ibdreg:
An R package for Genetic Linkage with Covariates

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ibdreg: outline

- Intro to Linkage (brief)
- Why do linkage with covariates?
- Methodology in ibdreg
- Demo of ibdreg on prostate cancer data

Linkage Intro:
Genetic Analysis Types

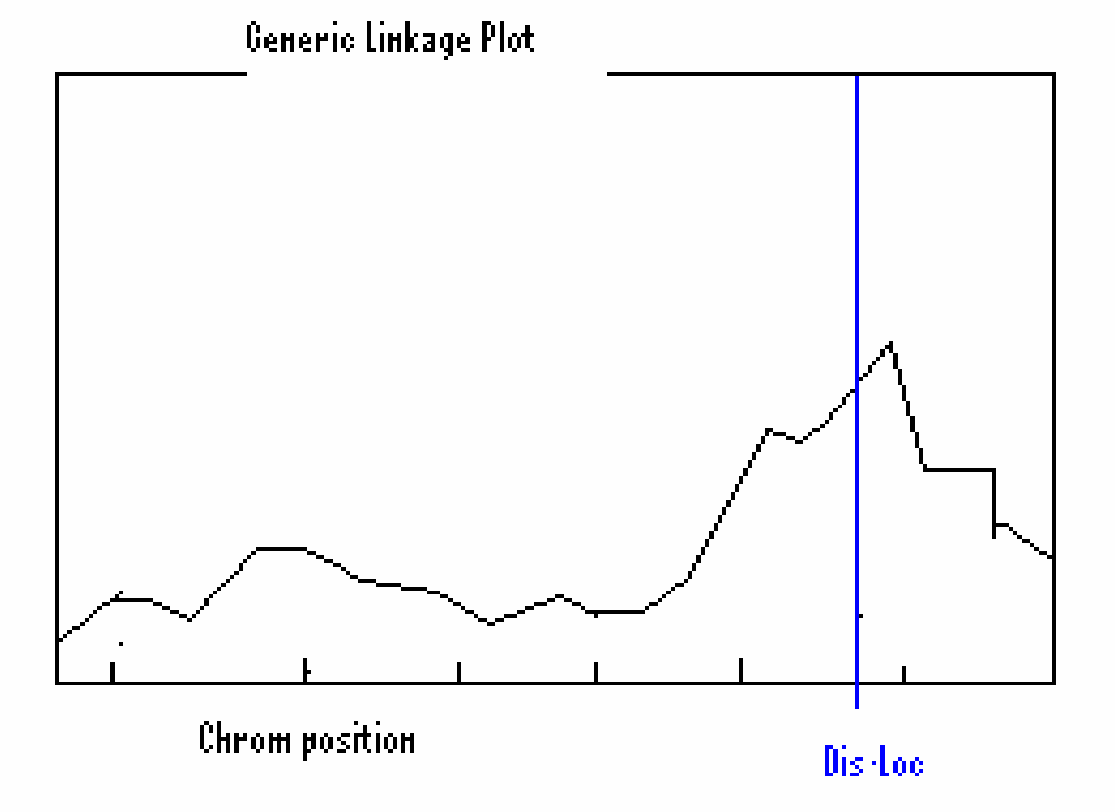
- Segregation Analysis - Use family data to determine the extent to which a disease is genetic
- Linkage Analysis – Use family data to locate a chromosome segment that is inherited jointly with a disease locus
- Association Analysis – Use unrelated data to find a genetic effect of a particular DNA marker(s) on a disease

Intro to Linkage: Recombination

- Mendel proposed genes (on peas) are inherited independently of one another
- Not quite right -- genes / markers which are "close" together show associations

- During meiosis, corresponding DNA segments can recombine in any location
- Recombination is somewhat constant over the whole genome
- In general, "close" markers have fewer recombinations

