

**IS THE PRESENCE OF RETINOPATHY OF PRACTICAL VALUE IN
DEFINING CASES OF DIABETIC NEPHROPATHY IN GENETIC
ASSOCIATION STUDIES? THE EXPERIENCE WITH THE
ANGIOTENSIN-I CONVERTING ENZYME I/D POLYMORPHISM BASED
ON 53 STUDIES COMPRISING 17,791 SUBJECTS**

Short running title: Defining diabetic nephropathy in genetic studies

Daniel P.K. Ng, PhD¹; Bee-Choo Tai, PhD¹; Lim Xiu Li, BSc (Hons)¹

¹Department of Community, Occupational and Family Medicine, National University of Singapore, Singapore

CORRESPONDING AUTHOR:

Daniel P K Ng, PhD
Department of Community, Occupational and Family Medicine
Yong Loo Lin School of Medicine
National University of Singapore
16 Medical Drive MD3
SINGAPORE 117597
Email: cofnpkd@nus.edu.sg

Received 29 April 2008 and accepted 28 May 2008.

Additional information for this article can be found in an online appendix at
<http://diabetes.diabetesjournals.org>.

This is an uncopyedited electronic version of an article accepted for publication in *Diabetes*. The American Diabetes Association, publisher of *Diabetes*, is not responsible for any errors or omissions in this version of the manuscript or any version derived from it by third parties. The definitive publisher-authenticated version will be available in a future issue of *Diabetes* in print and online at <http://diabetes.diabetesjournals.org>.

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

Objective: A key consideration when setting up genetic studies is the case definition. For diabetic nephropathy (DN), the case definition is typically defined based on the presence of albuminuria. However, it has been long debated whether DN cases defined in this way may have a high prevalence of non-diabetic kidney disease especially if diabetic retinopathy (DR) is absent.

Research Design and Methods: We performed a meta-analysis of 53 studies comprising 17,791 subjects investigating the angiotensin-I converting enzyme insertion/deletion (I/D) polymorphism, taking into account the requirement for DR in the case definition and assuming a random effects model.

Results: No publication bias was observed. The overall pooled odds ratio (OR) for all 53 studies was 0.78 (95%CI=0.70–0.87, $P<0.001$) which indicated a significant protection against DN for genotype II as compared with carriage of the D allele. The pooled OR for the 11 studies ($n = 3,413$) requiring DR in the case definition was 0.68 (95%CI=0.53-0.86, $P=0.002$) and this was not significantly different from the pooled OR of 0.81 (95%CI=0.71-0.92, $P=0.001$) obtained from the 42 remaining studies ($n = 14,378$) ($P=0.198$). This lack of any significant effect of DR was reiterated in subgroup analyses according to the type of diabetes present.

Conclusions: Stipulating the presence of DR in the case definition of DN did not appear to confer tangible benefits when detecting genetic associations. Besides reducing sample sizes, this stipulation makes the interpretation of genetic associations more difficult due to the potential confounding presence of DR.

KEYWORDS:

ACE polymorphism, diabetic nephropathy, diabetic retinopathy, misclassification, publication bias, random effects model

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

Diabetic nephropathy (DN) is the major cause of end stage renal disease in many developed countries despite pharmacological interventions (1). The efficacy of such interventions may be dependent on patient genotypes and epidemiological evidence firmly supports a role for genetic susceptibility in causing DN in both type 1 and type 2 diabetes (2). Identification of these genes holds the promise for greater insight into the pathophysiology of this debilitating complication, and may ultimately provide novel therapies for disease prevention and intervention.

A key consideration when setting up genetic studies for DN is the case definition. Since DN is rarely diagnosed using invasive kidney biopsies, the case definition of this complication in genetic studies is typically defined based on the presence of albuminuria (3). However, by accepting this case definition, it is plausible that there might be a substantial number of subjects who were classified as having DN but may actually have non-diabetic kidney disease instead. This misclassification in genetic studies will be expected to drive any true association towards the null. In an attempt to circumvent this problem, certain investigators have proposed that DN cases should be required to have diabetic retinopathy (DR) as well. The rationale for this proposal is that several studies have suggested that albuminuria can be attributed with confidence to DN if DR was present (4).

