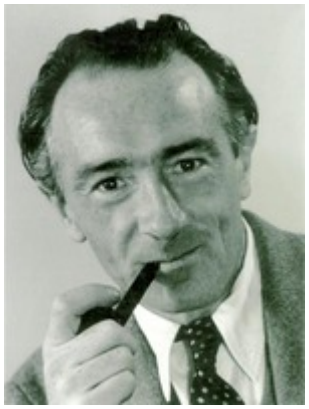
mortality; there in the *Dulag* I expected hundreds to die of diphtheria alone in the absence of specific therapy. In point of fact there were only four deaths, of which three were due to gunshot wounds inflicted by the Germans. This excellent result had, of course, nothing to do with the therapy they received or my clinical skill. It demonstrated, on the other hand, very clearly the relative unimportance of therapy in comparison with the recuperative power of the human body. On one occasion, when I was the only doctor there, I asked the German *Stabsarzt* for more doctors to help me cope with these fantastic problems. He replied: 'Nein! Ärtze sind überflüssig.' ('No! Doctors are superfluous.') I was furious and even wrote a poem about it; later I wondered if he was wise or cruel; he was certainly right.



Archibald Leman Cochrane 12/1/1909 – 18/6/1988

"Effectiveness and Efficiency: Random Reflections on Health Services". The Nuffield Provincial Hospital Trust.

UNIVERSITY OF OTAGO,
CHRISTCHURCH
Whāre Wānanga o Otāgo ki Ōtautahi

Meta analysis software

John F Pearson

Biostatistics and Computational Biology unit
University Otago Christchurch 14/5/2014

Software

Commercial

- Stata *(meta, metan, metareg)*

- SAS proc mixed, proc nlmixed, metadas macro
- SPSS
  (macro from David Wilson, calls R)
- Many specialist programs (CMA …)

Software

Free

- Revman (Cochrane.org)
- R
- OpenMeta-analyst (R, python)
- Many more

Software reviews

A systematic comparison of software dedicated to meta-analysis of causal studies

Leon Bax*[1,2], Ly-Mee Yu[3], Noriaki Ikeda[2] and Karel GM Moons[1]

Meta-analysis software – basic feature comparison

	CMA	MetAnalysis	MetaWin	MIX	RevMan	WEasyMA
General						
URL	*meta-analysis.com*	-	*metawinsoft.com*	*mix-for-meta-analysis.info*	*cc-ims.net/RevMan*	*weasyma.com*
Corporate single user price	~$1295.00	~$75.00	~$150.00	Free	$650	~$490.00
Student single user price	~$395.00	~$75.00	~$75.00	Free	Free	~$280.00
Download/program size	30 Mb	5 Mb	9 Mb	20 Mb/50 Mb	9 Mb	3 Mb
Compatibility	Windows	Windows	Windows	Windows	Windows	Windows
Last update	2006	2005	2002	2006	2005	2002
License	Single user	Single user	Single user	Open	Open	Single user
Input options						
Manual input	✓	✓	✓	✓	✓	✓
Copy & paste	✓		✓	✓	(✓)	

BMC Medical Research Methodology 2007, **7**:40 doi:10.1186/1471-2288-7-40

Example 1

Antidepressant response associated with a polymorphism

Study	RC	RT	NC	NT	RCC	RCT	RTT	NCC	NCT	NTT	Year
H et al. 2010	151	111	194	144	42	67	22	62	70	37	2010
P et al. 2007	874	670	322	322	253	368	151	74	174	74	2007
P et al. 2010	193	153	132	114	56	81	36	34	64	25	2010
Pu et al. 2013	349	59	114	36	147	55	2	42	30	3	2013
S et al. 2012	95	67	133	105	29	37	15	39	55	25	2012

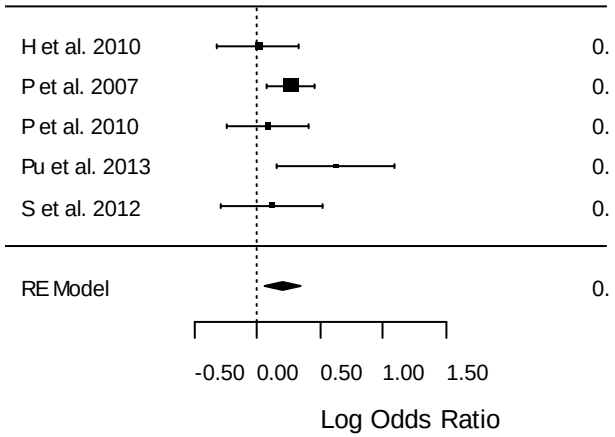
R responders
N non-responders
C/T alleles

Example 1

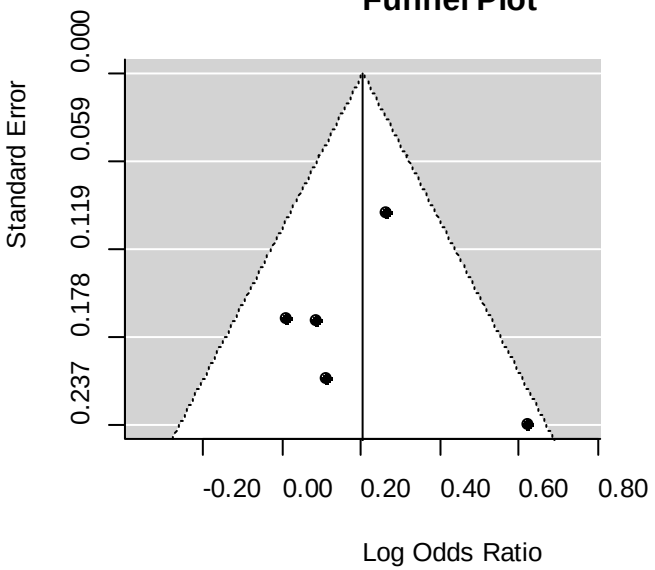
```
> setwd("H:/jpearson/Seminars/MetaAnalysis")
> require(metafor)
Loading required package: metafor
Loading required package: Formula

Loading 'metafor' package (version 1.9-2). For an overview
and introduction to the package please type: help(metafor).
> dat = read.table("ex1.csv",header=T,sep=",",as.is=TRUE)
> dat2 = escalc(measure="OR", ai = RC, bi = NC, ci = RT, di = NT,data=dat)
> res = rma(yi, vi, data=dat2,slab=Study)
> res

Random-Effects Model (k = 5; tau^2 estimator: REML)

tau^2 (estimated amount of total heterogeneity): 0.0032 (SE = 0.0192)
tau (square root of estimated tau^2 value):      0.0567
I^2 (total heterogeneity / total variability):   11.01%
H^2 (total variability / sampling variability):  1.12

Test for Heterogeneity:
Q(df = 4) = 5.6251, p-val = 0.2290

Model Results:

estimate       se     zval     pval    ci.lb    ci.ub
  0.2047   0.0733   2.7910   0.0053   0.0610   0.3485       **

---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

> plot(res)
```

Example 1

Forest Plot



Funnel Plot

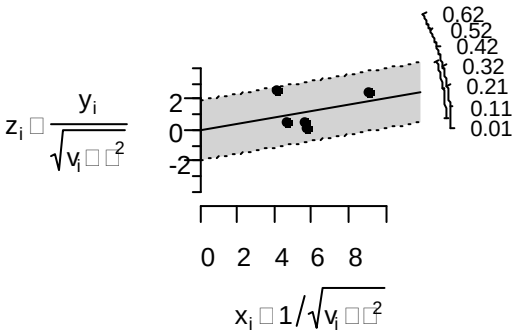


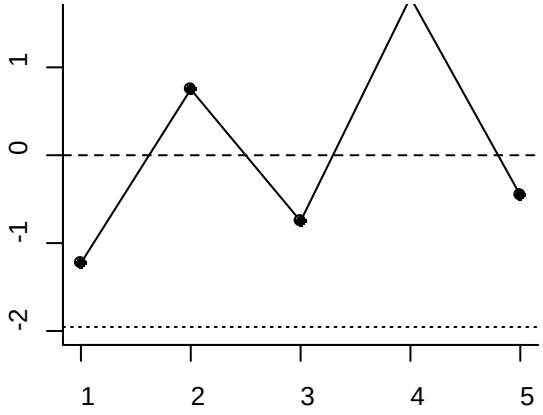
BMJ. 2006 Sep 16;333(7568):597-600.

The case of the misleading funnel plot.

Lau J[1], Ioannidis JP, Terrin N, Schmid CH, Olkin I.

