ELSEVIER

Journal of Clinical Epidemiology 61 (2008) 991–996

Journal of Clinical Epidemiology

Contour-enhanced meta-analysis funnel plots help distinguish publication bias from other causes of asymmetry

Jaime L. Peters[a,*], Alex J. Sutton[a], David R. Jones[a], Keith R. Abrams[a], Lesley Rushton[b]

[a]*Department of Health Sciences, University of Leicester, UK*
[b]*Department of Epidemiology and Public Health, Imperial College London*

Accepted 19 November 2007

Abstract

Objectives: To present the contour-enhanced funnel plot as an aid to differentiating asymmetry due to publication bias from that due to other factors.

Study Design and Setting: An enhancement to the usual funnel plot is proposed that allows the statistical significance of study estimates to be considered. Contour lines indicating conventional milestones in levels of statistical significance (e.g., <0.01, <0.05, <0.1) are added to funnel plots.

Results: This contour overlay aids the interpretation of the funnel plot. For example, if studies appear to be missing in areas of statistical nonsignificance, then this adds credence to the possibility that the asymmetry is due to publication bias. Conversely, if the supposed missing studies are in areas of higher statistical significance, this would suggest the cause of the asymmetry may be more likely to be due to factors other than publication bias, such as variable study quality.

Conclusions: We believe this enhancement to funnel plots (i) is simple to implement, (ii) is widely applicable, (iii) greatly improves interpretability, and (iv) should be used routinely.

Keywords: Publication bias; Funnel plots; Statistical significance; Contours; Meta-analysis; Asymmetry

1. Introduction

Publication bias describes the tendency for studies reporting uninteresting or unfavorable results to be less likely to be published [1]. Meta-analysis of published papers is likely to be affected by publication bias [2]. Although the precise mechanisms of publication bias are unknown, evidence suggests that statistical significance of the main outcome in a study is the most important factor; nonsignificant studies are less likely to be published [3–5]. Absence of statistical significance has also been identified as a factor in a related bias, outcome reporting bias: where multiple outcomes are investigated in a study, but only the outcomes with interesting or statistically significant results are reported in the paper [6–8].

The funnel plot is the simplest of all techniques to help assess possible publication bias. The effect estimate from each study in the meta-analysis is plotted against some measure of precision from that study [1]. Estimates of effect from smaller studies are more variable than those from the larger studies and so scatter more widely at the base of the plot creating, in the absence of bias, a symmetrical funnel shape. If smaller, nonstatistically significant studies tend to remain unpublished then an asymmetrical shape may be observed [9].

However, publication bias is not the only possible cause of asymmetry observed in a funnel plot [10]. Any factor which is associated with both study effect and study size could confound the true association and cause an asymmetric funnel. For example, if there is an indication of poorer study design in smaller studies [10] and this poor study design leads to systematic exaggeration of effect [11], this could manifest itself as asymmetry on a funnel plot because studies near the top of the plot (small bias) will have smaller effect sizes, on average, than those at the bottom (large bias). We introduce a graphical aid for the interpretation of funnel plots to help differentiate asymmetry caused by statistical significance related publication bias from that caused by other factors.

2. Funnel plots and contour-enhanced funnel plots

Contour-enhanced funnel plots display areas of statistical significance on a funnel plot. If it is assumed that the

* Corresponding author. Current address: School of Mathematical Sciences, Queensland University of Technology, Gardens Point Campus, Brisbane, QLD 4000, Australia. Tel.: +61-731-38-1756; fax: +61-731-38-2310.

E-mail address: j3.peters@qut.edu.au (J.L. Peters).

0895-4356/08/$ – see front matter
doi: 10.1016/j.jclinepi.2007.11.010

treatment effect in each study is normally distributed, then the significance of any effect size can be calculated from the effect size and the standard error. Because *effect size* and *standard error* (or some functions of it) are the two axes of a funnel plot, the (two-sided) statistical significance of any point on a funnel plot can be calculated. Thus, contours representing conventional ''milestone'' levels of statistical significance (e.g., <0.01, <0.05, <0.1) can be defined and regions associated with these significance levels plotted.

