A model to predict unstable carotid plaques in population with high risk of stroke

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Abstract
Background: Several models have been developed to predict asymptomatic carotid stenosis (ACS), however these models did not pay much attention to people with lower level of stenosis (50% or carotid plaques, especially instable carotid plaques) who might benefit from early interventions. Here, we developed a new model to predict unstable carotid plaques through systematic screening in population with high risk of stroke. Methods: Community residents who participated the China National Stroke Screening and Prevention Project (CNSSPP) were screened for their stroke risks. A total of 2841 individuals with high risk of stroke were enrolled in this study, 266 (9.4%) of them were found unstable carotid plaques. A total of 19 risk factors were included in this study. Subjects were randomly distributed into Derivation Set group or Validation Set group. According to their carotid ultrasonography records, subjects in derivation set group were further categorized into unstable plaque group or stable plaque group. Results: 174 cases and 1720 cases from Derivation Set group were categorized into unstable plaque group and stable plaque group respectively. The independent risk factors for carotid unstable plaque were: male (OR 1.966, 95%CI 1.406-2.749) older age (50-59, OR 6.012, 95%CI 1.410-25.629; 60-69, OR 13.915, 95%CI 3.381-57.267; ≥70, OR 31.267, 95%CI 7.472-130.83), married (OR 1.780, 95%CI 1.186-2.672), LDL-C (OR 2.015, 95%CI 1.443-2.814), and HDL-C (OR 2.130, 95%CI 1.360-3.338). A predictive scoring system was generated, ranging from 0-10. The cut-off value of this predictive scoring system is 6.5. The AUC value for derivation and validation set group were 0.738 and 0.737 respectively. Conclusions: For those individuals with high risk of stroke, we developed a new model which could identify those who have a higher chance to have unstable carotid plaques. When an individual’s predictive model score exceeds 6.5, the probability of having carotid unstable plaques is high, and carotid ultrasonography should be conducted accordingly. This model could be helpful in the primary prevention of stroke.

Background
Stroke is the third leading cause of lost disability adjusted life years worldwide [1]. It is well-known that atherosclerosis is the main risk factor of cardiovascular and cerebral vascular disease [2]. Asymptomatic carotid artery stenoses are very common among residents. So, it would be helpful to
screening carotid stenosis in selected population for the purpose of primary prevention of cardiovascular diseases (CVD) and stroke, although there is still controversy over it. The USPSTF recommended against screening for asymptomatic carotid stenosis (ACS) in the general adult population both in 2007 and 2014[3, 4]. The American Society of Neuroimaging has concluded that screening for ACS is appropriate if the prevalence is greater than 20% [5]. Several studies[6, 7] demonstrated older age, smoking, high blood pressure, diabetes and others were independent risk factors for carotid stenosis (>50% stenosis), however these models did not pay much attention to people with lower level of stenosis (≤50% or carotid plaques, especially unstable plaques) who might benefit from early intervention, such as statins, antiplatelet drugs.

In order to better understand the risk factors of unstable carotid plaques, we analyzed data of 2841 individuals from China National Stroke Screening and Prevention Project (CNSSPP) Database, including carotid duplex scan, demographic information, lifestyle risk factors, medical history, family history of stroke and blood tests, etc. The purpose of this study is to identify residents with a high probability of having unstable carotid plaques, for whom primary prevention therapy might be beneficial.

