINEII[4])
CRIT4←(LINEJJ[4]≥LINEII[3]) ∧ (LINEJJ[4]≤LINEII[4])

TEST2←CRIT1∨CRIT2∨CRIT3∨CRIT4
→(TEST2=0)/DOWN

OPAIR←2 1ρ(LINEII[12],LINEJJ[12])
TESTPAIR←+/, (MIDNUMS^.=OPAIR)
⊥(TESTPAIR=0)/'TEMP←OPAIR[2; 1],OPAIR[1;1] ◇ OPAIR←2 1ρTEMP'
TESTPAIR2←+/, (MIDNUMS^.=OPAIR)

⊥(TESTPAIR2=0)/'''TESTPAIR2 NOT IN DATASET''' ◇ →0'

OVERLAPS←OVERLAPS,[1](II,JJ,LINEII[12],LINEJJ[12])

DOWN:→(JJ<MAXJJ)/RETJJ

→(II<MAXII)/RETII

MAXK←1↑ρOVERLAPS

```

```

  A FOLLOWING VARIABLE HAS COLUMNS: COL1 RHE ON TRAITI RHE ON T
    RAITJ
RHE←0 4ρ0

SIZE←1↑ρOVERLAPS
→(SIZE=0)/DOWN4

K←0
RETK:K←K+1 A THIS LOOP ADDS RHE FOR OVERLAPPING QTLS

RHEVALS←(PART1[OVERLAPS[K;1];9],PART2[OVERLAPS[K;2];9],OVERLAP
S[K;3 4])

RHE←RHE,[1]RHEVALS

→(K<MAXK)/RETK

DOWN4:MAXL←1↑ρPART1
L←0
RETL:L←L+1

TEST←L∈OVERLAPS[;1]
→(TEST=1)/DOWN2

RHE←RHE,[1](PART1[L;9],0,PART1[L;12],0)

DOWN2:→(L<MAXL)/RETL

MAXLL←1↑ρPART2
LL←0
RETL:LL←LL+1

TEST←LL∈OVERLAPS[;2]
→(TEST=1)/DOWN3

RHE←RHE,[1](0,PART2[LL;9],0,PART2[LL;12])

DOWN3:→(LL<MAXLL)/RETL

CALCCOV2

GENCORRS[I;J]←R

IND1←(RHE[;1]≠0)/1↑ρRHE
TRHE1←+/RHE[IND1;1]
IND2←(RHE[;2]≠0)/1↑ρRHE
TRHE2←+/RHE[IND2;2]
AVETOTRHE←(TRHE1+TRHE2)÷2
AVETRHE[I;J]←AVETOTRHE

```



```

IND1←((RHE[;1]≠0)^(RHE[;2]≠0))/11↑ρRHE
A←ρIND1
B←+/RHE[;1]≠0
C←+/RHE[;2]≠0
OV←A÷(B+C-1)
OVERLAPMAT[I;J]←OV

→(J<MAXJ)/RETJ
→(I<MAXI)/RETI

TRIANGLE GENCORRS
⊥(ALLQTLS≠'N')/'DATA←ΔGENCORRS2,78 1ρVEC'
⊥(ALLQTLS='N')/'DATA←ΔGENCORRS2[NONSEEDIND;],45 1ρVEC'
BIAS←DATA[;2]-DATA[;1]
⊥(ALLQTLS≠'N')/'DATA←DATA,78 1ρBIAS'
⊥(ALLQTLS='N')/'DATA←DATA,45 1ρBIAS'
TRIANGLE AVETRHEs
⊥(ALLQTLS≠'N')/'DATA←DATA,78 1ρVEC'
⊥(ALLQTLS='N')/'DATA←DATA,45 1ρVEC'

TRIANGLE OVERLAPMAT
⊥(ALLQTLS≠'N')/'DATA←DATA,78 1ρVEC'
⊥(ALLQTLS='N')/'DATA←DATA,45 1ρVEC'

CALCCORR DATA[;1 2]
SHUFCORR1←CORR
CALCCORR DATA[;1 3]
SHUFCORR2←CORR
CALCCORR DATA[;3 4]
SHUFCORR3←CORR

CALCCORR DATA[;2 5]
SHUFCORR4←CORR

␣ 'PROGRAM PREDICTCORR FINISHED.  OUTPUT IN VARIABLES  ''GENCOR
RS'' AND ''AVETRHEs''.'

```

IV. Ascertainment Bias Programs

PROGRAMS ASCERTAIN11, ASCERTAIN10, AND ASCERTAIN10A
MODEL EFFECTS OF ASCERTAINMENT BIAS.

A. Program ASCERTAIN11

Code:

```
ASCERTAIN11
  THIS PROGRAM CALCULATES ASCERTAINMENT BIAS FOR 11-LOCUS TRAIT
  CALCULATES FALSE POSITIVE RATE FOR N CANDIDATE TRAITS, WHERE
    N  $\times$  1, 10, 25 50 AND 100.
  CALCULATES FPR FOR CHOOSING MOST DIVERGE TRAIT FROM CANDIDA
    TE LIST, AND FOR CHOOSING RANDOMLY FROM TOP HALF OF DIVERGE
    D TRAITS.

NTRAITS $\leftarrow$ 50  TOTAL NUMBER OF TRAITS TO CHOOSE FROM

SCORE1 $\leftarrow$ SCORE10TOP $\leftarrow$ SCORE10MID $\leftarrow$ SCORE25TOP $\leftarrow$ SCORE25MID $\leftarrow$ SCORE50TOP $\leftarrow$ 
SCORE50MID $\leftarrow$ RAND1 $\leftarrow$ RAND10 $\leftarrow$ RAND25 $\leftarrow$ RAND50 $\leftarrow$ RAND10TOP $\leftarrow$ RAND25TOP $\leftarrow$ RAND
50TOP $\leftarrow$ 0.0
SCORE100TOP $\leftarrow$ SCORE100MID $\leftarrow$ RAND100 $\leftarrow$ RAND100TOP $\leftarrow$ 0.0

NLOCI $\leftarrow$ 11

I $\leftarrow$ 0
RETI:I $\leftarrow$ I+1  REPLICATE LOOP

SCORES $\leftarrow$ 0.0  NUMBER OF 2 ALLELES
J $\leftarrow$ 0
RETJ:J $\leftarrow$ J+1  TRAIT LOOP

ALLELES $\leftarrow$ ?(NLOCI.2)

(16 $\geq$ (+/ALLELES))/'ALLELES $\leftarrow$ 1 (ALLELES-3)'  CHANGE IF CHANG
  E NUMBER OF TRIALS

SCORE $\leftarrow$ +/ (ALLELES=2)
SCORES $\leftarrow$ SCORES, SCORE

 $\rightarrow$ (J<100)/RETJ

SCORE1 $\leftarrow$ SCORE1, SCORES[1]
RAND1 $\leftarrow$ RAND1, SCORES[.50]

PART10 $\leftarrow$ SCORES[1.10]
PART10 $\leftarrow$ PART10[ $\nabla$ PART10]
SCORE10TOP $\leftarrow$ SCORE10TOP, PART10[1]
```

```

SCORE10MID←SCORE10MID,PART10[5]
RAND10←RAND10,PART10[?10]
RAND10TOP←RAND10TOP,PART10[?5]

PART25←SCORES[125]
PART25←PART25[▽PART25]
SCORE25TOP←SCORE25TOP,PART25[1]
SCORE25MID←SCORE25MID,PART25[13]
RAND25←RAND25,PART25[?25]
RAND25TOP←RAND25TOP,PART25[?13]

PART50←SCORES[150]
PART50←PART50[▽PART50]

SCORE50TOP←SCORE50TOP,PART50[1]
SCORE50MID←SCORE50MID,PART50[25]
RAND50←RAND50,PART50[?50]
RAND50TOP←RAND50TOP,PART50[?25]

SCORES←SCORES[▽SCORES]
SCORE100TOP←SCORE100TOP,SCORES[1]
SCORE100MID←SCORE100MID,SCORES[50]
RAND100←RAND100,SCORES[?100]
RAND100TOP←RAND100TOP,SCORES[?50]

→(I<10000)/RETI

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 1      ',(+/SCORE1
≥10)÷ρSCORE1
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 1  ',(+/SCORE1
≥10)÷ρSCORE1

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 10 TOP    ',(+/SCORE1
0TOP≥10)÷ρSCORE10TOP
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 10 MID    ',(+/SCORE1
0MID≥10)÷ρSCORE10MID
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 10 ',(+/RAND10
≥10)÷ρRAND10
'FALSE POSIT PROPORTION OFR RANDOM SAMPLE OF TOP 5 ',(+/RAND10
TOP≥10)÷ρRAND10TOP