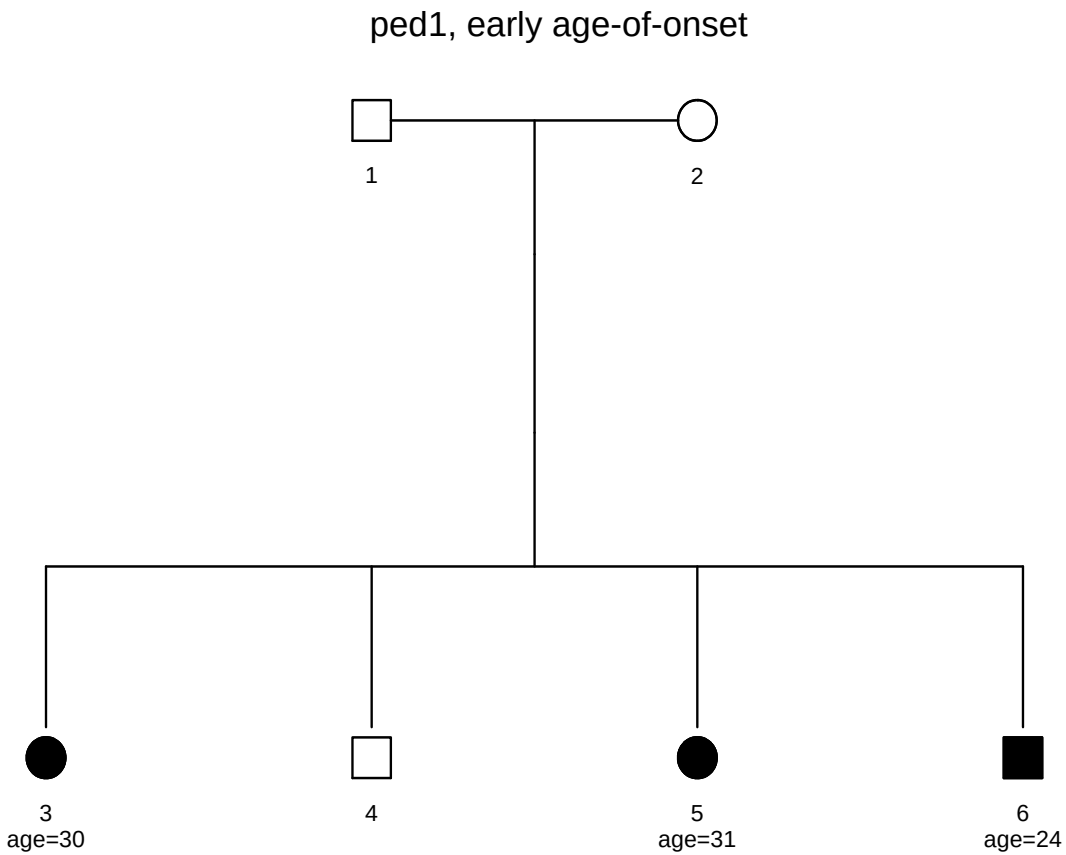
Intro to Linkage, basic plot



Linkage with Covariates: illustrated

Nuclear family with 4 children, 3 affected with disease

If genetic linkage:
- persons 3,5 share more alleles near disease locus
- persons 3,4 share fewer alleles at disease locus