Methods

**High risk group of stroke individuals**

Screening records ranging from 2012 to 2016 were provided by CNSSPP, a nationally ongoing community-based study, which was conducted by the National Project Office of Stroke Prevention and Control. Screenings were performed at 21 communities throughout the city of Nanjing. Individuals were selected for cluster sampling. They should meet these criterions: Age≥40 years old, living in the community for at least 9 month per year, and at least 85% population were included. According to the standard established by the CNSSPP committee[8], high-risk group of stroke were defined as follows: at least 40 years old, at least three of the following risk factors, including hypertension, atrial fibrillation, smoking, dyslipidemia, diabetes mellitus, physical inactivity, overweight or obesity (BMI ≥26 kg/m²), and family history of stroke. Individuals who had the history of stroke or transient ischemic attack were also considered at high risk.
Hypertension was defined as systolic blood pressure \( \geq 140 \) mm Hg, diastolic blood pressure \( \geq 90 \) mmHg, self-reported hypertension diagnosed by a physician, or use of antihypertensive medications [9]. Atrial fibrillation was defined as ECG examination indicated atrial fibrillation, self-reported diagnosis of atrial fibrillation, or use of anticoagulant medications. Smoking status was classified as smoking (current smoking or had a history of smoke for more than one year) or never smoking (never smoking or had a history of smoke for less than one year). Dyslipidemia was defined as having one or more of the following conditions: triglyceride \( \geq 2.26 \) mmol/L, total cholesterol \( \geq 6.22 \) mmol/L, high-density lipoprotein cholesterol \( < 1.04 \) mmol/L, low-density lipoprotein cholesterol \( \geq 4.14 \) mmol/L, self-reported diagnosis of dyslipidemia, or taking cholesterol-lowering medications [10]. Diabetes mellitus was defined as fasting plasma glucose \( \geq 7.0 \) mmol/L, self-reported diagnosis of diabetes mellitus, or use of oral antidiabetic agents or insulin injection. Physical activity was defined as regular physical exercise \( > 3 \) times/week for at least 30 minutes per session. Body mass index was calculated as body weight (in kg) divided by the square of height (in m; kg/m\(^2\)). Overweight or obesity was defined as body mass index \( \geq 26 \) kg/m\(^2\), according to the guidelines of the Working Group on Obesity in China [11]. A family history of stroke was defined as the occurrence of stroke in parents or siblings.

Before carotid duplex scans, subjects were asked to complete a standardized CNSSPP questionnaire, including demographic information, lifestyle risk factors, medical history, and family history of stroke, which were collected through face-to-face interviews by trained staff. The questionnaire of CNSSPP is provided in additional file 1.

**Blood test**

Fast venous blood (5mL) was collected, centrifugated at 3000g for 10 min and stored at -80°C Freezer. Levels of fasting plasma glucose (FPG), homocysteine (Hcy), Total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglyceride (TG) were determined using OLYMPUS AU5400 (OLYMPUS, Japan). EDTA anticoagulated whole blood samples (2ml) were collected to determine HbA1c level by TOSOH G8 (TOSOH, Japan).

**Carotid artery ultrasound screening protocol**
According to Chinese stroke vascular ultrasound examination guideline [12], the duplex scan consists of ultrasound imaging of the distal common carotid artery, bulb, and proximal internal and external carotid arteries, with evaluation of a Doppler signal for 3 to 5 beats in each location on both sides. Plaque is interpreted as greater than 1.5mm of IMT based on Doppler-derived [13]. Plaques with hypoechoic, mixed echoes, or ulceration are defined as unstable plaques. Carotid duplex examinations were performed by four experienced registered vascular technicians in Nanjing Brain Hospital, which is a Stroke Screening and Training Center. All of the vascular technicians were unaware of clinical information of subjects. Screening work was conducted in compliance with the protocol established by CNSSPP committee.

**Predictive model evaluation**

The whole sample was randomly divided into a model derivation set and a model validation set, which consisted of approximately two-thirds and one-third of the sample, respectively. A comparison was performed between the two groups with t-test for continuous variables and with χ² tests for categorical variables. Univariate logistic regression was carried out for each risk factor. When the value of P was less than 0.1, the variable was included in the multivariable logistic regression model (Stepwise forward). Variables with P values less than 0.05 are retained. According to the previous study [6], we also generated a scoring system based on the regression coefficients. The lowest coefficient in absolute value was used as denominator. The coefficient of each independent risk factor was divided by the absolute value of the lowest coefficient and then rounded up to the nearest integers. Each subject would have a score according to the model and then scores of all the subjects were used to plot receiver operator characteristic (ROC) curve, and to determine the prediction power of unstable carotid plaque, and the best cutoff score by Youden index.