The vital question however remains as to whether the stipulation of DR does indeed facilitate the identification of susceptibility genes for DN in real-life association studies. To address this issue, we have

presently performed a meta-analysis on the association between DN and the angiotensin-I converting enzyme insertion/deletion polymorphism (*ACE I/D*), taking DR status into account. This genetic marker is the most extensively studied polymorphism to date for DN and as such, data from 53 studies comprising 17,791 subjects was available for this meta-analysis.

RESEARCH DESIGN AND METHODS

Dataset

We used a pre-existing dataset based on 47 studies that were published from 1994 to March 2004 which examined the association between *ACE I/D* and DN (3). This dataset was subsequently expanded in 2006 to give a total of 53 studies comprising 17,791 subjects by the addition of 6 later studies (5-10). Briefly, studies were considered if they provided sufficient information to allow a comparison of the *ACE I/D* genotype distribution between cases and controls. Cases were type 1 or 2 diabetic subjects fulfilling the minimal criterion of microalbuminuria while controls were defined predominantly on the basis of normoalbuminuria. Of the 53 studies, 11 studies specifically required the concomitant presence of DR when defining cases of DN (Table 1).

Statistical analyses

Funnel plots of the effect estimate based on log-odds ratio were plotted against its standard error to evaluate the possibility of publication bias (11). The magnitude of the genetic association between *ACE I/D* and DN was obtained by calculating the odds ratio (OR) and its associated 95% confidence interval (95%CI). A random effects model was employed assuming that the studies represented a random

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

sample from the larger population of such studies, with each having its own underlying effect size. Under this model, it is assumed that there is a mean population effect size about which the study-specific OR varies. As the random effects model takes into account the inter-study heterogeneity such as differences in study design and case definitions for DN, it provides a more conservative evaluation of the significance of the association than one based on fixed effects (12).

RESULTS

A total of 53 studies (n=9,556 cases, 8,235 controls) fulfilled the criteria for inclusion in this review (Table 1). Twenty-one studies involved type 1 diabetic subjects (n=4,154), while the remaining 32 studies were conducted on patients with type 2 diabetes (n=13,637). The potential presence of publication bias was assessed using funnel plots of the estimate of log-odds ratio for the genotype II versus DD/ID against its standard error (Fig. 1a). Considerable scatter was observed around the pooled log-odds ratio estimate when the reciprocal of the standard error was small and approached convergence to form a symmetrical funnel as this reciprocal increased when all 53 studies were assessed. Similarly, there was no evidence of such bias when the 53 studies were analyzed separately depending on whether these required the concomitant DR in the case definitions (Fig. 1b,1c).

The overall pooled OR for all 53 studies was 0.78 (95%CI = 0.70–0.87, P<0.001) which indicated a significant protection against DN for genotype II as compared with carriage of the D allele (Fig. 2). The pooled OR for the 11 studies (n=3,413)

requiring DR in the case definition was 0.68 (95%CI=0.53-0.86, P=0.002) and this was not significantly different from the pooled OR of 0.81 (95%CI=0.71-0.92, P=0.001) obtained from the 42 remaining studies (n=14,378) that eschewed the corroborative presence of DR (P=0.198) (Fig. 2).

In subgroup analyses on 21 studies conducted on 4,154 type 1 diabetic patients, the overall pooled OR was 0.84 (95%CI=0.68-1.05, P=0.119). The pooled OR for 5 studies (n=1,759) requiring DR status in cases was 0.78 (95%CI=0.58-1.06, P=0.110) and this was similar to the pooled OR of 0.85 (95%CI=0.63-1.13, P=0.255) for the remaining 16 type 1 diabetes studies (n=2,395) (P=0.704). In corresponding subgroup analyses, the overall pooled OR was 0.75 (95%CI=0.66-0.86, P<0.001) for the 32 studies consisting of 13,637 type 2 diabetic patients. The pooled OR for 6 studies (n=1,654) requiring DR in the case definitions was 0.54 (95%CI=0.36-0.82, P=0.004) which was not significantly smaller than the pooled OR of 0.79 (95%CI=0.69-0.91, P=0.001) for the 26 remaining type 2 diabetes studies (n=11,983) (P=0.087).

