

## Original Article

# Carriage of *NBN* polymorphisms and acute leukemia risk

Yan Wang, Zhongliang Sun, Yuan Xu

Department of Hematology, Shandong Jining No. 1 People's Hospital, China

Received November 1, 2014; Accepted February 2, 2015; Epub March 15, 2015; Published March 30, 2015

**Abstract:** Background: Recent reports revealed a significant association of *NBN* polymorphisms with risk of acute leukemia among Chinese, but not among Europeans. The objective of this study was to obtain a more precise measure of acute leukemia risk associated with *NBN* rs1805794, rs2735383, rs709816 polymorphisms. Methods: Using PubMed, Embase, ISI Web of Science, and the Cochrane Library databases, we undertook a systematic literature search up to September 1, 2014. Eligible studies were singled out from 31 possibly related publications by two investigators. Based on the extracted *NBN* genotypes, we calculated pooled odds ratios (ORs) and 95% confidence intervals (95% CI) by use of the random effects model proposed by DerSimonian and Laird. Results: We finally derived 3,065 subjects for meta-analysis of rs1805794, and found that carriage of the CC genotype was associated with approximately 1.70-fold increased risk of acute leukemia (OR 1.66, 95% CI 1.17-2.36; OR 1.77, 95% CI 1.23-2.54). A 25% higher risk was also identified among the individuals with the C allele (OR 1.25, 95% CI 1.03-1.51). Among 1,553 subjects for rs2735383, no significant association was indicated in the investigated comparison models. Nor did the analysis of 1,485 samples for rs709816 suggest any noteworthy connection. Conclusions: Carriage of rs1805794 polymorphism in the *NBN* gene may be associated with the occurrence of acute leukemia. New clinical studies are needed to identify the genetic associations and thus facilitates an increased understanding of the molecular mechanisms of this malignancy.

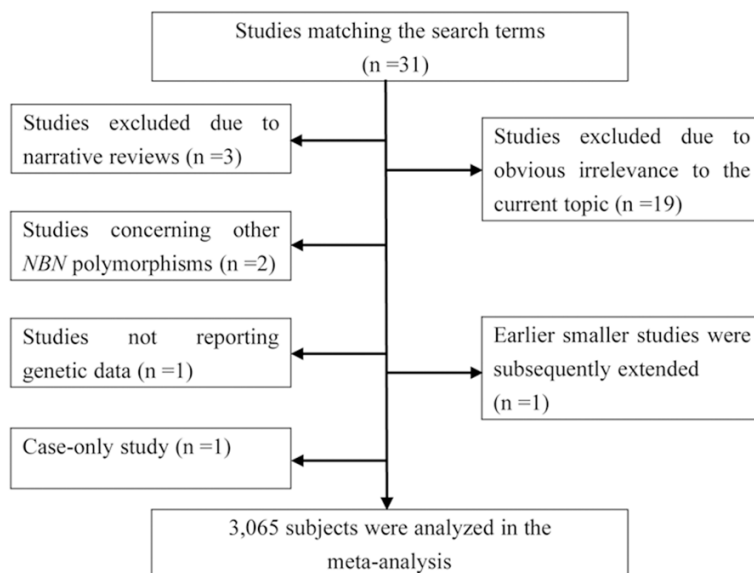
**Keywords:** Nibrin, polymorphisms, acute leukemia

### Introduction

Leukemia is a group of aggressive cancers that usually starts in the bone marrow and consequently causes a large quantity of incompletely developed white blood cells called leukemia cells or blasts [1, 2]. The incidence and mortality rate of leukemia, mainly composed of acute myeloid leukemia (AML), acute lymphoblastic leukemia (ALL), chronic myeloid leukemia (CML), and chronic lymphocytic leukemia (CLL), rapidly increases over the past decade: about 256,000 diagnoses and 209,000 deaths across the world in 2001, accounting for almost 3% of all cancer-related deaths that year [3]. The number of deaths increased to about 281,500 in 2010 [4]. This consistent increase along with the fact that etiology of leukemia is complex and the exact causes remain unidentified highlights the substantial importance to establish the susceptibility factors.