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 25 TOP    ',(+/SCORE2
5TOP≥10)÷ρSCORE25TOP
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 25 MID    ',(+/SCORE2
5MID≥10)÷ρSCORE25MID
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 25 ',(+/RAND25
≥10)÷ρRAND25

```

```
'FALSE POSI PROPORTION OFR RANDOM SAMPLE OF TOP 13 ',(+/RAND25
TOP≥10)÷ρRAND25TOP
```

```
□TCLF
```

```
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 50 TOP ',(+/SCORE5
0TOP≥10)÷ρSCORE50TOP
```

```
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 50 MID ',(+/SCORE5
0MID≥10)÷ρSCORE50MID
```

```
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 50 ',(+/RAND50
≥10)÷ρRAND50
```

```
'FALSE POSI PROPORTION OFR RANDOM SAMPLE OF TOP 25 ',(+/RAND50
TOP≥10)÷ρRAND50TOP
```

```
□TCLF
```

```
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 100 TOP ',(+/SCORE1
00TOP≥10)÷ρSCORE100TOP
```

```
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 100 MID ',(+/SCORE1
00MID≥10)÷ρSCORE100MID
```

```
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 100 ',(+/RAND1
00≥10)÷ρRAND100
```

```
'FALSE POSI PROPORTION OFR RANDOM SAMPLE OF TOP 50 ',(+/RAND10
0TOP≥10)÷ρRAND100TOP
```

```
□TCLF
```

```
□TCLF
```

```
'PROGRAM ASCERTAIN11 FINISHED.'
```

B. Program ASCERTAIN10

Code:

```
ASCERTAIN10
```

```
  # THIS PROGRAM CALCULATES ASCERTAINMENT BIAS
```

```
NTRAITS←50  # TOTAL NUMBER OF TRAITS TO CHOOSE FROM
```

```
SCORE1←SCORE10TOP←SCORE10MID←SCORE25TOP←SCORE25MID←SCORE50TOP←
SCORE50MID←RAND1←RAND10←RAND25←RAND50←RAND10TOP←RAND25TOP←RAND
50TOP←0ρ0
```

```
NLOCI←10
```

```
I←0
```

```
RETI:I←I+1  # REPLICATE LOOP
```

```
SCORES←0ρ0  # NUMBER OF 2 ALLELES
```

```
J←0
```

```
RETJ:J←J+1  # TRAIT LOOP
```

```

ALLELES←?(NLOCIP2)

(15≥(+/ALLELES))/'ALLELES←1(ALLELES-3)'      A CHANGE IF CHANGE
NUMBER OF TRIALS

SCORE←+/(ALLELES=2)
SCORES←SCORES,SCORE

→(J<50)/RETJ

SCORE1←SCORE1,SCORES[1]
RAND1←RAND1,SCORES[?50]

PART10←SCORES[110]
PART10←PART10[▽PART10]
SCORE10TOP←SCORE10TOP,PART10[1]
SCORE10MID←SCORE10MID,PART10[5]
RAND10←RAND10,PART10[?10]
RAND10TOP←RAND10TOP,PART10[?5]

PART25←SCORES[125]
PART25←PART25[▽PART25]
SCORE25TOP←SCORE25TOP,PART25[1]
SCORE25MID←SCORE25MID,PART25[13]
RAND25←RAND25,PART25[?25]
RAND25TOP←RAND25TOP,PART25[?13]

SCORES←SCORES[▽SCORES]
SCORE50TOP←SCORE50TOP,SCORES[1]
SCORE50MID←SCORE50MID,SCORES[25]
RAND50←RAND50,SCORES[?50]
RAND50TOP←RAND50TOP,SCORES[?25]

→(I<10000)/RETI

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 1      ',(+/SCORE1
≥9)÷ρSCORE1
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 1  ',(+/SCORE1
≥9)÷ρSCORE1

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 10 TOP    ',(+/SCORE1
0TOP≥9)÷ρSCORE10TOP
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 10 MID    ',(+/SCORE1
0MID≥9)÷ρSCORE10MID
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 10 ',(+/SCORE1
≥9)÷ρSCORE1
'FALSE POSIT PROPORTION OFR RANDOM SAMPLE OF TOP 5 ',(+/RAND10
TOP≥9)÷ρRAND10TOP

```

