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**Biomarkers and potential** **pathogenesis of colorectal cancer-related ischemic stroke**

Qin QX *et al.* Biomarkers and potential pathogenesis of CRCIS

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**Abstract**

***AIM***

To investigate the specific biomarkers and potential pathogenesis of colorectal cancer-related ischemic stroke (CRCIS).

***METHODS***

A retrospective study was conducted on CRCIS patients (colorectal cancer patients with ischemic stroke without conventional stroke risk factors) registered at seven centers between January 2007 and December 2017. Clinical data and laboratory and imaging findings were compared with age- and sex- matched patients with colorectal cancer (CRC) without ischemic stroke who admitted to the same hospital during the same period. Univariate and multivariate analysis were performed to analyze the independent risk factors for CRCIS. A receiver operator characteristic (ROC) curve was configured to calculate the optimal cut-off value of the products of the independent risk factors for CRCIS.

***RESULTS***

A total of 114 CRCIS patients and 114 CRC patients were included. Multiple lesions in multiple vascular territories were common in CRCIS patients (71, 62.28%). The levels of plasma D-dimer, carcinoembryonic antigen (CEA), cancer antigen l25 and neutrophil count (NC) were significantly higher in CRCIS patients than in CRC patients. Multiple logistic regression analysis revealed that plasma D-dimer levels [odds ratio (OR) = 1.002, 95% confidence interval (CI): 1.001–1.003, *P* < 0.001], CEA levels (OR = 1.011, 95%CI: 1.006–1.015, *P* < 0.001) and NC levels (OR = 1.626, 95%CI: 1.268–2.087, *P* < 0.001) were independent risk factors for CRCIS. In addition, ROC curve revealed that the area under curve for the products of plasma D-dimer, CEA and NC was 0.889 ± 0.022 (95%CI: 0.847–0.932, *P* < 0.001), and the optimal cut-off value for the product was 252.06, which was called CRCIS Index, with a sensitivity of 86.0% and specificity of 79.8%.

***CONCLUSION***

Hypercoagulability induced by elevated CEA and neutrophils may be an important cause of CRCIS, CRCIS index which serves as a biomarker of CRCIS needs further study.

**Key words:** Colorectal cancer; Ischemic stroke; Biomarker**;** Pathogenesis

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**Core tip:** Although cancer-related stroke has long been known, its biomarkers and underling pathogenesis are still unclear. It was hypothesized that a specific cancer type may affect the development of ischemic stroke (IS) according to its primary site, pathological type and growth stage. It is reported that colorectal cancer (CRC) can increase the risk of IS. We conducted a retrospective study on CRC-related IS (CRCIS) patients. We suggest that hypercoagulability induced by elevated carcinoembryonic antigen and increased neutrophil count are the main pathogenic factors in CRCIS, and CRCIS Index which serves as a biomarker of CRCIS needs further study.

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**INTRODUCTION**

Cancer and ischemic stroke (IS) are major causes of morbidity and mortality. In addition, cerebrovascular disease is a common complication in cancer patients, with 15% of patients experiencing thromboembolic events during their clinical course[1]. Moreover, up to 40% of cancer patients with IS lack conventional stroke risk factors, indicating that cancer itself may directly or indirectly lead to the development of IS[2,3]. Previous studies have suggested that cancer-related IS is characterized by elevated plasma D-dimer levels and multiple lesions in multiple arterial territories on diffusion-weighted magnetic resonance imaging (DWI)[4-6]. Although cancer is associated with IS through tumor compression, invasion of the blood vessels and left atrium, or nonbacterial thrombotic endocarditis[7-9], hypercoagulability is considered to be the most important pathogenic factor of cancer-related IS[10,11]. Furthermore, elevated levels of plasma D-dimer, high-sensitivity C-reactive protein (Hs-CRP), fibrinogen and pro-brain natriuretic peptide (pro-BNP) are thought to be potential biomarkers of cancer-related IS[12,13]. However, despite accumulating knowledge, the specific biomarkers and definitive pathogenesis of cancer-related IS have remained unclear. Most previous studies have been conducted on several types of cancer. The different characteristics of cancer, including primary growth zone, pathological type and growth stage, may affect IS in different ways[14]. We speculated that studies targeting on patients with a specific cancer may be more conducive to illuminate the specific biomarkers and pathogenesis of cancer-related IS.

