The Incidence of Raised Procoagulant Factors and Hyperhomocysteinaemia in Chinese Patients with Chronic Venous Insufficiency

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KEYWORDS
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Chronic venous insufficiency;
Thrombophilia;
Homocysteine;
Thrombosis

Abstract  Background: For reasons that are poorly understood, there appear to be differences in the prevalence of chronic venous insufficiency (CVI) and venous thromboembolism between Caucasians and Asians.
Objectives: To compare levels of procoagulant factors and homocysteine (Hcy) in Hong Kong (HK) Chinese and United Kingdom (UK) Caucasian populations of patients with CVI (patients of CEAP clinical stages C4 – C6).
Methods: HK Chinese and UK Caucasian patients with CEAP clinical grade 4–6 venous disease were enrolled. Patients with conditions known to be associated with thrombophilia (TP) were excluded. UK and HK patients were matched by gender, age (within 5 years) and by CEAP clinical grade. All subjects underwent clinical examination, venous duplex ultrasound, and measurement of Hcy and factors (F) VIII, IX and XI.
Results: 63 Patients were enrolled in each group: Mean age 64y (HK group); 67y (UK group). 37% were female; 19% had active venous ulceration. One-third of patients in each group had deep venous reflux. High Hcy, FIX and FXI were significantly more common in the UK group. Multiple TP was more common in the UK group: raised levels of ≥2 factors in 26 vs. 14 patients (P = 0.022, χ²). Median Hcy (14.3 vs. 10.8 μmol/L; P < 0.0005, Wilcoxon signed rank [WSR]), FIX (131 vs. 115%; P = 0.048), and FXI (114 vs. 97%; P = 0.002) were significantly higher in the UK group. There was no significant difference in FVIII levels.

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Introduction

For poorly understood genetic and environmental reasons, there appear to be significant differences in the prevalence of chronic venous insufficiency (CVI) between Caucasians and Asians. Lower limb deep vein thrombosis (DVT), one of the principal causes of CVI, appears to be much less common in Asians than in Caucasians. This difference may be, at least in part, because the most important genetically determined risk factors for DVT in Caucasians, namely the factor V Leiden (FVL) and prothrombin (PT) gene G20210A mutations, are very rare in Asians, and because Caucasians, especially those with CVI, have higher levels of homocysteine (Hcy) and coagulation factors (FVIII, FIX and FXI). The aim of the present study was to compare United Kingdom (UK) Caucasian and Hong Kong (HK) Chinese populations of patients with CVI in terms of homocysteine (Hcy) and coagulation factor levels.

Methods

Following ethics approval and obtaining informed written consent from all volunteers, HK Chinese and UK Caucasian CVI (CEAP clinical grade 4–6) patients who did not have a condition known to be associated with an acquired thrombophilic defect (Table 1) were recruited at Kwong Wah Hospital, Hong Kong, Peoples Republic of China (PRC) and Heart of England NHS Foundation Trust, Birmingham, UK, respectively. These were consecutive patients seen in outpatient clinics at both hospitals. UK and HK patients were matched by gender, age (within 5 years), and by CEAP clinical grade. All patients underwent duplex ultrasound examination of both lower limbs conducted by one of the authors (CF). Total plasma Hcy concentration was determined using high-pressure liquid chromatography (D530; Drew, Cumbria, England) as previously described. Factors VIII, IX and XI activities (% where 100% is equal to 100 IU/dl) were measured on a fully automated coagulation analyzer using standard reagents (MDA II, Biomerieux, Basingstoke, Hampshire, UK) by a one-step clotting assay using clotting factor-deficient plasma. Hyperhomocysteinaemia was defined as a total plasma Hcy level above 15 µmol/L (95th percentile of the general Caucasian population). Elevated factor activities were defined as FVIII > 150%, FIX > 129% and FXI > 121% (90th percentile values of the control Caucasian population in published studies). There is no published evidence that Hcy and clotting factor levels in the general Asian population differ from the general Caucasian population, and indeed the published studies on thrombophilia in Asian populations use similar reference ranges.

