Prognostic value of interleukin-6 in atrial fibrillation: A cohort study and meta-analysis

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Abstract

Objective: The prognostic value of interleukin-6 (IL-6) in patients with atrial fibrillation (AF) has not been fully elucidated. Therefore, we conducted a cohort study and a meta-analysis to assess the predictive value of IL-6 for stroke and mortality in patients with AF.

Methods: A cohort study was performed in newly diagnosed non-valvular patients with AF. A total of 217 patients with AF were followed up for a mean of 27 months. A multivariate Cox regression analysis was used to evaluate the association between IL-6 and stroke/all-cause mortality. The incremental value was also assessed by adding IL-6 to the CHA2DS2-VASc score. Besides, a meta-analysis of all reported cohort studies and our cohort study was conducted to validate the association of circulating IL-6 and stroke/mortality in patients with AF.

Results: Our cohort study showed that elevated plasma level of IL-6 was an independent risk factor for predicting stroke [hazard ratio (HR)=3.81; 95% confidence interval (CI), 1.11–13.05; p=0.033] and all-cause mortality (HR=3.11; 95% CI, 1.25–7.72; p=0.015) in patients with AF. Adding IL-6 levels to CHA2DS2-VASc score showed limited improvement of the predictive power for stroke [area under curve (AUC) from 0.81 to 0.88, p=0.006]. Meta-analysis confirmed that increased circulating level of IL-6 was significantly associated with increased risk of stroke (pooled HR=1.97; 95% CI, 1.22–3.17; p=0.006) and all-cause mortality (pooled HR=2.73; 95% CI, 2.29–3.25; p<0.001).

Conclusion: Increased circulating level of IL-6 was significantly associated with greater risk of stroke and all-cause mortality in patients with AF. Adding IL-6 biomarker to the CHA2DS2-VASc score may help to determine the management of AF treatment.

Keywords: atrial fibrillation, prognosis, interleukin-6, meta-analysis, stroke, mortality

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Introduction

Atrial fibrillation (AF) is the most common arrhythmia in clinical practice (1). The prevalence of AF is estimated to be 0.4% to 1.0% in the general population (2). AF increases the risk of stroke, heart failure, and overall mortality (3). In patients with AF, the annual incidence of ischemic stroke is about 5%, 2–7 times of that in patients without AF (4), and a doubling in mortality rate (5). Therefore, it is crucial to prevent the occurrence of adverse cardiovascular events and improve the prognosis of patients with AF.

Thromboembolism is the main cause of death and disability in patients with AF, and stroke is the most common type of

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HIGHLIGHTS

- The elevated plasma level of interleukin (IL) 6 was an independent risk factor for predicting stroke and all-cause mortality in patients with atrial fibrillation (AF).
- Adding IL-6 biomarker to the CHA2DS2-VASc score may help determine the management of AF treatment.
- IL-6 is a promising prognostic biomarker to help optimize the strategies for risk stratification in patients with AF.

manifestation (6). The mechanism of occurrence and development of AF is considered to be multifactorial. Several risk stratifications scoring schemes have been developed to determine which patients with AF have higher stroke risk. The most widely used scoring scheme is the CHA2DS2-VASC score (7). However, there remains a need for improving the predictive ability of CHA2DS2-VASc score. Studies have found that adding biomarkers such as von Willebrand factor, D-dimer, N-terminal proB-type natriuretic peptide, and high-sensitivity troponin T to the CHADS2 score improved the predictive accuracy (8, 9).

Although there is some evidence indicating that inflammation may be associated with AF (10), it may underlie pathogenesis of arrhythmia and vascular events (11). IL-6 is a pleiotropic cytokine, which can induce a pro-thrombotic state by increasing the expression of Şbrinogen, tissue factor, factor VIII, and von Willebrand factor, as well as by activating endothelial cells and increasing platelet production, which may contribute to the adverse outcome of AF (12, 13). Several studies have evaluated the associations between circulating level of IL-6 and stroke/mortality in patients with AF (14, 15), but the results lack consistency. Therefore, we conducted a prospective cohort study to determine the association of plasma level of IL-6 with stroke and all-cause mortality in patients with AF. In addition, we performed a meta-analysis to further validate the prognostic value of IL-6 in AF.

Methods

Cohort study

Patients

We consecutively recruited newly diagnosed patients with AF aged at least 18 years and hospitalized in Southwest Hospital of the Army Medical University in Chongqing, China, from December 2013 to August 2015. The diagnosis of AF was made according to the 2012 European Guidelines for Atrial Fibrillation (16). Exclusion criteria included mitral rheumatic valve disease or a prosthetic valve, infections, malignant tumors, connective tissue diseases (such as rheumatoid arthritis), other acute or chronic inflammatory diseases (such as giant cell arteritis). We collected demographic, epidemiological, and medical information from medical records and structured interviews. The study was approved by the Ethics Committee of Southwest Hospital of the Army Medical University. Written informed consent was obtained from all the patients.

Outcomes and follow up

Primary outcomes included all-cause mortality and stroke. All the patients were actively followed up annually by phone interviews. Stroke was defined as the first neurologic deficit that lasted for at least 24 hours and sub-classified as ischemic (with or without hemorrhagic conversion), hemorrhagic, or uncertain after discharge. The diagnosis of stroke was confirmed by screening the medical records. Next-of-kin reports, electronic medical notes, and the national register of death were used to verify the annual vital status and the cause of death of the patients. Patients known to be alive were censored at the time of last contact.

Measurement of IL-6

A fasting blood sample (5 mL) was drawn from each patient before any treatment. The plasma was separated into aliquots of 500-µL straws and frozen at –80°C immediately after processing until use. The plasma level of IL-6 was measured using MILLIPLEX MAP Human Th17 Magnetic Bead Panel kits (Millipore, Billerica, MA., USA) based on the Luminex xMAP technology (Luminex Corporation, Austin, TX., USA). Plates were run on the Luminex MagPix machine (Luminex Corporation). Raw data were collected using the Luminex xPONENT 4.2 software and analyzed using MILLIPLEX Analyst 5.1 software (Millipore). Concentrations of IL-6 were calculated using a standard curve. Two duplicate samples were run for quality control (replicate QC1 samples, low level; replicate QC2 samples, high level). The coefficients of variation of all repeated quality control samples were less than 10%.