 Colorectal cancer (CRC) is the third most common cancer among men and the second most common among women globally[15]. The incidence of CRC has shown a steady upward trend in China over the past decade, from 12.8 in 2003 to 16.8 per 100000 in 2011, and the incidence will reach 20.7 per 100000 in 2020[16]. Notably, previous studies have reported that CRC is associated with IS[17,18]. The risk of IS increased 1.61 times during the first 6 mo after diagnosis of CRC compared to that in the general population[19], indicating that CRC itself could lead to IS, so-called CRC-related IS (CRCIS). In the present study, we aimed to investigate the specific biomarkers and potential pathogenesis of CRCIS by comparing the clinical data of CRCIS patients with age- and sex-matched CRC patients, and calculating the CRCIS Index from its independent risk factors.

**MATERIALS AND METHODS**

***Patient selection***

This study was approved by the Guangxi Medical University Review Board. Patients with acute IS with the additional diagnosis of active CRC but without conventional stroke risk factors were recruited from seven centers in Guangxi Province between January 2007 and December 2017. Referring to the definition of active cancer in the study of Lee and his colleagues[20], active CRC was defined as a diagnosis of CRC within 6 month before enrollment, any treatment for CRC within the previous 6 month, or recurrent or metastatic CRC. The diagnosis of acute IS was based on the American Heart Association diagnostic criteria for stroke[21]. It was difficult to identify CRCIS in clinical practice. Referring to the definition of cancer-related stroke[2,3,5], CRCIS in present study was defined as patients with acute IS and active CRC without conventional stroke risk factors. Patients with primary or metastatic brain cancer or hematological malignancies were excluded, because these patients were considered to represent a subgroup with different underlying stroke mechanisms. Patients with cerebral hemorrhage or other cerebrovascular disease and those incomplete clinical data were also excluded. The control group was age- and sex- matched patients with CRC but without IS who admitted to the same hospital during the same period. The exclusion criteria included the presence of conventional stroke risk factors, brain metastasis and other primary cancers (Figure 1).

 To improve the power of the present study, the sample size need to estimate. Previous studies found that cancer-related IS was associated with elevated plasma D-dimer[10,11]. However, whether it was associated with CRCIS is not clearly. Therefore, we hypothesize that D-dimer may also be associated with the occurrence of CRCIS. It was reported that the positive rate of D-dimer in CRC was 15% to 31%. In our 1:1 matched case-control study, for sample size estimation, we assumed the positive rate of D-dimer in CRC patients was 20%, and risk of CRCIS was 3.5. Results showed that it needed 97 pairs when α = 0.05 and β = 0.2 (two-tailed).

***Clinical data collection***

Clinical data of all patients, including age, gender, stroke risk factors, pathological type of cancer, metastasis, treatment methods, IS etiology, IS lesion patterns, severity of focal neurological deficits and time interval from diagnosis of CRC to the occurrence of IS were collected consistently. In addition, laboratory findings including routine blood tests, blood biochemistry, coagulation function, and levels of plasma D-dimer and tumor markers such as carcinoembryonic antigen (CEA), cancer antigen (CA)125 and CA199 were collected. Findings of imaging examinations such as echocardiography, transcranial Doppler ultrasound, cranial computed tomography (CT), CT angiography, magnetic resonance imaging (MRI), and MR angiography were also collected. The severity of focal neurological deficits was assessed by the National Institutes of Health Stroke Scale (NIHSS).

***Statistical analysis***

Statistical analysis was performed using SPSS version 20.0 software. An independent sample *t* test was used to compare continuous variables between groups, while Pearson’s $χ$*2* or Fisher’s exact test was used to compare categorical variables. Multivariate logistic regression analysis was performed to identify the independent risk factors of IS in CRC patients. Variables with *P* < 0.05 in univariate analyses were considered explanatory variables and were entered to multivariate models. To explore the specific biomarkers of CRCIS, the CRCIS Index was calculated as follows. First, we calculated the products of the independent risk factors for IS in the two groups. Second, the cut-off value of the products of independent risk factors for IS that were used to identify IS in CRC patients was calculated by a receiver operator characteristic (ROC) curve. Finally, the optimum cut-off value of the products was determined and called the CRCIS Index. All *P-*values were two-sided, and *P* < 0.05 was considered statistically significant.

**RESULTS**

A total of 114 CRCIS patients [78 male, 36 female, mean age (65.33 ± 12.45) years] were finally included, accounting for 0.67% of 16915 CRC patients. There were 114 age- and sex-matched CRC patients that served as the control group. The most common pathological type of CRC in both groups was adenocarcinoma. There were no significant differences in cancer systemic metastasis and oncological treatment including surgery and chemoradiotherapy, and no treatment in CRCIS patients compared with CRC patients (Table 1).