Radial Plot





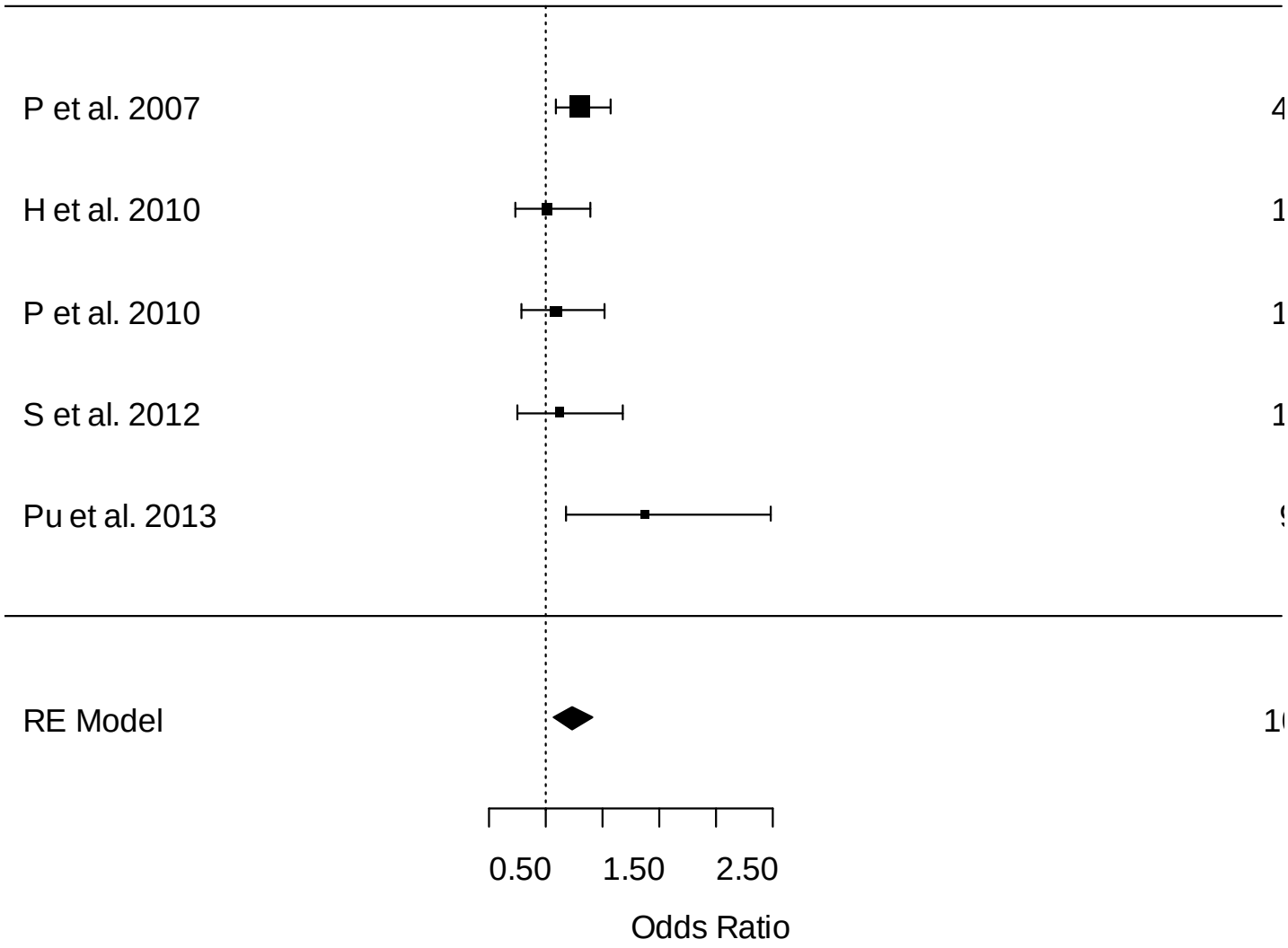
Example 1

```
> ranktest(res)

Rank Correlation Test for Funnel Plot Asymmetry

Kendall's tau = 0.4000, p = 0.4833

> leave1out(res)
                estimate     se   zval   pval   ci.lb  ci.ub      Q     Qp   tau2      I2     H2
H et al. 2010     0.2466 0.0726 3.3982 0.0007  0.1044 0.3888 3.9245 0.2697 0.0000  0.0025 1.0000
P et al. 2007     0.1670 0.1153 1.4482 0.1476 -0.0590 0.3931 4.8901 0.1800 0.0169 31.6901 1.4639
P et al. 2010     0.2280 0.0946 2.4116 0.0159  0.0427 0.4134 4.9837 0.1730 0.0105 28.0561 1.3900
Pu et al. 2013    0.1662 0.0744 2.2334 0.0255  0.0203 0.3120 2.2921 0.5140 0.0021  8.3821 1.0915
S et al. 2012     0.2153 0.0939 2.2941 0.0218  0.0314 0.3993 5.3830 0.1458 0.0118 32.9569 1.4916

> influence(res)
$inf
                rstudent  dffits cook.d  cov.r tau2.del QE.del    hat  weight inf
H et al. 2010    -1.3041 -0.6032 0.3261 0.9788   0.0000 3.9245 0.1739 17.3862
P et al. 2007     0.5001  0.3519 0.2640 2.4725   0.0169 4.8901 0.4461 44.6139
P et al. 2010    -0.6536 -0.2868 0.1013 1.6623   0.0105 4.9837 0.1720 17.2003
Pu et al. 2013    1.8134  0.5304 0.2760 1.0290   0.0021 2.2921 0.0903  9.0297
S et al. 2012    -0.4081 -0.1327 0.0209 1.6374   0.0118 5.3830 0.1177 11.7700

$dfb
                intrcpt
H et al. 2010   -0.6296
P et al. 2007    0.4025
P et al. 2010   -0.2740
Pu et al. 2013   0.5419
S et al. 2012   -0.1218
```

```
> ?influence.rma.uni
```

Example 1

```
> forest(res,trans=exp,refline=1,order=c(2,1,3,5,4),showweight=TRUE)
```