Statistical analysis

Frequency counts and proportions were used to report categorical data. For the description of continuous variables, means and standard deviation were used for normally distributed data, and median and interquartile ranges (25^{th}–75^{th} percentile) were used for non-normally distributed data. The best cutoff value of IL-6 was determined by X-tile software, which is a free software available from Yale University School of Medicine that can determine a cutoff point for continuous data with a time-dependent outcome (17). Univariate and multivariate Cox proportional hazards regression models were used to evaluate the association of IL-6 with stroke and all-cause mortality in patients with AF. Baseline variables with $p<0.05$ in univariate Cox regression model were included as covariates in the multivariate Cox regression model. Hazard ratios (HR) and 95% confidence interval (CI) were estimated. Kaplan-Meier curves were used to depict the cumulative risk of stroke and all-cause mortality in patients with AF by different IL-6 levels, and the significance of their differences was assessed using log-rank tests. A CHA2DS2-VASc stroke risk score (including congestive heart failure; hypertension; age ≥75 years;

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diabetes; stroke, transient ischemic attack, or thromboembolism; vascular disease; age 65–74 years, and sex) was assigned to each patient. However, these schemes have only a modest predictive value for predicting "high-risk" patients. C-statistics were estimated to compare the predictive power between modified models, using methods recommended by Pencina et al. (18). Besides, we used the time-dependent area under the receiver operating characteristic curve (AUC) to compare the predictive ability between modified models (adding IL6 to the CHA2DS2-VASc score) (19). A 2-sided p value <0.05 was considered to be statistically significant. All these statistical analyses were conducted using the Statistical Package for the Social Sciences statistical software (version 20.0; IBM Corp., Armonk, NY, USA) and R 3.5.2 (R Foundation for Statistical Computing, Vienna, Austria).

Search strategy and inclusion criteria of meta-analysis

We searched PubMed, EMBASE, Springer Link, Web of Science, CNKI, and supplemented with Google scholar search engine for relevant studies published from database establishment to January 2020. We used the following keywords, "interleukin 6," "inflammatory markers," "biomarker," "prognosis," and "atrial fibrillation." In addition, relevant review articles were also cross-referenced. The included studies had to satisfy the following predefined criteria: (i) the study was a cohort study; (ii) the study enrolled patients with AF; (iii) stroke or mortality events were assessed as outcomes; (iv) the study investigated the association between IL-6 and outcome events. If studies were duplicated, the one with the most complete data was chosen.

Data extraction and quality assessment of meta-analysis

Two investigators (Xiaoyue Jia and Na Wu) independently extracted the data using a standard data extraction form. We extracted general data (first author's name, year of publication), study characteristics (country and study design), types of AF, mean age of participants, mean follow-up time, adverse outcomes, and information related to effect size like HR, 95% CI, and cutoff values. If both univariate analysis and multivariate analysis were available for the studies, the adjusted HR (95% CI) from the multivariate analysis with a maximum number of adjusted variables were extracted priority.

The quality of studies was independently assessed by two reviewers (Xiaoyue Jia and Xi Cheng). The assessment was done using the primary criteria for non-randomized studies described in the Newcastle-Ottawa scale. The total scores ranged from 0 to 9 for these studies (Supplementary Table 1). Any disagreement between the reviewers was discussed and resolved by consensus.

Statistical analysis of meta-analysis

Statistical heterogeneity across studies was assessed using the I^2 statistic, and heterogeneity was considered to be significant if I^2 >50%. A fixed-effects model was used to calculate pooled effect sizes when I^2 ≤50%. Otherwise, a random-effects model was applied. Funnel plot with or without contour enhancement was applied to detect publication bias owing to small study effects. RevMan 5.3 software (version 5.3, Cochrane Collaboration, Oxford, United Kingdom) was used to perform the meta-analysis.

Results

Cohort study

A total of 232 patients with newly diagnosed non-valvular AF were enrolled in the cohort, and 217 (54.38% men) patients with a mean age of 63.41 years were followed up. The median follow-up time was 27 months (IQR 23–30 months). During follow-up, all-cause mortality rate was 11.06% and the incidence rate of stroke was 6.91% (Table 1). The optimal IL-6 cut-off values was 55.20 pg/mL.

Age and left atrial diameter (LAD) in patients with AF and stroke or death was significantly larger than that without stroke or survivor (both p<0.05) (Supplementary Table 2). According to the result of univariate Cox regression analysis, age, history of coronary heart disease, and heart failure were significantly associated with all-cause mortality. Age, history of stroke, and diabetes were significant risk factors of stroke (Supplementary Tables 3, 4). These significant variables were further adjusted in the multivariate Cox regression model. The results of multivariate cox regression analysis indicated that the increased plasma level of IL-6 was significantly associated with increased risk of all-cause mortality and stroke with adjusted HR 3.81 (95% CI 1.11–13.05; p=0.033) and 3.11 (95% CI 1.25–7.72; p=0.015), respectively (Table 2). The Kaplan-Meier curve showed that patients with elevated IL-6 had a higher risk of all-cause mortality (p<0.05) and for stroke (p>0.05) (Fig. 1 and 2).

We further evaluated the predictive power of the model by adding IL-6 bio-markers to the CHA2DS2-VASc score. Adding high IL-6 to the CHA2DS2-VASC score had a better predictive power for stroke, AUC was improved from 0.81 to 0.88 significantly at 12-month follow-up time (p=0.006), whereas the improvement was

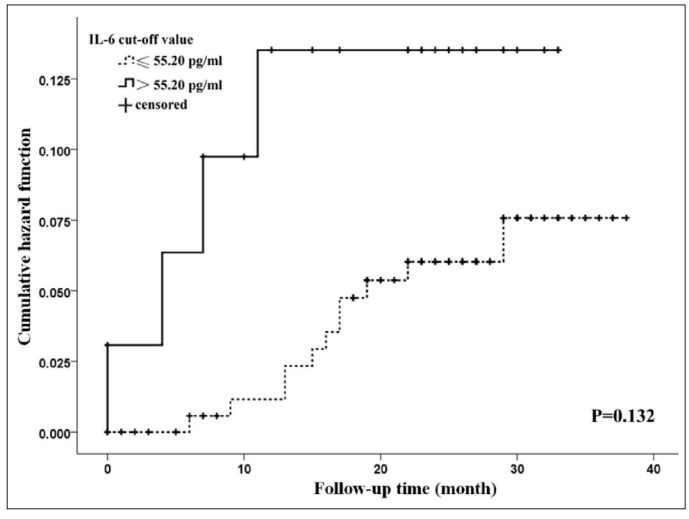

Figure 1. Cumulative risk of stroke in patients with atrial fibrillation at different plasma interleukin-6 levels