We considered the possibility that this lack of effect of DR may be due to the fact that cases in some studies may have had a high prevalence of this eye complication even though it was not explicitly required in the case definition. Since these studies would have been placed under the category of studies not requiring DR in the preceding analyses, the anticipated outcome would have been to drive any apparent effect of DR towards the null. To clarify this issue, we scrutinized the published reports and found that of the 42 studies that did not

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

specifically stipulate DR in the case definition, 25 reports did provide sufficient clinical information which allowed us to determine the prevalence of DR among the cases (Table 1). 7 studies were selected from among these in which DR was present in at least a majority (80%) of cases (Table 1). These studies were combined with the 11 studies that specified DR in their case definitions for comparison with the other remaining studies. The overall pooled ORs for these 18 studies ($n=4,414$) was 0.71 (95%CI=0.58-0.87, $P=0.001$) compared to 0.82 (95%CI=0.72-0.94, $P=0.003$) for the 35 remaining studies ($n=13,377$) ($P=0.249$) (Supplementary Fig. 1). Confining our analyses to just the 36 studies which provided information about DR also yield similar findings (data not shown). No significant differences associated with the requirement for DR were observed in either patients with type 1 ($P=0.448$) or type 2 diabetes ($P=0.236$).

DISCUSSION

The promise of new insights into the pathogenesis of diabetic nephropathy is fuelling intense efforts to identify genes conferring risk to DN (13-15). While much of the attention has been placed on attaining large sample sizes to provide power for detecting small effects, another key consideration is the case definition of DN. In this study, we reviewed the literature on the association of *ACE* I/D and DN and found evidence which suggested that stipulating the concomitant presence of DR in order to corroborate a diagnosis of DN is unlikely to yield significant benefits when searching for genetic associations.

The inclusion of DR in the case definition is commonplace in the published literature on *ACE* I/D. 21% of the 53

studies imposed this requirement and this was comparable in studies focussing on either type 1 or type 2 diabetic patients (24% and 19% respectively). This practice is likely to have originated based on several studies which found that only a subset of patients with proteinuria and/or azotemia have kidney biopsies that substantiated a diagnosis of diabetic glomerulopathy and this has subsequently been taken to mean that proteinuria per se is insufficient as conclusive evidence of diabetic nephropathy (16-20). However, in a systematic review of 9 published reports as well as their own data (21), Oslén and Mogensen had deliberated on this issue and proposed that a very likely reason for the high prevalence of non-diabetic kidney disease was due to the fact that most of the reports were based on biased groups of patients who were inadvertently selected for such non-diabetic kidney conditions (21). Another potential explanatory factor was the application of a differing criterion for diagnosing glomerulonephritis, a major contributor of non-diabetic kidney disease (21).

In our literature review, several points emerged which may be highlighted. Of the 11 studies that required DR in the case definition, 9 studies did not insist that their controls should have DR as well. It was also striking that one study deliberately specified that its controls had to be free of DR when all its cases had this eye complication (Table 1). Understandably, one will be hard pressed to conclude whether any observed association was truly between *ACE* I/D and DN, DR or possibly even a combination of both complications.

In practical terms, the requirement for DR in controls will inadvertently diminish the

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

overall size which has already become more limited since cases must have DR. Unfortunately, on the basis of our present results, this drop in sample size and consequently power, comes without any tangible reciprocal benefit which would be potentially expected if disease misclassification among cases had been rampant in the absence of DR as previously suggested (20). Moreover, since recent studies suggested that the majority (70-74%) of albuminuric type 2 diabetic patients do indeed have diabetic glomerulopathy even in the absence of DR (22,23), it becomes questionable whether genetic associations found in studies employing DR can be readily extrapolated to these DN patients. Nevertheless, it is still noteworthy that the overall pooled OR was slightly but consistently of a larger magnitude in studies where DR was prevalent although even with the large dataset under review, this difference failed to reach statistical significance. One may thus consider the possibility that including DR might help in the identification of potential genetic factors for common underlying traits which may manifest itself as a joint retinal-renal phenotype.