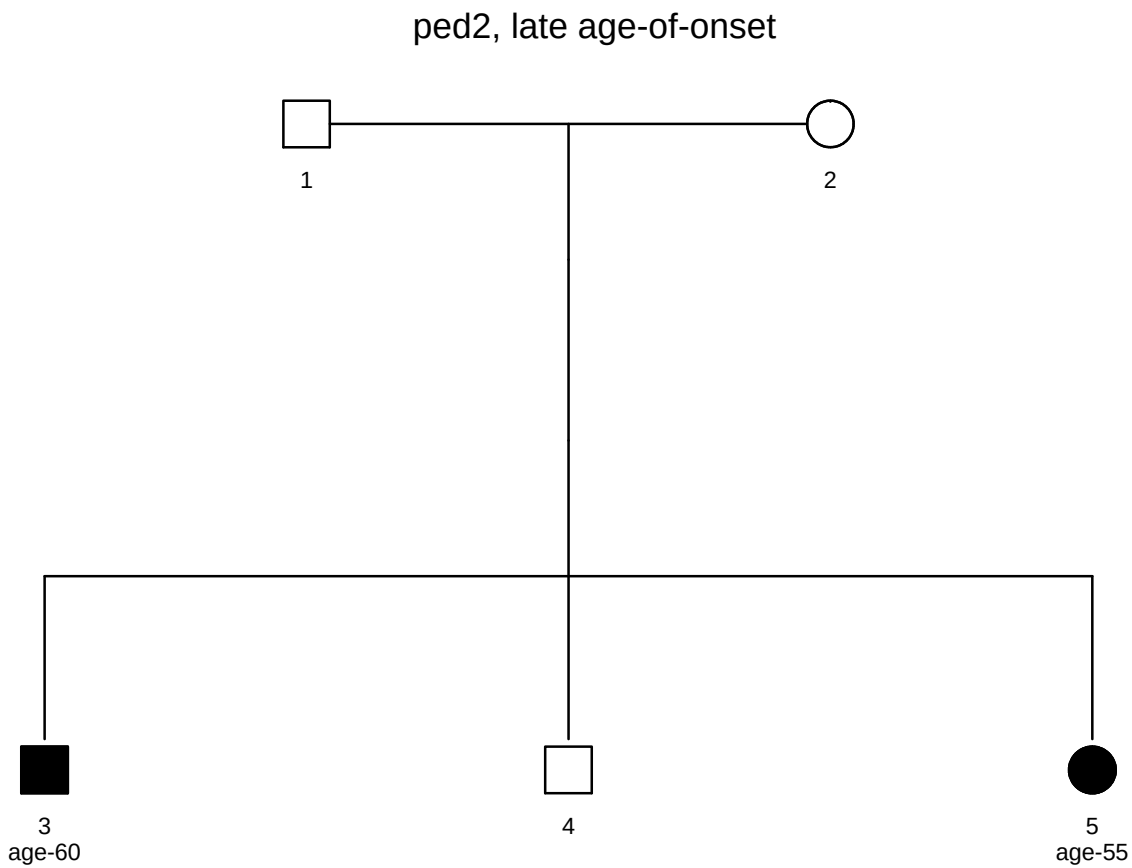
Linkage with Covariates: IBD Sharing

- Identity-By-Descent (IBD): alleles that have been inherited from the same ancestor
- Estimate probability of sharing 0, 1, and 2 alleles IBD: f_0, f_1, f_2
- Use Merlin, Genehunter, etc.
- Estimated IBD sharing for a relative pair: $$s_r = 2 f_{r,2} + 1 f_{r,1}$$

Linkage with Covariates: illustrated

Nuclear family, two affected siblings

Late-onset form of the disease



Linkage with Covariates

- Ped1 and Ped2 could be two forms of disease: early and late onset
- Mixture of gene and environment effects, i.e. disease heterogeneity
- Assume the early onset is genetically linked at locus **L**
- IBD sharing at **L** is:
  - ped1: greater than null
  - ped2: somewhere near null

Linkage with Covariates

- The more alike the covariates, the more allele sharing is expected
- Consider a function of the age of onset for affected siblings:
  1. age1 + age2
  2. age1 * age2
  3. $(|age1-60| * |age2-60|)^2$
- Define any function that applies to the disease and covariate impact