Table 1. Baseline characteristics of patients with AF and factors associated with death or stroke

Patient characteristics	Number of patients (%)
Age, mean ± SD, years	63.41±12.20
Sex	
Male	118 (54.38)
Female	99 (45.62)
BMI, mean ± SD, kg/m^2	24.32±3.59
Education	
Junior school and below	173 (79.72)
High school and above	44 (20.27)
Income (10,000 RMB/year)	
<2.5	110 (50.69)
≥2.5	107 (49.31)
AF type	
Paroxysmal	68 (31.34)
Persistent	149 (68.66)
Smoking status	
Former/current	73 (33.64)
Never	144 (66.36)
Drinking status	
Former/current	76 (35.02)
Never	141 (64.98)
Warfarin treatment	
Yes	65 (29.96)
No	152 (70.04)
Statin treatment	
Yes	97 (44.70)
No	120 (55.30)
History of combined diseases	
Hypertension	109 (50.23)
Diabetes	33 (15.21)
Coronary heart disease	83 (38.25)
Cardiomyopathy	22 (10.14)
Heart failure	78 (35.94)
TIA	9 (4.15)
Vascular diseases	14 (6.45)
Previous stroke	18 (8.29)
IL-6, median (range), pg/mL	28.3 (0.3–216.2)
≤55.2	184 (84.79)
>55.2	33 (15.21)
CHA2DS2-VASc score	
<2	68 (32.72)
≥2	146 (67.28)
Follow-up time, mean (IQR), month	27 (23–30)
stroke	15 (6.91)
All-cause mortality	24 (11.06)

AF - atrial fibrillation; BMI - body mass index; IL - interleukin; SD - standard deviation; TIA - transient ischemic attack

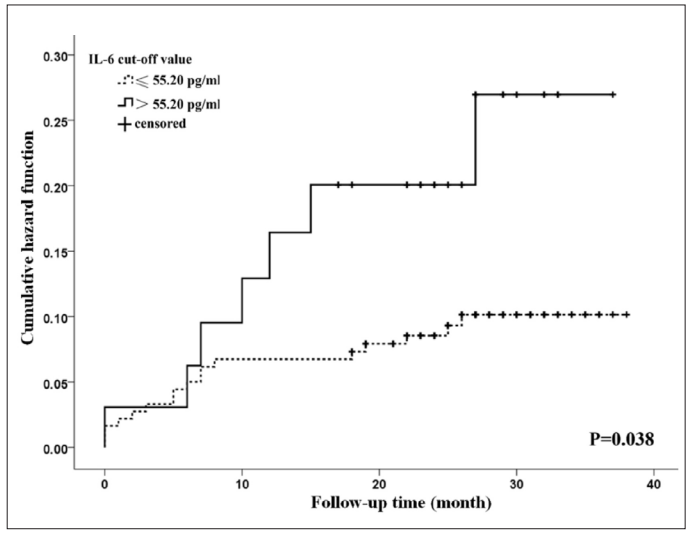

Figure 2. Cumulative risk of all-cause mortality in patients with atrial fibrillation at different plasma interleukin-6 levels

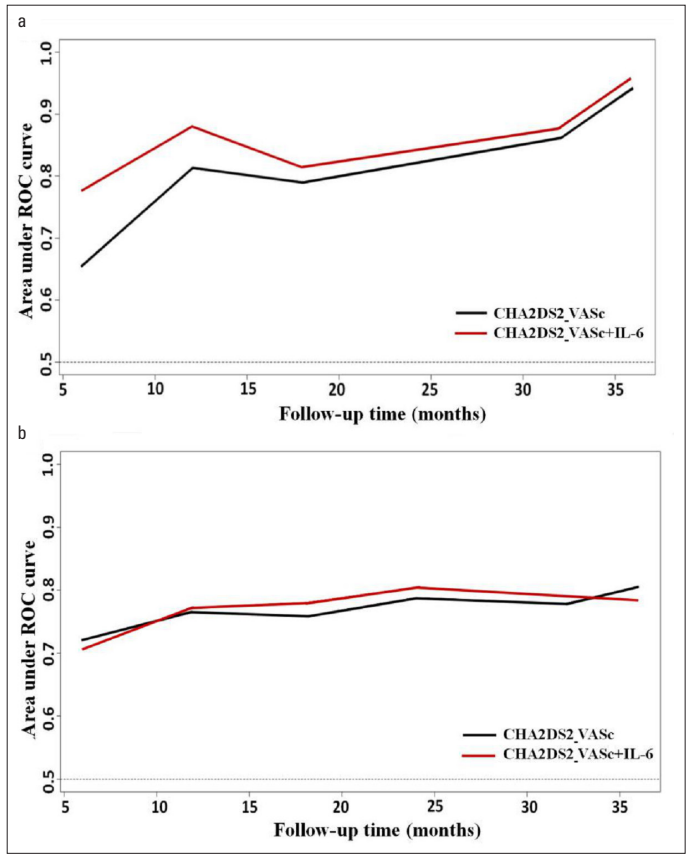

Figure 3. Area under the receiver operating curves (AUC) using a time-dependent receiver operating curve analysis. (a) Time-dependent AUC of the two scores for stroke. (b) Time-dependent AUC of the two scores for all-cause mortality

non-statistically significant in all-cause mortality (AUC from 0.76 to 0.77, p=0.635) (Fig. 3). For stroke, the c-statistic increased from 0.79 (95% CI, 0.69–0.88) to 0.83 (95% CI, 0.74–0.93) when high IL-6 was added (p=0.213). As for the all-cause mortality, the discrimination index c-statistic was 0.75 (95% CI, 0.68–0.82) in the prediction

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Table 2. Associations between plasma IL-6 level and stroke and all-cause mortality in patients with AF