In addition to environmental risk factors (high-dose ionizing radiation, prior chemotherapy, and chemical compounds) and poor lifestyles (heavy tobacco smoking), inherited factors are considered as important components in the development of leukemia [5, 6]. Nibrin (*NBN*, *Nbs1*), a member of MRN complex, play a central role in regulating activity of *NBN* protein related to immunoglobulin class shifting, telomere sustainment, meiotic recombination and reconstruction of DNA damage such as double strand breaks (DSBs) [7-9], a major cause of chromosomal damage and subsequent formation of various diseases. In exon 5 of *NBN*, there locates a common non-synonymous single-nucleotide polymorphism with a C to G conversion (rs1805794). It has previously been described that rs1805794 has an influence on *NBN* protein function and the interactions with many other proteins [10]. A series of meta-analyses demonstrated strong evidence of significantly increased risk of breast

## NBN and acute leukemia risk



**Figure 1.** Study flow chart for the process of selecting the eligible studies.

cancer, lung cancer, and urinary system cancer ascribed to *NBN* polymorphisms including rs1805794 [11-14]. Nonetheless, the role of *NBN* polymorphisms in the development of acute leukemia remains unidentified, due to the inconsistent results yielded in epidemiological and molecular studies representing distinct populations [15-18].

Herein, we targeted three polymorphisms (rs1805794, rs2735383, rs709816) in the *NBN* gene, and performed a meta-analysis to better define the association between *NBN* and risk of developing acute leukemia.

### Methods

#### Publication search

To identify the publications reporting on association of *NBN* gene polymorphisms and risk of acute leukemia, we undertook a systematic literature search up to September 1, 2014, using PubMed, Embase, ISI Web of Science, and the Cochrane Library databases, without limits on language. The search terms included acute myeloid leukemia, acute lymphoblastic leukemia, leukemia, polymorphism, variant, *NBN*, and *NBS1*. In order to derive as many usable data as possible, we additionally scrutinized the references quoted in narrative reviews and epidemiological studies examining the association under investigation.

#### Inclusion criteria, exclusion criteria and data extraction

We pre-defined the following conditions to single out all eligible studies from the possibly relevant publications: (1) case group consisted of AML or ALL patients, (2) collecting healthy subjects as reference group, (3) evaluating risk of acute leukemia in association with at least one of the *NBN* gene polymorphisms being investigated, (4) providing genotype data in detail to calculate odds ratios (ORs), (5) genotype distribution in control populations must be consistent with Hardy-Weinberg equilibrium, and (6) the subjects must be unique. In case of two or more

publications where the same patients were included, we considered the publication with the largest sample size.

We excluded the studies when a smaller study was subsequently updated by an extended study by the same group of authors, essential genotyping data were not provided and unavailable even after having contacted the major authors, published as a case-case or case-only study, or deviation from Hardy-Weinberg equilibrium was detected in controls.

For the studies included in the meta-analysis, two experienced investigators separately extracted data on first author, publication year, study location (country), ethnicity (racial origin), genotyped patients and controls, subtype (AML or ALL), mean age, minor allele frequency (MAF), and count of wild-type, heterozygous, and homozygous genotypes. We categorized ethnic populations as Chinese or European. Disparities, if any, were settled via discussion.

#### Quality assessment

The methodological quality of each study was separately assessed by two investigators who completed data extraction. The assessment was carried out according to the Newcastle-Ottawa quality assessment scale (NOS) [23]. This scale consists of three parts including a total of nine items (1 point for each item): com-

**Table 1.** Characteristics of studies assessing effects of *NBN* polymorphisms on acute leukemia risk

Author and year	Study location	Study population	Number of cases/controls	<i>NBN</i> genotypes						Subtype
				Cases			Controls			
rs1805794				GG	GC	CC	GG	GC	CC	
Li 2013 [15]	China	Chinese	428/600	63	210	155	192	298	110	AML
Jiang 2011 [16]	China	Chinese	175/350	26	83	66	119	181	50	ALL
Smolkova 2014 [17]	Norway	European	460/545	211	203	46	252	241	52	ALL
Mosor 2013 [18]	Poland	European	232/275	96	92	44	111	134	30	ALL, AML
rs2735383				GG	GC	CC	GG	GC	CC	
Li 2013 [15]	China	Chinese	428/600	157	208	63	202	290	108	AML
Jiang 2011 [16]	China	Chinese	175/350	61	80	34	122	174	54	ALL
rs709816				TT	TC	CC	TT	TC	CC	
Smolkova 2014 [17]	Norway	European	458/545	183	214	61	216	258	71	ALL
Mosor 2013 [18]	Poland	European	207/275	71	96	40	96	129	50	ALL, AML

AML, acute myeloid leukemia; ALL, acute lymphoblastic leukemia.

parability (2 items), exposure (3 items), and selection (4 items).

#### Quantitative data synthesis

Deviation from Hardy-Weinberg equilibrium was examined by use of the goodness-of-fit  $\chi^2$ -test in control groups. Statistical data were performed using the Stata software package v.12.0 (Stata Corporation, College 162 Station, TX, USA).  $P < 0.05$  was considered significant, unless stated otherwise.

Data on wild-type, heterozygous and homozygous genotypes of *NBN* polymorphisms were used to assess the risk of developing acute leukemia [ORs and 95% confidence intervals (CIs)]. We calculated the pooled OR and 95% CIs assuming the homozygous comparison model, the recessive comparison model, and the allele comparison model to investigate the association between carriage of two minor alleles or one minor allele alone and risk of acute leukemia.

