Gene-Environment Interactions

What is gene-environment interaction?

“A different effect of an environmental exposure on disease risk in persons with different genotypes," or, alternatively, *"a different effect of a genotype on disease risk in persons with different environmental exposures.”*

Ottman, Prev Med 1996

Five different models describing how genotype and the environment can work together to alter risk

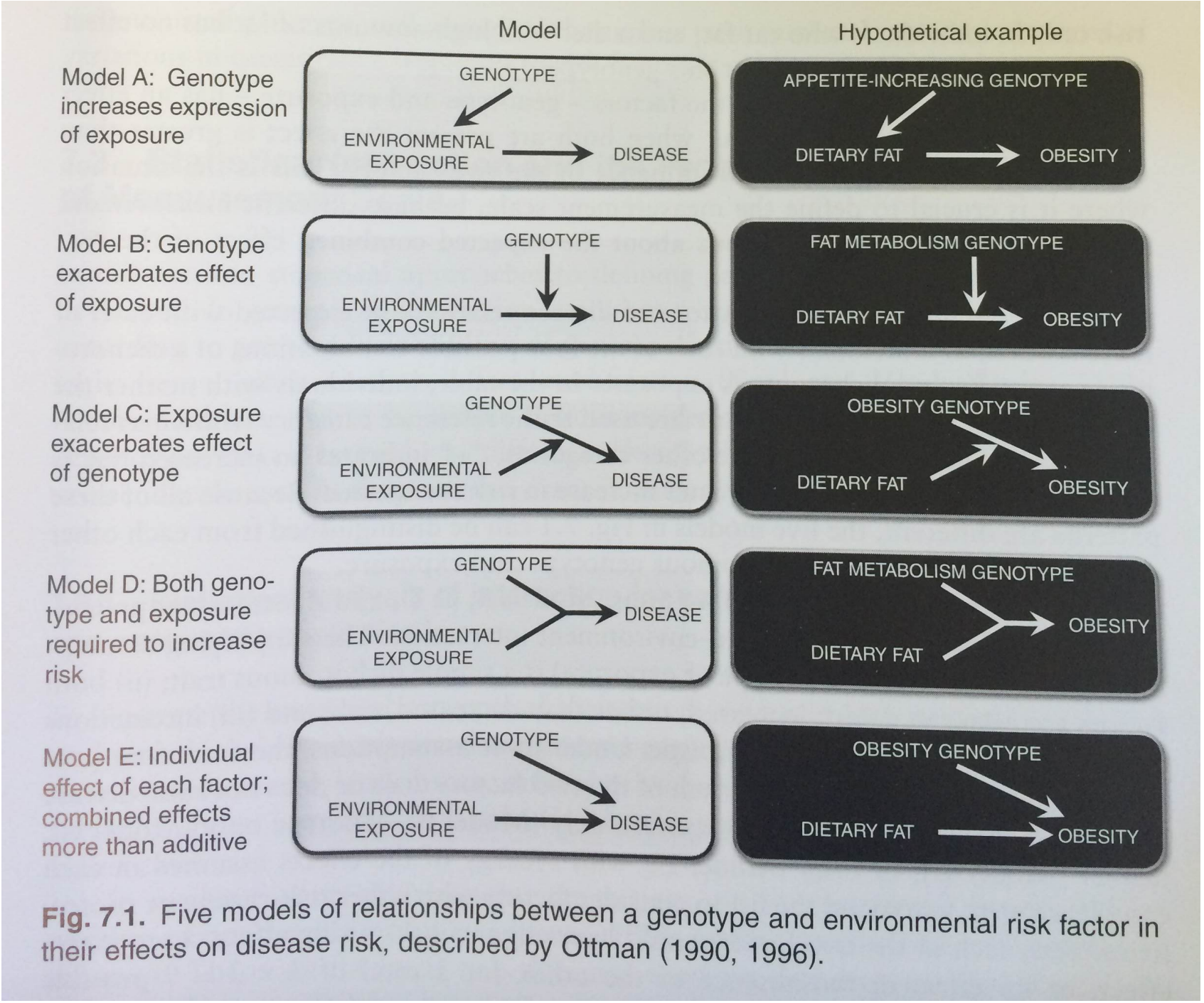
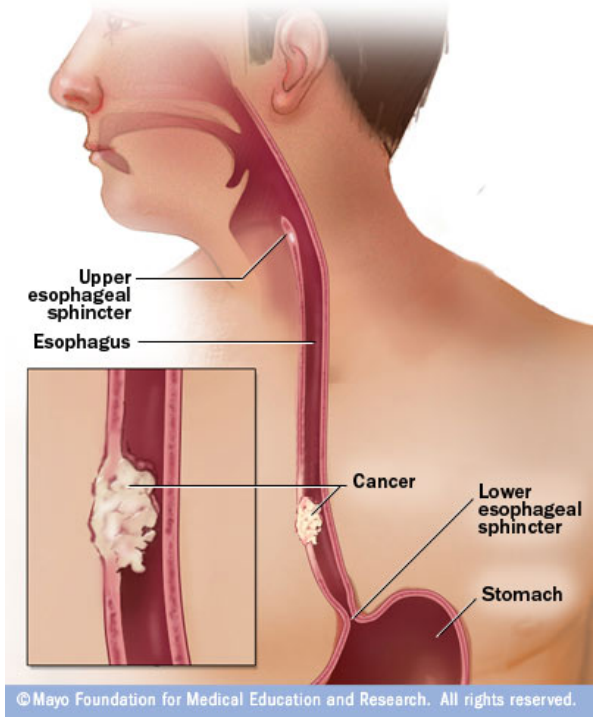


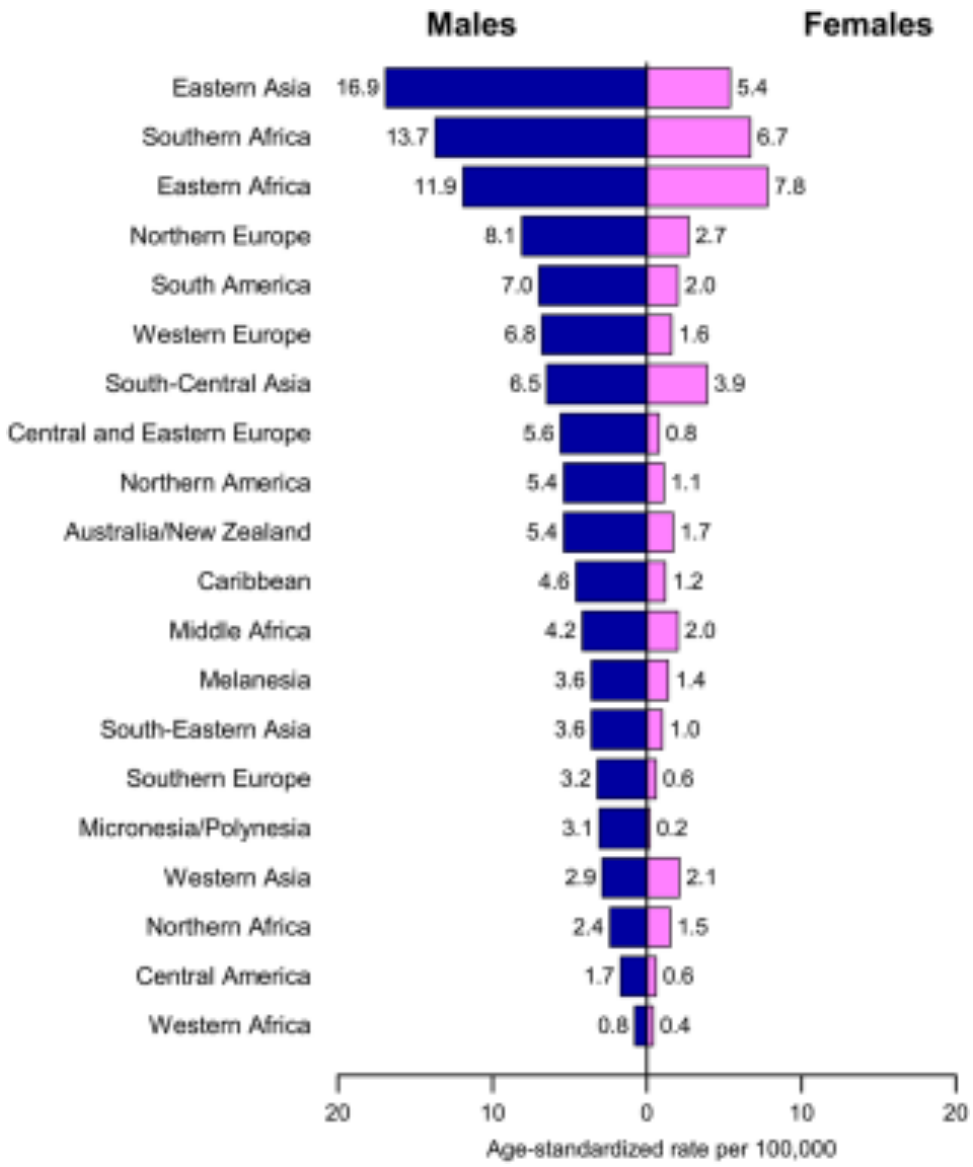
Fig. 7.1. Five models of relationships between a genotype and environmental risk factor in their effects on disease risk, described by Ottman (1990, 1996).