	Univariate Cox regression		Multivariate Cox regression	
Variable	**HR (95% CI)**	***P*-value**	**HR (95% CI)**	***P*-value**
Stroke				
IL-6 (pg/mL)				
≤55.2	Reference		Reference	
>55.2	1.69 (0.61, 4.66)	0.312	3.81 (1.11, 13.05)	0.033
All-cause mortality				
IL-6 (pg/mL)				
≤55.2	Reference		Reference	
>55.2	2.46 (1.02, 5.93)	0.045*	3.11 (1.25, 7.72)	0.015*

*$P<0.05$, the difference was statistically significant.
Cox proportional hazards model for stroke adjusted for age, types of AF, history of diabetes and history of stroke; for all-cause mortality, adjusted for age, types of AF, history of coronary disease and history of heart failure; using a backward selection strategy.
AF - atrial fibrillation; CI - confidence interval; HR - hazard ratio; IL - interleukin

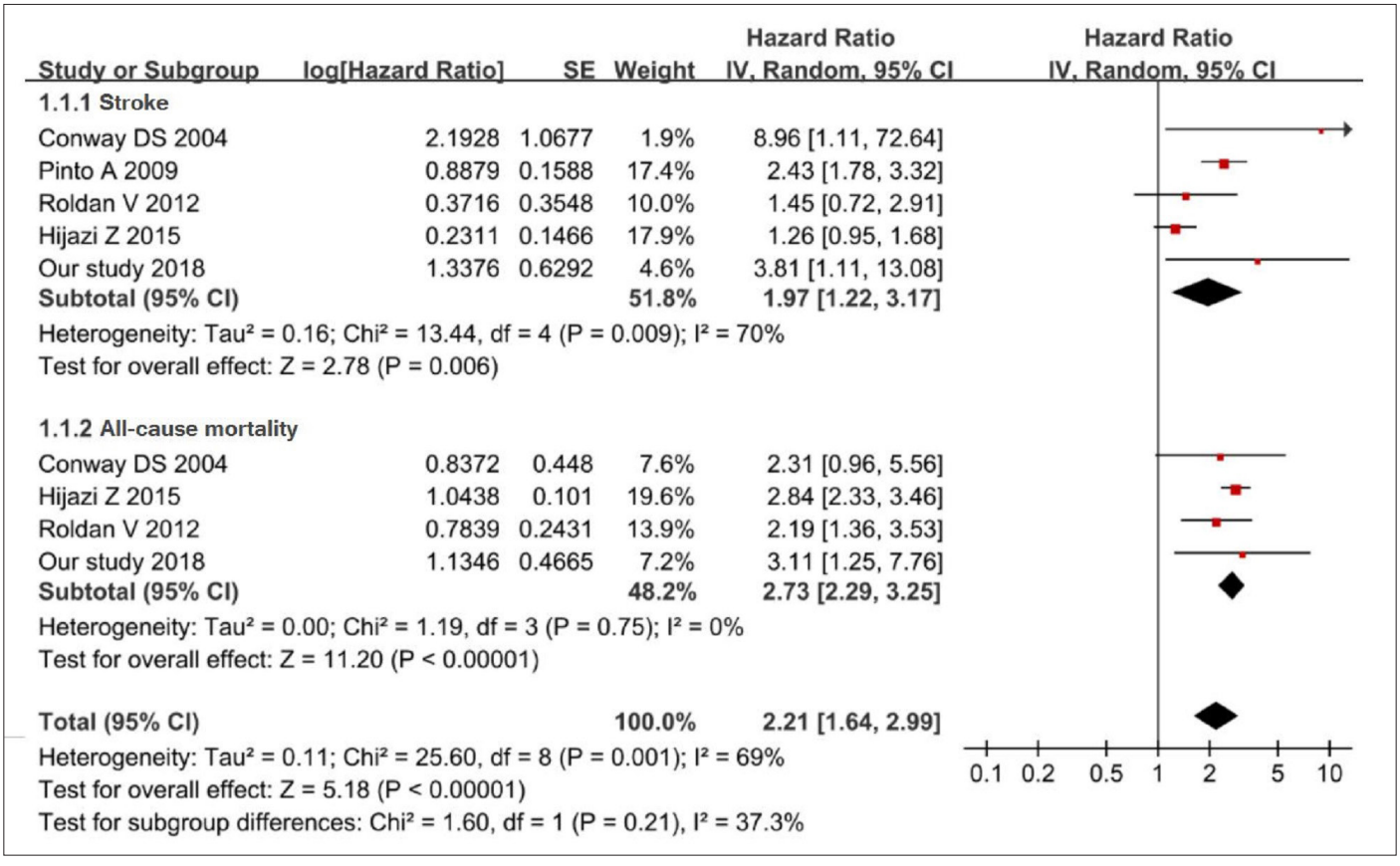

Figure 4. Forest plot of the relationship between the interleukin-6 level and outcomes. The squares and horizontal lines represent the hazards ratio (HR) and 95% confidence interval (CI), respectively. The area of the squares reflects the weight of each study. The diamond represents the pooled HR and 95% CI

model consisting only of the CHA2DS2-VASc score. Adding high IL-6 to the score increased the c-statistic to 0.77 (95% CI, 0.69-0.84), while it was not statistically significant (p=0.460).

Meta-analysis

A total of 299 studies were retrieved after initial search. After screening the titles and abstracts, 270 studies were excluded. The remaining 29 studies were retrieved for the full text. Finally, 4 studies (20-23) met the inclusion criteria and a total of 16,334 participants were included in the final analysis (Supplementary Fig. 1). All the 4 included cohort studies investigated the predictive role of IL-6 for stroke, and 3 studies (20, 21, 23) with 15,961 patients investigated the predictive role of IL-6 for all-cause mortality.

The mean age ranged from 63.0 to 76.0 years. The duration of follow-up for cohort studies ranged from 1.8 to 6.4 years, and all studies had a quality score of at least 5. The detailed characteristics and NOS score are depicted in Supplementary Table 5. Meta-analysis of 4 published studies and our cohort study showed that higher level of IL-6 was significantly associated with an increased risk of stroke events. The pooled HR is 1.97 (95% CI, 1.22-3.17; $p<0.05$) with a medium heterogeneity ($I^2=70\%$, $p=0.001$) across the studies by random effects model (Fig. 4). A sensitivity analysis was performed, and the result showed that the study of Hijazi et al. (23) might be the source of heterogeneity. Compared with other studies, the study by Hijazi et al. (23) had shorter follow-up time (less than 2 two years), and there is one more inclusion criterion of patients with AF (at least one of the risk factors for stroke was required), which may lead to clinical heterogeneity. After excluding the study of Hijazi et al. (23), the pooled HR of stroke was 2.34 (95% CI, 1.78–3.09; $p<0.05$) with a low heterogeneity ($I^2=26\%$, $p=0.001$) (Supplementary Fig. 2).