 After diagnosis of CRC, 60 (52.63%), 19 (16.67%) and 13 (11.40%) patients experienced acute IS in the first 6 month, 6 month to 1 year, and > 1 year, respectively. In addition, 22 (19.30%) patients presented with IS as the initial manifestation of occult CRC, which was confirmed as CRC during anti-stroke therapy. Multiple lesions in multiple arterial territories in the brain on DWI were observed in 71 (62.28%) CRCIS patients. NIHSS score ranged from 0 to 21 on the day of IS onset (Table 2 and Figure 2).

 In terms of laboratory findings, there was no significant difference in most items in the two groups. However, the levels of plasma D-dimer, NC, CEA and CA125 were significantly higher in the CRCIS than in CRC patients (*P* < 0.05). Multiple logistic regression analysis revealed that the D-dimer levels [odds ratio (OR) = 1.002, 95% confidence interval (CI): 1.001–1.003, *P* < 0.001], CEA levels (OR = 1.011, 95%CI: 1.006–1.015, *P* < 0.001) and NC levels (OR = 1.626, 95%CI: 1.268–2.087, *P* < 0.001) were independent risk factors of CRCIS (Table 3).

 The ROC curves for identifying CRCIS from the products of D-dimer, CEA and NC were shown in Figure 1. The mean ± standard error of the area under curve (AUC) for the product was 0.889 ± 0.022 (95%CI: 0.847–0.932, *P* < 0.001), indicating good overall accuracy of the test. The optimum diagnostic cut-off value for the products calculated from the ROC was 252.06, which was called the CRCIS Index, with a sensitivity of 84.3% and specificity of 80.9% (Figure 3).

**DISCUSSION**

IS is a common complication in patients with cancer and it has received increasing attention[22,23]. Previous studies showed that the incidence of IS was 1.6 times higher in cancer patients than in patients without cancer, especially during the first 6 mo after cancer diagnosis[19]. In the present study, the incidence of IS in hospitalized CRC patients was 0.61%, and most of them (52.63%) developed IS within the first 6 mo after diagnosis of CRC. It is suggested that as soon as CRC diagnosis is established, measures should be taken to prevent IS. In addition, IS as an initial manifestation of occult cancer has been reported in previous studies[17,24]. In the present study, there were 22 (19.3%) patients who were hospitalized for acute IS and then diagnosis of CRC was confirmed, indicating that measures should be taken to screen out occult cancer, including CRC, in patients with unexplained IS.

 Previous studies have suggested that certain clinical, laboratory and radiological features could help to distinguish cancer-related IS from stroke of other etiology[3,25]. Many studies have demonstrated that cancer-related IS is characterized by markedly elevated plasma D-dimer levels and multiple lesions in multiple arterial territories on DWI[4-6]. In the present study, CRCIS patients had similar features to general cancer-related IS such as elevated plasma D-dimer levels and multiple lesions in multiple cerebral arterial territories, as well as distinctive features including elevated plasma CEA and increased NC, which may be the useful clues to identify CRCIS.

 The unveiling of underlying pathogenesis of cancer-related IS is an important issue, because pathogenesis may influence the choice of management, stroke prognosis, and risk of recurrence. In some previous studies, cancer was found to be associated with IS through tumor compression, invasion of the blood vessels and left atrium, and nonbacterial thrombotic endocarditis[7-9]. However, increasing studies have suggested that cancer-related IS were characterized by elevated plasma D-dimer levels[4-6]. Recently, Wang and his colleagues found that the plasma D-dimer value of 2.785 μg/mL was the cutoff in identifying cancer-related IS patients[26]. Moreover, the frequency of microembolic signals in the internal carotid arteries on transcranial Doppler images correlates linearly with D-dimer levels in patients with IS and cancer, which indicates that elevated D-dimer levels are an independent predictor for the detection of embolic signals[11]. Moreover, the D-dimer level is a direct measure of activated coagulation and has been used as a biomarker of hypercoagulability[27]. The elevated D-dimer level suggests that hypercoagulability plays a major role in the pathogenesis of cancer-related IS. In the present study, plasma D-dimer level was significantly higher in CRCIS patients, indicating that hypercoagulability is an important factor in the pathogenesis of CRCIS.