Poll: Why study Gene-Environment interactions?

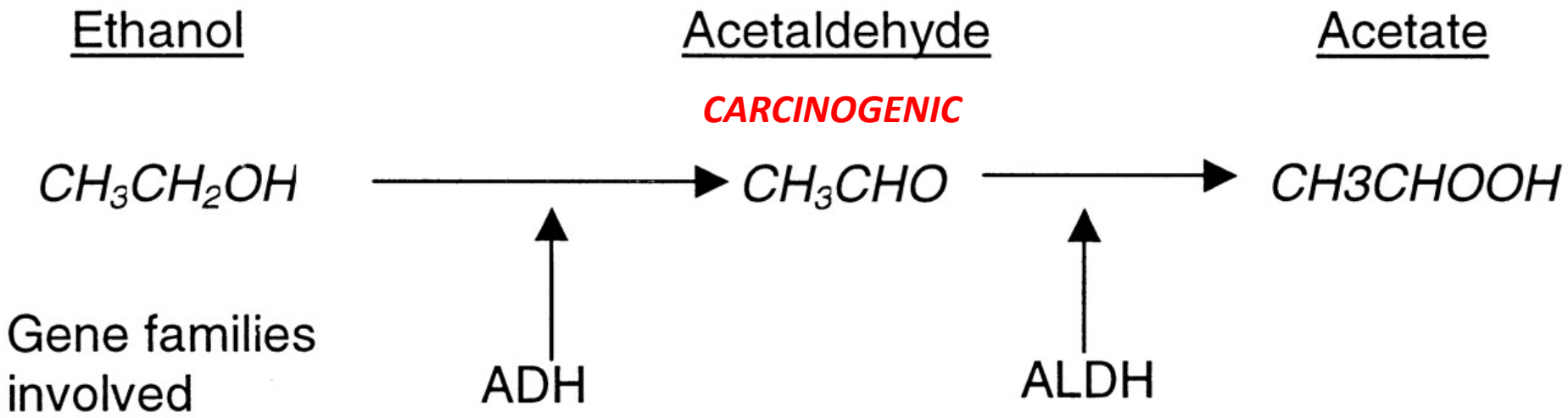
Example: Esophageal cancer

- Risk factors: alcohol intake, tobacco use, being a man, Barrett Syndrome, obesity

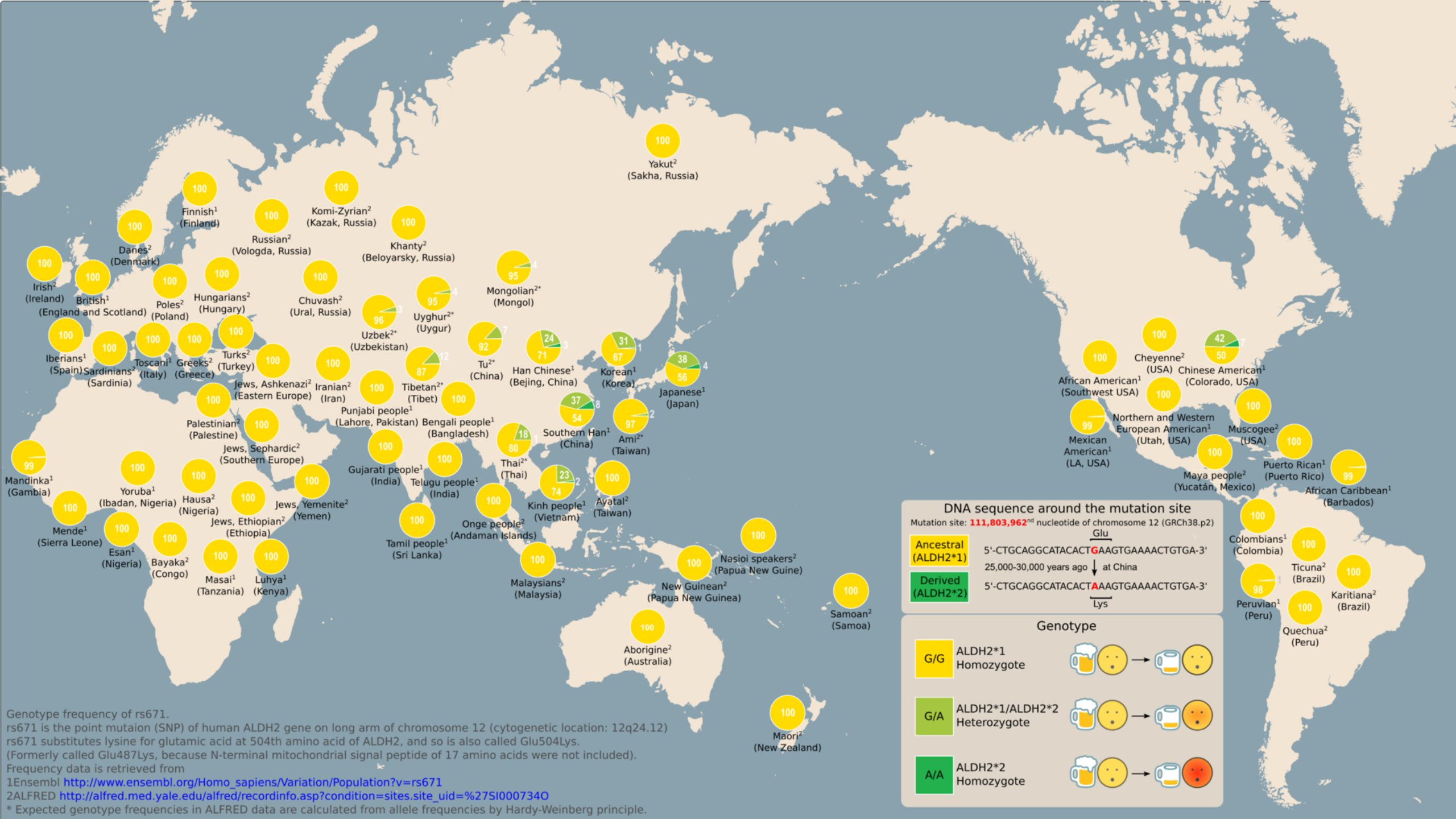




Metabolism of alcohol (ethanol) to acetaldehyde (ethanal) and then acetic acid (ethanoic acid)