We used validation set to evaluate the ability of the predictive model to discriminate between subjects with and without unstable carotid plaque, which was also assessed using a ROC curve by SYSSTAT (SPSS Inc, Chicago, IL). A ROC curve plots the true-positive rate (test sensitivity) for a given threshold on the y-axis and the corresponding false-positive rate (one-test specificity) on the x-axis. The area under the resulting fitted curve represents the discriminating ability of that particular
screening method and is assumed to be normally distributed. An area of 50% represents a non-discriminant screening test in which the true-positive rate equals the false-positive rate. The area under the ROC curve for excellent test and poor test approximates 100% and 50%, respectively. After checking the normality of all continuous variables, continuous variables were presented as means (Standard deviation, SD), and categorical variables were presented as percentages. According to empirical formula of sample estimation based on multi factor analysis, more than 228 cases were needed in this study. All statistical analyses were performed using the SPSS version 20.0 software for Windows (SPSS, Inc, Chicago, IL, USA). In all statistical analyses, a P value <0.05 was considered statistically significant.

Results
During 2012-2016, 34227 residents were enrolled in CNSSPP. According to the risk factor screening, 5250 residents were at high risk, and were screened carotid arteries by ultrasonography. A total of 2309 residents were excluded because of previous stroke or transient ischemic attack, 100 residents were excluded because of incomplete data, as shown in Figure 1. Of the total 2841 subjects included in this study, the prevalence of carotid plaque was 35.2% (n=1000), the prevalence of unstable carotid plaque was 9.4% (n=266). All the 2841 subjects were randomly distributed into the derivation set group (n=1894) and the validation set group (n=947). Characteristics of these subjects are provided in Table 1.

The derivation set included 1894 subjects (mean age =60.7±8.6 years), 174 (9.2%) of whom had unstable carotid plaques. The clinical characteristics of this set are provided in Table 2. Men were more likely to have unstable carotid plaques. A higher proportion of subjects older than 60 had unstable carotid plaques. Subjects who were married or had a history of diabetes mellitus would have more chance to have unstable carotid plaques. Subjects with unstable carotid plaques were more likely to have a higher level of FPG, Hcy, TC, LDL-C, a lower level of HDL-C, and overweight or obesity. No significant differences were found between groups with regard to education level, atrial fibrillation, hypertension, hypercholesterolemia, smoking, and lack of physical activity, family history of stroke, HbA1c, or TG.
Table 3 shows the results of multivariable logistic regression analysis. Five variables were significantly associated with unstable carotid plaque: male (OR 1.966, 95%CI 1.406-2.749); older age (50-59, OR 6.012, 95%CI 1.410-25.629; 60-69, OR 13.915, 95%CI 3.813-57.267; ≥70, OR 31.267, 95%CI 7.472-130.83), married (OR 1.780, 95%CI 1.186-2.672), LDL-C (OR 2.015, 95%CI 1.443-2.814), and HDL-C (OR 2.130, 95%CI 1.360-3.338). According to the coefficients generated from the multivariable logistic regression analysis, ROC curves were plotted. The area under the curve (AUC) was 0.741, as shown in Figure 2.

For these 5 risk factors, marriage (married) had the lowest regression coefficients 0.577, we scored it 1 point. Other 4 factors’ scores were calculated by dividing coefficients by 0.577 and then rounded to the nearest integers. Using this scoring system, we got a score for each subject from derivation set. Scores of all the subjects were used to plot ROC curve, and determine the prediction power of unstable carotid plaque and the best cutoff score by Youden index. The AUC was 0.738. The Yoden index of score was calculated by sensitivity plus specificity. The best predictive value of score was 6.5, sensitivity was 71.8%, and specificity was 63.0%, as shown in Figure 2.