Four studies (including our study) examined the association between IL-6 and all-cause mortality. The pooled HR of all-cause mortality was 2.73 (95% CI, 2.29–3.25; $p<0.001$) with no significant heterogeneity ($I^2=0\%$, $p=0.75$) across the studies, indicating that elevated plasma level of IL-6 was significantly associated with increased risk of all-cause mortality.

Discussion

We explored the association between IL-6 and the prognosis of patients with AF in a cohort study and meta-analysis. Our results showed that elevated IL-6 levels were significantly associated with higher risk of stroke and all-cause mortality in patients with AF. Adding plasma level of IL-6 could limit the improvement of the predictive power of CHA2DS2-VASc scores for stroke and all-cause mortality.

At present, the association of IL-6 with AF prognosis remains controversial. Most studies show that the increase of IL-6 is an independent risk factor for stroke or mortality in patients with AF (24, 25). However, a few studies (23) reported that IL-6 level was not related to the risk of stroke in patients with AF. Our cohort study and meta-analysis consistently revealed that elevated IL-6 level was related to higher risk of stroke and death, independent of established clinical risk factors.

The pathophysiological mechanism involved in inflammation and the prognosis of AF remains undetermined. IL-6 is an established inflammatory biomarker, which is a circulating cytokine produced by monocytes, T lymphocytes, and epithelial cells (26). IL-6 also increases platelet production and platelet sensitivity to thrombin, stimulates transcription of fibrinogen, and is linked to endothelial cell activation and damage (27, 28). As a pleiotropic cytokine, IL-6 can induce CRP production by hepatocytes and is involved in immuno-inflammatory reaction by promoting neutrophil adhesion, atherosclerotic plaque rupture, and thrombus formation (29). It is therefore plausible that IL-6 inducing inflammation might mediate the prothrombic or hypercoagulable state that exists in AF (30), with consequences for left atrial thrombosis, stroke, and vascular events.

On the basis of clinical risk factors, the CHA2DS2-VASc score is the most commonly used stratification scheme to assess the risk of stroke in patients with AF and guide the use of anticoagulants, although its assessment ability and predictive accuracy is modest (31, 32). To increase prediction ability, incorporation of inflammatory biomarkers to CHA2DS2-VASc score become a hot research field (33, 34). Our cohort study indicated that modifying the CHA2DS2-VASc score by adding IL-6 could limit the improvement of the prediction ability for stroke and all-cause mortality in patients with AF. Further epidemiological studies with large samples are needed to verify the clinical significance of our results.

Study limitations

Several limitations of this study should be considered in interpreting our results. Heterogeneity tests showed that there was significant heterogeneity for some studies investigating the predictive role of IL-6 for stroke, and no significant heterogeneity for studies examining the association between IL-6 and all-cause mortality. Because of the limited number of studies, our sensitivity analysis showed that the study of Hijazi et al. (23) might be the potential source of heterogeneity. Comparing the characteristics of the included studies, we found that mean age of participants, mean follow-up time, time points of the sample assay, and cut-off point varied among studies. In previous studies of meta-analysis, the cut-off values of IL-6 for predicting adverse outcomes were 3.35, 20, 0.89, and 2.3 pg/mL, respectively, (20-23) lower than our study. The cut-off value of IL6 in Hijazi et al. (23) and Roldan et al. (20) studies was the median of anticoagulant patients. The difference in assay methods might also partly contribute to the high heterogeneity among the studies. As a single-center study, our cohort study might have selection bias. Because of anticoagulant treatment rate in China is relatively low (35, 36), only 65 (30.0%) patients received warfarin anti-coagulation in our cohort study, which may also introduce bias. Another limitation is the small sample size and insufficient follow-up time, which may have led to insufficient statistical power. Finally, although funnel plots showed no obvious publication bias (Supplementary Fig. 3) in our meta-analysis, the efficiency of publishing bias test is low because of the limited number of studies. Lau et al. (37) doubted that the widely used method of the funnel plot accurately predicts publication bias. A true standard measure of publication bias would require prospective registries of studies with detailed knowledge of which studies had been published and which were unpublished. Therefore, publication bias cannot be completely excluded.

Conclusion

In conclusion, our study suggested that high levels of IL-6 were significantly associated with stroke and death and other adverse outcomes in patients with AF. Moreover, compared to

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the classical CHADS-VASC score, adding IL-6 levels to CHADS-VASC score showed limited improvement of the predictive power. IL-6 is a promising prognostic biomarker to help optimize the strategies for risk stratification, treatment, and prevention of adverse outcomes in patients with AF.

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Author contributions: Concept – L.Z., Y.L.; Design – X.J., X.C., N.W., L.Z., Y.L.; Supervision – X.J., X.C., N.W., L.Z., Y.L.; Fundings – N.W.; Materials – X.C., B.X.; Data collection &/or processing – X.J., X.C., N.W., C.L., Z.Z., S.T.; Analysis &/or interpretation – X.J., X.C., N.W., Y. X.; Literature search – Y. X., L.W.; Writing – X.J., X.C., N.W.; Critical review – L.Z., Y.L.

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Supplementary Table 1. Quality assessment score scale

Item	Score
NOS score scale for cohort studies*	
Selection	
(1) Representativeness of the exposed cohort	
Truly representative of the average status in the community	1
Somewhat representative of the average status in the community	1
Selected group of users (e.g. nurses, volunteers)	0
No description of the derivation of the cohort	0
(2) Selection of the non-exposed cohort	
Drawn from the same community as the exposed cohort	1
Drawn from a different source	0
No description of the derivation of the non-exposed cohort	0
(3) Ascertainment of exposure	
Secure record (e.g. surgical records)	1
Structured interview	1
Written self-report	0
No description	0
(4) Demonstration that outcome of interest was not present at the start of study	
Yes	1
No	0
Comparability	
Comparability of cohorts on the basis of the design or analysis	
Study controls for the most important factor	1
Study controls for any additional factor	1
Outcome	
(1) Assessment of outcome	
Independent blind assessment	1
Record linkage	1
Self-report	0
No description	0
(2) Was follow-up long enough for outcomes to occur?	
Yes	1
No	0
(3) Adequacy of follow up of cohorts	
Complete follow-up - all subjects accounted for	1
Subjects lost to follow-up unlikely to introduce bias	1
Follow-up rate is low and no description of those lost	0
No statement	0

*A study can be awarded a maximum of one score for each numbered item within the Selection and Outcome categories. A maximum of two scores can be given for Comparability.