Several strengths and limitations of our study may be discussed. On a positive note, the meta-analysis was conducted on a substantial dataset comprising 17,791 patients from 53 studies. Moreover, there was no overt sign of publication bias which would argue against the validity of our results with funnel plot analyses indicating that small negative studies were as likely to be published as large studies with positive findings. Secondly, we performed subgroup analyses according to whether the patients had type 1 or type 2 diabetes. This distinction was relevant

since it has been debated as to whether the presence of non-diabetic kidney disease was more common in albuminuric patients with type 2 diabetes compared to those with type 1 diabetes (16,24).

A main limitation is that our study was restricted solely to *ACE* I/D. This was borne of necessity since *ACE* I/D is the most extensively studied polymorphism to date with regards to DN and there is currently a severe lack of extensive studies into other genetic markers. Given this situation, our study has managed to render a first critical insight into this issue. Finally, reports of late have provided evidence that DN may be associated with specific risk haplotypes at the *ACE* locus. However, a meta-analysis on *ACE* haplotypes is precluded due to a paucity of such reports (10, 25).

In conclusion, our study using real-life association data has suggested that the presence of DR may be of limited practical value for defining cases of DN when seeking genetic associations. In addition, the reduced sample sizes arising from such a stipulation may make it harder to detect these associations. Interpretation of the results from such studies could also be hampered by the possible confounding presence of DR if left uncontrolled.

ACKNOWLEDGEMENTS

This study was supported by the National Medical Research Council, Singapore (NMRC/0850/2003, NMRC/1018/2005). We thank Siti Nurbaya (National University of Singapore) for assisting with the literature review.

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

REFERENCES

1. Jones CA, Krolewski AS, Rogus J, et al. Epidemic of end-stage renal disease in people with diabetes in the United States population: do we know the cause? *Kidney Int* 67: 1684-1691, 2005
2. Ng DPK, Krolewski AS. Molecular genetic approaches for studying the etiology of diabetic nephropathy. *Current Mol Med* 5: 511-527, 2005
3. Ng DPK, Tai BC, Koh D, et al. Angiotensin-I Converting Enzyme Insertion/Deletion Polymorphism and Diabetic Nephropathy: A Meta-analysis of Studies reported during 1994-2004 and comprising 14,727 Subjects. *Diabetologia* 48:1008-1016, 2005
4. Remuzzi G, Schieppati A, Ruggenenti P. Clinical practice. Nephropathy in patients with type 2 diabetes. *N Engl J Med* 346:1145-1151, 2002
5. Arzu Ergen H, Hatemi H, Agachan B, et al. Angiotensin-I converting enzyme gene polymorphism in Turkish type 2 diabetic patients. *Exp Mol Med* 36: 345-350, 2004
6. Degirmenci I, Kebapci N, Basaran A, et al. Frequency of angiotensin-converting enzyme gene polymorphism in Turkish type 2 diabetic subjects. *Int J Clin Pract* 59:1137-1142, 2005
7. Shestakova MV, Vikulova OK, Nosikov VV. Role of genetic factors and arterial hypertension in development and progression of diabetic nephropathy (DN) in type 1 diabetes mellitus. *Diabetes* 54(Suppl 1):A195, 2005
8. Canani LH, Costa LA, Crispim D, et al. The presence of allele D of angiotensin-converting enzyme polymorphism is associated with diabetic nephropathy in patients with less than 10 years duration of type 2 diabetes. *Diabet Med* 22:1167-1172, 2005
9. Wang Y, Ng MCY, So WY, et al. Prognostic effect of insertion/deletion polymorphism of the ACE gene on renal and cardiovascular clinical outcomes in Chinese patients with type 2 diabetes. *Diabetes Care* 28:348-354, 2005
10. Ng DPK, Placha G, Choo S, et al. A disease haplotype for advanced nephropathy in type 2 diabetes at the ACE locus. *Diabetes* 55: 2660-2663, 2006
11. Whitehead A. Meta-analysis of controlled clinical trials. Chichester: John Wiley & Sons:197-213, 2002
12. Fleiss JL. The statistical basis of meta-analysis. *Statistical Methods in Medical Research* 2:121-145, 1993
13. Mueller PW, Rogus JJ, Cleary PA, et al. Genetics of Kidneys in Diabetes (GoKinD) study: a genetics collection available for identifying genetic susceptibility factors for diabetic nephropathy in type 1 diabetes. *J Am Soc Nephrol* 17:1782-1790, 2006
14. Knowler WC, Coresh J, Elston RC, et al. Family Investigation of Nephropathy and Diabetes Research Group. The Family Investigation of Nephropathy and Diabetes (FIND): design and methods. *J Diabetes Complications* 19:1-9, 2005
15. Tarnow L, Groop PH, Hadjadj S, et al. European rational approach for the genetics of diabetic complications--EURAGEDIC: patient populations and strategy. *Nephrol Dial Transplant* 23:161-168, 2008