Between-study heterogeneity was assessed by the  $\chi^2$ -based  $Q$  test and we considered  $P < 0.05$  statistically significant. We also utilized the  $I^2$  metric to quantify the proportion of total variation across studies [19], with 0%, 0-25%, 25-50%, 50-100% indicating no, low, moderate, and large heterogeneity, respectively. The random-effects model (the DerSimonian and Laird), an analytical method prone to provide wider 95% CIs, was performed to estimate values of the single studies as well as the pooled ORs [20].

Sensitivity analysis was performed by sequentially omitting the included epidemiological studies, to check whether the independent data sets exerted significant influence on the combined effect sizes. Potential publication bias was determined using the funnel plot and Egger's linear regression test, with an asymmetric funnel plot suggesting presence of substantial publication bias [21, 22].

## Results

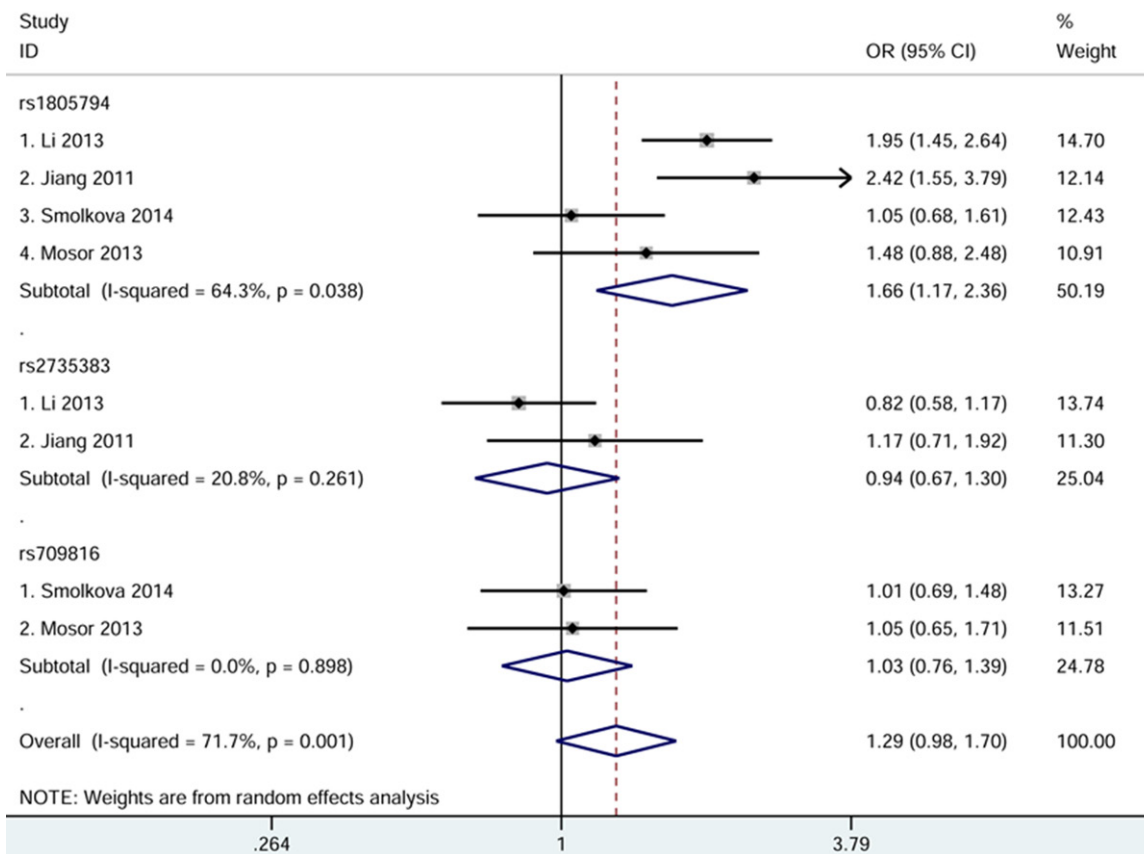
### Study exclusion and inclusion

Systematically searching the electronic databases yielded 31 related publications in total. All titles were screened and 19 publications were excluded due to obvious irrelevance to the current topic. For the remaining 12 studies, we scanned the abstracts and deleted 5 studies due to narrative reviews or concerning other *NBN* polymorphisms (each polymorphism being investigated in less than two studies of acute leukemia). We then retrieved 7 full-length studies and examined their eligibility according to the inclusion criteria, excluding 3 studies owing to lack of essential data, being updated by following larger studies and case-only study. The reasons of study exclusion and inclusion are detailed in **Figure 1**.