Linkage with Covariates

- Intuitively, affected relative pairs with similar covariates share more alleles than the null
- Linear relationship for IBD sharing and pair-specific covariates (X):

$$m_r = m_r^0 + c_r X\beta = X_r^* \beta$$

m_r, m_r^o: Expected s_r under linkage, no linkage

c_r = scaling factor for the relationship types

$$X_r^* = c_r[1 \mid X]$$

ibdreg method

Quasi-likelihood score function:

$$U = \sum_{i=1}^{n} D_i^T V_i^{-1} (S_i - M_i)$$

where

i = 1..n pedigrees

S_i, M_i: vectors of estimated and expected (H_o) allele sharing

D_i: derivative of M_i with respect to β

V_i: covariance matrix of the S_i vector

ibdreg method

Develop a chi-square test:

$$T = U^T V_u^{-1} U$$

where $V_u = \sum_{i=1}^{n} X_i^{*T} V_{0,i}^{-1} X_i^*$

With q degrees of freedom:

- q=1 for intercept-only (linkage)
- q=2 for intercept with 1 covariate
- etc.

ibdreg method

Available tests (degrees of freedom)

1. Linkage without covariates (q=1)
2. Linkage with p covariates (q=1+p)
3. Effect of p covariates on IBD sharing, adjusting for linkage (p)