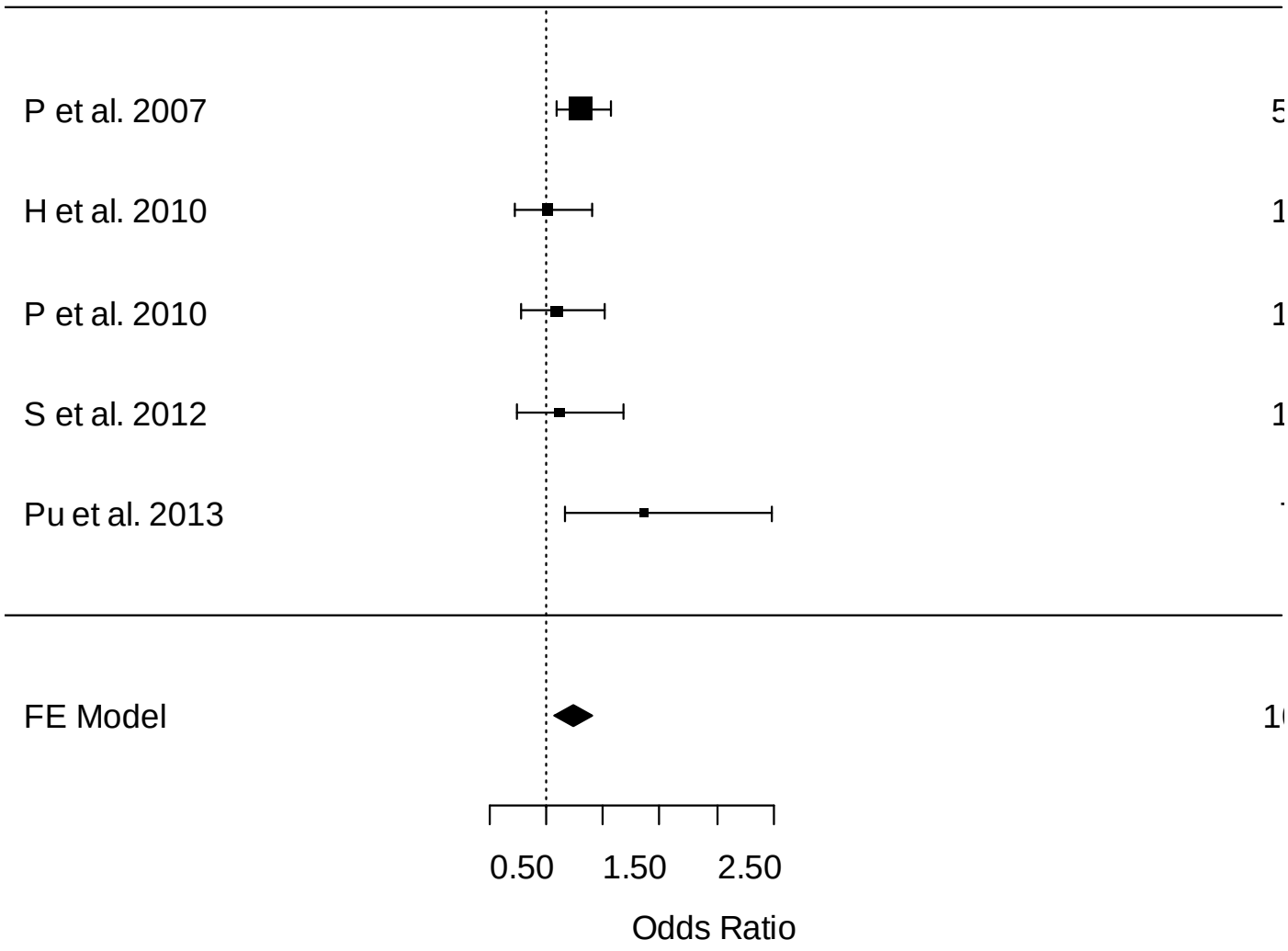


Example 1 (fixed effect model)

```
> res = rma(yi, vi, data=dat2,slab=Study,method="FE")
> res

Fixed-Effects Model (k = 5)

Test for Heterogeneity:
Q(df = 4) = 5.6251, p-val = 0.2290

Model Results:

estimate       se     zval     pval    ci.lb    ci.ub
  0.2088   0.0665   3.1387   0.0017   0.0784   0.3392       **

---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Example 1 (fixed effect model)

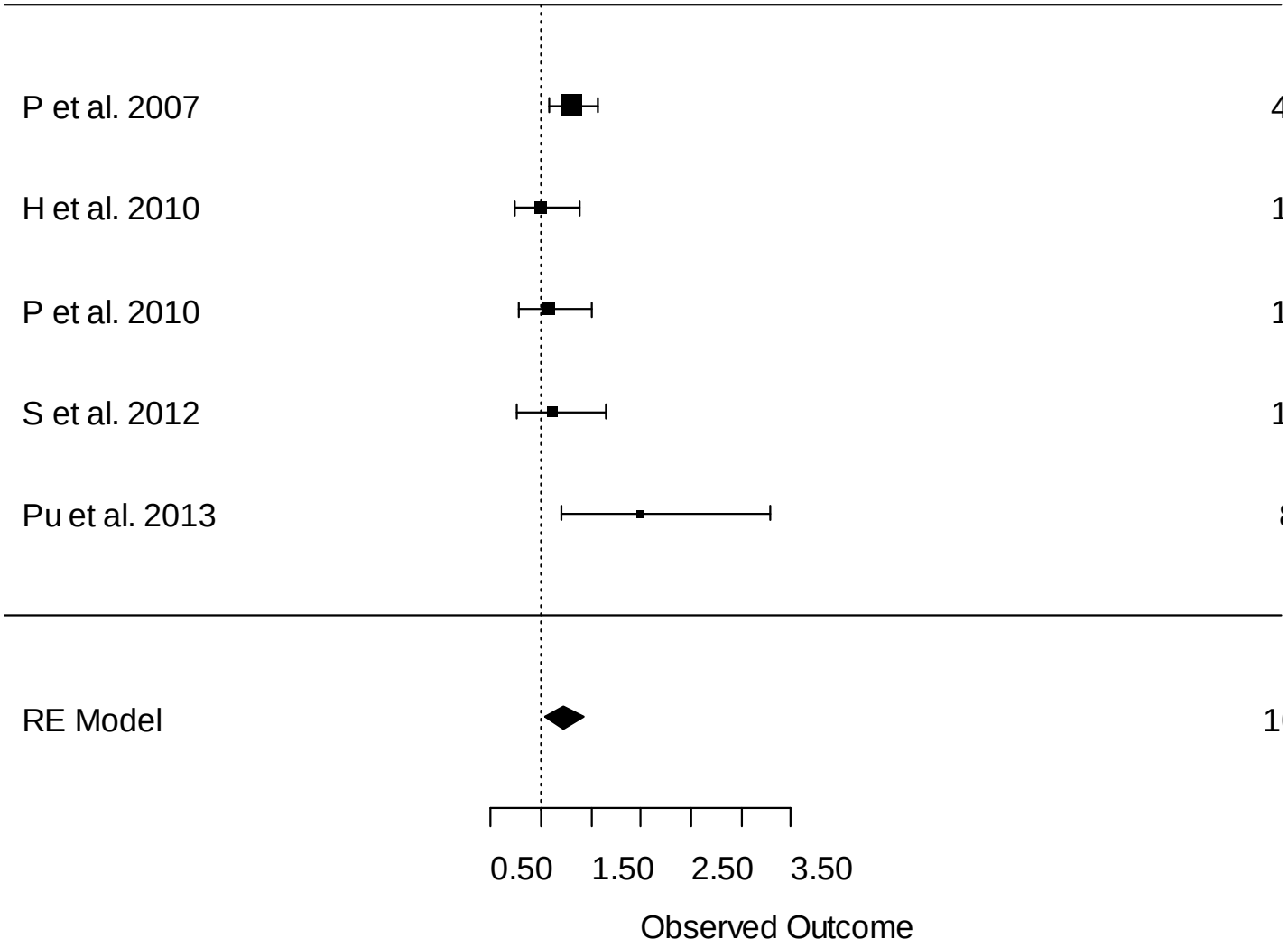
```
> forest(res,trans=exp,refline=1,order=c(2,1,3,5,4),showweight=TRUE)
```



Example 1 (additive inheritance)

```
> # Function to fit additive genetic model
> AMG = function(R,NotR,add = c(2,1,0)){
+   RNR = cbind(as.numeric(R),as.numeric(NotR))                          # Counts of Responders ar
+   am  = glm(RNR~add,family=binomial())                                 # Fit logistic regressior
+   sam = summary(am)                                                    # extract parameters fron
+   return(c(exp(c(OR = coefficients(am)["add"],confint(am)["add",])), # OR with 95% CI
+          p = sam$coefficients["add","Pr(>|z|)"],                       # P value
+  r = coefficients(am)["add"],                                  # effect for meta analysis
+  v = diag(sam$cov.scaled)["add"]                               # variance for meta analysis
+  ))
+ }
> # Fit additive model to each study
> addmodels = NULL
> for(i in 1:5){addmodels = rbind(addmodels,AMG(dat[i,6:8],dat[i,9:11]))}
Waiting for profiling to be done...
Waiting for profiling to be done...
Waiting for profiling to be done...
Waiting for profiling to be done...
Waiting for profiling to be done...
> round(addmodels,2)
     OR.add 2.5 % 97.5 %    p r.add v.add
[1,]   1.01  0.74   1.39 0.95  0.01  0.03
[2,]   1.31  1.09   1.57 0.00  0.27  0.01
[3,]   1.09  0.78   1.51 0.61  0.08  0.03
[4,]   2.00  1.21   3.30 0.01  0.69  0.06
[5,]   1.11  0.75   1.65 0.60  0.11  0.04
> res <- rma(r.add, v.add, data=addmodels,slab=dat$Study)
> forest(res,trans=exp,refline=1,order=c(2,1,3,5,4),showweight=TRUE)
```

Example 1 (additive inheritance)



Example 2 BCG

13 studies examining the effectiveness of the Bacillus Calmette-Guerin (BCG) vaccine for preventing tuberculosis

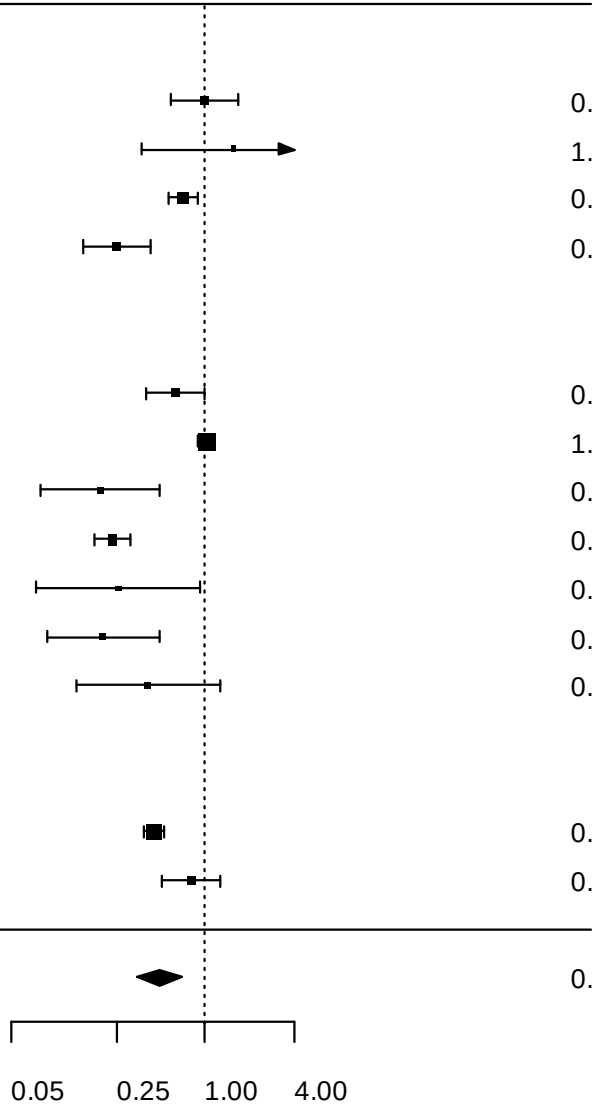
```
> dat.bcg
   trial                author year tpos  tneg cpos  cneg ablat       alloc
1      1             Aronson 1948    4   119   11   128    44      random
2      2      Ferguson & Simes 1949    6   300   29   274    55      random
3      3       Rosenthal et al 1960    3   228   11   209    42      random
4      4     Hart & Sutherland 1977   62 13536  248 12619    52      random
5      5 Frimodt-Moller et al 1973   33  5036   47  5761    13  alternate
6      6       Stein & Aronson 1953  180  1361  372  1079    44  alternate
7      7      Vandiviere et al 1973    8  2537   10   619    19      random
8      8            TPT Madras 1980  505 87886  499 87892    13      random
9      9      Coetzee & Berjak 1968   29  7470   45  7232    27      random
10    10       Rosenthal et al 1961   17  1699   65  1600    42 systematic
11    11         Comstock et al 1974  186 50448  141 27197    18 systematic
12    12    Comstock & Webster 1969    5  2493    3  2338    33 systematic
13    13         Comstock et al 1976   27 16886   29 17825    33 systematic
```