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

16. Richards NT, Greaves I, Lee SJ, et al. Increased prevalence of renal biopsy findings other than diabetic glomerulopathy in type II diabetes mellitus. *Nephrol Dial Transplant* 7:397-399, 1992
17. Amoah E, Glickman JL, Malchoff CD, et al. Clinical identification of nondiabetic renal disease in diabetic patients with type I and type II disease presenting with renal dysfunction. *Am J Nephrol* 8:204-211, 1988
18. Suzuki D, Takano H, Toyoda M, et al. Evaluation of renal biopsy samples of patients with diabetic nephropathy. *Intern Med* 40:1077-1084, 2001
19. Parving HH, Gall MA, Skott P, et al. Prevalence and causes of albuminuria in non-insulin-dependent diabetic patients. *Kidney Int* 41:758-762, 1992
20. John GT, Date A, Korula A, et al. Nondiabetic renal disease in noninsulin-dependent diabetics in a south Indian Hospital. *Nephron*. 1994;67(4):441-3.
21. Olsen S, Mogensen CE. How often is NIDDM complicated with non-diabetic renal disease? An analysis of renal biopsies and the literature. *Diabetologia* 39:1638-1645, 1996
22. Serra A, Romero R, Bayes B, et al. Is there a need for changes in renal biopsy criteria in proteinuria in type 2 diabetes? *Diabetes Res Clin Pract* 58:149-153, 2002
23. Christensen PK, Larsen S, Horn T, et al. Causes of albuminuria in patients with type 2 diabetes without diabetic retinopathy. *Kidney Int* 58:1719-1731, 2000
24. Mauer SM, Steffes MW, Ellis EN, et al. Structural-functional relationships in diabetic nephropathy. *J Clin Invest* 74:1143-1155, 1984
25. Hadjadj S, Tarnow L, Forsblom C, et al. Association between angiotensin-converting enzyme gene polymorphisms and diabetic nephropathy: case-control, haplotype, and family-based study in three European populations. *J Am Soc Nephrol* 18:1284-1291, 2007