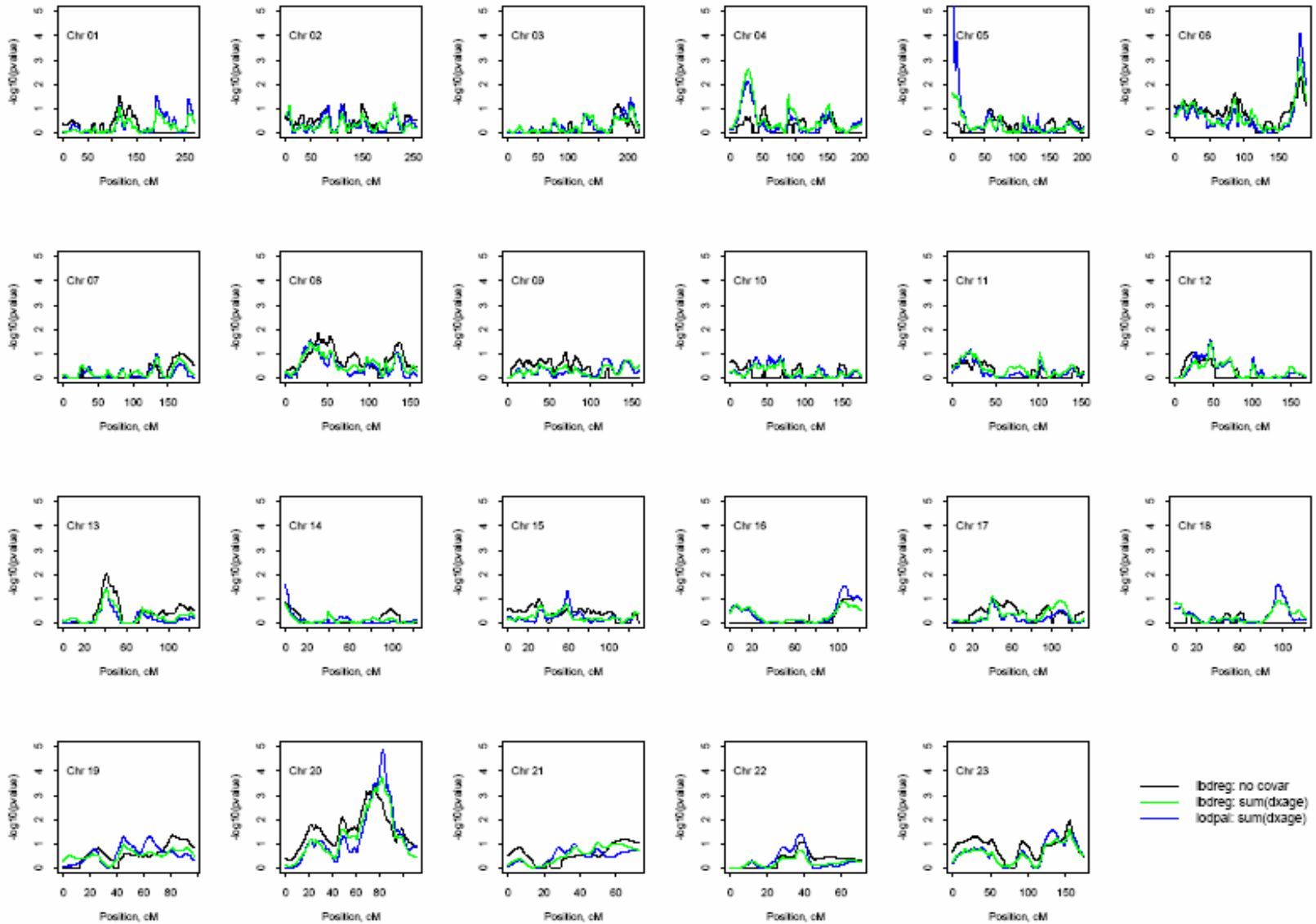
ibdreg applied

- Data: sample of pedigrees ascertained at Mayo Clinic (SE MN) for having multiple cases of prostate cancer
- 159 pedigrees (max 21 people)
- 495 affected relative pairs (ARPs)
- Covariate: age-of-onset (dxAge)

ibdreg applied

- LODPAL, of S.A.G.E., approximates a pseudo-likelihood of IBD sharing probabilities by a trinomial logistic regression model (**blue line**)
- Compare to ibdreg (**green line**), both using sum(dxAge) as a covariate
- Also include ibdreg, linkage-only (**black line**)
- Same c scale (genetic effect between dominant and recessive)

Figure 1


Chr 01
Chr 02
Chr 03
Chr 04
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Chr 15
Chr 16
Chr 17
Chr 18
Chr 19
Chr 20
Chr 21
Chr 22
Chr 23
-log10(pvalue)
Position, cM
lbdreg: no covar
lbdreg: sum(dxage)
lodpal: sum(dxage)

ibdreg applied

For figure above, note a few things:

- LODPAL and ibdreg generally close
- Differences:
  1. Narrow peaks (ch6, ch20)
  2. Ends of chromosomes (ch5)
- Both may be attributable to instability of maximizing pseudo-likelihood of LODPAL

ibdreg applied: R code

```
# load the library
R> library(ibdreg)

# load covariate data, containing ped id and person id to match file
R> cov.data <- read.table("cov.data.csv", sep=",")

# create data.frame with id (ped and person) and IBD data with Merlin results
R> ibd.dat.obj <-
create.ibd.dat(postfile="chr20.post.ibd",
                        priorfile="chr1.prior.ibd")

# simulate ibd.var object using "gene-dropping"
# requires a .pre file for pedigree specs
R> ibd.var.obj <- sim.ibd.var("ch.20.pre",
                           n.sim=10000)
```

ibdreg applied: R code

```
# define a function for the sum of covariates
R> pairSum <- function(cov1,cov2) {cov1+cov2}

# run ibdreg for AA relatives,
# with covariate sum(dxage), minimax c scaling
R> sum.dxage.AA <-
ibdreg(formula=~pairSum(dxage),
   status.method="AA", c.scale="minimax",
   status=pcstat, ped.id=ped.id,
   person.id=person.id, data=cov.data,
   ibd.dat=ibd.dat.obj, ibd.var=ibd.var.obj)
```