In a review of 48 meta-analyses from the Cochrane Library [12], Sutton et al. [13] assessed evidence of publication bias using the trim and fill method [14,15]. Trim and fill is an iterative nonparametric method used to investigate the number of ''missing'' studies in a meta-analysis due to funnel plot asymmetry and offers an ''adjusted'' pooled estimate as a sensitivity analysis: its ability to detect publication bias has been found to be variable [14,16,17]. The funnel plots of these 48 meta-analyses are available on the BMJ web site. Assessment of publication bias using standard funnel plots and the trim-and-fill method makes no explicit consideration of the levels of statistical significance of the observed study estimates or the significance levels of regions in which studies are suspected to be missing in a funnel plot. In Fig. 1, the contour-enhanced funnel plots of the 48 meta-analyses used in Sutton et al. [13] are presented (using the same identifying number for each meta-analysis as in previous articles [13,18]).

Because assessment of Fig. 1 allows consideration of the statistical significance of the studies in the 48 meta-analyses, this in turn aids interpretation of any observed asymmetry within a meta-analysis. (Note that for 12 meta-analyses in Fig. 1 [plots 7, 12, 15, 17, 23, 32–36, and 42] the ''desirable'' results [e.g., that a treatment is better than control] is on the right-hand side of the funnel plot. For the remaining meta-analyses the ''desirable'' result is on the left-hand side of the funnel plot.) More specifically, consider meta-analysis 11, which investigates the effectiveness of injected cholera vaccines against placebo in terms of the number of cases of cholera up to seven months after administration [19]. The usual funnel plot of the 18 trials (Fig. 2a) suggests asymmetry, but more interestingly, assessment of the contour-enhanced funnel plot (Fig. 2b) indicates that the areas where studies seem to be ''missing'' (as indicated by the ellipse) are areas where nonsignificant studies would be plotted (i.e., the area without shading). This adds further credence to the possibility that the asymmetry observed in Fig. 2a,b is caused by publication bias based on statistical significance.

If, however, the ''missing'' studies were in areas of statistical significance (those with darker shading), this would lead one to suggest that the observed asymmetry is more likely to be due to factors other than publication bias based on statistical significance (e.g., variable study quality or even nonstatistical significance based publication bias mechanisms). Such an example is apparent with meta-analysis 38, which investigates the acceptability of chlorpromazine for treatment of schizophrenia as measured by the length of stay in the study for patients on chlorpromazine compared to placebo in 18 trials [20]. In Fig. 3a, the usual funnel plot is shown for meta-analysis 38 suggesting evidence of asymmetry. However, the contour-enhanced funnel plot (Fig. 3b) shows that the ''missing'' studies are expected to lie in areas of high statistical significance (whereas the majority of available studies are nonsignificant), indicating that the observed asymmetry may not be due to publication bias based on statistical significance. Had the contour-enhanced funnel plot not been considered, it is likely that publication bias would have been suspected. Instead, other factors need to be considered as possible explanations unmeasured or unknown confounding factor.