Accumulation of acetaldehyde leads to alcohol flush reaction



Genotype frequency of rs671.
rs671 is the point mutaion (SNP) of human ALDH2 gene on long arm of chromosome 12 (cytogenetic location: 12q24.12)
rs671 substitutes lysine for glutamic acid at 504th amino acid of ALDH2, and so is also called Glu504Lys.
(Formerly called Glu487Lys, because N-terminal mitochondrial signal peptide of 17 amino acids were not included).
Frequency data is retrieved from
1Ensembl http://www.ensembl.org/Homo_sapiens/Variation/Population?v=rs671
2ALFRED http://alfred.med.yale.edu/alfred/recordinfo.asp?condition=sites.site_uid=%27SI000734O
* Expected genotype frequencies in ALFRED data are calculated from allele frequencies by Hardy-Weinberg principle.

Interaction between alcohol intake and *ADH1B* and *ALDH2* genotypes in Esophageal squamous-cell carcinoma

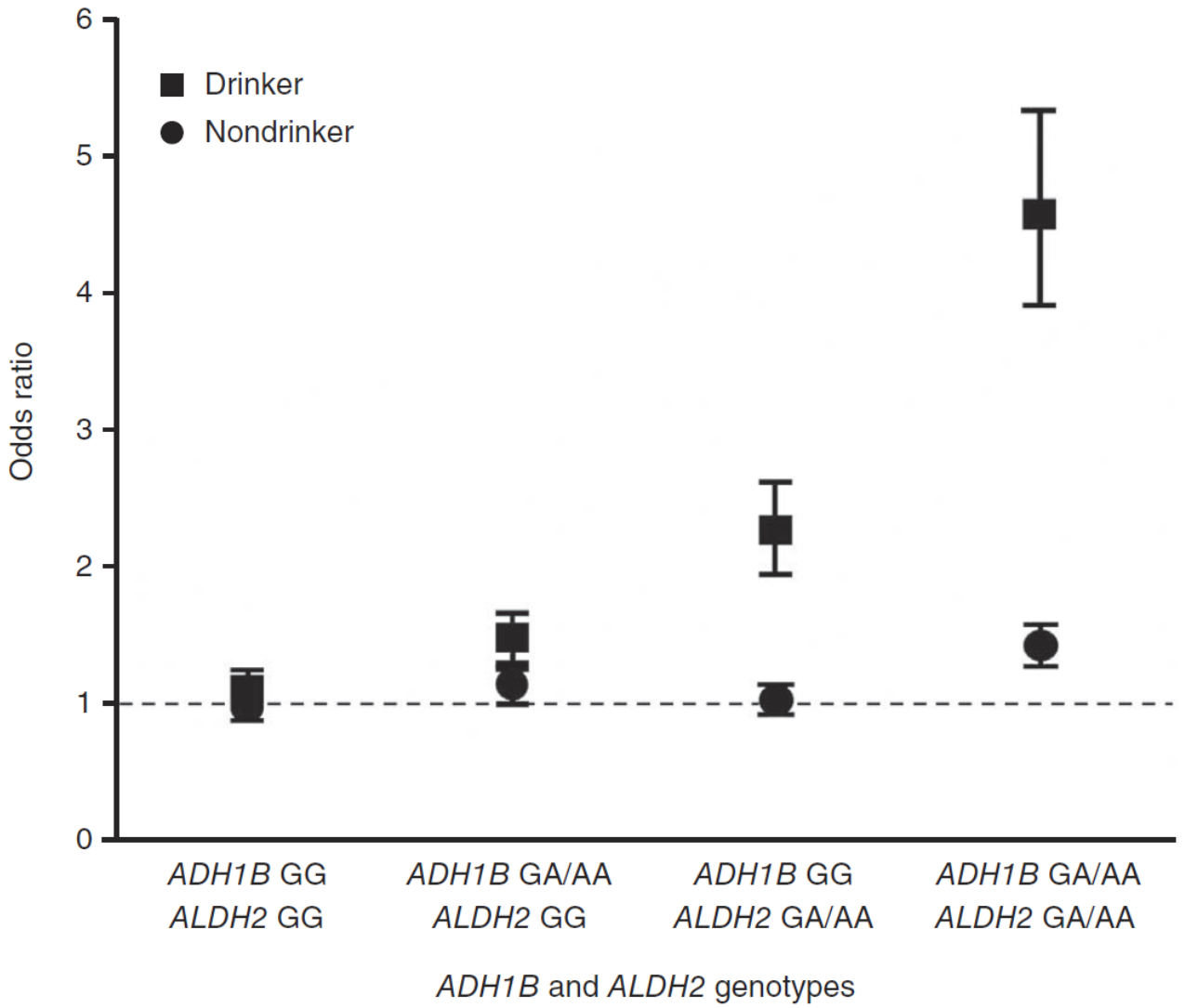


Figure 2 Plots showing the ORs for ESCC in alcohol drinkers and nondrinkers with different *ADH1B* rs1042026 and *ALDH2* rs11066015 genotypes. The vertical bars represent the 95% CIs. The horizontal dashed line indicates the null value (OR = 1.0).

Wu et al. (2012) Nat Genet

GE interactions and statistical power

- Rule of thumb:

You need **four** times as many individuals to detect an interaction effect compared to main effect analysis

The 4-by-2 table: Interaction on the multiplicative scale

	Case	Control	OR	
G=0,E=0	N_{100}	N_{000}	1	Reference
G=1,E=0	N_{110}	N_{010}	$\frac{N_{110}N_{000}}{N_{010}N_{100}}$	Risk among unexposed carriers
G=0,E=1	N_{101}	N_{001}	$\frac{N_{101}N_{000}}{N_{001}N_{100}}$	Risk among exposed non-carriers
G=1,E=1	N_{111}	N_{011}	$\frac{N_{111}N_{000}}{N_{011}N_{100}}$	Risk among exposed carriers

Often when people talk about interaction, they talk about departure from the multiplicative scale

$$OR_{INT} = \frac{OR_{11}}{OR_{10}OR_{01}}$$

Interaction exists when observed effect of G & E together is not a simple function of their individual effects

H_0: $OR_{GE}=OR_GOR_E$ vs. H_A: $OR_{GE}\neq OR_GOR_E$

In practice, we often test for interaction on the multiplicative scale

$$\text{logit}\,\mathrm{P}(D=1) = \beta + \beta_g G + \beta_e E + \beta_{ge} GE$$

$$Test: \beta_{ge} \neq 0$$

Thomas, 2004

The Joint Interaction test – a tool for gene discovery

- Is this gene associated with disease risk in any of the exposure sub-groups?
- Compare “main effect of E only” model to “main effects plus interaction” model

Null model: $\text{logit P}(D=1) = \beta + \beta_e E$

Alternative model: $\text{logit P}(D=1) = \beta + \beta_g G + \beta_e E + \beta_{ge} GE$

Compare -2 log L_{null} +2 log L_{alt} to chi-square 2 d.f.