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

Table 1. Summary of 53 studies on ACE I/D and DN*

First Author	Year	Diabetes	Case definition requires DR?	Cases with DR (%)	Control definition requires DR?	Control with DR (%)	Case genotypes (n)			Control genotypes (n)		
							DD	ID	II	DD	ID	II
Doria	1994	Type 1	No	70.0	No	21.0	24	35	15	16	41	20
Powrie	1994	Type 1	No	NA	No	NA	7	8	4	24	37	24
Dudley	1995	Type 2	No	22.1	No	19.0	47	85	31	70	148	49
Fujisawa	1995	Type 2	No	NA	No	NA	7	23	24	6	12	17
Mizuiri	1995	Type 2	Yes	100.0	No	NA	19	50	11	9	11	11
Panagiotopoulos	1995	Type 2	No	NA	No	NA	15	25	10	42	44	29
Schmidt	1995	Type 1	No	74.6	No	63.9	52	38	24	55	55	23
Tarnow	1995	Type 1	Yes	100.0	No	65.0	63	95	40	67	77	46
Rabensteiner	1995	Type 1	No	NA	No	NA	16	39	9	8	33	15
Chowdhury	1996	Type 1	Yes	100.0	No	NA	78	124	40	55	79	32
Doi	1996	Type 2	No	93.9	No	69.4	29	85	50	12	56	56
Nakajima	1996	Type 2	No	NA	No	NA	14	50	37	4	19	18
Oh	1996	Type 1	No	83.9	No	42.9	10	9	12	7	10	11
Ohno	1996	Type 2	No	58.2	No	37.7	15	38	26	5	15	33
Yoshida	1996	Type 2	Yes	100.0	No	48.0	19	28	25	7	46	43
Barnas	1997	Type 1	No	100.0	No	78.0	14	27	9	4	21	15
Hibberd	1997	Type 1	Yes	100.0	No	46.5	21	42	9	36	43	7
Jeffers	1997	Type 2	No	NA	No	NA	23	20	7	139	218	102
Marre	1997	Type 1	Yes	100.0	Yes	100.0	119	168	50	48	69	40
Ringel	1997	Type 1	No	41.0	No	20.4	35	68	31	57	130	39
Ringel	1997	Type 2	No	35.4	No	15.0	44	84	33	35	69	36
Demurov	1997	Type 1	No	NA	No	NA	24	29	3	24	32	20
Schmidt	1997	Type 2	No	64.7	No	35.3	121	129	61	131	154	62
Pfohl	1998	Type 1	No	87.0	No	87.0	17	15	8	15	18	7
Freire	1998	Type 1	No	38.0	No	10.0	33	32	12	34	45	10
Grzeszczak	1998	Type 2	No	48.9	No	39.2	129	230	103	73	118	63
Hanyu	1998	Type 2	Yes	100.0	Yes	100.0	4	13	7	2	5	14
Huang	1998	Type 2	No	NA	No	NA	11	16	2	20	25	9
Wu	1998	Type 2	No	NA	No	NA	12	18	21	1	11	6
Bouhanick	1999	Type 1	No	NA	No	NA	4	5	4	19	34	10
De Cosmo	1999	Type 1	Yes	100.0	No	NA	73	79	23	65	53	18
Kuramoto	1999	Type 2	No	42.4	No	13.8	9	16	8	3	13	13
Miura	1999	Type 1	No	71.4	No	44.7	13	49	36	10	58	35
Vleming	1999	Type 1	No	100.0	No	NA	39	24	16	26	34	22
Wong	1999	Type 2	No	96.0	No	30.0	7	30	43	12	40	36
Hsieh	2000	Type 2	No	NA	No	NA	40	59	80	21	50	86
van Ittersum	2000	Type 1	No	71.0	No	28.2	13	33	23	49	86	53
Araz	2001	Type 2	No	70.0	No	31.7	34	64	18	43	57	23
Azar	2001	Type 1	No	NA	No	NA	23	27	2	1	7	2
Gohda	2001	Type 2	No	NA	No	NA	85	222	229	31	92	89
Taniwaki	2001	Type 2	No	84.9	No	72.5	14	40	32	12	26	31
Viswanathan	2001	Type 2	Yes	100.0	No†	0.0	24	45	17	5	8	10
Fradin	2002	Type 2	No	35.0	No	19.5	38	61	18	44	54	20

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

Lee	2002	Type 2	No	NA	No	NA	40	137	117	39	170	208
Ha	2003	Type 2	Yes	100.0	No	39.4	43	62	35	9	57	33
Hadjadj	2003	Type 2	No	4.5	No	2.0	1119	146	552	208	282	115
								8				
Okuno	2003	Type 2	No	50.0	No	26.3	3	8	1	5	12	21
Arzu Ergen	2004	Type 2	No	16.0	No	22.0	9	11	5	24	21	5
Degirmenci	2005	Type 2	No	NA	No	NA	12	25	6	30	47	19
Shestakova	2005	Type 1	No	NA	No	NA	13	35	15	12	30	24
Canani	2005	Type 2	Yes	100.0	No	NA	126	181	66	181	308	120
Wang	2005	Type 2	No	77.9	No	NA	19	43	36	128	496	559
Ng	2006	Type 2	No	NA	No	NA	96	148	47	52	83	32

*The first 47 studies have been previously referenced (3).