Supplementary Table 2. Baseline information of patients with AF with different outcomes

	Death outcome			Stroke outcome		
Variable	**Death (24)**	**Survivor (193)**	***P*-value**	**Stroke (15)**	**Without stroke (202)**	***P*-value**
Age (years)	73.71±8.73	62.13±11.97	<0.001	69.93±9.77	62.93±12.24	0.032
Sex			0.982			0.993
Male	13 (54.17%)	105 (44.40%)		8 (53.33%)	110 (54.46%)	
Female	11 (45.83%)	88 (45.60%)		7(46.67%)	92 (45.54%)	
BMI (kg/m^2)	25.06±4.29	24.23±3.50	0.289	23.18±3.24	24.40±3.61	0.205
Income (10,000 RMB/year)	0.170			0.391		
<2.5	9 (37.50%)	101 (52.33%)		6 (40.00%)	104 (51.49%)	
≥2.5	15 (62.50%)	92 (47.27%)		9 (60.00%)	98 (48.51%)	
Smoking	7 (29.17%)	66 (34.20%)	0.623	3 (20.00%)	70 (34.65%)	0.246
Drinking	8 (33.33%)	68 (35.23%)	0.854	3 (20.00%)	73 (36.14%)	0.206
Treatment						
Warfarin	8 (33.33%)	57 (29.53%)	0.702	7 (46.67%)	58 (28.71%)	0.153
Statins	14 (58.33%)	83 (43.01%)	0.154	8 (53.33%)	89 (44.06%)	0.486
Comorbidity						
Hypertension	16(66.67%)	93 (48.19%)	0.088	10 (66.67%)	99 (49.01%)	0.187
Diabetes	5 (20.83%)	28 (14.51%)	0.379	6 (40.00%)	27 (13.37%)	0.014
Coronary heart disease	14(58.33%)	69 (35.75%)	0.032	7 (46.67%)	76 (37.62%)	0.487
Cardiomyopathy	4 (16.67%)	18 (9.33%)	0.278	0 (0.00%)	22 (10.89%)	0.374
Heart failure	19(79.17%)	59 (30.57%)	<0.001	7 (46.67%)	71 (35.15%)	0.370
TIA	0 (0.00%)	9 (4.66%)	0.602	2 (13.33%)	7 (3.47%)	0.121
Vascular diseases	2 (8.33%)	12 (6.22%)	0.657	2 (13.33%)	12 (5.94%)	0.250
Previous stroke	2 (8.33%)	16 (8.29%)	1.000	5 (33.33%)	13 (6.44%)	0.004
Echocardiograhic parameters						
LAD, mm	49.74±8.37	44.28±9.76	0.001	50.2±11.14	44.47±9.54	0.028
LVEDD, mm	57.47±10.69	50.63±8.23	0.007	51.07±8.09	51.43±8.85	0.879
Left ventricular EF, %	45.52±12.94	56.65±12.81	<0.001	55.87±13.26	55.37±13.30	0.888

AF - atrial fibrillation; BMI - body mass index; EF - ejection fraction; LAD - left atrial diameter; LVEDD - left ventricular end diastolic diameter; TIA - transient ischemic attack

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Supplementary Table 3. Univariate Cox regression analysis on association between plasma IL-6 level and stroke and all-cause mortality in patients with AF

	Stroke		All-cause mortality	
Variable	**HR (95% CI)**	***P*-value**	**HR (95% CI)**	***P*-value**
Age (years)	2.02 (1.10, 3.71)	0.024*	3.29 (1.94, 5.60)	<0.001*
<65	Reference		Reference	
65~74	1.72 (0.46, 6.41)	0.170	3.16 (0.89, 11.21)	0.074
≥75	4.03 (1.22, 13.32)	0.022*	10.71 (3.51, 32.63)	< 0.001*
Sex				
Male	Reference		Reference	
Female	1.09 (0.47, 2.51)	0.848	1.02 (0.46, 2.28)	0.955
BMI (kg/m^2)				
<24	Reference		Reference	
≥24	0.34 (0.11, 1.08)	0.067	0.96 (0.43, 2.15)	0.929
AF type				
Paroxysmal	Reference		Reference	
Persistent	1.36 (0.43, 4.28)	0.597	3.35 (0.99, 11.22)	0.050
Education				
Junior school and below	Reference		Reference	
High school and above	1.05 (0.29, 3.77)	0.940	0.81 (0.28, 2.40)	0.709
Income (10,000 RMB/year)				
<2	Reference		Reference	
≥2	1.61 (0.57, 4.53)	0.365	1.79 (0.78, 4.09)	0.169
Smoker				
No	Reference		Reference	
Yes	0.29 (0.07, 1.31)	0.107	0.81 (0.34, 1.96)	0.640
Drinking				
No	Reference		Reference	
Yes	0.29 (0.07,1.29)	0.103	0.94 (0.40, 2.20)	0.942
Warfarin				
No	Reference		Reference	
Yes	2.09 (0.76, 5.75)	0.156	1.15 (0.49, 2.69)	0.748
Statins				
No	Reference		Reference	
Yes	1.48 (0.54, 4.08)	0.450	1.77 (0.79, 3.99)	0.167
Comorbidity				
Hypertension	2.08 (0.71, 6.09)	0.181	2.06 (0.88, 4.82)	0.095
Diabetes	4.16 (1.48, 11.69)	0.007*	1.52 (0.56, 4.06)	0.409
Coronary disease	1.54 (0.56, 4.25)	0.404	2.40 (1.07, 5.41)	0.034*
Cardiomyopathy	0.04 (0.00, 66.41)	0.399	1.82 (0.62, 5.33)	0.274
Heart failure	1.79 (0.65, 4.94)	0.260	7.45 (2.78, 9.97)	<0.001*
TIA	1.86 (0.88, 3.92)	0.102	0.22 (0.01, 15.60)	0.482
Vascular diseases	2.57 (0.58, 11.41)	0.214	1.40 (0.33, 5.95)	0.650
Stroke	2.41 (1.41, 4.12)	0.001*	0.98 (0.47, 2.01)	0.949
IL-6 (pg/mL)				
≤55.2	Reference		Reference	
>55.2	1.69 (0.61, 4.66)	0.312	2.46 (1.02, 5.93)	0.045*