Visual assessment of these 48 contour-enhanced funnel plots has highlighted a further interesting feature in some of these meta-analyses—a ''tunnel'' effect, where the majority of studies in the meta-analysis appear to have statistically significant effect sizes in either direction, whereas few studies exist in the nonsignificant central core of the funnel. This is inconsistent with the expected distribution of study estimates. A potential example of this can be seen with meta-analysis 41 in Fig. 1. Here, a number of the studies in the meta-analysis lie on the margins of the regions of conventional statistical significance (5% level) on both sides of the funnel plot, indicating that several studies may be missing from the central core. This ''tunnel'' effect suggests that the nonsignificant studies are suppressed, regardless of their direction, whereas statistically significant studies, whether they indicate harms or benefits are being published and incorporated in systematic reviews. The ''tunnel'' effect could also arise, if, in a study, an outcome, which is marginally statistically significant, has been selected from several possible outcomes or analysis methods, or if data are ''manipulated'' until a significant result is obtained. A meta-analysis with a ''tunnel'' effect is unlikely to lead to a very biased pooled estimate because studies are approximately symmetrical in the funnel plot, but estimation of between-study heterogeneity will be affected. Thus, one should be cautious about interpretations of the variability of a pooled effect from a meta-analysis displaying this effect because of its impact on the between-study heterogeneity estimate. Jackson has discussed further the effect of publication bias on estimation of the between-study heterogeneity parameter [21]. However, this ''tunnel'' effect could just be due to sampling error, so one must be cautious about overinterpreting this effect.

Pseudo 95% confidence interval (CI) guidelines have also been suggested as an enhancement to the funnel plot [24]. These pseudo 95% CI guidelines illustrate the expected 95% CI around the fixed effects pooled estimate from the meta-analysis for different standard errors (i.e., pooled estimate $\pm$ 1.96 $\times$ standard error). A clear distinction should be made between the pseudo 95% CIs and the contours of statistical significance presented in this paper.

