Factor V Leiden is not commonly associated with idiopathic portal vein thrombosis in southern India

Abraham Koshy, Mary Jeyakumari

Department of Gastroenterology and Molecular Biology Unit, Institute of Population Health and Clinical Research, St. John’s Medical College Hospital, Bangalore 560 034

Background: Factor V Leiden has been reported in 2%-30% of patients with portal vein thrombosis. This wide variation makes it difficult to assess the importance of factor V Leiden as a predisposing factor.

Methods: Factor V Leiden was determined by restriction fragment length polymorphism in 112 patients with portal vein thrombosis, 104 with deep vein thrombosis and 98 control subjects.

Results: Only 3/112 (3%) patients with portal vein thrombosis had factor V Leiden, compared to 1/98 (1%) controls and 16/104 (15%) with deep vein thrombosis; of these, 3, 1 and 15, respectively, were heterozygous for this mutation.

Conclusion: Factor V Leiden contributes little, if at all, to the development of portal vein thrombosis in southern India.

Activated protein C resistance is associated with recurrent venous thrombosis, cerebral thrombosis and bad obstetric history. The most common cause of inherited activated protein C resistance is factor V Leiden (FVL). FVL is due to a single G1691A, R506Q mutation in exon 10 of the factor V gene. In the West, the prevalence of FVL in the general population is 0%-7% and that in patients with recurrent venous thrombosis is 3%-35%. The role of FVL as a predisposing factor for PVT is controversial. Studies from the Netherlands, Turkey, Egypt and Mumbai suggest that it is a predisposing factor, whereas studies from the UK, France, Italy and northern India suggest otherwise.

We determined the frequency of FVL in patients with PVT in southern India. An attempt was also made to detect clinical clues to distinguish patients with PVT who have FVL from those who do not.

Methods

Consecutive patients with PVT diagnosed by Doppler and/or CT examination of the abdomen between January 2001 and June 2005 were included in the study. Patients with underlying cirrhosis or pancreatitis were excluded. Cirrhosis was excluded on the basis of history, physical findings and ultrasonography. Acute pancreatitis was excluded on history, physical findings, serum amylase and lipase levels, and chronic pancreatitis on the basis of CT findings. One patient with combined hepatic and portal vein thrombosis who presented with clinical features of hepatic vein thrombosis was excluded. One hundred and four patients with deep vein thrombosis of the limbs (n=41) or cerebral vessels (n=63) but without portal or hepatic vein thrombosis were studied as disease controls. In addition, 98 healthy adults residing in two villages of Palamner Taluk, Chittoor District, Andhra Pradesh, comprising an ethnic mix of people from Andhra Pradesh, Karnataka and Tamilnadu states of southern India were selected from a community-based study.

Research guidelines of the institute Ethical Committee for the protection of human subjects were followed. All study subjects gave informed consent.

G1691A FVL mutation

Buffy coat was obtained from blood samples, and genomic DNA was extracted from it using QIAamp DNA Midi Blood kits (Qiagen GmbH, Hilden, Germany). DNA (100 ng) was amplified in 50 µL volume containing 200 pmol of primers, 2.5 U of Taq polymerase and 2.5 mmol/L MgCl2 in a thermal cycler, using specific primers (5’-TGCCCAGTGCTTAACAAGACCA and 5’-TGTTATCACACTGGTGCTAA).12

After 1 min at 94°C, samples were subjected to 35 cycles for 30 s each, at 94°C-56°C-72°C. The resulting 267-bp amplicon of the factor V gene including nucleotide 1691 (15 µL) was digested with 1 U Mn1. The digested fragments were run on 3% agarose gel for 60 min. The 200-bp (Leiden allele), 163-bp (wild type allele), 67-bp (both alleles) and 37-bp (wild type allele) bands were identified.

Statistical analysis

Inter-group comparisons of qualitative data were done using Fisher’s exact test.
## Results

Of 112 patients with PVT, 88 (79%) presented with upper gastrointestinal (GI) bleeding, 10 (9%) with abdominal pain, 8 (7%) with splenomegaly, 4 with fatigue, and one with jaundice due to portal hypertensive biliopathy; the remaining one patient was asymptomatic and was diagnosed during a routine check-up.