The ability of the predictive model to discriminate between subjects with and without unstable carotid plaque was evaluated in a separate validation set comprising 947 persons (mean age = 60.4±8.7 years), 92 (9.7%) of whom had unstable carotid plaques. The AUC was 0.743. According to the scoring system, we got a score for each subject from validation set. The scores of all the subjects were used to generate ROC curve. The AUC was 0.737. The Yoden index of score was calculated by sensitivity plus specificity. The best predictive value of score was still 6.5, sensitivity was 76.1%, and specificity was 63.6%, as shown in Figure 3.

Discussion
This study explored the risk factors of unstable carotid plaque in asymptomatic patients with high risk of stroke and developed a scoring system. This is the first time that a quantitative scale was developed to assess the risk of unstable carotid plaques. Previous studies focused on CIMT or carotid artery stenosis. Traditional risk factors such as hypertension, diabetes, smoking, etc, were often used in various prediction systems, however, serological indicators and personal history (education level,
marriage) were rarely included. Patients with moderate to severe stenosis may benefit from carotid endarterectomy and carotid artery stenting, however they are relatively rare. So, if we pay more attention to unstable carotid plaques, we may identify more high-risk patients with lower level of stenosis who may benefit from aggressive primary interventions, such as healthy lifestyle, control of chronic diseases and risk-reducing medications [14].

In this study, marriage was associated with an increased prevalence of unstable carotid plaque both by univariate and multivariable analysis. It is the first time that marriage was associated with unstable carotid plaque. Yue W et al investigated potential association between carotid artery stenosis and cognitive impairment among patients with acute ischemic stroke, including one subgroup analysis of the relationship between marriage and severity of carotid stenosis. The result showed that marriage was closely associated with severe stenosis of carotid artery, and the association was statistically significant (P=0.0471) [15]. We noticed that this study was from Tianjin, China. This association may have some relationship to the Chinese tradition, as marriage can improve living quality, especially in terms of eating habits. Another possible reason is that subjects filled out questionnaires by themselves, which might not be very accurate.

Gender is another independent risk factor for unstable carotid plaque in individuals with high risk of stroke. Our study indicated male was a risk factor for unstable carotid plaque in individuals with high risk of stroke, which was similar to the outcome of a cohort from Taiwan [16]. On the other hand, a cohort from America indicated that men and women had equal risk for ACS [6]. Such discrepancies may be attributed to the racial difference in these studies, or the small sample size of our study.

Previous studies [17, 18] founded that older age, smoking, peripheral arterial disease, hypercholesterolemia, hypertension, Diabetes mellitus, and coronary artery disease were associated with occult carotid stenosis of >50% or 60%. Our study found that diabetes mellitus was a risk factor for unstable carotid plaque by univariate analysis, however, our analysis showed that other traditional risk factors such as hypertension, HbA1c, homocysteine and smoking were not associated with unstable carotid plaque. The cause of stroke is complicated, carotid thromboembolism is only part of it (accounting for up to 20%), which is caused by carotid plaque or stenosis, especially unstable
carotid plaque [19]. For that reason, the traditional risk factors may cause stroke, but may not necessarily cause unstable carotid plaque in this study.

The Framingham Heart Study showed that the occurrence of atherosclerosis was negatively correlated with the level of HDL-C and positively correlated with the level of LDL-C [20]. This is similar to our research. Unstable plaque is a special type of atherosclerosis. LDL-C can damage endothelial cells and smooth muscle cells through oxidation, modification and glycosylation, start and maintain the inflammatory response of vascular wall, so as to develop atherosclerosis [21]. HDL-C can transport cholesterol from surrounding tissues (including macrophages and atherosclerotic plaques) to the liver for recycling or excretion in the form of cholic acid. This process is called reverse cholesterol transport (RCT). Through RCT, it can reduce the deposition of lipids in the blood vessel wall, thus reducing the cholesterol level in plasma and blood vessel wall and reducing the occurrence of atherosclerosis [22]. Hill [23] reported that the prevalence of asymptomatic carotid artery stenosis was estimated to range from 2% to 8% for stenosis 50% or greater and from 1% to 2% for stenosis 80% or greater in general population. Due to the low prevalence of carotid artery disease, numerous cost-evaluation models had failed to demonstrate a benefit in screening general population [17]. On the other hand, Jones [24] reported that the prevalence of carotid plaque was 21.0% in a total of 173 middle-aged subjects, which was similar to our findings. In this study, our subject’s average age was 60 years old, and the prevalence of carotid plaque and unstable carotid plaque was 35.2% and 9.4%, respectively.