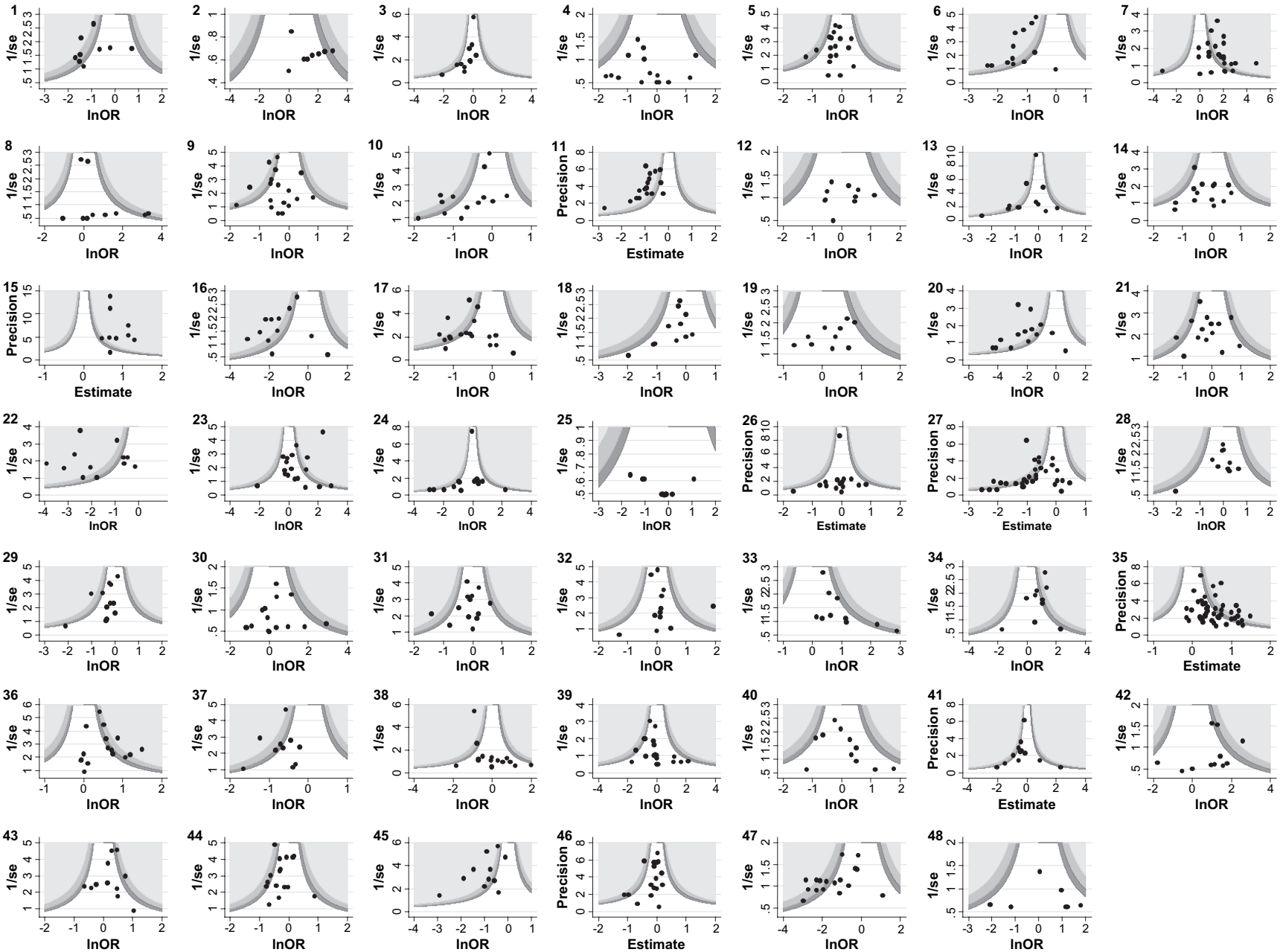


Fig. 1. Contour-enhanced funnel plots of 48 meta-analyses from the Cochrane Library.

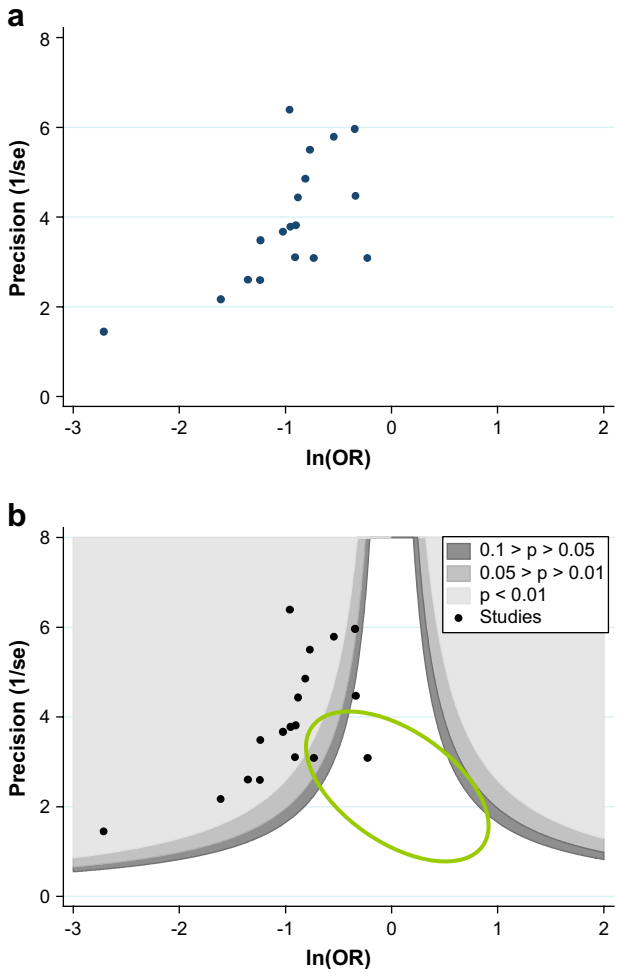


Fig. 2. (a) Funnel plot of meta-analysis 11(A negative ln(OR) corresponds to favoring vaccine as opposed to placebo.). (b) Contour-funnel plot of meta-analysis 11 (the ellipse indicates likely areas where "missing" studies are expected).