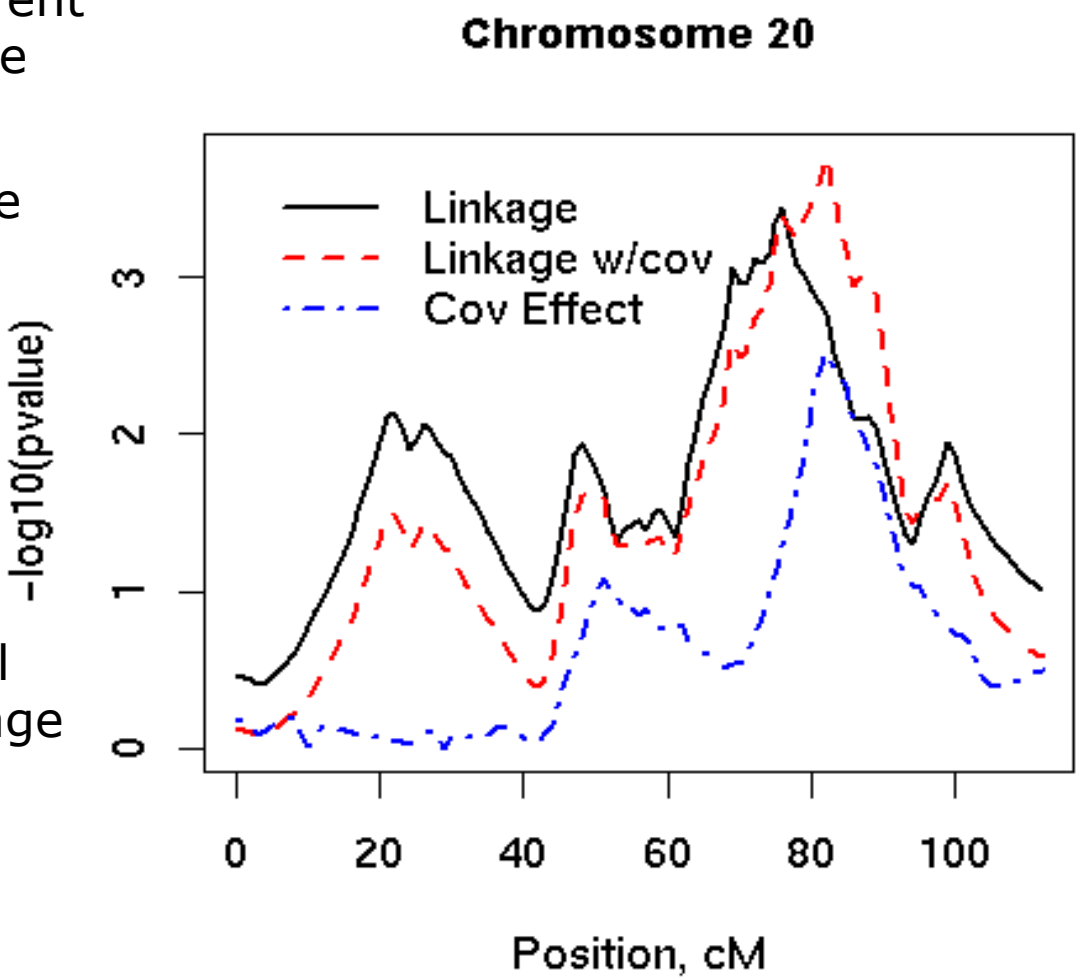
ibdreg applied: chromosome 20

Linkage signal different location from linkage w/cov

In non-peak, linkage w/cov is smaller

--1 more deg. of freedom

Cov Effect has small contribution to linkage signal



ibdreg applied: all ARPs

- Disregarding covariates, we can test linkage on all 495 relative pairs in the example, which contain sub-groups
  - AA: Affected-Affected (429)
  - UU: Unaffected-Unaffected (7)
  - AU: Affected-Unaffected (59)