Our study developed a simple and handy scoring system. When individuals with high risk of stroke get a score of more than 6.5 points, carotid artery ultrasound screening should be recommended. Previous prediction models [6, 7] were only generated through medical history inquiry. In this study, objective blood indicators were included in the prediction model, which reduced subjective bias and made it more accurate.

However, certain inadequacies of this study cannot be ignored. First, although this study was carried out throughout 21 Communities in Nanjing, it’s still a single-center cross-sectional study, therefore cannot reflect the national prevalence and cannot tell the progression of unstable carotid plaque. Second, for the lack of time and staff, the sizes of the plaques were not measured in detail, and the
plaque characteristics were not evaluated by 3D probe either. Third, some biomarkers are not fully included in this study, such as hs-CRP, which is a very important indicator of CV disease. Lastly, follow-up data of this population are being compiled, especially with regard to interventions (medication and surgery) and the occurrence of stroke. Regardless of these limitations, this study still provided solid evidence of the risk of unstable carotid plaque in individuals with high risk of stroke, and would be perfected in the future.

Conclusions
The prevalence of carotid artery plaque as well as unstable plaque is high in population with high risk of stroke. It is necessary to screen carotid artery by Doppler ultrasonography in this population for early intervention. We developed a scoring system ranging from 0 to 10. When a resident's score exceeds 6.5, the probability of having unstable carotid plaque is high, thus Doppler ultrasound examination of the carotid artery should be conducted as soon as possible and interventions should be introduced according to the guidelines.

Abbreviations
ACS: asymptomatic carotid stenosis; CNSSPP: China National Stroke Screening and Prevention Project; CVD: cardiovascular diseases; FPG: fasting plasma glucose; Hcy: homocysteine; TC: Total cholesterol; HDL-C: high-density lipoprotein cholesterol; LDL-C: low-density lipoprotein cholesterol; HbA1C: Hemoglobin A1C; TG: triglyceride; ROC: receiver operator characteristic; AUC: area under the curve

Declarations
**Ethics approval and consent to participate**
The study was approved by the Ethics Committee of the Nanjing Brain Hospital Institutional Review Board, Nanjing Medical University (registration no 2017-kyy119-01). All participants received information on the study and provided written informed consent to participate.

**Consent to publish**
Not applicable.

**Availability of data and materials**
The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.
Competing Interests

The authors have no conflicts of interest to disclose.

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Authors’ contributions

XW participated in study design, performed the statistical analyses and revised the manuscript. JY, CY, HL, MD and FS contributed to the conception of the work. JY, CY, LW and CW contributed to the acquisition of data. JY drafted the first version of the manuscript. All authors critically revised the manuscript. All authors read and approved the final manuscript.

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Not Applicable.