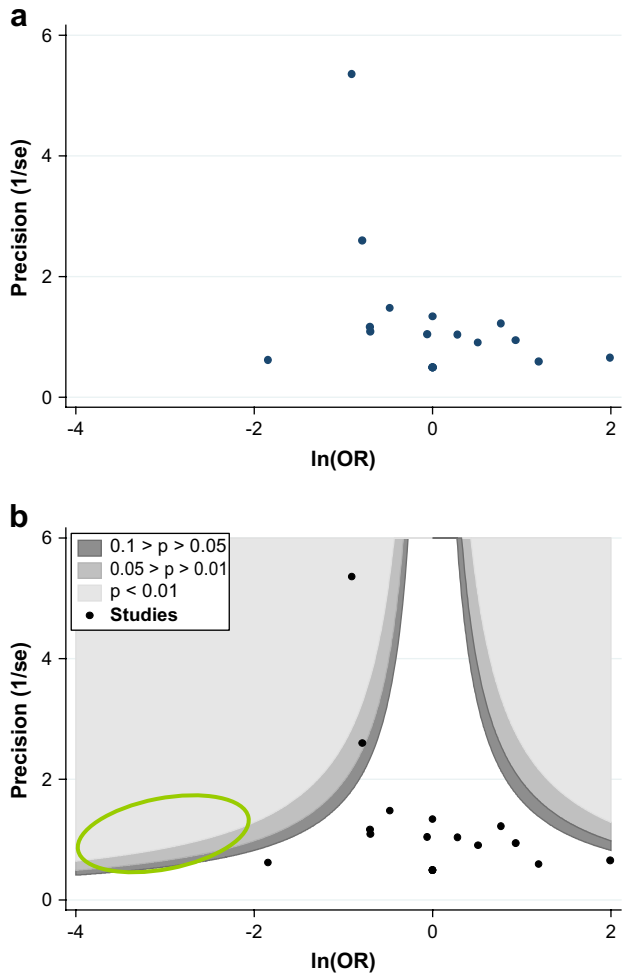


Fig. 3. (a) Funnel plot of meta-analysis 38 (A negative ln(OR) corresponds to favoring chlorpromazine as opposed to placebo.). (b) Contour-funnel plot of meta-analysis 38 (the ellipse indicates likely areas where "missing" studies are expected).

The 95% CIs are used to indicate the extent of between-study heterogeneity in addition to assessing funnel asymmetry [24] and are determined by the studies in the meta-analysis and the pooled estimate. Since the pseudo 95% CIs are based on the observed fixed effects pooled estimate they may be misleading if the meta-analysis is subject to publication bias (as the observed pooled estimate forming the basis for these 95% CIs will be biased). The contours displayed on funnel plots indicate levels of statistical significance for the primary studies in the meta-analysis and are independent of the pooled estimate, therefore, if the pooled estimate is biased, the contours are not affected.

A further difference between the pseudo 95% CIs and the contour lines is in interpretation: for the pseudo 95% CIs we are judging whether a study lies within expected limits on the funnel plot to help assess and interpret asymmetry and between-study heterogeneity; for the contour-funnel plot, we are interested in the distribution of the studies to assess whether observed asymmetry is likely to be due to publication bias based on statistical significance. It is worthwhile noting that the pseudo 95% CIs and $P = 0.05$ contour lines will coincide if and only if the fixed effects pooled estimate in a meta-analysis is exactly the null (e.g., Odds Ratio = 1). Although the traditional 95% CI guidelines can help to assess heterogeneity and funnel asymmetry, they do not help to identify the cause of any funnel asymmetry. In Fig. 4 the pseudo 95% CIs are displayed on the contour-enhanced funnel plot for meta-analysis 38.