Kraft et al, Hum Hered. 2007

Case-Only Analysis

Based on genotype-exposure table in CASES

	Carrier	Non-carrier
Exposed	N_{11}	N_{12}
Unexposed	N_{21}	N_{22}

Genotypic odds ratios for exposure from this table are equal to interaction relative risks only if genotypes and exposure are not correlated in general population.

Assuming G and E are independent in the source population, then if G and E are associated in the cases, this indicates a departure from a multiplicative odds model. (i.e. regress E on G in cases—if correlated, there is an "interaction.")

Can be much more powerful than traditional logistic regression analysis!

Piegorsch et al, Stat Med 1994

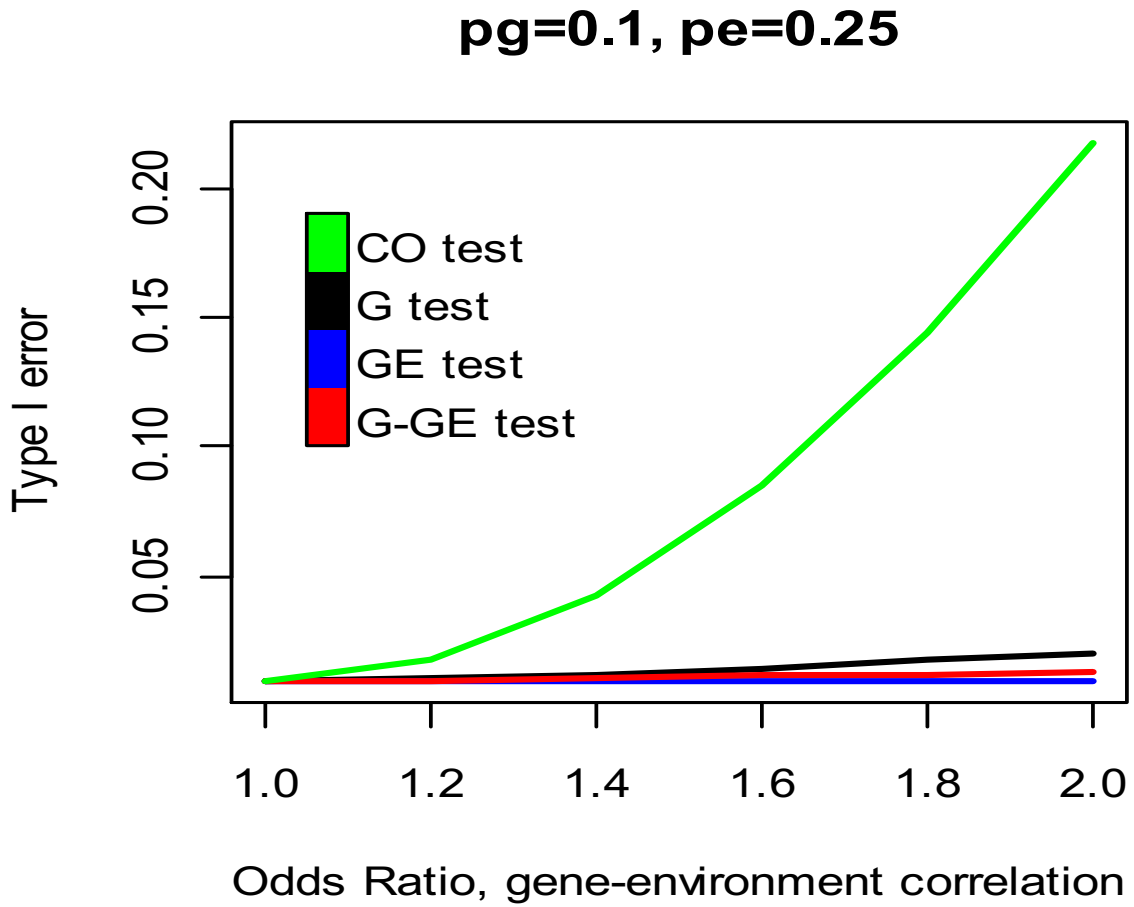
GREAT!!

Does this mean I can throw away all my controls (and decrease genotyping cost)?

- Well, the increase in power is not due to the restriction to cases per se, rather the additional assumption of G-E independence (which you can test in your controls)

- Data on controls allow for estimation of G and E main effects in addition to the interaction effect and will also allow for calculation of joint G-E-stratum-specific ORs

What if G and E are (positively) correlated?



Type I error rates as a function of GE dependence.

Sensitivity= 0.6

Specificity = 0.9

OR(E)= 1.6

Lindstrom et al, Hum Hered. 2009

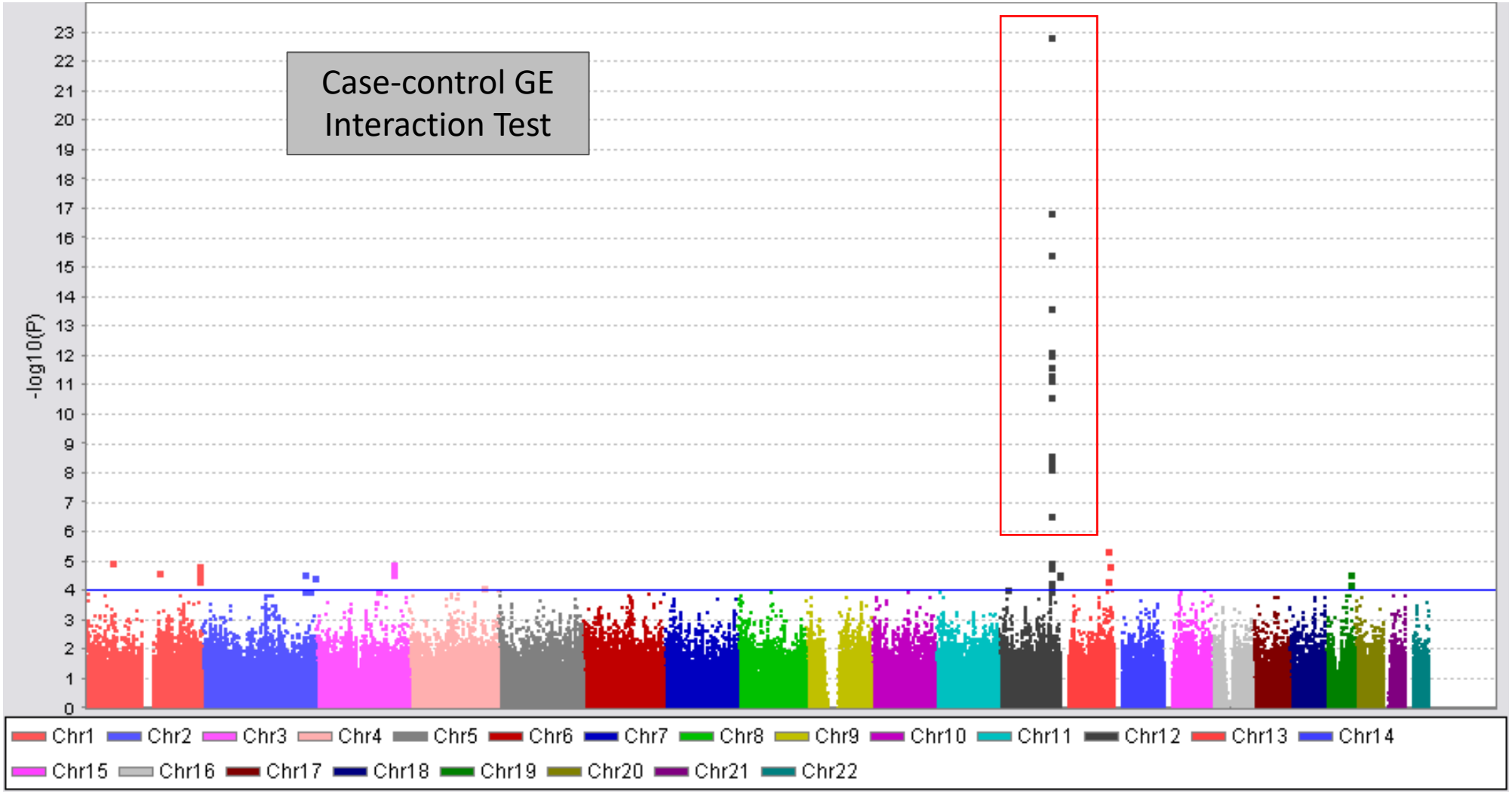
What if there is a negative correlation between G and E?