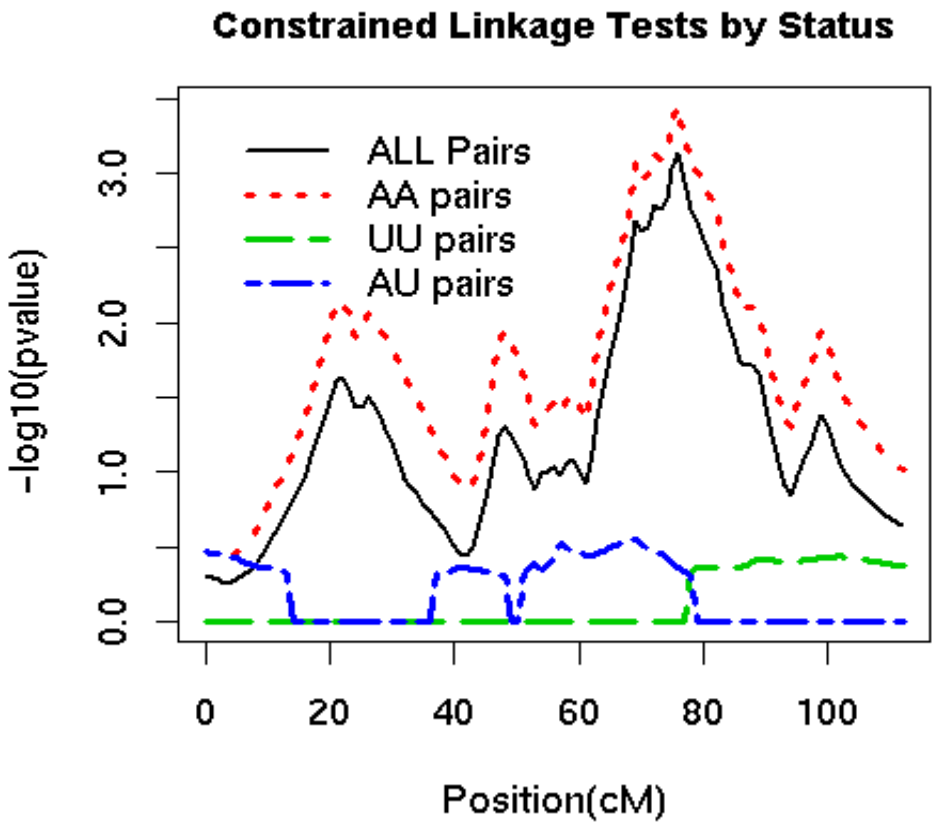
- Sharing under linkage is expected as
  - AA: More than null
  - UU: More than null, but less than AA
  - UU: Less than null

Ibdreg applied, all ARPs

Linkage tests for all relative pairs

Test for linkage in direction expected under linkage:
- AA-more
- UU-more
- AU-less

Overall linkage is driven by AA pairs



ibdreg applied: unexpected sharing

- IBD allele sharing can be invalid by:
  1. Mis-specify marker allele freq when parents missing
  2. Incorrect pedigree relationships
  3. Unaccounted inbreeding
- The χ^2 test for linkage given as both unconstrained and constrained to favor linkage (1-sided, smaller p-value)
- If $p_{unconstrained} < p_{constrained}$, indicates unexpected IBD sharing (chroms 1, 7, 12)