### Study characteristics

Our meta-analysis finally incorporated four studies [15-18], providing 3,065 subjects for rs1805794, 1,553 subjects for rs2735383, and 1,485 samples for rs709816. All studies

## NBN and acute leukemia risk



**Figure 2.** Meta-analysis for *NBN* polymorphisms and acute leukemia risk in the homozygous comparison model. Each study was shown by a point estimate of the effect size (OR) (size inversely proportional to its variance) and its 95% confidence interval (95% CI) (horizontal lines). The diamond denotes the pooled OR.

reported detailed genetic data and showed consistency with Hardy-Weinberg equilibrium ( $P > 0.05$ ). These four studies were composed of two ALL studies, one AML study and one study investigating both subtypes without reporting separate genotyping data. In addition, two studies were conducted in Chinese subjects and two in European subjects. The main characteristics of studies assessing the effects of *NBN* polymorphisms on acute leukemia risk are shown in **Table 1**.

### Quantitative analysis

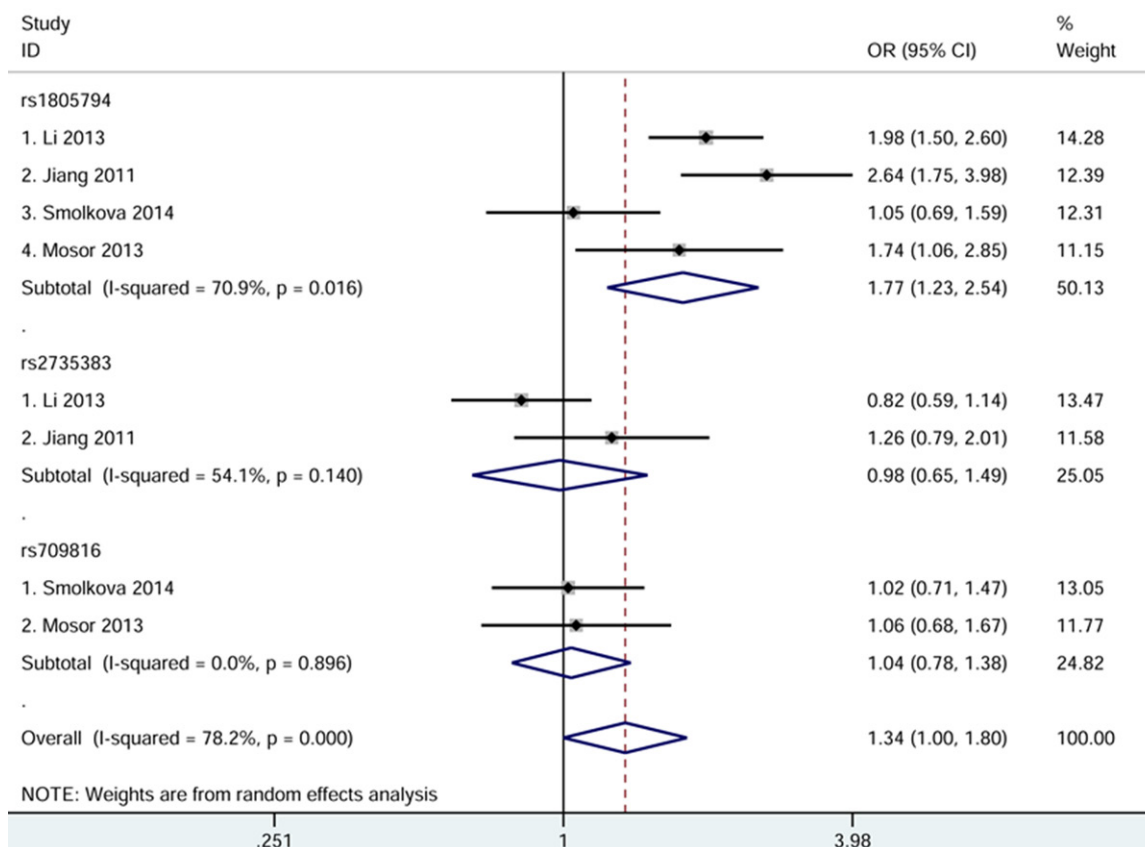
**Association of *NBN* rs1805794 polymorphism with acute leukemia:** In the meta-analysis of rs1805794, we found that carriage of the CC genotype, compared with carriage of the GG genotype, was associated with 1.66-fold increased risk of acute leukemia (OR 1.66, 95% CI 1.17-2.36, **Figure 2**). A slightly higher risk was detected under the recessive comparison model (OR 1.77, 95% CI 1.23-2.54, **Figure 3**). When using the allele comparison model, we

found a 25% higher risk among the individuals with a single C allele (OR 1.25, 95% CI 1.03-1.51, **Figure 4**).