```
> ?dat.bcg
```

Colditz, G. A., Brewer, T. F., Berkey, C. S., Wilson, M. E., Burdick, E., Fineberg, H. V., & Mosteller, F. (1994) Efficacy of BCG vaccine in the prevention of tuberculosis: Meta-analysis of the published literature.*Journal of the American Medical Association*, **271**, 698–702.

BCG

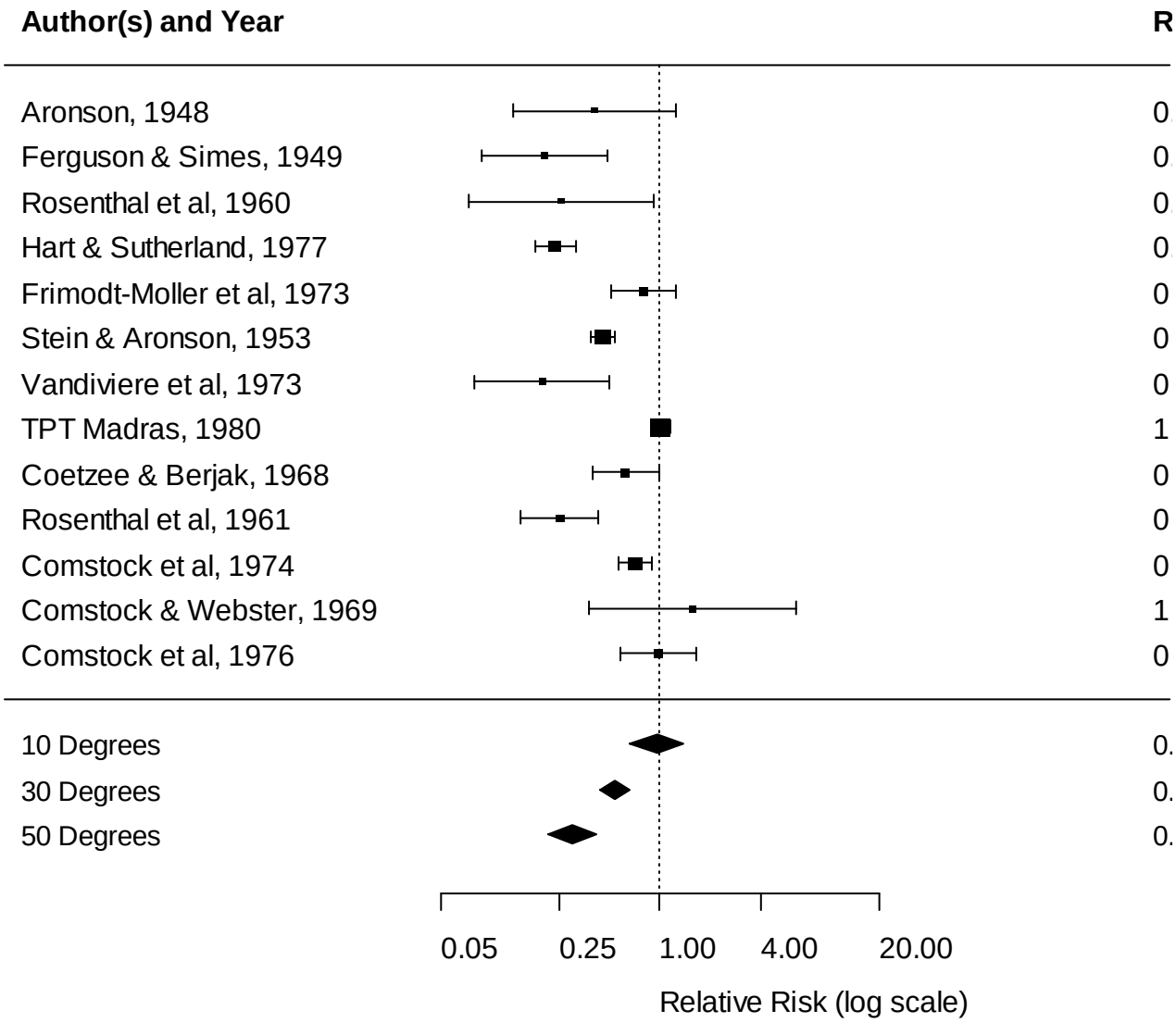
Author(s) and Year	Vaccinated TB+	Vaccinated TB-	Control TB+	Control TB-	R
Systematic Allocation					
Comstock et al, 1976	27	16886	29	17825	0.
Comstock & Webster, 1969	5	2493	3	2338	1.
Comstock et al, 1974	186	50448	141	27197	0.
Rosenthal et al, 1961	17	1699	65	1600	0.
Random Allocation					
Coetzee & Berjak, 1968	29	7470	45	7232	0.
TPT Madras, 1980	505	87886	499	87892	1.
Vandiviere et al, 1973	8	2537	10	619	0.
Hart & Sutherland, 1977	62	13536	248	12619	0.
Rosenthal et al, 1960	3	228	11	209	0.
Ferguson & Simes, 1949	6	300	29	274	0.
Aronson, 1948	4	119	11	128	0.
Alternate Allocation					
Stein & Aronson, 1953	180	1361	372	1079	0.
Frimodt-Moller et al, 1973	33	5036	47	5761	0.
RE Model					0.



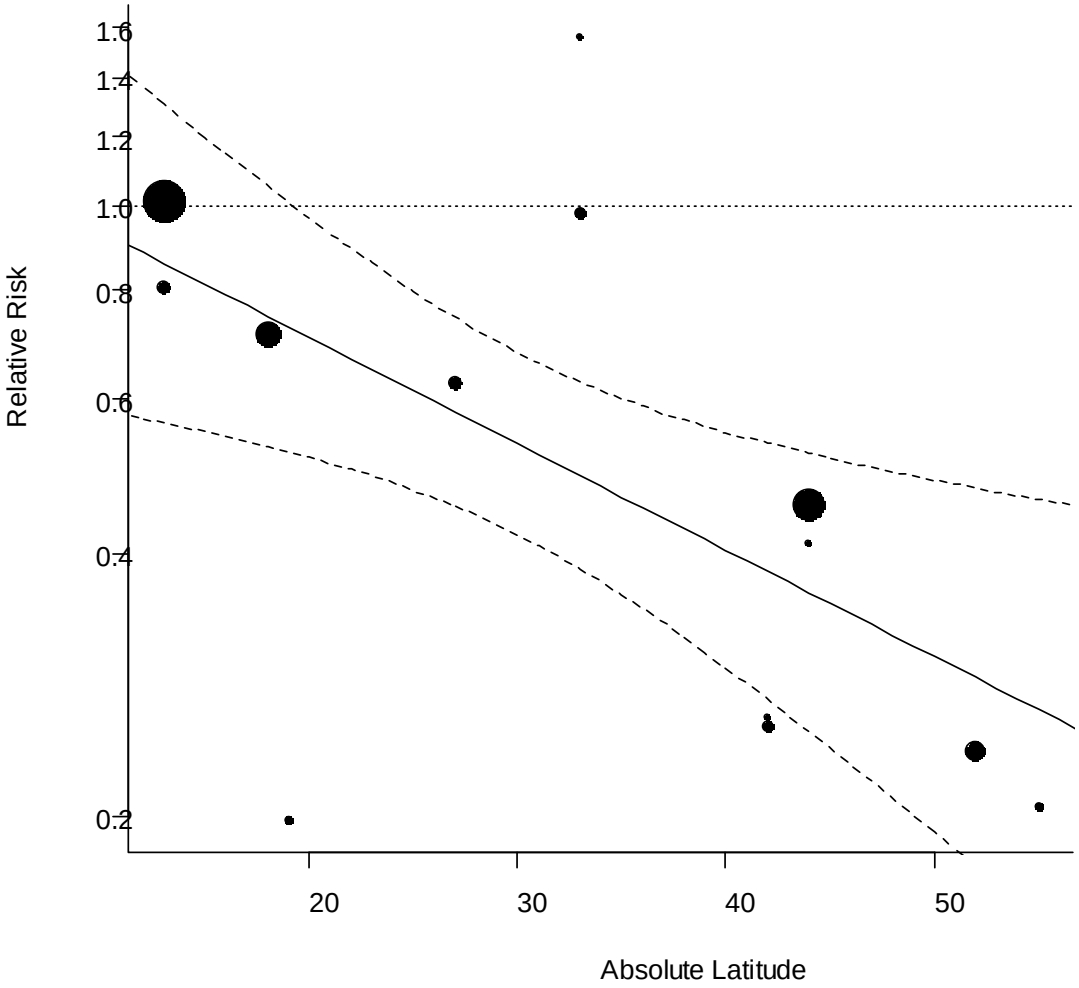