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Tables
Table 1. Demographic and Risk Factor Profiles in the Derivation and Validation Sets
| Variable                                | Derivation Set | Validation Set | P      |
|-----------------------------------------|----------------|----------------|--------|
|                                        | n=1894         | n=947          |        |
| Sex (male)                              | 87446.1%       | 45147.6%       | 0.457  |
| Mean age (years)                        | 60.7±8.6       | 60.4±8.7       | 0.321  |
| Education level (Primary school or below) | 73038.5%       | 40242.4%       | 0.045  |
| Marriage(married)                       | 133070.2%      | 69973.8%       | 0.102  |
| Atrial fibrillation                     | 1306.9%        | 576.0%         | 0.392  |
| Diabetes mellitus                       | 45724.1%       | 25126.5%       | 0.168  |
| Hypertension                            | 150379.4%      | 74378.5%       | 0.579  |
| Hypercholesterolemia                    | 74139.1%       | 34936.9%       | 0.241  |
| Overweight or obesity                   | 84744.7%       | 40843.1%       | 0.408  |
| Smoking                                 | 63433.5%       | 33935.8%       | 0.219  |
| Lack of Physical activity               | 144376.2%      | 72776.8%       | 0.731  |
| Family history of strok                 | 60231.8%       | 28229.8%       | 0.276  |
| FPG(mmol/L)                             | 6.1±1.8        | 6.2±2.0        | 0.079  |
| HbA1c (%)                               | 6.6±2.5        | 6.6±1.9        | 0.872  |
| Hcy(mmol/L)                             | 12.2±7.5       | 12.0±6.6       | 0.611  |
| TC(mmol/L)                              | 5.1±1.0        | 5.1±1.0        | 0.386  |
| LDL-C(mmol/L)                           | 3.0±0.8        | 3.0±0.8        | 0.944  |
| HDL-C(mmol/L)                           | 1.6±0.8        | 1.5±0.7        | 0.600  |
| TG(mmol/L)                              | 1.7±1.3        | 1.7±1.4        | 0.699  |
| Carotid plaque                          | 66034.8%       | 34035.9%       | 0.579  |
| Carotid instability plaque              | 1749.2%        | 929.7%         | 0.649  |

Table 2. Comparison of Demographic and Risk Factor Profiles in Participants with and Without Carotid Instability Plaque in the Derivation Set
| Variable                        | With Carotid Vulnerable Plaque n=174 | Without Carotid Vulnerable Plaque n=1720 | P Value | OR 95%CI          |
|--------------------------------|-------------------------------------|------------------------------------------|---------|------------------|
| Sex (male)                     | 10962.6%                           | 76544.5%                                 | 0.001   | 2.0931.518-2.887 |
| Mean age (years)               |                                    |                                          |         |                  |
| 40-49                          | 14(8.0%)                           | 207(12.0%)                               | 0.001   | 1.00Reference    |
| 50-59                          | 35(20.1%)                          | 512(29.8%)                               | 0.018   | 5.6861.340-24.117|
| 60-69                          | 87(50.0%)                          | 765(44.5%)                               | 0.001   | 12.9333.160-52.934|
| ≥70                            | 38(21.8%)                          | 236(13.7%)                               | 0.001   | 27.5006.626-114.142|
| Education level                |                                    |                                          |         |                  |
| (Primary school or below)      | 6738.5%                            | 66338.5%                                 | 0.992   | 0.9980.725-1.375 |
| Marriage(married)              | 14181.0%                           | 118969.1%                                | 0.001   | 1.9081.289-2.826 |
| Atrial fibrillation            | 137.5%                             | 1176.8%                                  | 0.740   | 1.1060.610-2.007 |
| Diabetes mellitus              | 5431.0%                            | 40323.4%                                 | 0.026   | 1.4711.047-2.066 |
| Hypertension                   | 14583.3%                           | 135879.0%                                | 0.175   | 1.3330.880-2.019 |
| Hypercholesteremia             | 6738.5%                            | 67439.2%                                 | 0.861   | 0.9720.705-1.339 |
| FPG(mmol/L)                    |                                    |                                          |         |                  |
| ≤6.1                           | 101(58.0%)                         | 1192(69.3%)                              | 0.007   | 1.0Reference     |
| 6.11~6.99                      | 29(16.7%)                          | 235(13.7%)                               | 0.091   | 1.4560.942-2.252 |
| ≥7.0                           | 44(25.3%)                          | 293(17.0%)                               | 0.003   | 1.7721.216-2.582 |
| HbA1c ( >6.5%)                 | 14(8.0%)                           | 125(7.3%)                                | 0.959   | 1.1090.496-2.094 |
| Hcy(>15mmol/L)                 | 30(17.2%)                          | 212(12.3%)                               | 0.122   | 1.3930.915-2.120 |
| TC(>5.2mmol/L)                 | 91(52.3%)                          | 716(41.6%)                               | 0.007   | 1.5371.125-2.101 |
| LDL-C(>3.12mmol/L)             | 97(55.7%)                          | 704(40.9%)                               | 0.001   | 1.4671.223-1.761 |
| HDL-C                           | 32(18.4%)                          | 194(11.3%)                               | 0.006   | 1.7731.175-2.675 |
| (<1.04mmol/L)                  |                                    |                                          |         |                  |
| TG(>1.7mmol/L)                 | 59(33.9%)                          | 574(33.4%)                                | 0.886   | 1.0240.737-1.424 |
Table 3. Determinants of Carotid Plaque Derived from Stepwise multivariable Logistic Regression Analysis