 Although hypercoagulability may be associated with cancer-related IS, its underlying mechanism remains unclear. Interestingly, recent studies have suggested that mucins generated from mucinous cancer are associated with hypercoagulability and increase the risk of IS[28-30]. A study by Jovin *et al*[28] reported four patients with metastatic cancer, brain infarcts, and markedly elevated mucinous serum marker CA125 levels, and suggested a possible association between this protein and stroke. Moreover, the relationship between mucins and hypercoagulability were further confirmed by necropsy evidences from mucinous cancer patients with widespread intracranial arteriovenous thrombosis and multiple cerebral infarctions, in which the mucin within vessels and in microthrombus in the regions of infarction were found by microscopic examination[29]. Furthermore, animal experiments demonstrated that mucins secreted by cancer cells could trigger the reciprocal activation of platelets and neutrophils and lead to the formation of thrombus in the blood[30]. These findings indicated that mucins associated hypercoagulability played an important role in cancer-related IS. In the present study, CRCIS patients have higher CEA and CA125, and elevated CEA was an independent risk factor for IS. CEA, a general oncofetal antigen, is also a mucinous marker similar to CA125[31]. Therefore, we speculated that CEA, a mucinous substance generated from CRC cells may also lead to hypercoagulability by triggering the reciprocal activation of platelets and neutrophils.

 Additionally, neutrophil extracellular traps (NETs) that generated from neutrophils were also found to associate with hypercoagulability and thrombosis diseases (including IS) in patients with cancer[32,33]. In solid tumor models, it was found that cancer can induce an increase of peripheral blood neutrophils. And increased neutrophils may release NETs in the vascular, which may promote hypercoagulability by stimulating platelet activation[34]. Therefore, NETs generated from neutrophils was considered to be a novel coagulation-promoting mechanism in cancer patients[35,36]. In the present study, NC was significantly increased in CRCIS patients, and multivariate logistic regression analysis revealed that the increased NC may independently increase the risk of IS. Therefore, the increase in neutrophils may also lead to hypercoagulability by releasing NETs to stimulate the activation of platelets and thus increase the risk of IS in CRC patients.

 In the present study, multivariate logistic regression analysis revealed that elevated plasma D-dimer and CEA and increased NC were independent risk factors for CRCIS, indicating that these parameters could use as potential biomarkers for CRCIS. However, as a common coagulation marker, plasma D-dimer lack specificity and sensitivity. Elevated levels of CEA are also found in other cancers and increased NC is common in systemic inflammation. Therefore, elevated plasma D-dimer and CEA and increased NC were not specific biomarkers for CRCIS. Considering that development of CRCIS may be caused by the combined effects of elevated plasma D-dimer and CEA levels and increased NC, we calculated the CRCIS Index. As the area under the ROC curve of the CRCIS Index was highly accurate, and the sensitivity and specificity of the CRCIS Index were high, we suggest that CRCIS Index could serve as a potential biomarker. However, due to the retrospective design of the present study, the other biomarkers mentioned in previous studies that may be related to cancer-related IS were not studied[12,13]. The role of CRCIS Index in CRCIS patients may need to be confirmed in future studies with larger samples and more biomarkers. Nevertheless, our study provided a meaningful method to the further investigation about the index of CRCIS in future studies.”

 The strengths of our study were multicenter enrollment and comparison with CRC patients without IS. The main limitations of this study include the relative small sample size and some uncontrollable setting. Still, our results could promote further prospective larger population studies, which would be better to illuminate the biomarkers and pathogenesis of IS in CRC patients.

 In summary, our findings suggest that hypercoagulability induced by elevated CEA and neutrophils may be an important cause of CRCIS. CRCIS index which serves as a novel biomarker of CRCIS needs to be confirmed in future study.

**Article Highlights**

***Research background***

Cancer is associated with an increased risk of ischemic stroke (IS) and IS can be the first manifestation of an occult cancer. Several biomarkers and mechanisms in cancer related-IS have been reported. However, most previous studies had been conducted on several types of cancer, few studies targeted on a certain cancer. The specific biomarkers and mechanisms of colorectal cancer related-IS (CRCIS) were not fully investigated yet. Therefore, the aim of the study is to investigate the specific biomarkers and potential pathogenesis of CRCIS, which may contribute to the establishment of CRCIS therapeutic strategy.