2. Discussion

Tools to aid interpretation of funnel plots are clearly needed [22]. A recent study suggests that correct identification of the presence or absence of publication bias using funnel plots is poor [23] with only 52.5% of funnel plots correctly assessed for publication bias in simulated meta-analysis data sets. We believe contour-enhanced

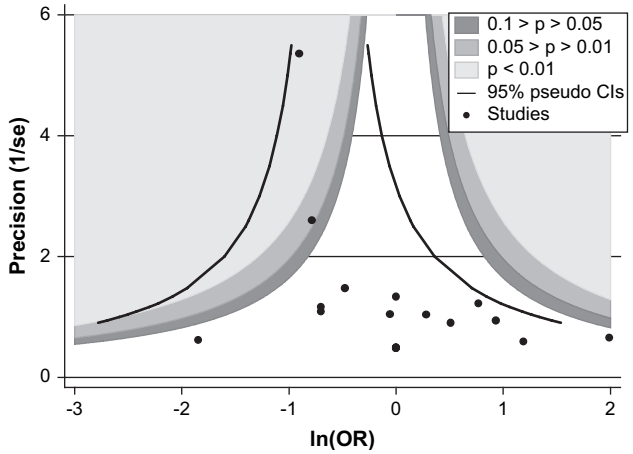


Fig. 4. Contour-funnel plot of meta-analysis 38 with 95% pseudo CIs displayed.

funnel plots greatly help interpretation of funnel plot asymmetry.

A number of points need consideration regarding the contour-enhanced funnel plots presented here:

1 Publication bias may be induced by mechanisms other than those based exclusively on statistical significance (e.g., sample size [25,26] and effect size [27]). This possibility should be considered when interpreting the contour-enhanced funnel plot.
2 The *P*-value determining the publication (or suppression) of a primary study may be based on a different measurement outcome to that used in the funnel plot, although this is likely to have little impact in practice on the conclusions made from assessment of the contour-enhanced funnel plot.
3 The contour-enhanced funnel plots presented here assume a two-sided significance test is used in the original studies and study suppression is assumed to be based on this *P*-value. A contour-enhanced funnel plot could also be constructed to display contours on one side of the funnel plot, if it was believed that suppression was based only on one-sided *P*-values.
4 Alternative choices of *y*-axis scales for funnel plots have received some discussion [24,28], and the use of other scales (i.e., standard error and variance) is shown at http://www2.le.ac.uk/Members/drj/supplementary-materials-for-papers. Sample size, or a function of sample size, cannot be used as the *y*-axis because unique *P*-values cannot be defined using a sample size scale.
5 As with all methods for detecting publication bias, observed asymmetry may be some artifact of sampling variation and nothing more.

While the plots presented in this paper display contours for multiple levels of statistical significants, for the purpose of assessing publication bias, it may be sufficient just to distinguish between the significant (at the traditional 5% level) and nonsignificant regions of the plot.

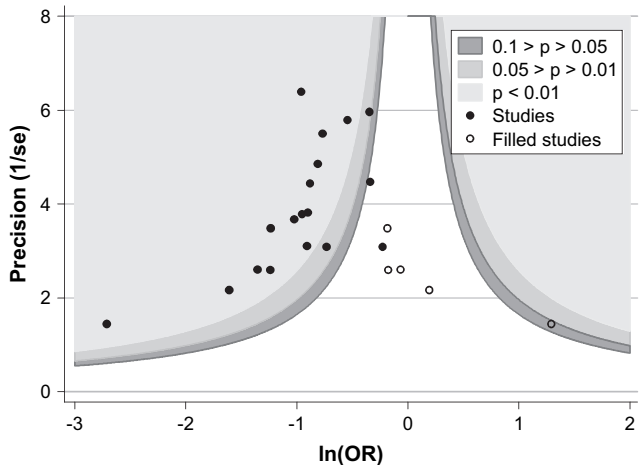
Neither statistical tests [10,29–32], nor the trim-and-fill method [14,15] consider asymmetry *in the context* of statistical significance, but rather should strictly be seen as methods to address asymmetry, and not necessarily publication bias [33]. The contour-funnel plot should *complement* their use. For example, the contour-funnel plot could be used naturally in conjunction with the trim-and-fill method because the latter informs the likely location of missing studies. In contrast, selection-modeling techniques [34–36] model study selection as a function of *P*-value; the contour-enhanced funnel plot goes some way to reconciling such an approach with a visual inspection of the data.