†Absence of DR specifically required in Controls of this study

NA, information on DR not available

DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

FIGURE LEGENDS

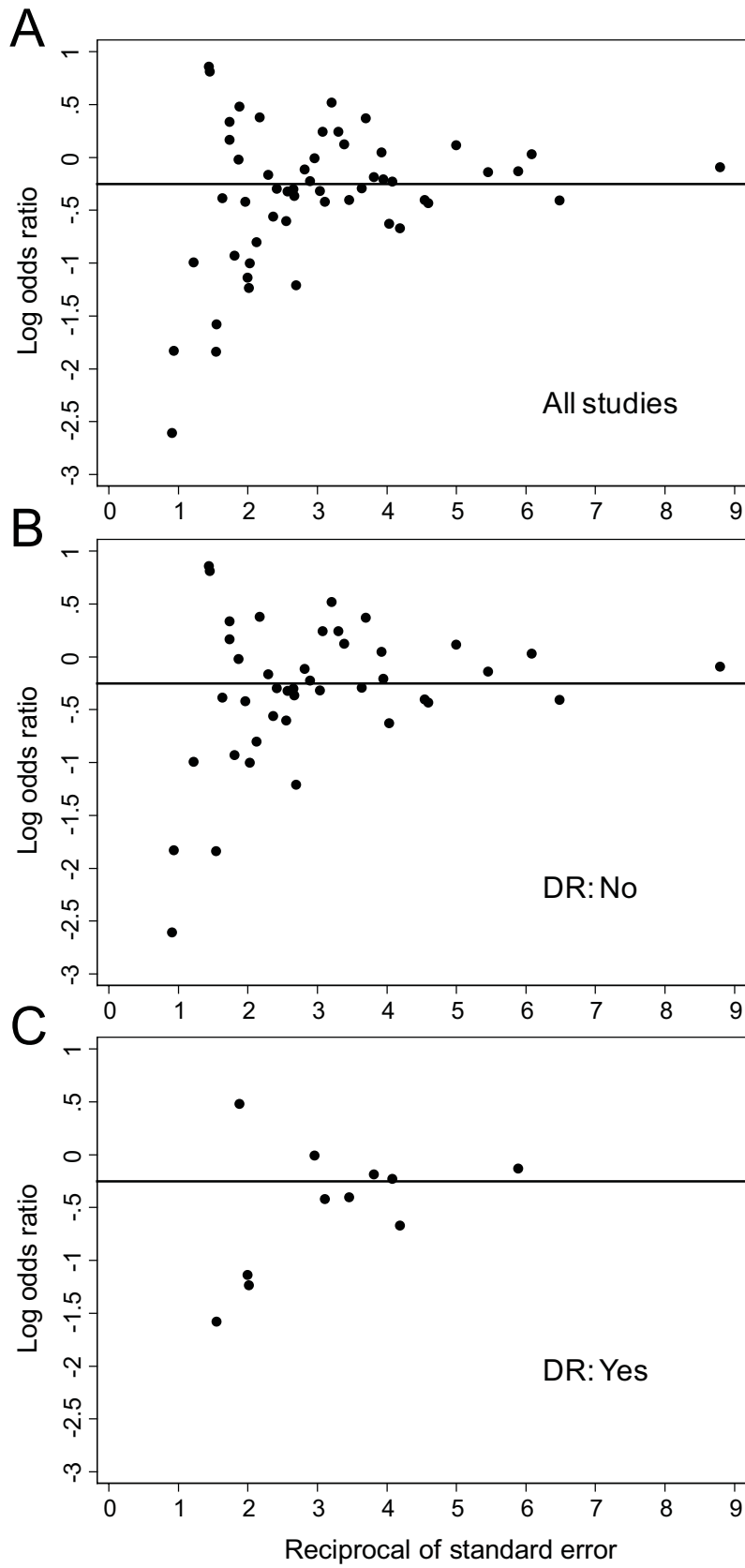
Figure 1

Funnel plot for the evaluation of publication bias in studies of association of *ACE* I/D for a) all 53 studies, b) 42 studies not requiring retinopathy in the case definition as well as c) 11 studies requiring DR to corroborate the presence of DN.

Figure 2

Odds ratio and the associated 95% confidence interval for comparing *ACE* II with ID/DD genotypes in all 53 studies which comprised 11 studies requiring DR in case definition and 42 studies not requiring DR.

Figure 1



DEFINING DIABETIC NEPHROPATHY IN GENETIC STUDIES

Figure 2

All studies