***Research motivation***

Cancer related-IS has received widespread attention. However, biomarker and pathogenesis of CRCIS remain unclear. The key problems to be solved are how to identify CRCIS patients in clinical practice and whether hypercoagulability is the major cause and mechanism underlying hypercoagulability. To identify the biomarkers of CRCIS patients, which combined with clinical manifestation may be helpful to distinguish patients with other stroke etiology and other types of cancer-related stroke. Understanding the specific mechanisms in stroke of CRC patients is crucial to its therapeutic strategy.

***Research objectives***

The main objective of the retrospective study was to investigate the specific biomarkers and potential pathogenesis of CRCIS. The main findings were that hypercoagulability induced by elevated CEA and neutrophils may be an important cause of CRCIS, and the CRCIS index could serve as a potential biomarker. Future studies are needed to confirm these findings.

***Research methods***

The clinical data of 114 CRC patients with IS but without conventional stroke risk factors were retrospectively analyzed and compared with those of CRC patients without IS. Univariate and multivariate analyses were used to identify independent risk factors for CRCIS. The products of the independent risk factors in the two groups were calculated respectively. The ROC curve was used to determine the area under the curve and the optimal value of the products.

***Research results***

Our study found that to multiple lesions in multiple vascular territories on DWI were common in CRCIS patients (71, 62.28%). In addition, the level of plasma D-dimer, neutrophil count (NC) and carcinoembryonic antigen (CEA) and cancer antigen l25 were significantly higher in CRCIS patients than that in CRC patients. And D-dimer and NC and CEA were found to independently associate with CRCIS. Considering the combined effects of D-dimer and NC and CEA may be the major cause of hypercoagulability, the products of them were calculated. The area under the curve of the product was 0.889 ± 0.022, optimal cut-off value of the product was 252.06, which was called CRCIS Index in the study, with a sensitivity of 86.0% and specificity of 79.8%.

***Research conclusions***

Neutrophil extracellular traps generated from active neutrophils and CEA secreted by CRC cells that resulted in hypercoagulability may be the major cause of CRCIS.The CRCIS index which serves as a biomarker for CRCIS may need to be confirmed by future study.

***Research perspectives***

In the future, the detail mechanism of hypercoagulability in CRCIS patients may need to further illuminate. And CRCIS index that serves as a useful biomarker for the identification of CRCIS should be confirmed in prospective larger population studies.

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**Table 1 Clinical characteristics of the patients in the two groups**

|  |  |  |  |
| --- | --- | --- | --- |
| **Characteristics** | **CRCIS (*n* = 114)** | **CRC (*n* = 114)** | ***P* value** |
| Age  | 65.33 ± 12.45 | 63.67 ± 9.18 | 0.748a |
| Gender |  |  |  |
| Male, *n* (%) | 78 (68.42) | 78 (68.42) | 1.000b |
| Female, *n* (%) | 36 (31.58) | 36 (31.58) |  |
| Blood tests |  |  |  |
| WBC (× 109/L) | 7.71 ± 2.27 | 7.18 ± 1.97 | 0.058a |
| HGB (g/L) | 114.96 ± 23.09 | 122.57 ± 85.55 | 0.360a |
| PLT (109/L) | 242.28 ± 116.46 | 235.51 ± 82.35 | 0.613a |
| NC (109/L) | 5.65 ± 2.28 | 4.16 ± 1.49 | < 0.001a |
| PT (s) | 10.94 ± 1.40 | 10.82 ± 0.98 | 0.474a |
| INR | 0.95 ± 0.13 | 0.92 ± 0.08 | 0.137a |
| APTT (s) | 29.52 ± 6.79 | 30.11 ± 6.20 | 0.496a |
| FIB (g/L) | 5.42 ± 4.42 | 5.32 ± 4.74 | 0.862a |
| D-dimer (μg/mL) | 1.40 ± 0.93 | 0.56± 0.47 | < 0.001a |
| CRP (mg/L) | 33.54 ±35.50 | 28.79 ± 34.38 | 0.431a |
| CEA (U/mL) | 222.31 ± 175.85 | 88.48 ± 55.04 | < 0.001a |
| CA 125 (U/mL) | 47.13 ± 54.32 | 22.70 ± 34.61 | < 0.001a |
| CA 199 (U/mL) | 293.41 ± 1428.18 | 24.83 ± 34.212 | 0.066a |
| Type of CRC, *n* (%) |  |  |  |
| Adenocarcinoma | 107 | 110 | 0.235 b |
| Nonadenocarcinoma  | 7 | 4 |  |
| Systemic metastasis (n,%) |  |  | 0.080b |
| Yes  | 74 (64.9) | 61 (53.5) |  |
| No | 40 (35.1) | 53 (46.5) |  |
| Methods of therapy (n, %) |  |  |  |
| Surgery  | 76 (66.7) | 87(76.3) | 0.107b |
| Chemotherapy | 46 (40.4) | 33 (28.9) | 0.070b |
| No treatment | 28 (24.6) | 24 (21.2) | 0.528b |