A sample size calculation indicated that up to 27 patients would be required in each group to detect a difference of at least 30% in the activities of factors VIII, IX and XI, and of 4 µmol/L in plasma Hcy levels, with two-tailed 5% significance level and a power of 80% (specific sample sizes required were 27, 23, 19 and 17 for factors VIII, IX and XI and homocysteine respectively). Wilcoxon signed ranks test and Chi-squared test were used to determine differences between the UK and HK groups. The Statistical Package for the Social Sciences (SPSS), version 17.0 software (SPSS, Chicago, IL, USA) was used for data analysis.

Results

There were 63 HK Chinese patients and 63 UK Caucasian patients enrolled in the study. The characteristics and basic
CEAP classification of the two patient groups are shown in Table 2. One-third of patients in each group had deep venous reflux, but none had evidence of post-thrombotic deep vein damage on duplex ultrasound scan.

The percentage of patients with abnormally high levels of each procoagulant factor according to published reference ranges (detailed above) were calculated for both groups. Fig. 1 shows that raised levels of each procoagulant factor were much more common in the UK group compared with the HK group: 41% of the UK group had Hcy levels >15 μmol/L compared with 9% of the HK group ($P = 0.0001$, $χ^2$); FVIII levels were raised (>150% activity) in 19% of the UK group and 14% of the HK group ($P = 0.47$, $χ^2$); raised FIX levels (>129% activity) were found in 52% of the UK group and 32% of the HK group ($P = 0.029$, $χ^2$); and FXI levels were raised (>121% activity) in 40% of the UK group and 19% of the HK group ($P = 0.011$, $χ^2$).

Using the same published reference ranges Fig. 2 shows the number of patients with raised levels of no, one, or two or more of the four factors studied. Multiple TP was also more common in the UK group, with 26 of the patients having raised levels of two or more of the four factors studied, compared with 14 of the HK group ($P = 0.022$, $χ^2$).

Hcy levels were significantly higher in the UK group (median 14.3 μmol/L; interquartile range [IQR] 11.2–16.0) than the HK group (median 10.8 μmol/L; IQR 8.9–13.1; $P < 0.0005$, Wilcoxon signed ranks test) (Fig. 3). FVIII activity showed no significant difference between the groups (Fig. 4). FIX activity was significantly higher in the UK group (median 131%; IQR 114-143) than the HK group (median 115%; IQR 103-131; $P = 0.048$) (Fig. 5). Factor XI activity was also significantly higher in the UK group (median 114%; IQR 91-135) compared with the HK group (median 97%; IQR 83-116; $P = 0.002$) (Fig. 6).

**Discussion**

The main findings of this study are that levels of the procoagulants FIX and FXI and of Hcy are significantly higher in the Caucasian (UK) population with CVI compared with an age and sex-matched Hong Kong Chinese population also with CVI.

Venous thromboembolism (VTE) has been traditionally found to be less common in Asian populations than in Caucasians, although recent data suggest that the prevalence is increasing.5,24–26 Studies comparing the risk factors for VTE have consistently found differences between Caucasian and Asian populations in the genetic determinants of VTE. While the mutations FVL and PT20210A account for a large number of VTE cases in Caucasians,3,4 these mutations are rarely found in Asians.5–8 Conversely, the prevalence of deficiencies of the naturally occurring coagulation inhibitors, protein C, protein S and antithrombin III are higher in the general Thai, Chinese and Japanese populations compared with Caucasians.5,7,8,24

![Figure 1](image1.png) The percentage of patients with a high level of each procoagulant factor. ($P$ values obtained from chi-squared test).