*P<0.05, the difference was statistically significant
AF - atrial fibrillation; BMI - body mass index; CI - confidence interval; HR - hazards ratio; IL - interleukin; TIA - transient ischemic attack

Supplementary Table 4. Multivariate Cox regression analysis on association between plasma IL-6 level and stroke and all-cause mortality in patients with AF

	Stroke		All-cause mortality	
Variable	**HR (95% CI)**	***P*-value**	**HR (95% CI)**	***P*-value**
Age (years)	2.06 (1.08, 3.90)	0.027*	3.10 (1.70, 5.67)	0.001*
<65	Reference		Reference	
65~74	1.55 (0.41, 5.84)	0.519	2.41 (0.67, 8.68)	0.180
≥75	4.22 (1.23, 14.54)	0.022*	8.91 (2.64, 30.03)	0.001*
Comorbidity				
Diabetes	3.34 (1.13, 9.82)	0.029*	-	-
Coronary disease	-	-	1.10 (0.45, 2.64)	0.833
Heart failure	-	-	5.41 (1.99, 14.65)	0.001*
Stroke	2.11 (1.21, 3.70)	0.009*	-	-
IL-6 (pg/mL)				
≤55.2	Reference		Reference	
>55.2	3.81 (1.11, 13.05)	0.033*	3.11 (1.25, 7.72)	0.015*

*$P<0.05$, the difference was statistically significant; - unavailable.
Cox proportional hazards model for stroke adjusted for age, types of AF, history of diabetes and history of stroke; for all-cause mortality, adjusted for age, types of AF, history of coronary disease and history of heart failure; using a backward selection strategy.
AF - atrial fibrillation; CI - confidence interval; HR - hazard ratio; IL - interleukin

Supplementary Table 5. Characteristics of studies included for meta-analysis

First author	Year	Country	Study design	AF patients (n)	Male (%)	Mean age	Mean follow-up time (days)	Adverse outcome	Cut-off value	NOS score
Roldan et al. (20)	2012	UK	Cohort	930	470 (50.5)	76	975	Stroke/TIA, Heart failure, Cardiovascular events	3.35 pg/mL	8
Conway et al. (21)	2004	UK	Cohort	77	44 (57.1)	68	2305	Stroke, death, heart failure	20 pg/mL	6
Pinto et al. (22)	2009	Italy	Cohort	373	237 (63.5)	66.08	1095	stroke	0.89 pg/mL	9
Hijazi et al. (23)	2015	Sweden	Cohort	14954	9630 (67.1)	70	694	Stroke/embolism, Myocardial infarctions bleeding, death	2.3 pg/mL	6
Our study	2018	China	Cohort	217	118 (54.4)	63.41	760	Stroke, death	55.20 pg/mL	7

NOS - Newcastle-Ottawa scale; AF - atrial fibrillation; IL-6 - interleukin-6; TIA - transient ischemic attack