We believe the contour-enhanced funnel plot is a significant advancement in methods to address publication bias in meta-analyses of comparative effects, as it can aid assessment of publication bias due to statistical significance. Because it can enhance interpretation of a funnel plot, it should be used routinely for assessing possible publication bias (and complimentary to existing methods). We encourage its implementation in common statistical software used to carry out meta-analysis, and a Stata macro is available from the corresponding author to construct such plots.

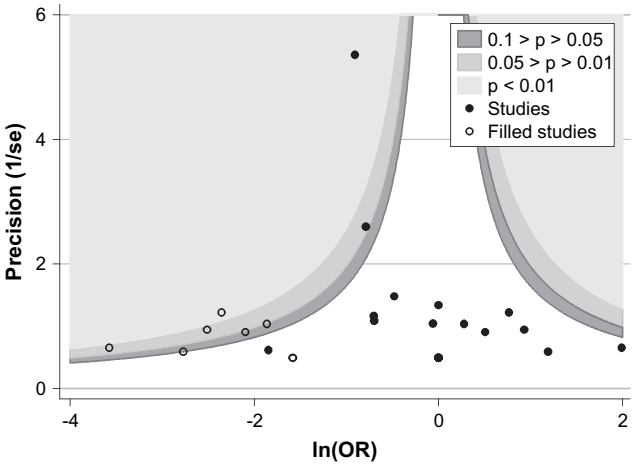
Acknowledgements

JP was funded through a UK Department of Health Evidence Synthesis Award while undertaking this work. The funding source had no role in any aspect of this study.

Appendix



Appendix Fig. 1. Contour-funnel plot of meta-analysis 11 with "filled" studies (open circle) estimated from the trim-and-fill method plotted.



Appendix Fig. 2. Contour-funnel plot of meta-analysis 38 with ''filled'' studies (open circle) estimated from the trim-and-fill method plotted.