**Table 2** Demographic data and basic CEAP classification. a

<table>
<thead>
<tr>
<th></th>
<th>Hong Kong Chinese</th>
<th>United Kingdom Caucasian</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean (range) in yrs</td>
<td>64 (34–87)</td>
<td>67 (31–84)</td>
</tr>
<tr>
<td><strong>Gender:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>23 (37)</td>
<td>23 (37)</td>
</tr>
<tr>
<td><strong>CEAP clinical grade of worst limb:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C4</td>
<td>27 (43)</td>
<td>26 (41)</td>
</tr>
<tr>
<td>C5</td>
<td>24 (38)</td>
<td>25 (40)</td>
</tr>
<tr>
<td>C6</td>
<td>12 (19)</td>
<td>12 (19)</td>
</tr>
<tr>
<td><strong>Etiology:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Primary (EP)</td>
<td>63 (100)</td>
<td>63 (100)</td>
</tr>
<tr>
<td>Secondary (ES)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td><strong>Anatomical patterns of venous reflux:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Superficial only (A5)</td>
<td>40 (64)</td>
<td>42 (67)</td>
</tr>
<tr>
<td>Deep only (A6)</td>
<td>4 (6)</td>
<td>7 (11)</td>
</tr>
<tr>
<td>Both superficial and deep (A56)</td>
<td>19 (30)</td>
<td>14 (22)</td>
</tr>
<tr>
<td><strong>Pathophysiological classification:</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reflux (Pr)</td>
<td>63 (100)</td>
<td>63 (100)</td>
</tr>
<tr>
<td>Obstruction (Po)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
</tbody>
</table>

*a Values in table are numbers with percentages in brackets unless otherwise stated.*
The genetic risk profile of VTE in Asians appears, therefore, to be very different to Caucasians. More recently, a number of acquired procoagulant thrombophilic defects, have been found to be associated with an increased risk of VTE in Caucasians. Previously published work from our group supports the hypothesis that patients with raised levels of procoagulants may be predisposed to thrombosis on both a macrovascular and microvascular level which may contribute to the skin changes of CVI. Specifically, we found significantly higher levels of FVIII, FIX, FXI and Hcy when compared with age and sex-matched controls with no clinical or duplex evidence of venous disease. The aim of the current study was to determine whether these acquired procoagulants are also raised in Chinese patients with CVI.

### Homocysteine

Hyperhomocysteinaemia (HHcy) has been associated with both arterial and venous thrombosis and is often defined as a level >15 μmol/L, a value determined from the 95th percentile of the general Caucasian population. It is believed that an increase of 5 μmol/L is associated with a 2–3-fold increase in VTE risk. A meta-analysis found the prevalence of HHcy in Caucasian VTE patients ranged from 5.7 to 34.8% and in healthy controls from 0.0 to 7.1%. The pooled odds ratio (OR) for VTE in the presence of HHcy was 2.95. In studies of healthy Chinese populations HHcy has been found to have a similar, if not higher, prevalence to that seen in Caucasian healthy populations. Gao et al. found HHcy in 44% of men (mean level 12.6 μmol/L) and 22% of women (mean level 8.4 μmol/L) with a mean age of 42 years, in an urban population in Beijing, China. However, their definition of HHcy was much lower than in other published studies (>12 μmol/L for men, and >11 μmol/L for women). Their mean Hcy values were however similar to another study of Hcy in a Chinese population; mean Hcy 15.4 μmol/L in men of 35–64 years and 12.2 μmol/L in women aged 35–64 years. Their prevalence rate of HHcy was 15.3%. The Singapore Chinese Health Study found mean Hcy levels of 11.2 μmol/L in men (mean age 58 years) and 9.1 μmol/L in women (mean age 57 years) with 95th percentile values of 17.4 and 16.5 μmol/L respectively.