a With two independent samples *t*-test. b With chi-square test. CRCIS: Colorectal cancer related-ischemic stroke; CRC: Colorectal cancer; WBC: White blood cell; HGB: Hemoglobin; PLT: Platelet; NC: Neutrophil count; PT: Prothrombin time; INR: International normalized ratio; APTT: Activated partial thromboplastin time; FIB: Fibrinogen; CRP: C-reactive protein; CEA: Carcinoembryonic antigen; CA: Cancer antigen.

**Table 2 Data of ischemic stroke onset *n* (%)**

|  |  |
| --- | --- |
| **Characteristics** | **No. of patients****(*n* =114)** |
| IS lesion pattern  |  |
| Single arterial lesion  | 43 (37.72) |
| Multiple arterial lesions  | 71 (62.28) |
| NIHSS scores at the day of IS onset  |  |
| 0-5 | 31 (27.19) |
| 6-15 | 66 (57.90) |
| 16-20  | 13 (11.40) |
| > 20 | 4 (3.51) |
| Time interval between CRC diagnosis and IS onset |  |
| IS as the first manifestation of CRC | 22 (19.30) |
| IS onset after CRC diagnosis |  |
| 0-6 mo | 60 (52.63) |
| 7-12 mo | 19(16.67) |
| > 12 mo | 13 (11.40) |

IS: Ischemic stroke; NIHSS: National Institutes of Health Stroke Scale; CRC: Colorectal cancer.

**Table 3 Multivariate logistic regression analysis**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Factors** | **β** | **SE** | **Wals (β)** | **Df** | ***P* value** | **OR** | **95%CI** |
| D-dimer | 0.002 | 0.001 | 26.660 | 1 | < 0.001 | 1.002 | 1.001-1.003 |
| NC | 0.486 | 0.127 | 14.617 | 1 | < 0.001 | 1.626 | 1.268-2.087 |
| CEA | 0.011 | 0.002 | 20.104 | 1 | < 0.001 | 1.011 | 1.006-1.015 |
| CA125 | 0.007 | 0.004 | 2.595 | 1 | 0.107 | 1.007 | 0.998-1.016 |
| ConstantConstant | -5.843 | 0.866 | 45.484 | 1 | < 0.001 | 0.003 |  |

NC: Neutrophil count; CEA: Carcinoembryonic antigen; CA125: Cancer antigen 125; SE: Standard error; OR: Odds ratio; CI: Confidence interval.

**CRC patients registered at 7 centers (n = 16 915) between January 2007 and December 2017**

(With histological evidence and diagnosed by oncologist expert)

*Excluded* (n=16 502)

Patients without acuteischemic stroke

**Colorectal cancer patients with acute ischemic stroke (n = 413)**

*Excluded* (n=299)

-Patients with conventional stroke risk factors

- Primary or metastatic brain cancer

-Other primary cancer or haematological cancer
- Not an active colorectal cancer

- Incomplete clinical data

**Colorectal cancer related-ischemic stroke patients**

**(CRCIS group: n= 114)**

**Active colorectal cancer patients alone (CRC group: n=114)**

**(**Age- and sex-matched with colorectal cancer related ischemic stroke**)**

**Figure 1 Patient selection.** CRC: Colorectal cancer.

**Figure 2 A case of a typical presentation of ischemic stroke in a colorectal cancer patient without conventional stroke mechanism.** A 47-year-old male patient experienced acute ischemic stroke in the first 3 mo after diagnosis of colorectal cancer. Diffusion-weighted magnetic resonance imaging showed high signal changes in the middle cerebral and anterior cerebral and anterior choroidal arteries. Echocardiography results were normal.

**Figure 3** **A receiver operator characteristic curves analysis of the product of D-dimer, carcinoembryonic antigen and neutrophil count.** The area under curve of the products was 0.889 ± 0.022 (95%CI: 0.847–0.932, *P* < 0.001). The optimum cut-off point was 252.06. At this cut-off value, the sensitivity was 86.0% and the specificity was 79.8%. ROC: Receiver operator characteristic.