| Variable                        | B     | Odds Ratio (95% CI)         | P Value | Score |
|---------------------------------|-------|----------------------------|---------|-------|
| Sex male                        | 0.676 | 1.9661.406-2.749           | 0.001   | 1     |
| age                             |       |                            |         |       |
| 40-49                           | NA    | 1.0Reference               | 0.001   | 0     |
| 50-59                           | 1.794 | 6.0121.410-25.629          | 0.015   | 3     |
| 60-69                           | 2.633 | 13.9153.381-57.267         | 0.001   | 5     |
| ≥70                             | 3.443 | 31.2677.472-130.83         | 0.001   | 6     |
| marriage(married)               | 0.577 | 1.7801.186-2.672           | 0.005   | 1     |
| Diabetes mellitus               | 0.205 | 1.227 (0.785-1.920)        | 0.370   | NA    |
| Overweight or obesity           | -0.248| 0.780(0.555-1.098)         | 0.154   | NA    |
| FPG (mmol/L)                    |       |                            |         |       |
| ≤6.1                            | NA    |                            | 0.547   | NA    |
| 6.11–6.99                       | 0.189 | 1.208 (0.751-1.945)        | 0.436   | NA    |
| ≥7.0                            | 0.253 | 1.287 (0.788-2.102)        | 0.313   | NA    |
| TC (>5.2mmol/L)                 | 0.364 | 1.439 (0.933-2.221)        | 0.100   | NA    |
| LDL-C (3.12mmol/L)              | 0.701 | 2.0151.443-2.814           | 0.001   | 1     |
| HDL-C (1.04mmol/L)              | 0.756 | 2.1301.360-3.338           | 0.001   | 1     |

Additional Files
- Additional file 1
- The questionnaire of CNSSPP

- This is the questionnaire of CNSSPP. It includes the following contents: demographic information, preliminary screening information, and re-screening information. After preliminary screening, it was suggested that the high-risk population (Including previous stroke and TIA individuals) should be re-screened, otherwise the screening would be terminated.

- Additional file 2
- The results of gender age matched control group

- This is the result of the study on the gender age matched control group (1:1), which can completely correct the influencing factors of gender and age. The result show that whether or not adjusted for age and gender, married, a higher level of LDL-C and a lower level of HLD-C were both the independent risk factors of carotid unstable plaque.

Figures
Residents participated in the screening (2012-2016): 34227 cases

Fill in the questionnaire:

Low-risk population: 24530 cases
Middle risk population: 4401 cases
Unrated population: 46 cases

High-risk population (Including stroke and TIA):

Blood examination and carotid ultrasound screening:

5250 cases

Stroke population: 923 cases
TIA population: 1386 cases

High-risk population:

2941 cases

Incomplete data:

100 cases

High risk population included in this study:

2841 cases

Figure 1
Flowchart illustrating the inclusion/exclusion of individuals in the study.

Figure 2
The ROC curve of Derivation Set group for risk factors and predictive model Scoring in high risk group of strokes.

Figure 3
The ROC curve of Validation Set group for risk factors and predictive model Scoring in high risk group of strokes.

Supplementary Files
This is a list of supplementary files associated with this preprint. Click to download.
The questionnaire of CNSSPP.pdf
The results of gender age matched control group.pdf