Two published studies have looked at HHcy as a risk factor for VTE in Asian patients. The first, a case-control study of Taiwanese Chinese individuals with VTE and normal controls found neither plasma Hcy level (OR 1.07) nor HHcy (>15 μmol/L; OR 1.65) were significantly associated with VTE. Mean Hcy level in the cases was 10.5 μmol/L and 8.9% had HHcy (mean age 51 years). HHcy was also not associated with recurrent VTE in this study. The second study was a series of 105 consecutive Thai patients with VTE. Of the 85 patients in the series without malignancy (mean age 43 years), only 5.3% had HHcy (defined as >15 μmol/L). Apart from two previous studies from our group which found HHcy to be prevalent in Caucasian patients with CVI (up to 65% in patients with active ulceration) there are no published studies looking at Hcy, or indeed factors VIII, IX or XI, and CVI. In this study we found that total homocysteine levels were significantly lower in the Chinese patients compared with the Caucasian controls and HHcy was significantly less common in Chinese than Caucasians (9.5% vs. 40%). The median Hcy level in the Chinese patients was 10.8 μmol/L, similar to that found in the literature for healthy Asian patients and those with VTE. It doesn’t therefore appear that HHcy is associated with VTE in Asians or indeed with CVI in Chinese patients.
Factor VIII

An elevated FVIII (defined as >150 IU/dl, or 150% activity) is present in 10% of the general Caucasian population, and has been found to be present in 20–25% of Caucasian VTE patients.15,30,31 Risk of first VTE episode has been found to increase by 10% with each 10% increase in FVIII activity, even when values remain within the normal range.9,32 Mean FVIII activity is higher in one study in Caucasians and Japanese Americans compared with rural and urban Japanese,33 and Chinese patients have been found to have similar FVIII levels to Caucasians.18 Two studies have looked at FVIII levels in Asian VTE patients. The first, a case series of 50 consecutive Taiwanese Chinese patients with VTE found no patients had a raised FVIII level.34 The second study was a case series of 85 Thai patients with VTE and without malignancy.30 30.4% of the patients had a raised FVIII level (>150 IU/dL).19 In the present study, high FVIII activity levels were found in 14 and 19% of Chinese and Caucasian patients respectively. There was no significant difference in FVIII activity between Chinese and Caucasian patients with CVI. FVIII activity does not appear in this study to be related to CVI in Chinese patients.

Factor IX

Elevated FIX levels of >129% activity are found in 10% of the general Caucasian population and are associated with an almost 3-fold increase in the risk of VTE.16,35–37 No studies were available that looked at the prevalence of raised FIX in Asian patients, with or without VTE. In this study we found raised FIX levels in over 30% of the Chinese patients and over 50% of the Caucasian patients with CVI. We also found significantly higher FIX levels in the Caucasian patients compared with the Chinese patients.

Factor XI

Elevated FXI levels of >121% activity are found in 10% of the general Caucasian population and are associated with a 2-fold increase in the risk of VTE.16,35–37 No studies were available that looked at the prevalence of raised FXI in Asian patients, with or without VTE. In this study we found raised FXI levels in over 20% of the Chinese patients and over 50% of the Caucasian patients with CVI. We also found significantly higher FXI levels in the Caucasian patients compared with the Chinese patients.

**Figure 4** Box and whisker plot indicating factor VIII activity for UK Caucasian vs. HK Chinese patients.

**Figure 5** Box and whisker plot indicating factor IX activity for UK Caucasian vs. HK Chinese patients.

**Figure 6** Box and whisker plot indicating factor XI activity for UK Caucasian vs. HK Chinese patients.
increase in VTE risk. Angchaisuksiri et al. found a raised FXI in 26.8% of 85 Thai VTE patients without malignancy (defined as >140 IU/dL) and concluded that high FXI is a risk factor for VTE in Thais. In this study FXI activity was significantly higher in the Caucasian patients when compared with the Chinese patients.

The main weakness of this study is that we have no nonvenous control population for the Hong Kong patient group. The reason for this is manpower and financial constraints. However, there is no evidence from the literature that the reference ranges of Hcy, and factors VIII, IX and XI are any different in Chinese patients compared with Caucasian populations. In the absence of different reference ranges it is reasonable to use those that have been published, and similar to those used in VTE studies in many Asian populations.

In conclusion, Hong Kong Chinese patients with CVI do not appear to have the high levels of homocysteine and factors IX and XI that are seen in Caucasian patients with CVI. As with the inherited thrombophilias, the pattern of raised procoagulant factors in Chinese patients appears to differ from Caucasians.

Conflict of Interest/Funding

None.

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