```
> ?forest
```

BCG (meta regression)

```
> res <- rma(yi, vi, mods = ~ ablat + year, data=dat, method="REML")
```



BCG (meta regression)



Baseline risk

- bivariate model

Berkey, C. S., et al.(1995). A random-effects regression model for meta-analysis. *Statistics in Medicine*, **14**, 395–411

- ML vs Baysian (… ML OK (asymptotically unbiased estimates)
- very limited improvement

Arends L et al. Baseline risk as predictor of treatment benefit: three clinical meta-re-analyses. Statistics in Medicine 2000; 19(24):3497– 3518.

BCG

Pediatrics. 2007 Nov;120(5):e1269-77.

Is childhood vaccination associated with asthma? A meta-analysis of observational studies.

Balicer RD[1], Grotto I, Mimouni M, Mimouni D.

	OR	95% CI
Mommers et al[15]	1.17	0.63–2.19
Marks et al[29]	1.06	0.93–1.23
Alm et al[21]	0.83	0.48–1.43
Grüber et al[19]	0.85	0.71–1.00
Grüber et al[22]	1.13	0.71–1.80
Total	0.98	0.88–1.08

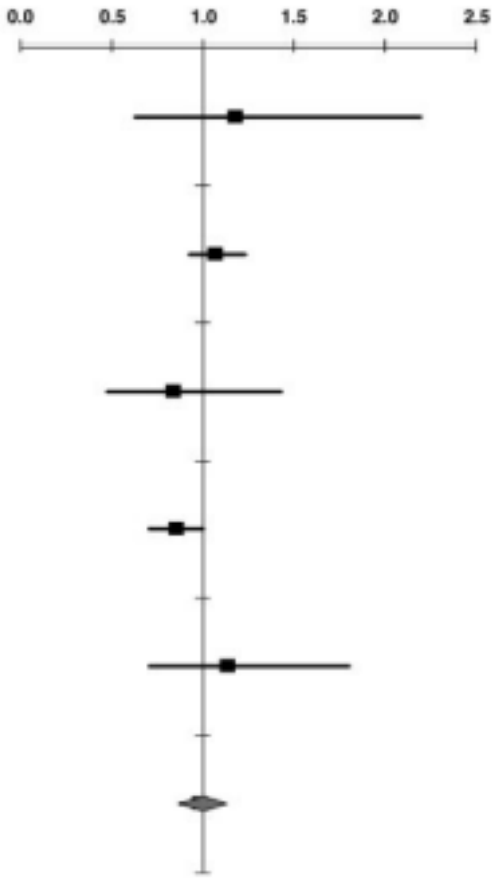
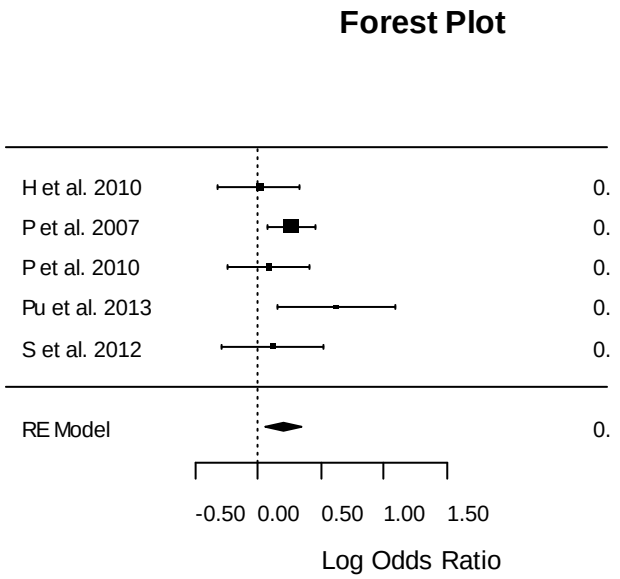


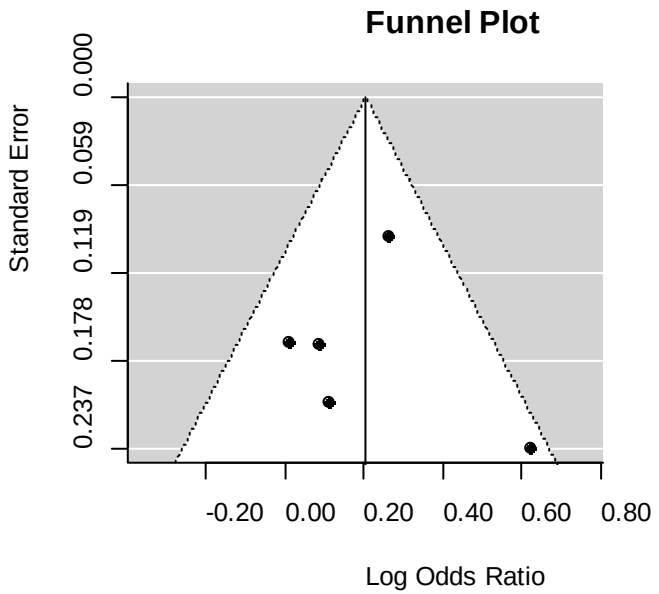
FIGURE 1
ORs for the association of asthma with BCG vaccination in 5 studies. The horizontal bars represent the 95% CIs.

BMJ. 2006 Sep 16;333(7568):597-600.

The case of the misleading funnel plot.

Lau J[1], Ioannidis JP, Terrin N, Schmid CH, Olkin I.





Bias in meta-analysis detected by a simple, graphical test

Matthias Egger; George Davey Smith; Martin Schneider; Christoph Minder

British Medical Journal; Sep 13, 1997; 315, 7109; Academic Research Library
pg. 629

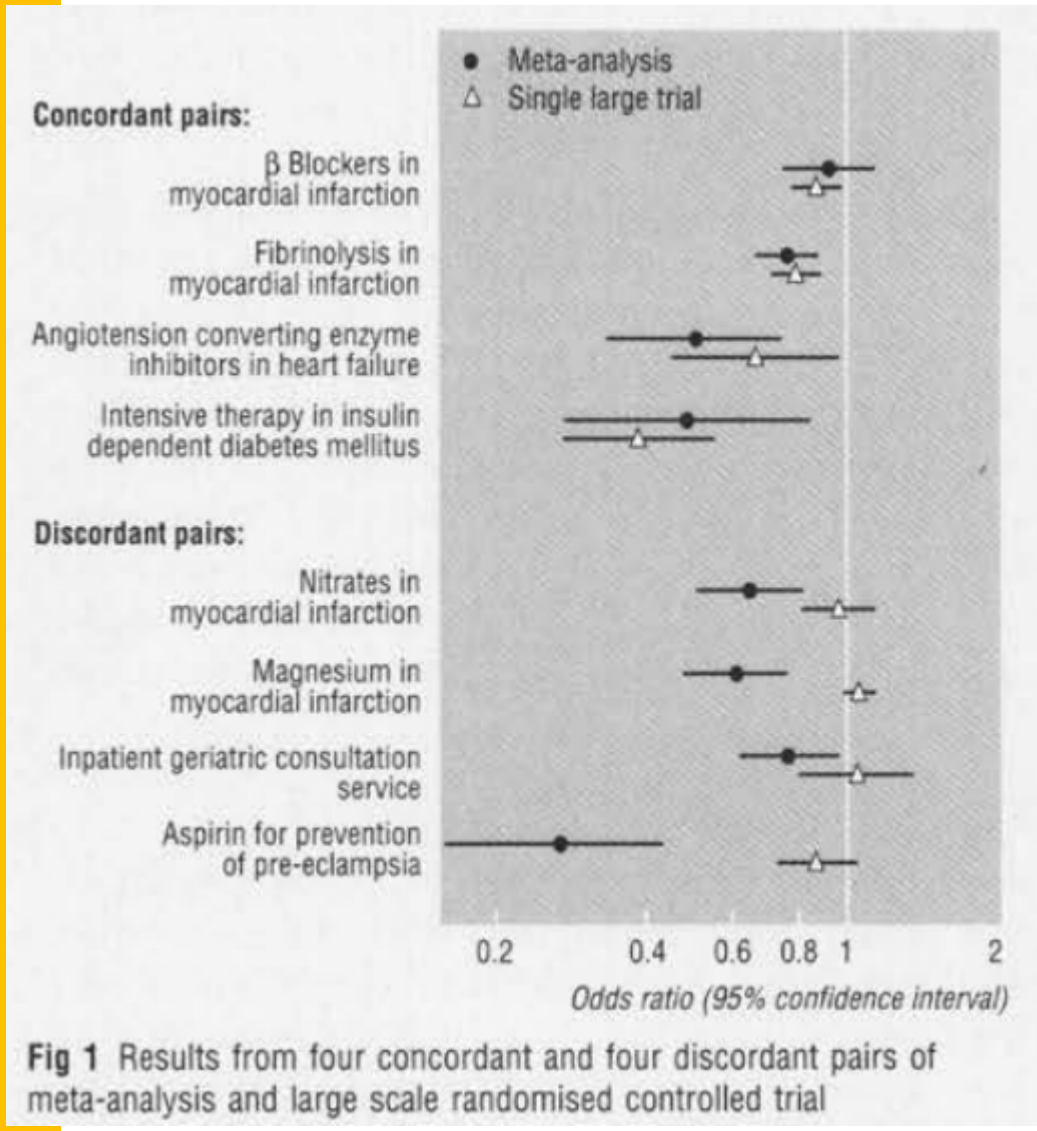
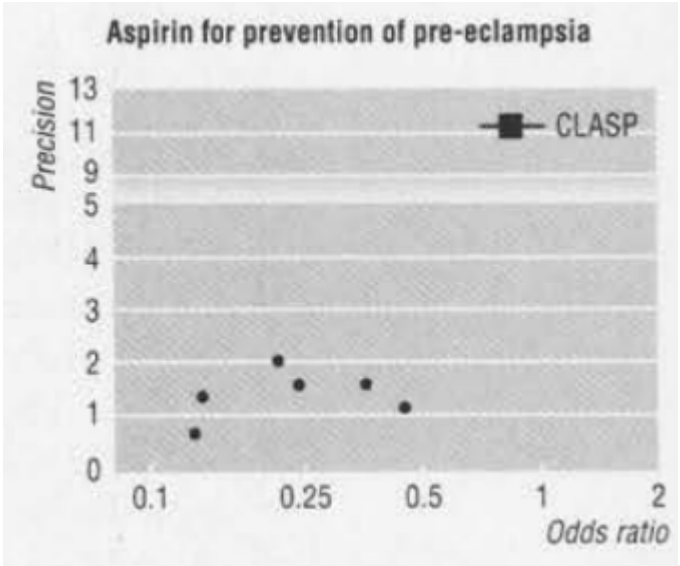
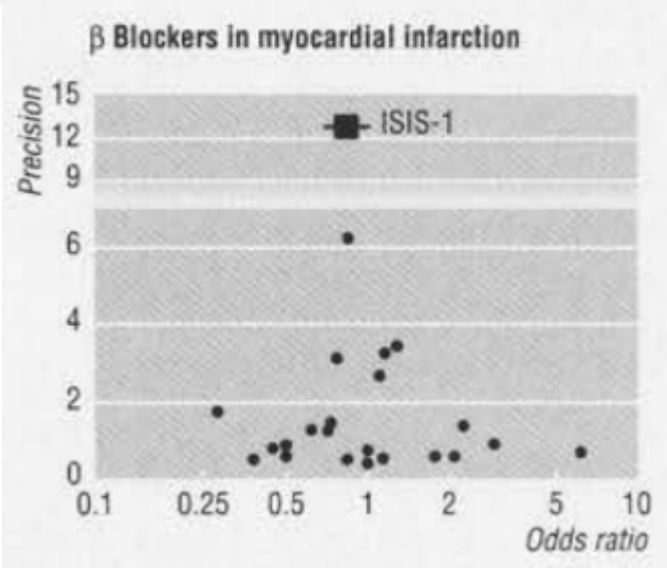


Fig 1 Results from four concordant and four discordant pairs of meta-analysis and large scale randomised controlled trial





BMJ. 2006 Sep 16;333(7568):597-600.

The case of the misleading funnel plot.

Lau J[1], Ioannidis JP, Terrin N, Schmid CH, Olkin I.

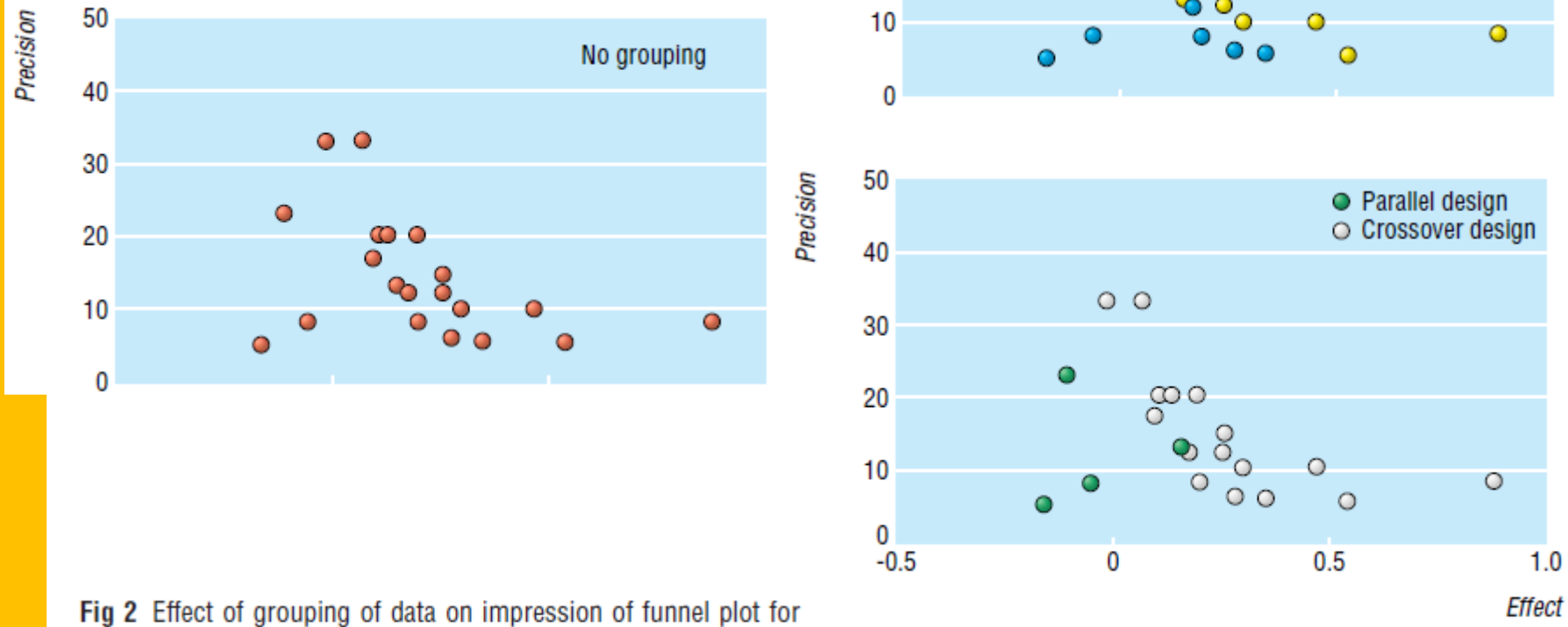