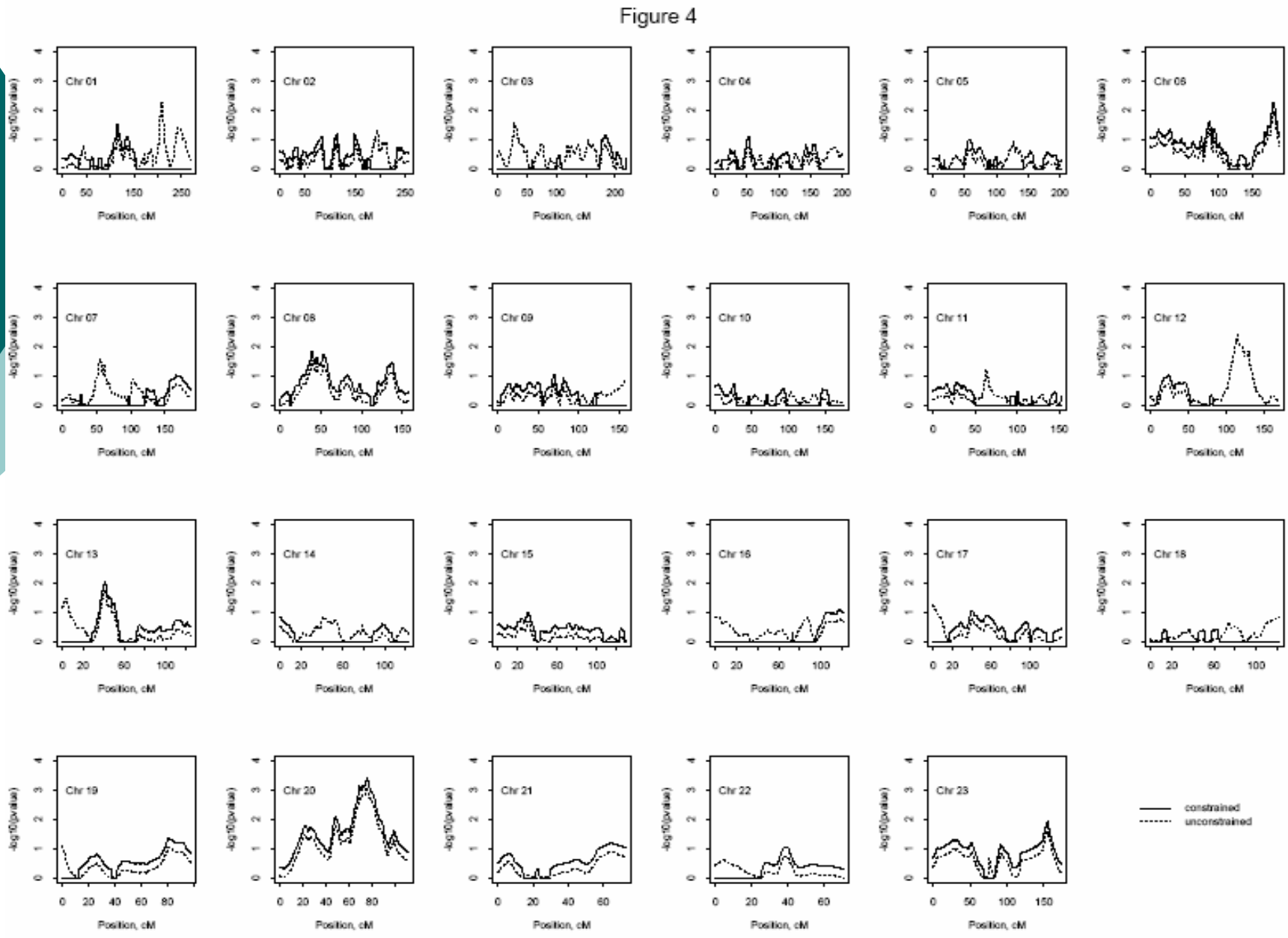


Figure 4
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Chr 16
Chr 17
Chr 18
Chr 19
Chr 20
Chr 21
Chr 22
Chr 23
-log10(pvalue)
Position, cM
constrained
unconstrained

ibdreg, concluded

- Allows multiple tests for linkage
- Provides options for specifying covariates and scaling (c_r)
- Scaling avoids fitting new regression line for each relationship type
- Linear regression is easy to fit, and easy to apply weights to account for dependence of variance on covariates
- Returned S3 object with print and plot methods
- Perl scripts supplied to work with Merlin.

Thank You

For more details and references see:

Schaid DJ, Sinnwell JP, Thibodeau SN. Testing genetic linkage with relative pairs and covariates by quasi-likelihood score statistics.
Hum Hered. 2007;64(4):220-33. Epub 2007 Jun 12.