**Association of *NBN* rs2735383 polymorphism with acute leukemia:** We assessed the effects of *NBN* rs2735383 polymorphism on acute leukemia occurrence among 1,553 subjects. The pooled OR in the homozygous comparison model showed no significant association (OR 0.94, 95% CI 0.67-1.30), as displayed in **Figure 2**. The non-significant association persisted in the recessive comparison model (**Figure 3**) and the allele comparison model (**Figure 4**).

**Association of *NBN* 709816 polymorphism with acute leukemia:** In the 1,485 samples for rs709816, we did not find any significant association in any of the comparison models tested (OR 1.03, 95% CI 0.76-1.39 for the homozygous model, **Figure 2**; OR 1.04, 95% CI 0.78-1.38 for the recessive model, **Figure 3**; OR 1.01, 95% CI 0.88-1.16 for the allele model, **Figure 4**).

## NBN and acute leukemia risk



**Figure 3.** Meta-analysis for NBN polymorphisms and acute leukemia risk in the recessive comparison model. Each study was shown by a point estimate of the effect size (OR) (size inversely proportional to its variance) and its 95% confidence interval (95% CI) (horizontal lines). The diamond denotes the pooled OR.

### Heterogeneity test

We detected significant between-study heterogeneity across the studies focusing on rs1805794 and acute leukemia risk in all genetic models adopted ( $P < 0.05$ ). We attempted to explore the source of heterogeneity and found ethnicity was an important factor that affected the homogeneity (data not shown).

### Sensitivity analyses

We performed sensitivity analyses by repeating the meta-analysis while removing one study, finding that the recalculated ORs were not quantitatively altered (data not shown). The procedures indicated that the results of our meta-analysis are reliable and robust.

### Discussion

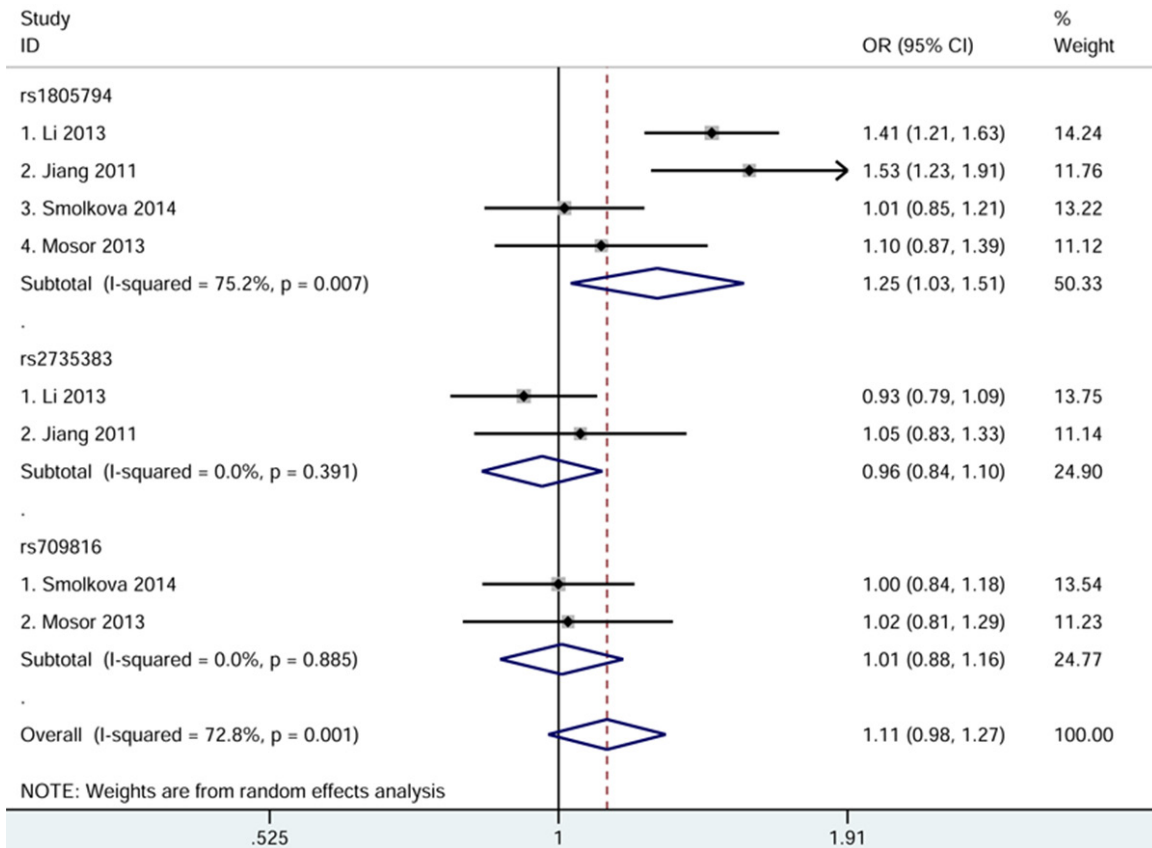
To our knowledge, this is the first meta-analysis that has examined the association of NBN polymorphisms with acute leukemia. Based on the