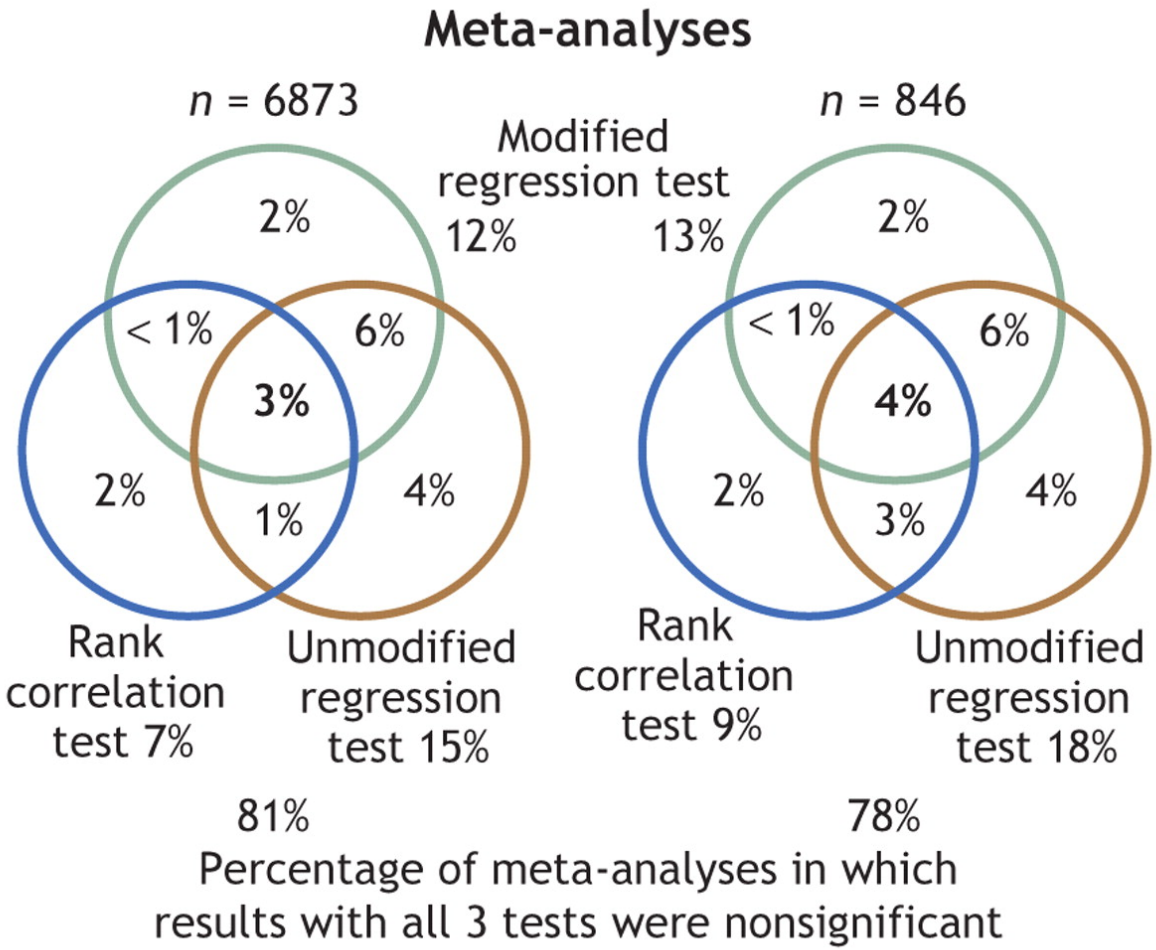


Fig 2 Effect of grouping of data on impression of funnel plot for meta-analysis of inhaled disodium cromoglicate that found significant heterogeneity[24]

Fig. 2: Venn diagrams disclosing modest concordance in the application of the 3 funnel-plot asymmetry tests to statistically significant results in the wider data set of 6873 meta-analyses (left) and in the restricted data set of 846 meta-analyses (right).



Ioannidis J P , and Trikalinos T A CMAJ 2007;176:1091-1096

GWAS (…and NGS)

Review

Journal of Human Genetics (2009) **54,** 615–623; doi:10.1038/jhg.2009.95; published online 23 October 2009

Meta-analysis of genetic association studies: methodologies, between-study heterogeneity and winner's curse

Hirofumi Nakaoka[1,2] and Ituro Inoue[1]

Specific issues
(i) assessment of Hardy–Weinberg equilibrium (HWE) and
(ii) definition of genetic models.

85% of genetic variation is accounted for by within-population inter-individual differences, not by differences between groups

'racial' differences in genetic risks may be spurious interpretations of the data. Small sample size, study design flaws or other biases[13, 14, 20] may be more common reasons than true 'racial' heterogeneity for the observed discrepancies between studies addressing genetic risks.

John P A Ioannidis *Nature Genetics* - **36**, 1312 - 1318 (2004)

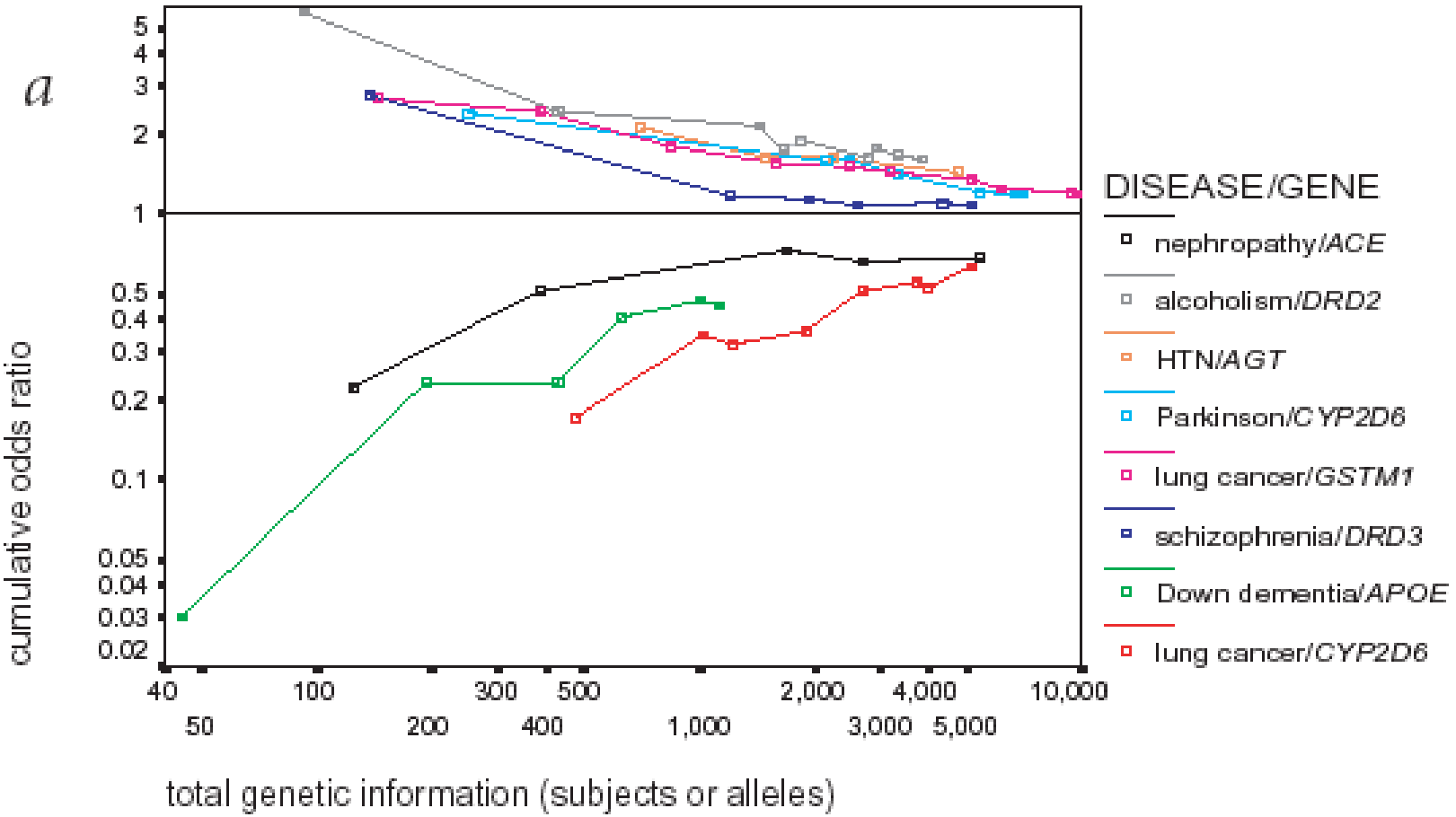


Figure 2. Evolution of the strength of an association as more information is accumulated.
The strength of the association is shown as an estimate of the odds ratio (OR) without confidence intervals. ***a***, Eight topics in which the results of the first study or studies differed beyond chance ($P<0.05$) when compared with the results of the subsequent studies. ***b***, Eight topics in which the first study or studies did not claim formal statistical significance for the genetic association but formal significance was reached by the end of the meta-analysis. Each trajectory starts at the OR of the first