The clinical profile of patients with PVT is shown in the Table. Twenty-six patients had associated splenic vein thrombosis and ten had associated superior mesenteric vein thrombosis. Twenty-one (19%) patients were born of consanguineous parents; all consanguineous parents were 2nd degree relatives. One patient (without FVL) gave a history of deep vein thrombosis at initial presentation with upper GI bleed. Two patients had resection of gangrenous ileum at initial presentation; one of them died 1 week after resection of the gangrenous ileum. None of the patients gave family history of thrombosis. One patient was 7 months' pregnant, one delivered one month before detection of PVT, and one had tubectomy 2 days prior to initial presentation with upper GI bleed. One patient had essential thrombocythemia on bone marrow examination, with platelet count of 550 x10^9/L. One patient with platelet count of 780 x10^9/L had undergone splenectomy 15 years earlier. Two patients who had upper GI bleed and had received multiple transfusions were HBsAg positive. All other patients tested negative for HBsAg, anti-HCV and anti-HIV.

Three patients with PVT were heterozygous and none homozygous for FVL. Fifteen of 104 (14%) patients with deep vein thrombosis without PVT were heterozygous for FVL and one was homozygous. One of 98 (1%) healthy controls was heterozygous and none homozygous for FVL.

Among patients with PVT, the prevalence of FVL was higher in those who also had splenic vein thrombosis (3/26) as compared to those who did not (0/86; p=0.02, Fisher’s exact test).

## Discussion

The prevalence of FVL in the general population varies from 0% to 7%, being almost absent in Africa and the Middle East and common in Europe. In patients with venous thrombosis, the prevalence is higher at 19% in the Netherlands and 39% in northern India, but is only 3% in Mumbai. In patients with PVT, the prevalence is 7.6% in the Netherlands and 8% in Mumbai. The highest prevalence of 30% has been reported in a study of Egyptian children with PVT.

The present study is the second largest series of patients with PVT in whom FVL has been reported. Only three of 112 patients had FVL. Our results are similar to the largest series reported from France but are unlike the reports from the Netherlands and from western India (Mumbai). Our data also show that the low prevalence of FVL in southern Indian patients with PVT is not due to its absence in the general population since its prevalence in patients with venous thrombosis at other sites was moderately high.

We found that FVL was associated with splenic vein thrombosis. However, this association requires confirmation in a larger group of patients. Patients with PVT may have multiple predisposing factors. A subset of the patient group presented here was also tested for prothrombin G20210A gene variant as a predisposing factor but none were positive.

In conclusion, FVL is only occasionally associated with idiopathic portal vein thrombosis in southern India.

## Table: Clinical characteristics of 112 patients with portal vein thrombosis

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Median</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>23</td>
<td>1-65</td>
</tr>
<tr>
<td>M:F</td>
<td>73:39</td>
<td></td>
</tr>
<tr>
<td>Duration of disease (years)</td>
<td>2.5</td>
<td>0.1-42.0</td>
</tr>
<tr>
<td>Liver span (cm)</td>
<td>9.5</td>
<td>4-16</td>
</tr>
<tr>
<td>Spleen below costal margin (cm)</td>
<td>2</td>
<td>0-20</td>
</tr>
<tr>
<td>Serum albumin (g/L)</td>
<td>35</td>
<td>23-46</td>
</tr>
<tr>
<td>Serum bilirubin (mg/dL)</td>
<td>0.8</td>
<td>0.2-30.0</td>
</tr>
<tr>
<td>ALT (x upper limit of normal)</td>
<td>0.5</td>
<td>0.2-2.9</td>
</tr>
<tr>
<td>Platelet count (x 10^9/L)</td>
<td>115</td>
<td>25-780</td>
</tr>
<tr>
<td>INR</td>
<td>1.2</td>
<td>0.8-2.8</td>
</tr>
<tr>
<td>APTT (s)</td>
<td>27</td>
<td>24-63</td>
</tr>
<tr>
<td>Varices grade (0-4)</td>
<td>2</td>
<td>0-4</td>
</tr>
</tbody>
</table>

## References


Correspondence to: Professor Koshy, Lakeshore Hospital, Kochi 682304. Fax: (484)2701996. E-mail: koshyabe@yahoo.com

Acknowledgement: We thank Dr Auburn Jacob, Emmaus Swiss Leprosy Project, for access to the community and Drs Mario Vaz and Anura Kurpad for advice and assistance

Received November 24, 2005. Received in final revised form February 6, 2006. Accepted April 8, 2006