epidemiological data published to date, we demonstrated evidence supporting the notion that presence of the CC genotype or the C allele of NBN rs1805794 conferred significantly increased susceptibility to acute leukemia. In contrast, we did not find any evidence of an association for s2735383 and rs709816 polymorphisms. These findings should be evaluated with caution in the light of sample inadequacy and additional larger studies are clearly needed to identify the association between NBN polymorphisms and acute leukemia susceptibility.

DSB is a common form of DNA damage especially hazardous to human cells because of the serious consequences if left unrepaired, such as genomic instability, genome rearrangements and tumorigenesis. Previously identified mechanisms involved in the repair of DSBs include microhomology-mediated end joining (MMEJ), homologous recombination, and non-homologous end joining (NHEJ) [25]. The fact that the



## NBN and acute leukemia risk



**Figure 4.** Meta-analysis for *NBN* polymorphisms and acute leukemia risk in the allele comparison model. Each study was shown by a point estimate of the effect size (OR) (size inversely proportional to its variance) and its 95% confidence interval (95% CI) (horizontal lines). The diamond denotes the pooled OR.

MREII/RAD50/*NBN* complex has a key role in recognition and response to distinct types of DNA damage suggests the importance of *NBN* during the process of DSBs repair [26]. Mutations in the *NBN* gene result in defective response to DNA DSBs which has been associated with a chromosomal instability disorder-Nijmegen breakage syndrome [27]. People with this *NBN* mutation-related disease are more likely to suffer a variety of cancers, such as breast cancer, bladder cancer, lung cancer and acute leukemia [24, 28-30]. The causal associations between *NBN* polymorphisms and the former three common cancers have recently been confirmed in multiple meta-analyses where a sufficient number of epidemiologic studies were incorporated [11-13].

Considering the functional importance of *NBN* polymorphisms, and most importantly the status quo that no previous meta-analysis has examined the association of these polymorphisms with risk of acute leukemia, we decided

to use the statistical method to derive a higher statistical power for the measure of interest. rs1805794, but not rs2735383, rs709816, showed a strong association with the risk of developing acute leukaemia, an observation consistent with earlier clinical studies among subjects of Chinese origin [15, 16], and contrary to the studies in European samples [17, 18]. A possible reason for the inconsistency may relate to the sampling variance. The sample is obviously expanded when all data sets are pooled together, contributing to the identification of a significant association that the single studies failed to identify. Another possible explanation is that *NBN* polymorphisms predispose to acute leukemia in an ethnic-specific manner and do not function in European populations. These hypotheses remain to be investigated in future larger studies.

It is believed that different kinds of leukemia have different etiologies. In the present study, however, we did not separately investigate the

risk of ALL and AML associated with *NBN* polymorphisms because of data insufficiency. This constitutes the first weakness of this analysis. Furthermore, the lack of an association between rs2735383, rs709816 and acute leukemia was unexpected. In principle, a study with a limited number is usually underpowered and thereby unlikely to detect the true association. The third weakness refers to the indicated heterogeneity that may arise from differences in intervention, methodology, subject population, and experimental design of the studies concerning rs1805794. The aforementioned points should be noted in order to better understand the current findings.

In conclusion, we examined the association of three polymorphisms in the *NBN* gene with risk of acute leukemia and demonstrated a significant increase in relation to rs1805794. Replication studies containing a large number of samples with different ethnic origins are warranted to validate the genetic susceptibility conferred by *NBN* polymorphisms, and to facilitate a clearer understanding of the mechanisms underlying acute leukemia.

#### Disclosure of conflict of interest

None.