Nat Genet. 2001 Nov;29(3):306-9.

Replication validity of genetic association studies.

Ioannidis JP[1], Ntzani EE, Trikalinos TA, Contopoulos-Ioannidis DG.

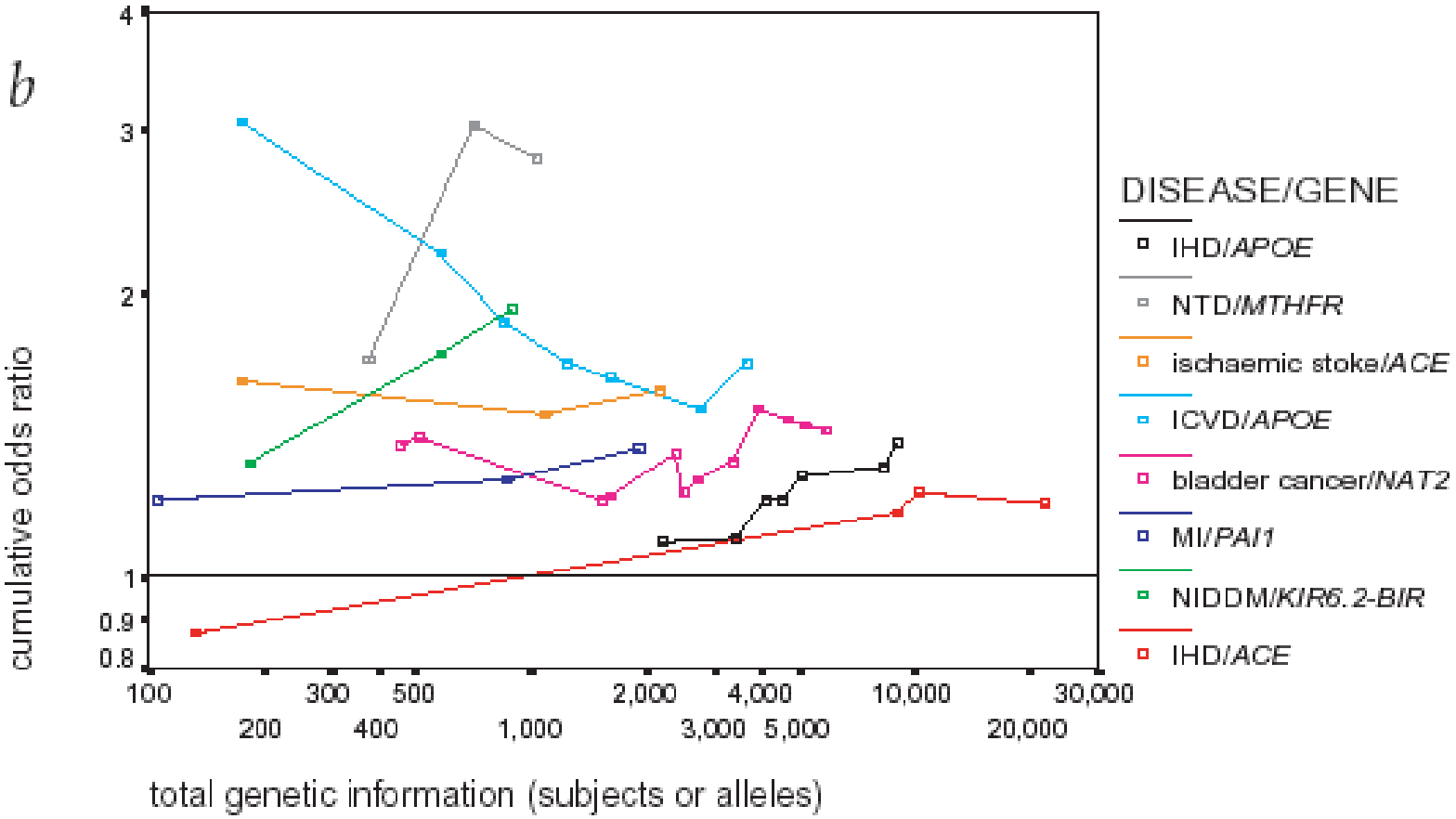


Figure 2. Evolution of the strength of an association as more information is accumulated.
The strength of the association is shown as an estimate of the odds ratio (OR) without confidence intervals. ***a***, Eight topics in which the results of the first study or studies differed beyond chance ($P<0.05$) when compared with the results of the subsequent studies. ***b***, Eight topics in which the first study or studies did not claim formal statistical significance for the genetic association but formal significance was reached by the end of the meta-analysis. Each trajectory starts at the OR of the first

Nat Genet. 2001 Nov;29(3):306-9.

Replication validity of genetic association studies.

Ioannidis JP[1], Ntzani EE, Trikalinos TA, Contopoulos-Ioannidis DG.

GWAS (…and NGS)

Review

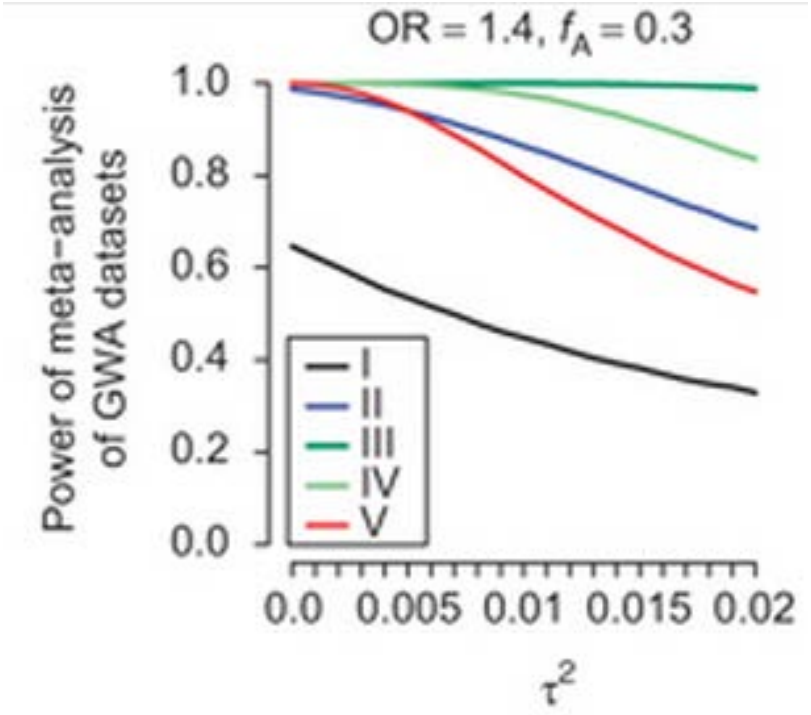
Journal of Human Genetics (2009) **54,** 615–623; doi:10.1038/jhg.2009.95; published online 23 October 2009

Meta-analysis of genetic association studies: methodologies, between-study heterogeneity and winner's curse

Hirofumi Nakaoka[1,2] and Ituro Inoue[1]

Simulations of 5 meta analyses

Scenario	k	n_{case} / $n_{control}$
I	5	500/500
II	10	500/500
III	20	500/500
IV	10	1000/1000
V	5	2000/2000



Pharmacogenomics February 2009, Vol. 10, No. 2, Pages

Meta-analysis in genome-wide association studies

Eleftheria Zeggini[1,2] & John PA Ioannidis[3,4,5†]

T2D

Stage	Description	Number of samples	Number of evaluated SNPs	Main findings
1	Three-way GWA scan meta-analysis	10,128	2,202,892	Clear deviation from the null hypothesis of no association
2	Follow-up of interesting signals in independent samples	22,426	69	Excess of T2D-associated loci in the same direction as stage 1