Esophageal cancer, *ALDH2* and Alcohol Intake

	OR_{E-G}	OR_{GxE}
rs670 (*ALDH*2*)	0.23	2.69

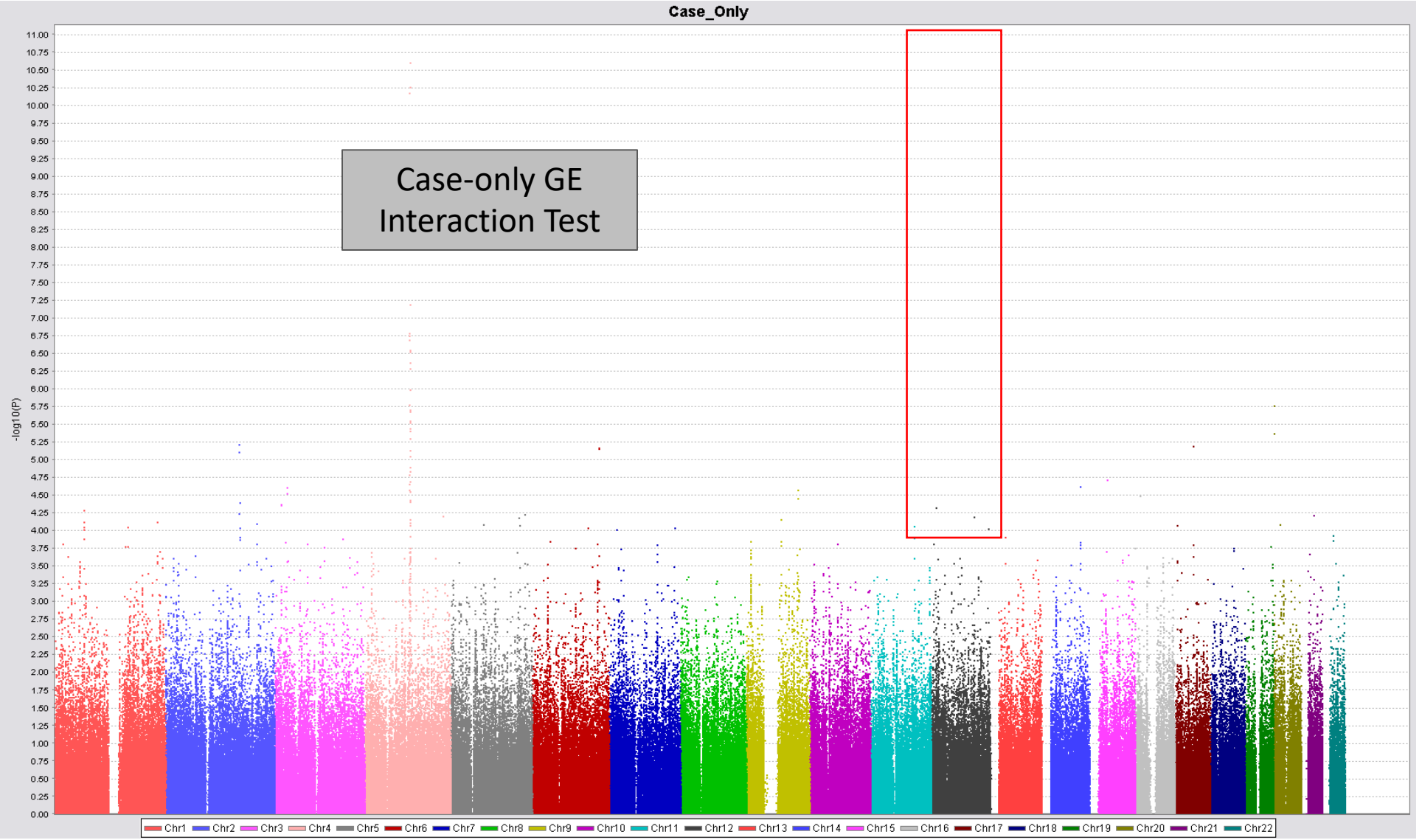
The risk allele is associated with a decreased risk of heavy drinking in the general population, and an increase in the effect of alcohol on esophageal cancer risk

Example: ESCC, *ALDH2* and Alcohol Intake



Courtesy of Chen Wu

Example: ESCC, *ALDH2* and Alcohol Intake



Courtesy of Chen Wu

Empirical Bayes Estimator

- If G and E are independent– Case-only test. Otherwise GxE interaction tests in a case-control setting (1 df)

- Trade-off between bias and efficiency:

$$\hat{\beta}_{EB} = \frac{\hat{\sigma}^2_{CC}}{(\hat{\tau}^2 + \hat{\sigma}^2_{CC})}\hat{\beta}_{CO} + \frac{\hat{\tau}^2}{(\hat{\tau}^2 + \hat{\sigma}^2_{CC})}\hat{\beta}_{CC}$$

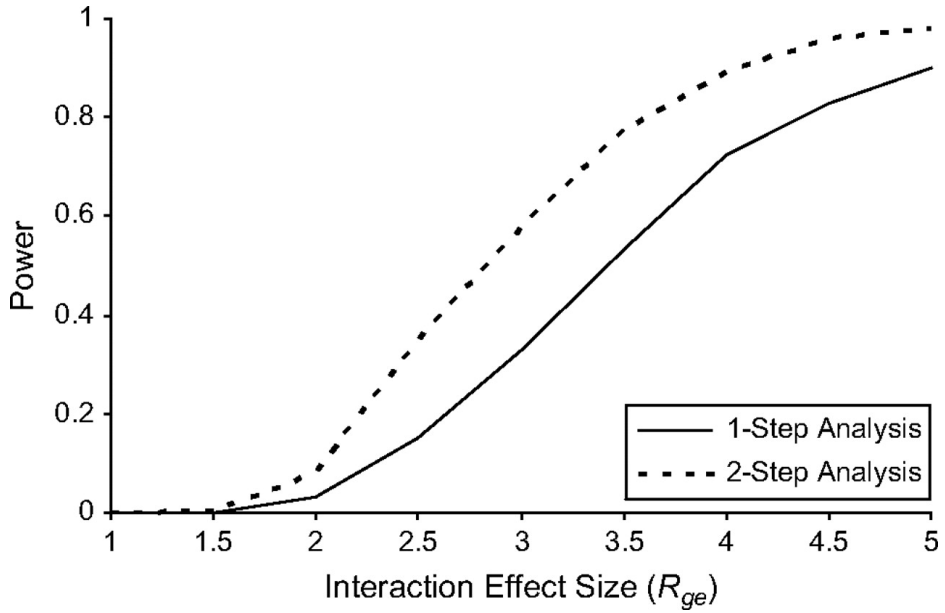
- $\hat{\tau}^2$ is an estimate of the G-E dependence θ^2_{GE}