References

[1] Sutton AJ, Abrams KR, Jones DR, Sheldon TA, Song F. Methods for meta-analysis in medical research. Chichester: Wiley; 2000.
[2] Rothstein HR, Sutton AJ, Borenstein M. Publication bias in meta-analysis. Chichester: Wiley; 2005.
[3] Dickersin K. How important is publication bias? A synthesis of available data. AIDS Educ Prev 1997;9:15–21.
[4] Easterbrook PJ, Berlin JA, Gopalan R, Matthews DR. Publication bias in clinical research. Lancet 1991;337:867–72.
[5] Ioannidis JPA. Effect of the statistical significance of results on the time to completion and publication of randomized efficacy trials. JAMA 1998;279:281–6.
[6] Chan AW, Altman DG. Identifying outcome reporting bias in randomised trials on PubMed: review of publications and survey of authors. BMJ 2005;330:753–6.
[7] Williamson PR, Gamble C, Altman DG, Hutton JL. Outcome selection bias in meta-analysis. Stat Methods Med Res 2005;14: 515–24.
[8] Chan AW, Hrobjartsson A, Haahr MT, Gotzsche PC, Altman DG. Empirical evidence for selective reporting of outcomes in randomized trials—Comparison of Protocols to published articles. JAMA 2004;291:2457–65.
[9] Light RJ, Pillemar DB. Summing up: the science of reviewing research. Cambridge, MA: Harvard University Press; 1984.
[10] Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. BMJ 1997;315:629–34.
[11] Schulz KF, Chalmers I, Hayes RJ, Altman DG. Empirical evidence of bias: dimensions of methodological quality associated with estimates of treatment effects in controlled trials. JAMA 1995;273:408–12.
[12] Cochrane Database of Systematic Reviews. In: The Cochrane Collaboration. Cochrane Library, 1998; Issue 3, Oxford.
[13] Sutton AJ, Duval SJ, Tweedie RL, Abrams KR, Jones DR. Empirical assessment of effect of publication bias on meta-analyses. BMJ 2000;320:1574–7.
[14] Duval S, Tweedie R. Trim and fill: a simple funnel-plot-based method of testing and adjusting for publication bias in meta-analysis. Biometrics 2000;56:455–63.
[15] Duval S, Tweedie RL. A nonparametric ''trim and fill'' method of accounting for publication bias in meta-analysis. J Am Stat Soc 2000;95:89–98.
[16] Terrin N, Schmid CH, Lau J, Olkin I. Adjusting for publication bias in the presence of heterogeneity. Stat Med 2003;22:2113–26.
[17] Peters JL, Sutton AJ, Jones DR, Abrams KR, Rushton L. Performance of the trim and fill method in the presence of publication bias and between-study heterogeneity. Stat Med 2007;26:4544–62.
[18] Williamson PR, Gamble C. Identification and impact of outcome selection bias in meta-analysis. Stat Med 2005;24:1547–61.
[19] Graves P, Deeks J, Demicheli V, Pratt M, Jefferson T. The effectiveness and safety of cholera vaccines. The Cochrane Library: The Cochrane Collaboration; 1998.
[20] Thornley B, Awad G, Adams CE. Chlorpromazine versus placebo for schizophrenia. The Cochrane Library: The Cochrane Collaboration; 1998.
[21] Jackson D. The implications of publication bias for meta-analysis' other parameter. Stat Med 2006;25:2911–21.
[22] Lau J, Ioannidis JPA, Terrin N, Schmid CH, Olkin I. The case of the misleading funnel plot. BMJ 2006;333:597–600.
[23] Terrin N, Schmid CH, Lau J. In an empirical evaluation of the funnel plot, researchers could not visually identify publication bias. J Clin Epidemiol 2005;58:894–901.
[24] Sterne JAC, Egger M. Funnel plots for detecting bias in meta-analysis: guidelines on choice of axis. J Clin Epidemiol 2001;54:1046–55.
[25] Hedges LV. Estimation of effect size under nonrandom sampling: the effects of censoring studies yielding statistically insignificant mean differences. J Educ Stat 1984;9:61–85.
[26] Begg CB. Publication bias. In: The handbook of research synthesis. New York; Russel Sage Foundation; 1994, pp. 399-409.
[27] Taubes G. Epidemiology faces its limits. Science 1995;269:164–9.
[28] Tang J-L, Liu JLY. Misleading funnel plots for detection of bias in meta-analysis. J Clin Epidemiol 2000;53:477–84.
[29] Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. Biometrics 1994;50:1088–101.
[30] Peters JL, Sutton AJ, Jones DR, Abrams KR, Rushton L. Comparison of two methods to detect publication bias in meta-analysis. JAMA 2006;295:676–80.
[31] Macaskill P, Walter SD, Irwig L. A comparison of methods to detect publication bias in meta-analysis. Stat Med 2001;20:641–54.
[32] Harbord RM, Egger M, Sterne JAC. A modified test for small-study effects in meta-analyses of controlled trials with binary endpoints. Stat Med 2006;25:3443–57.
[33] Sterne JAC, Gavaghan D, Egger M. Publication and related bias in meta-analysis: power of statistical tests and prevalence in the literature. J Clin Epidemiol 2000;53:1119–29.
[34] Hedges LV, Vevea JL. Selection method approaches. In: Rothstein H, Sutton AJ, Borenstein M, editors. Publication bias in meta-analysis: Prevention, assessment and adjustments. Chichester: Wiley; 2005.
[35] Hedges LV, Vevea JL. Estimating effect size under publication bias: small sample properties and robustness of a random effects selection model. J Educ Behav Stat 1996;21:299–332.
[36] Copas J. What works? Selectivity models and meta-analysis. J Royal Stat Soc Series A, Stat Soc 1999;162:95–109.