```

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 25 TOP      ',(+/SCORE2
5TOP≥9)÷ρSCORE25TOP
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 25 MID      ',(+/SCORE2
5MID≥9)÷ρSCORE25MID
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 25  ',(+/SCORE1
≥9)÷ρSCORE1
'FALSE POSI PROPORTION OFR RANDOM SAMPLE OF TOP 13  ',(+/RAND25
TOP≥9)÷ρRAND25TOP

```

```

□TCLF
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 50 TOP      ',(+/SCORE5
0TOP≥9)÷ρSCORE50TOP
'FALSE POSITIVE PROPORTION FOR SAMPLE OF 50 MID      ',(+/SCORE5
0MID≥9)÷ρSCORE50MID
'FALSE POSITIVE PROPORTION FOR RANDOM SAMPLE OF 50  ',(+/SCORE1
≥9)÷ρSCORE1
'FALSE POSI PROPORTION OFR RANDOM SAMPLE OF TOP 25  ',(+/RAND50
TOP≥9)÷ρRAND50TOP

```

```

□TCLF
□TCLF
'PROGRAM ASCERTAIN10 FINISHED.'

```

C. Program ASCERTAIN11A

Code:

```

ASCERTAIN11A
  A THIS PROGRAM CALCULATES ASCERTAINMENT BIAS FOR CHOOSING RAND
    ONLY FROM X MOST DIVERGED TRAITS OUT OF 100 TRAITS.

NTRAITS←50  A TOTAL NUMBER OF TRAITS TO CHOOSE FROM

SCORE1←SCORE10TOP←SCORE10MID←SCORE25TOP←SCORE25MID←SCORE50TOP←
SCORE50MID←RAND1←RAND10←RAND25←RAND50←RAND10TOP←RAND25TOP←RAND
50TOP←0ρ0
SCORE100TOP←SCORE100MID←RAND100←RAND100TOP←0ρ0

TSCORE100←TSCORE90←TSCORE80←TSCORE70←TSCORE60←TSCORE50←TSCORE4
0←TSCORE30←TSCORE20←TSCORE10←0ρ0

NLOCI←11

I←0
RETI:I←I+1  A REPLICATE LOOP

SCORES←0ρ0  A NUMBER OF 2 ALLELES

```

```

J←0
RETJ:J←J+1  A TRAIT LOOP

ALLELES←?(NLOCIP2)  A GENERATE ALLELES FOR TRAIT (1 OR 2 AT E
ACH LOCUS)

Φ(16≥(+/ALLELES))/'ALLELES←1(ALLELES-3)'  A SWITCH 1 AND 2
ALLELES IF MORE 1 THAN 2 ALLELES

SCORE←+/(ALLELES=2)  A NUMBER OF 2 ALLELES IN GENOTYPE
SCORES←SCORES,SCORE

→(J<100)/RETJ

SCORES←SCORES[▽SCORES]  A ORDER TRAITS BY NUMBER OF
2 ALLELES, HIGHEST TO LOWEST

TSCORE100←TSCORE100,SCORES[?100]  A PICK STUDY TRAIT RANDOMLY
FROM AMONG ALL CANDIDATE TRAITS
TSCORE90←TSCORE90,SCORES[?90]  A PICK STUDY TRAIT RANDOMLY
FROM AMONG MOST-DIVERGED 90 PERCENT OF CANDIDATE TRAITS
TSCORE80←TSCORE80,SCORES[?80]  A ETC.
TSCORE70←TSCORE70,SCORES[?70]
TSCORE60←TSCORE60,SCORES[?60]
TSCORE50←TSCORE50,SCORES[?50]
TSCORE40←TSCORE40,SCORES[?40]
TSCORE30←TSCORE30,SCORES[?30]
TSCORE20←TSCORE20,SCORES[?20]
TSCORE10←TSCORE10,SCORES[?10]

→(I<10000)/RETI

MAT←0 10000ρ0  A MAKE A MATRIX OUT OF THE TSCORE VARIABLES
MAT←MAT,[1]TSCORE100
MAT←MAT,[1]TSCORE90
MAT←MAT,[1]TSCORE80
MAT←MAT,[1]TSCORE70
MAT←MAT,[1]TSCORE60
MAT←MAT,[1]TSCORE50
MAT←MAT,[1]TSCORE40
MAT←MAT,[1]TSCORE30
MAT←MAT,[1]TSCORE20
MAT←MAT,[1]TSCORE10