Mukherjee & Chatterjee, Biometrics 2008

Genome-wide G-E Interaction analysis: 2-step approaches

1. Test for GxE dependence and/or associations between the SNP and your outcome in your entire dataset. Select SNPs with p<α_1

2. Take m SNPs from stage 1 and perform traditional GxE interaction tests in a case-control setting (1 df). All SNPs with p<α/m are declared significant



Murcray et al, Am J Epi 2009, Genet Epi 2011

Table 3. Genome-wide significance of tests for gene-environment interaction for rs11066015 (12q24) and rs3805322 (4q23)

	Genome-wide Significant? ($\alpha=5\times10^{-8}$)	
	ALDH2 rs11066015[a]	*ADH* rs3805322[b]
Standard case-control test	**Yes**	no
Case-only test	No	**Yes**
Empirical Bayes test	**Yes**	no
Hybrid two-step approach	**Yes**	no
Cocktail 1	**Yes**	**Yes**
Cocktail 2	**Yes**	**Yes**

[a] Empirical Bayes estimate of $OR_{G\times E}=3.66$ (2.79,4.80); for the screening stage of the hybrid test, both G-E association and marginal G-D tests were significant with $p_A=6.0\times10^{-14}<\alpha_A$ and $p_M=7.3\times10^{-8}<\alpha_M$, and the standard test of G×E interaction at the second stage was quite significant ($p<10^{-16}$); for the cocktail methods, $p^{screen}=p_M$ for cocktail 1 and $p^{screen}=p_A$ for cocktail 2, both of these pass the first stage threshold, and the second stage tests (the Empirical Bayes test for Cocktail 1 and standard case-control test for Cocktail 2) are both very significant ($p<10^{-16}$).

[b] Empirical Bayes estimate of $OR_{G\times E}=1.70$ (1.36,2.20), $p=5.4\times10^{-5}$; for the screening stage of the hybrid test, both G-E association and marginal G-D tests were significant with $p_A=1.1\times10^{-9}<\alpha_A$ and $p_M=9.3\times10^{-13}<\alpha_M$, however, the standard test of G×E interaction at the second stage did not meet the second stage threshold ($\sim 4.2\times10^{-4}$); for the cocktail methods, $p^{screen}=p_M$ for cocktail 1 and 2, which passes the first stage threshold, and the second stage test (the Empirical Bayes test for both) meets the second stage threshold ($\sim 4.2\times10^{-4}$).

Wu et al, Genet Epi 2014

GxE interaction studies require large sample sizes

- A common approach is to pool data from multiple studies within large international consortia.

- Although this will result in greatly increases sample size, it introduces challenges for harmonizing data across studies. This is often the most difficult and time-consuming part of a multi-study GxE interaction study

Exercise:

Harmonizing E N=25,050

What are the sample sizes for these derived variables?

Cigarettes per day
Packs per day
Former smoker
Ever smoker
Current smoker

(a)

Study (*N*)	Smoking-related questions	Possible responses
Study 1 (2,500)	1. Do you currently smoke cigarettes?	Y/N
	2. If yes, how many cigarettes per day?	###
Study 2 (1,200)	1. Have you smoked more than 100 cigarettes in your lifetime?	Y/N
	2. If yes, do you currently smoke?	Y/N
	3. If yes, how many packs per day do you smoke?	###
Study 3 (8,500)	1. Have you ever smoked?	Y/N
Study 4 (1,250)	1. Do you currently smoke?	Y/N
Study 5 (4,200)	1. Do you smoke?	Y/N
	2. When did you first start smoking regularly?	Past year; 1–5 years ago; >5 years ago
Study 6 (6,600)	1. Have you smoked tobacco in the past month?	Y/N
Study 7 (800)	1. Have you ever smoked regularly?	Y/N
	2. If yes, do you still smoke?	Y/N
	3. If yes, how much do you smoke a day?	1–10 cigarettes, 11–20 cigarettes, 21–30 cigarettes, >30 cigarettes

Bennett SN, et al. Genet Epidemiol. 2011

Practical issues in gene-environment interaction studies

Measurement Errors

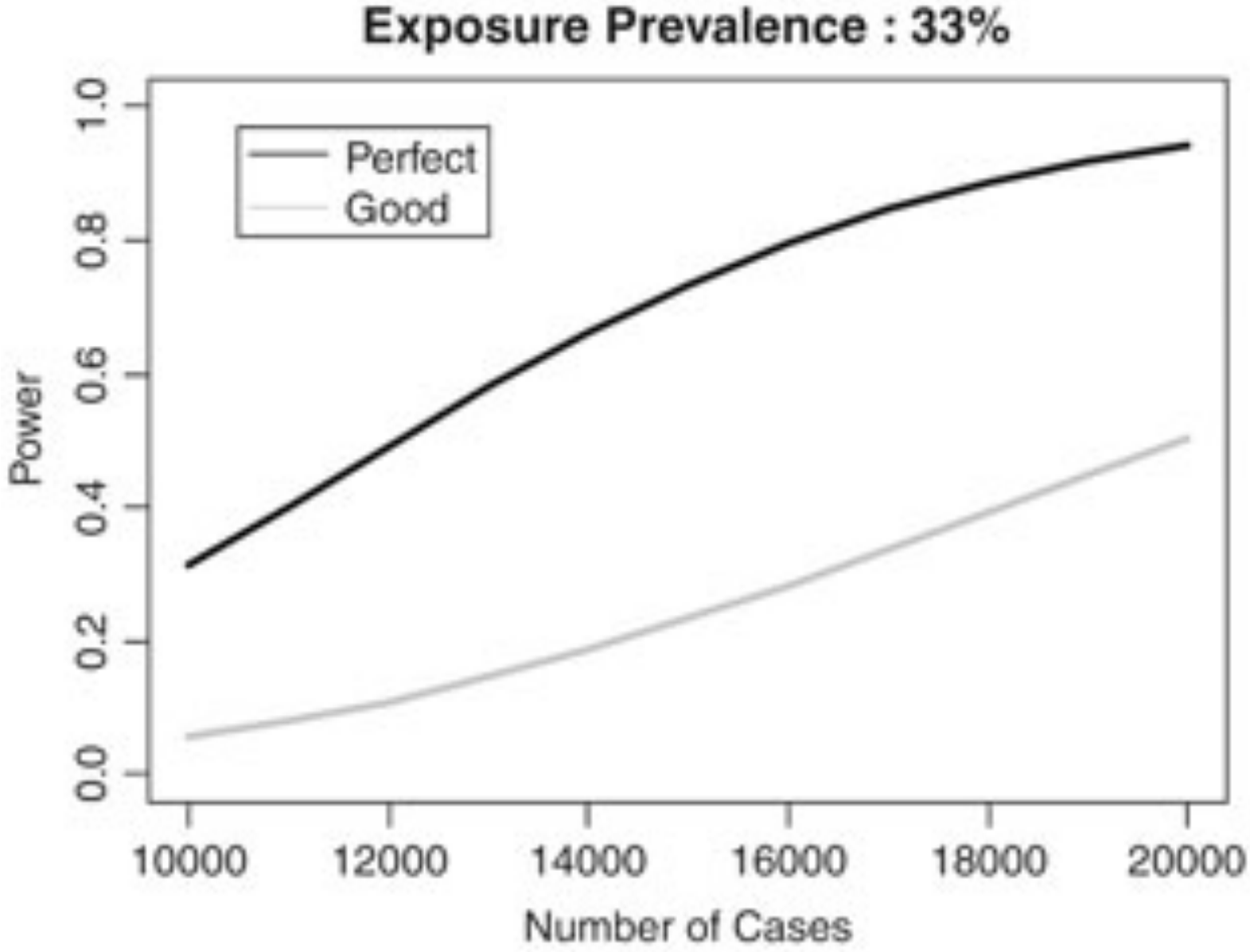
Distribution of E/Replication

Modeling E

Software

Even small measurement errors can greatly decrease power to detect gene-environment interactions