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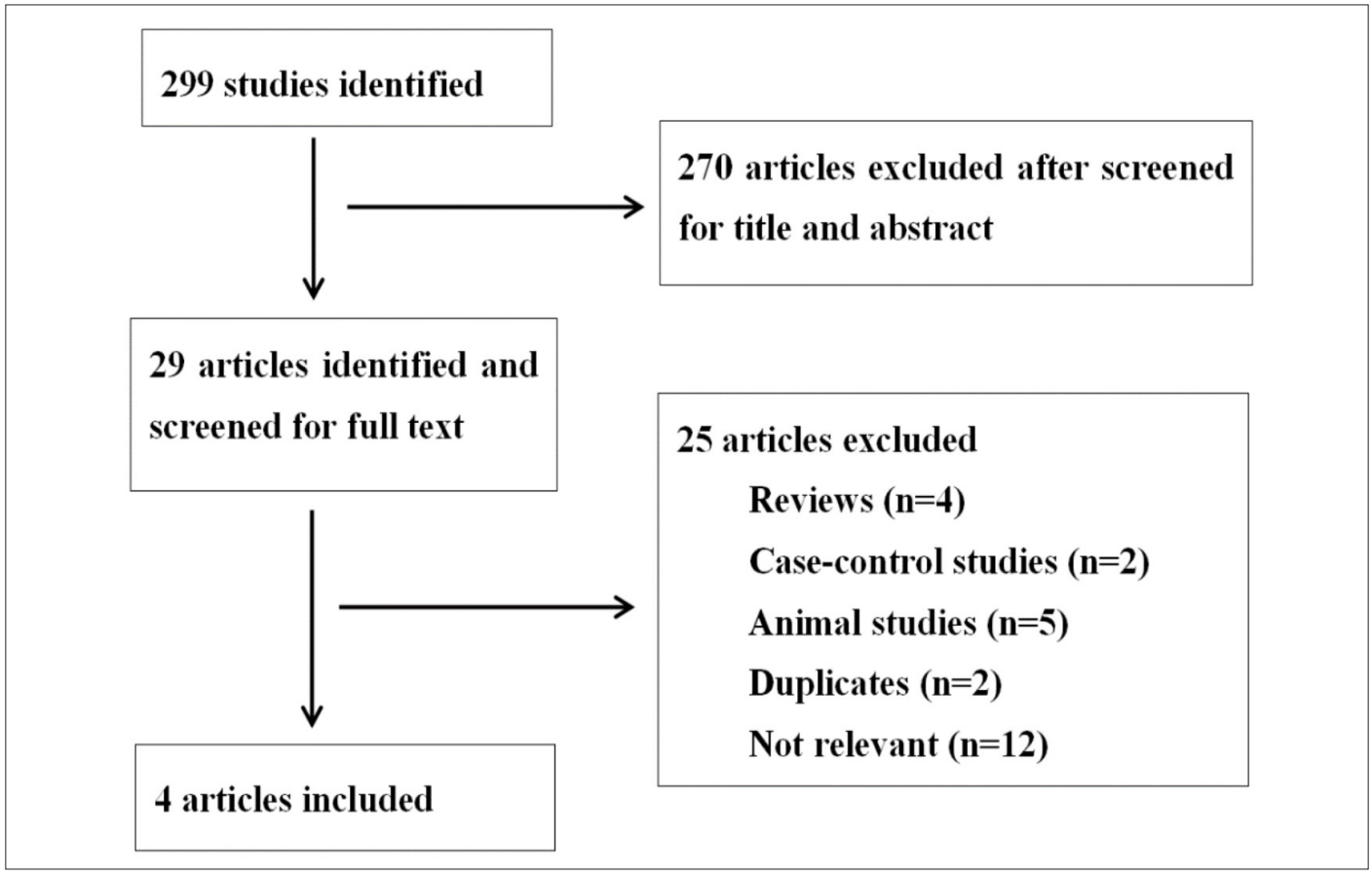
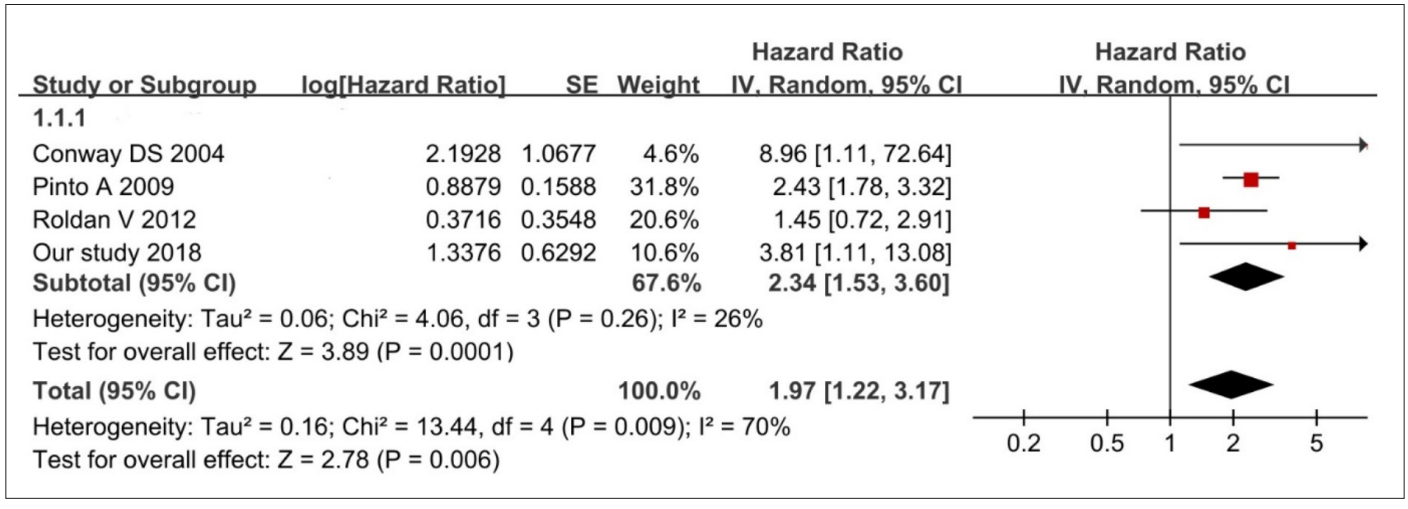
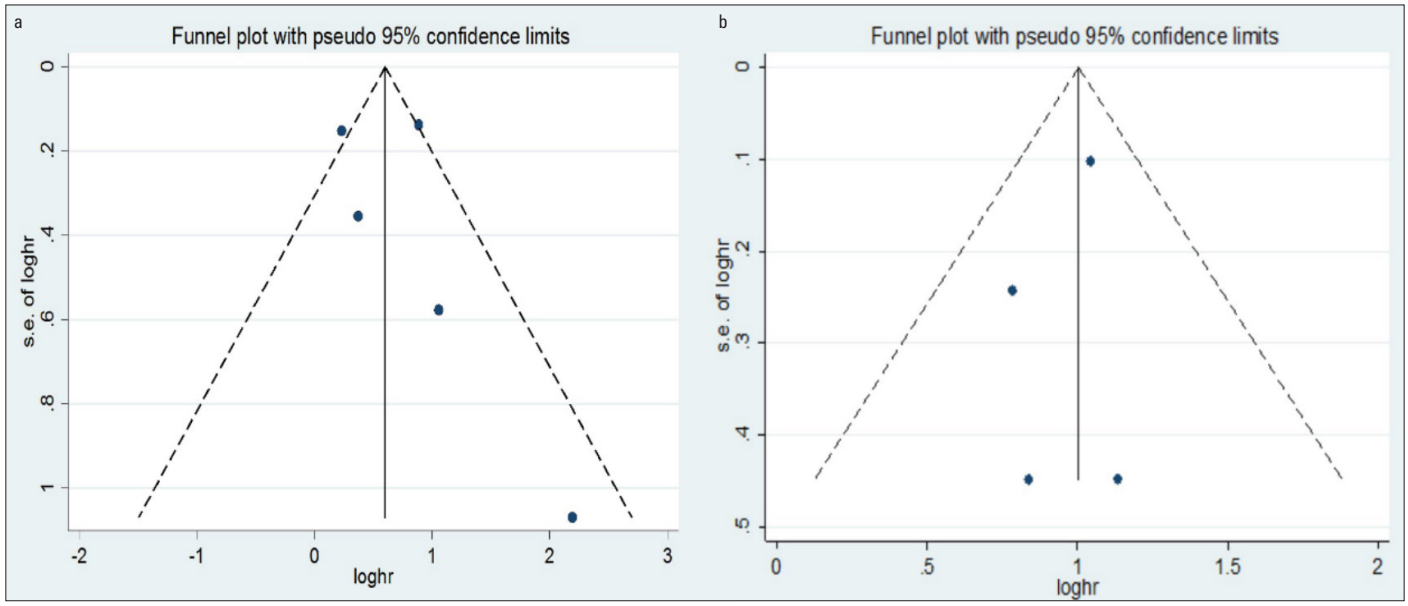


Supplementary Figure 1. Flowcharts presenting the pipeline of the search and data extraction



Supplementary Figure 2. Sensitivity analysis on association between plasma interleukin-6 level and stroke events



Supplementary Figure 3. Funnel plot for publication bias analysis. a) Stroke events and b) all-cause mortality events