3	Further follow-up of promising signals in independent samples	57,366	11	Six novel loci surpassing specified genome-wide significance levels

GWA: Genome-wide association; T2D: Type 2 diabetes.

Pharmacogenomics February 2009, Vol. 10, No. 2, Pages

Meta-analysis in genome-wide association studies

Eleftheria Zeggini[1,2] & John PA Ioannidis[3,4,5†]

T2D

Representative SNP	Chromosome	Position	Region/gene	Identification
rs10010131	4	6343816	*WFS1*	Non-GWA
rs1801282	3	12368125	*PPARG*	Non-GWA
rs757210	17	33170628	*TCF2*	Non-GWA
rs5219	11	17365206	*KCNJ11*	Non-GWA
rs7901695	10	114744078	*TCF7L2*	Non-GWA
rs10811661	9	22124094	*CDKN2A/B*	GWA
rs10946398	6	20769013	*CDKAL1*	GWA
rs13266634	8	118253964	*SLC30A8*	GWA
rs4402960	3	186994389	*IGF2BP2*	GWA
rs5015480	10	94455539	*HHEX/IDE*	GWA
rs8050136	16	52373776	*FTO*[*]	GWA
rs2237895	11	2813770	*KCNQ1*	GWA
rs10923931	1	120230001	*NOTCH2*	GWA meta-analysis
rs12779790	10	12368016	*CDC123/CAMK1D*	GWA meta-analysis
rs4607103	3	64686944	*ADAMTS9*	GWA meta-analysis
rs7578597	2	43644474	*THADA*	GWA meta-analysis
rs7961581	12	69949369	*TSPAN8/LGR5*	GWA meta-analysis
rs864745	7	27953796	*JAZF1*	GWA meta-analysis

[*] *Reflecting an association with BMI and obesity risk rather than Type 2 diabetes per se. GWA: Genome-wide association.*

GWAS (…and NGS)

- Meta-analysis of genome-wide association studies has contributed to the discovery of most of the recently identified genetic risk factors for complex diseases.
- Common meta-analytical approaches have been successfully applied; however, novel methods have been …
- Heterogeneity in meta-analysis can be introduced from various sources and should not be disregarded. Several methods have …
- … Tools other than *P* values may be useful for inference.

Take homes

- Meta analysis available on many platforms.
- Don’t over analyse small (<10) studies
- Encouraging use of ES as well as significance
- Winner’s curse: small p ~ big IF