"Good"
Sensitivity=77%
Specificity=99%



Bennett SN, et al. Genet Epidemiol. 2011

How, where, and when you measure the exposure have consequences for gene-environment interaction studies

Example: FTO, Physical Activity and Obesity

- Meta-analysis of 218,166 European-ancestry subjects
- Risk of Obesity (BMI ≥ 30 vs. BMI < 25 kg/m^2) for *FTO* SNP rs9939609

	OR (95% CI)
Inactive	1.30 (1.24-1.36)
Active	1.22 (1.19-1.25)
rs9939609 x physical activity interaction	0.92 (0.88-0.97)
	P-value = 0.0010

Slide courtesy of Leah Mechanic

Kilpelainen, 2011

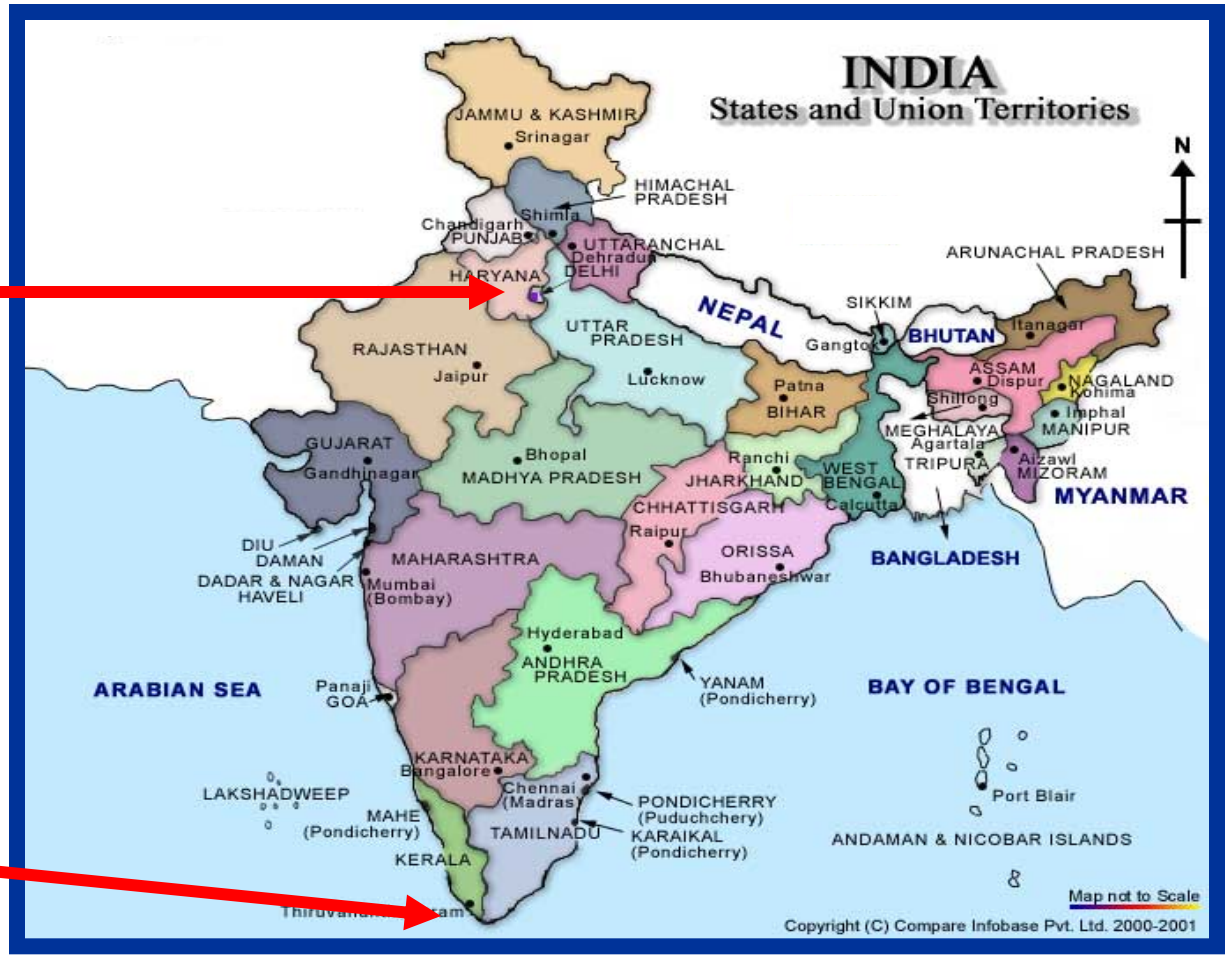
Slides courtesy of N Chatterjee

India health study

Interaction between *FTO*, physical activity and obesity

New Delhi

Trivandrum



Moore, Obesity 2012

Participant characteristics by region

Characteristic	New Delhi	Trivandrum
Total (n=1,313)	n=619	n=694
Age, years (mean, SD)	47.4 ± 10.0	48.8 ± 9.2
Household monthly income, %		
<5,000 rupees	7.1	71.9
>10,000 rupees	76.7	3.1
Household items, %		
Car	25	7
Refrigerator	87	58
Washing machine	79	14
Total physical activity, MET-hr/wk	42.5 ± 43.8	147.3 ± 85.2
Vigorous physical activity, MET-hr/wk	0.6 ± 6.8	26.2 ± 51.4
Sitting, hr/day	10.4 ± 2.0	5.0 ± 2.3
Centrally obese, %	82.1	60.2