VEC←(+/MAT≥10)÷10000  A COUNT PROPORTION OF STUDY TRAITS
IN MATRIX SIGNIFICANT AT NOMINAL LEVEL (E.G. HAS EITHER 10
OR 11 2 ALLELES)--THIS IS FALSE DISCOVERY RATE
OBSRATIO←VEC÷VEC[1]  A CALCULATE RATIO OF FDR FOR A GIVEN
BETA TO FDR FOR BETA=1 ('FDR RATIO')
Q←19

```

```

QQ←1÷(1-.1×Q)          a EXPECTED FDR RATIO (EQUAL TO 1÷BETA)
QQ←1,QQ

OUT←(10 1ρVEC),10 1ρQQ
OUT←OUT,10 1ρOBSRATIO
OUT←OUT,10 1ρ(100,90,80,70,60,50,40,30,20,10)

□TCLF
'FALSE POSITIVE RATOS '
□TCLF
' OBS  OBSRATIO EXPRATIO  PROPORTION'
OUT

□TCLF
□TCLF
'PROGRAM ASCERTAIN11A FINISHED.'
```


IV. Counting antagonistic QTLs for assessing selection

PROGRAM CONTRA-DIRECTIONAL QTL COUNTS, FOR EACH FLORAL AND NECTAR TRAIT (NOT TRANSFORMED):

- (1) NUMBER OF CONTRA QTLs (QTLs WITH EFFECTS IN OPPOSITE DIRECTION AS SPECIES DIFFERENCE)
- (2) NUMBER OF THESE OVERLAPPING CONSISTENT FLORAL QTLs (CONSISTENT MEANS EFFECT IN SAME DIRECTION AS SPECIES DIFFERENCE)
- (3) NUMBER OF THESE OVERLAPPING CONSISTENT NECTAR QTLs
- (4) NUMBER OF THESE OVERLAPPING EITHER CONSISTENT FLORAL QTLs, CONSISTENT NECTAR QTLs, OR BOTH

A. Program ANTAGQTL

Code:

ANTAGQTL

 THIS PROGRAM IDENTIFIES QTL'S WITH ANTAGONISTIC EFFECTS ON
 DIFFERENT TRAITS

THREES←3.1,3.2,3.3

MATCHES←0 10ρ0

MAXJ←1↑ρΔMASTERDATA

MAXI←MAXJ-1

I←0

RETI:I←I+1 A LOOP FOR FIRST QTL

J←I

RETJ:J←J+1

LINEI←ΔMASTERDATA[I;]

LINEJ←ΔMASTERDATA[J;]

TEST←LINEI[1]=LINEJ[1]

→(TEST=1)/DOWN A SKIP IF QTLs AFFECT SAME TRAIT

TEST←LINEI[2]≠LINEJ[2]

→(TEST=1)/DOWN A SKIP ON DIFFERENT CHROMOSOME

TEST←((LINEI[4]=LINEJ[3])∨(LINEJ[4]=LINEI[3]))

→(TEST=1)/DOWN

CRIT1←(LINEI[3]≥LINEJ[3]) ∧ (LINEI[3]≤LINEJ[4]) A THESE
 TWO LINES TEST WHETHER THERE IS OVERLAP

CRIT2←(LINEI[4]≥LINEJ[3]) ∧ (LINEI[4]≤LINEJ[4])

CRIT3←(LINEJ[3]≥LINEI[3]) ∧ (LINEJ[3]≤LINEI[4])

CRIT4←(LINEJ[4]≥LINEI[3]) ∧ (LINEJ[4]≤LINEI[4])

```
TEST2←CRIT1∨CRIT2∨CRIT3∨CRIT4  
→(TEST2=0)/DOWN
```

```
TEST1←((LINEI[9]>0)^(LINEJ[9]<0)) ∨ ((LINEI[9]<0)^(LINEJ[9]>0)  
)
```

```
→(TEST1=0)/DOWN
```

```
TEST←(LINEI[1]∈THREES)∨(LINEJ[1]∈THREES)  
→(TEST=1)/DOWN
```

```
NEWLINE←LINEI[14],LINEJ[14],LINEI[9],LINEJ[9]  
MATCHES←MATCHES,[1]NEWLINE
```

```
DOWN:→(J<MAXJ)/RETJ
```

```
→(I<MAXI)/RETI
```