**Address correspondence to:** Yan Wang, Department of Hematology, Shandong Jining No. 1 People's Hospital, Shandong Province, China. E-mail: wangyan862@126.com

#### References

- [1] ab. Leukemia. National Cancer Institute. <http://www.cancer.gov/cancertopics/types/leukemia/>
- [2] ac. What You Need To Know About Leukemia. National Cancer Institute (NCI). <http://www.cancer.gov/cancertopics/wyntk/leukemia/page2>
- [3] Mathers CD, Cynthia Boschi-Pinto, Alan D Lopez and Christopher JL Murray. Cancer incidence, mortality and survival by site for 14 regions of the world. Global Programme on Evidence for Health Policy Discussion Paper No. 13 (World Health Organization) 2001.
- [4] Lozano R, Naghavi M, Foreman K, Lim S, Shibuya K, Aboyans V, Abraham J, Adair T, Aggarwal R, Ahn SY, Alvarado M, Anderson HR, Anderson LM, Andrews KG, Atkinson C, Badour LM, Barker-Collo S, Bartels DH, Bell ML, Benjamin EJ, Bennett D, Bhalla K, Bikbov B, Bin Abdulhak A, Birbeck G, Blyth F, Bolliger I, Boufous S, Bucello C, Burch M, Burney P, Carapetis J, Chen H, Chou D, Chugh SS, Coffeng LE, Colan SD, Colquhoun S, Colson KE, Condon J, Connor MD, Cooper LT, Corriere M, Cortinovis M, de Vaccaro KC, Couser W, Cowie BC, Criqui MH, Cross M, Dabhadkar KC, Dahodwala N, De Leo D, Degenhardt L, Delossantos A, Denenberg J, Des Jarlais DC, Dharmaratne SD, Dorsey ER, Driscoll T, Duber H, Ebel B, Erwin PJ, Espindola P, Ezzati M, Feigin V, Flaxman AD, Forouzanfar MH, Fowkes FG, Franklin R, Fransen M, Freeman MK, Gabriel SE, Gakidou E, Gaspari F, Gillum RF, Gonzalez-Medina D, Halasa YA, Haring D, Harrison JE, Havmoeller R, Hay RJ, Hoen B, Hotez PJ, Hoy D, Jacobsen KH, James SL, Jasrasaria R, Jayaraman S, Johns N, Karthikeyan G, Kassebaum N, Keren A, Khoo JP, Knowlton LM, Kobusingye O, Koranteng A, Krishnamurthi R, Lipnick M, Lipshultz SE, Ohno SL, Mabweijano J, Mactntyre MF, Mallinger L, March L, Marks GB, Marks R, Matsumori A, Matzopoulos R, Mayosi BM, McAnulty JH, McDermott MM, McGrath J, Mensah GA, Merriman TR, Michaud C, Miller M, Miller TR, Mock C, Mocumbi AO, Mokdad AA, Moran A, Mulholland K, Nair MN, Naldi L, Narayan KM, Nasser K, Norman P, O'Donnell M, Omer SB, Ortblad K, Osborne R, Ozgediz D, Pahari B, Pandian JD, Rivero AP, Padilla RP, Perez-Ruiz F, Perico N, Phillips D, Pierce K, Pope CA 3rd, Porrini E, Pourmalek F, Raju M, Ranganathan D, Rehm JT, Rein DB, Remuzzi G, Rivara FP, Roberts T, De Leon FR, Rosenfeld LC, Rushton L, Sacco RL, Salomon JA, Sampson U, Sanman E, Schwebel DC, Segui-Gomez M, Shepard DS, Singh D, Singleton J, Sliwa K, Smith E, Steer A, Taylor JA, Thomas B, Tleyjeh IM, Towbin JA, Truelsen T, Undurraga EA, Venketasubramanian N, Vijayakumar L, Vos T, Wagner GR, Wang M, Wang W, Watt K, Weinstein MA, Weintraub R, Wilkinson JD, Woolf AD, Wulf S, Yeh PH, Yip P, Zabetian A, Zheng ZJ, Lopez AD, Murray CJ, AlMazroa MA and Memish ZA. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet* 2012; 380: 2095-2128.
- [5] ad. <http://www.cancer.gov/researchandfunding/snapshots/leukemia> A Snapshot of Leukemia. National Cancer Institute (NCI).
- [6] Hutter JJ. Childhood leukemia. *Pediatr Rev* 2010; 31: 234-241.
- [7] Tauchi H, Matsuura S, Kobayashi J, Sakamoto S and Komatsu K. Nijmegen breakage syndrome gene, NBS1, and molecular links to factors for genome stability. *Oncogene* 2002; 21: 8967-8980.