Moore, Obesity 2012

Association of *FTO* rs3751812 with waist circumference

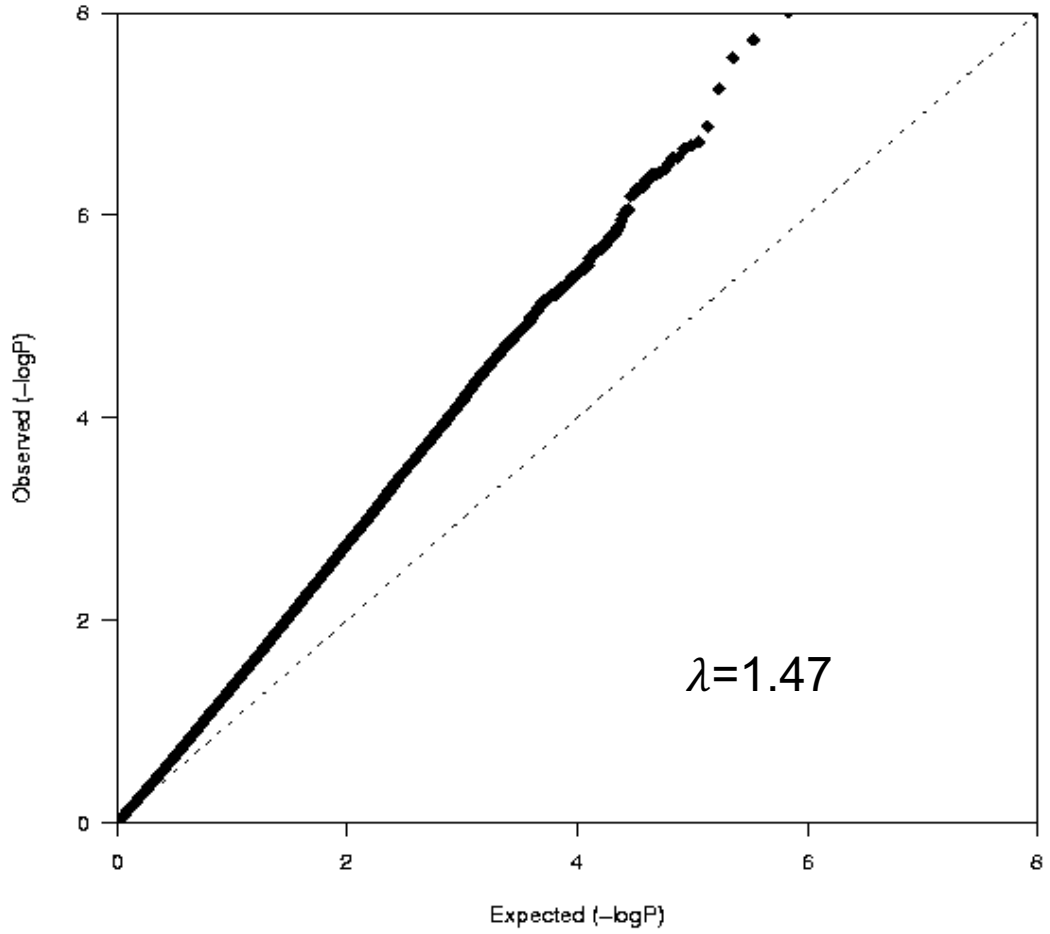
Characteristic	N	**Effect size per T allele (95% CI)**	P_{trend}	**Interaction by PA**
Overall	1,209	**+1.61 cm (0.67, 2.55)**	0.0008	**0.009**
New Delhi				
Overall	578	**+2.53 cm (1.08, 3.97)**	0.0006	**0.59**
By PA				
≤ 91 MET-hrs/wk	517	**+2.36 cm (0.82, 3.89)**	0.003	
92-151 MET-hrs/wk	32	**+6.39 cm (1.94, 10.85)**	0.005	
152-217 MET-hrs/wk	24	**-0.95 cm (-7.33, 5.42)**	0.77	
218+ MET-hrs/wk	5	**N/A**	N/A	
Trivandrum				
Overall	574	**+0.87 cm (-0.35, 2.08)**	0.16	**0.004**
By PA				
≤ 91 MET-hrs/wk	170	**+3.50 cm (0.90, 6.10)**	0.008	
92-151 MET-hrs/wk	132	**+1.13 cm (-1.08, 3.33)**	0.32	
152-217 MET-hrs/wk	141	**+1.04 cm (-1.63, 3.70)**	0.45	
218+ MET-hrs/wk	131	**-2.32 cm (-4.82, 0.18)**	0.07	

Moore, Obesity 2012

A note about modeling “E” (i)

Genome-wide GxE interaction study of BMI and Type II Diabetes

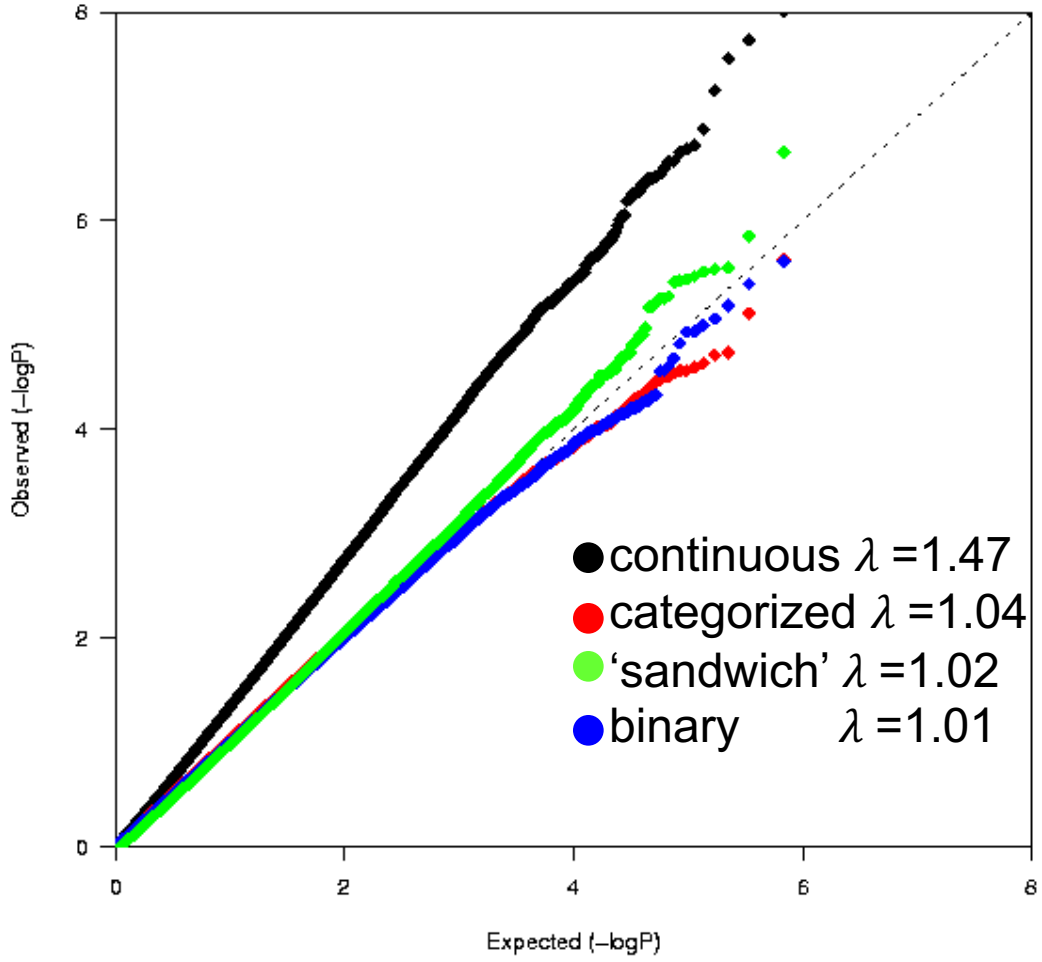
Standard 1 df case-control test for GxE Interaction



Slide courtesy Marilyn Cornelis

Tchetgen Tchetgen and Kraft, Epidemiology, 2011

A note about modeling “E” (ii)



Slide courtesy Marilyn Cornelis

Tchetgen Tchetgen and Kraft, Epidemiology, 2011

Software for analysis

Software	Good for	URL
PLINK	GWAS, data handling, 1df GxE test, joint test	http://pngu.mgh.harvard.edu/~purcell/plink/
GxEscan	R script incorporating multiple genome-wide GxE interaction tests	http://biostats.usc.edu/software
R	Flexible, write your own scripts	http://www.r-project.org/
METAL	Meta-analysis	http://www.sph.umich.edu/csg/abecasis/metal/
CGEN	R package, additive interaction	https://rdrr.io/bioc/CGEN/man/additive.test.html

Software for power calculations

Software	Good for	URL
Quanto	Joint test, GE test, family-based designs, case-control, continuous outcome	http://hydra.usc.edu/gxe/
Power	Additive interaction	http://dceg.cancer.gov/tools/design/power
Gemis	Misclassification in E	http://www.hsph.harvard.edu/peter-kraft/software/