## NBN and acute leukemia risk

- [8] Petrini JH and Stracker TH. The cellular response to DNA double-strand breaks: defining the sensors and mediators. *Trends Cell Biol* 2003; 13: 458-462.
- [9] Lu CS, Truong LN, Aslanian A, Shi LZ, Li Y, Hwang PY, Koh KH, Hunter T, Yates JR 3rd, Berns MW and Wu X. The RING finger protein RNF8 ubiquitinates Nbs1 to promote DNA double-strand break repair by homologous recombination. *J Biol Chem* 2012; 287: 43984-43994.
- [10] Tauchi H. Positional cloning and functional analysis of the gene responsible for Nijmegen breakage syndrome, NBS1. *J Radiat Res* 2000; 41: 9-17.
- [11] Wang Z, Cui D and Lu W. NBS1 8360G > C polymorphism is associated with breast cancer risk: a meta-analysis. *Breast Cancer Res Treat* 2010; 123: 557-561.
- [12] Wang L, Cheng J, Gao J, Wang J, Liu X and Xiong L. Association between the NBS1 Glu185Gln polymorphism and lung cancer risk: a systemic review and meta-analysis. *Mol Biol Rep* 2013; 40: 2711-2715.
- [13] Zhang Y, Huang YS, Lin WQ, Zhang SD, Li QW, Hu YZ, Zheng RL, Tang T, Li XZ and Zheng XH. NBS1 Glu185Gln polymorphism and susceptibility to urinary system cancer: a meta-analysis. *Tumour Biol* 2014; 35: 10723-9.
- [14] Zhang G, Zeng Y, Liu Z and Wei W. Significant association between Nijmegen breakage syndrome 1 657del5 polymorphism and breast cancer risk. *Tumour Biol* 2013; 34: 2753-2757.
- [15] Li N, Xu Y, Zheng J, Jiang L, You Y, Wu H, Li W, Wu D and Zhou Y. NBS1 rs1805794G>C polymorphism is associated with decreased risk of acute myeloid leukemia in a Chinese population. *Mol Biol Rep* 2013; 40: 3749-3756.
- [16] Jiang L, Liang J, Jiang M, Yu X, Zheng J, Liu H, Wu D and Zhou Y. Functional polymorphisms in the NBS1 gene and acute lymphoblastic leukemia susceptibility in a Chinese population. *Eur J Haematol* 2011; 86: 199-205.
- [17] Smolkova B, Dusinska M and Hemminki K. NBN and XRCC3 genetic variants in childhood acute lymphoblastic leukaemia. *Cancer Epidemiol* 2014; 38: 563-568.
- [18] Mosor M, Ziolkowska-Suchanek I, Nowicka K, Dzikiewicz-Krawczyk A, Januszkiewicz-Lewandowska D and Nowak J. Germline variants in MRE11/RAD50/NBN complex genes in childhood leukemia. *BMC Cancer* 2013; 13: 457.
- [19] Higgins JP and Thompson SG. Quantifying heterogeneity in a meta-analysis. *Stat Med* 2002; 21: 1539-1558.
- [20] DerSimonian R and Kacker R. Random-effects model for meta-analysis of clinical trials: an update. *Contemp Clin Trials* 2007; 28: 105-114.
- [21] Ioannidis JP and Trikalinos TA. The appropriateness of asymmetry tests for publication bias in meta-analyses: a large survey. *CMAJ* 2007; 176: 1091-1096.
- [22] Egger M, Davey Smith G, Schneider M and Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ* 1997; 315: 629-634.
- [23] Wells GA, Shea B and O'Connell D. Newcastle-Ottawascale. Available from: URL: [http://www.ohri.ca/programs/clinical\\_epidemiology/oxford.htm](http://www.ohri.ca/programs/clinical_epidemiology/oxford.htm). 2006.
- [24] Mosor M, Ziolkowska I, Januszkiewicz-Lewandowska D and Nowak J. Polymorphisms and haplotypes of the NBS1 gene in childhood acute leukaemia. *Eur J Cancer* 2008; 44: 2226-2232.
- [25] Watson JD BT, Bell SP, Gann A, Levine M, Losick R. 5th edition. *Molecular Biology of the Gene*, ch. 9 and 10. Peason Benjamin Cummings: CSHL Press; 2004.
- [26] Matsuura S, Kobayashi J, Tauchi H and Komatsu K. Nijmegen breakage syndrome and DNA double strand break repair by NBS1 complex. *Adv Biophys* 2004; 38: 65-80.
- [27] Digweed M and Sperling K. Nijmegen breakage syndrome: clinical manifestation of defective response to DNA double-strand breaks. *DNA Repair Amst* 2004; 3: 1207-1217.
- [28] Silva SN, Tomar M, Paulo C, Gomes BC, Azevedo AP, Teixeira V, Pina JE, Rueff J and Gaspar JF. Breast cancer risk and common single nucleotide polymorphisms in homologous recombination DNA repair pathway genes XRCC2, XRCC3, NBS1 and RAD51. *Cancer Epidemiol* 2010; 34: 85-92.
- [29] Choudhury A, Elliott F, Iles MM, Churchman M, Bristow RG, Bishop DT and Kiltie AE. Analysis of variants in DNA damage signalling genes in bladder cancer. *BMC Med Genet* 2008; 9: 69.
- [30] Qiu Y, Lin Y. Association of smoking, residential environment and polymorphisms with lung cancer. *Occup Med* 2011; 28: 133-136-140.