CLINICAL VALIDITY

Question 18: How often is the test positive when the disorder is present (i.e. sensitivity)?
Question 19: How often is the test negative when the disorder is not present (i.e. specificity)?
Question 20: Are there methods to resolve clinical false positive results in a timely manner?
Question 21: What is the prevalence of the disorder in this setting?
Question 22: Has the test been adequately validated on all populations to which it may be offered?
Question 23: What are the positive and negative predictive values?
Question 24: What are the genotype/phenotype relationships?
Question 25: What are the genetic, environmental or other modifiers?

CLINICAL VALIDITY
Question 18: How often is the test positive when the disorder is present (i.e. sensitivity)?
Question 19: How often is the test negative when the disorder is not present (i.e. specificity)?

Summary

Disorder/Setting

- The specific clinical disorder is recurrent venous thrombosis in individuals with an inherited clotting disorder, and the setting for offering the DNA testing is a confirmed recent episode of deep venous thrombosis in adults (Questions 1 and 3).

Factor V Leiden

- Five studies satisfy the criteria of the present analysis for determining clinical sensitivity and specificity.
- Clinical sensitivity of factor V Leiden testing answers the following question: For every 100 individuals with a recurrent episode of deep venous thrombosis, how many will carry a factor V Leiden mutation?
- The overall clinical sensitivity is 28 percent, with a 95 percent CI of 12.9-34.6%.
- Clinical false positive rate of factor V Leiden testing answers the following question: For every 100 individuals who do not experience a recurrent episode of deep venous thrombosis, how many will carry a factor V Leiden mutation?
- The overall clinical false positive rate (1-specificity) is 19 percent, with a 95 percent CI of 14.1-26.7%.
- The overall likelihood ratio for a recurrent episode of venous thrombosis among factor V Leiden carriers is 1.5.

Prothrombin G20210A

- Four studies satisfy the criteria of the present analysis for determining clinical sensitivity and specificity.
- Clinical sensitivity of prothrombin G20210A testing answers the following question: For every 100 individuals with a recurrent episode of deep venous thrombosis, how many will carry a prothrombin G20210A mutation?
- The overall clinical sensitivity is 11 percent, with a 95 percent CI of 6.2-21.1%.

- Clinical false positive rate of prothrombin G20210A testing answers the following question: For every 100 individuals who do not experience a recurrent episode of deep venous thrombosis, how many will carry a prothrombin G20210A mutation?
- The overall clinical false positive rate (1-specificity) is 6 percent, with a 95 percent CI of 5.6-6.8%.
- The overall likelihood ratio for a recurrent episode of venous thrombosis among prothrombin G20210A carriers is 1.8.

Introduction

In order to answer Question 18 in this review, it is necessary to document factor V Leiden or prothrombin G20210A mutation status in a cohort of individuals with an initial episode of venous thrombosis and then follow the cohort for a period of time to determine who develops a recurrence. Selection criteria for including published studies in the present analysis require a mean of at least four years' follow-up. Five studies satisfy these criteria for factor V Leiden, along with four studies for prothrombin G20210A. All of these studies include only Caucasians (both Hispanic and non-Hispanic). Rates are likely to be different for blacks (Question 22), as the prevalence of factor V Leiden is close to zero.

Sensitivity and Specificity of Factor V Leiden Testing

Question 18 asks how often a positive test for factor V Leiden (FVL) is associated with a recurrent episode of venous thrombosis among individuals who have been diagnosed with an initial episode. This defines the clinical sensitivity of a DNA test (see Appendix for description of 2 x 2 tables and the calculation of sensitivity and specificity). Figure 18-1 shows the mean clinical sensitivity and 95 percent confidence interval among the five selected studies. The confidence interval around the group estimate is calculated using the DerSimonian and Laird random effects model with an accompanying test of heterogeneity. According to the chi-square analysis, these studies are not statistically heterogeneous in their estimate of sensitivity. A total of 1637 individuals was followed for a minimum of 4 years, 385 of whom had a factor V Leiden mutation. The overall sensitivity of factor V Leiden mutation testing for predicting a recurrent episode of venous thrombosis in these studies is 28 percent. Table 18-1 shows the sizes of the individual studies, their respective sensitivities and the 95 percent confidence intervals. Details of the studies used for these calculations are in the Appendix. The Appendix also lists studies excluded from the present analyses because study subjects were not fully confirmed to have a first thrombotic event.

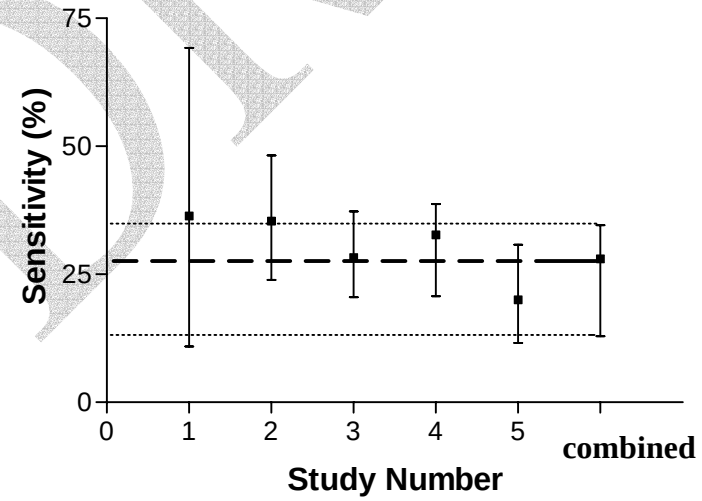

Figure 18-1. Clinical Sensitivity of the Factor V Leiden Mutation Testing. Study reference numbers are from Table 18-1. The individual and overall mean sensitivities are

shown as filled-in squares. Error bars are the 95 percent CI. The combined mean sensitivity is 28 percent (95 percent CI 13-35%). The chi-square for the test for heterogeneity is 3.43 and $p > 0.05$.

Question 19 asks how often a positive test for factor V Leiden is not present among individuals who do not experience a recurrence of venous thrombosis. This defines the clinical specificity of each DNA test. This aspect of clinical test performance can also be expressed as the false positive rate, which is 1-specificity. The false positive rate contributes to understanding test performance by directly expressing how often a mutation will be found among individuals who will not experience a recurrence of venous thrombosis. The false positive rates are shown graphically in Figure 18-2 for the five factor V Leiden studies, and the data on which they are based are shown in Table 18-1. For the five factor V Leiden studies, the overall false positive rate, adjusted for heterogeneity, is 19 percent. The chi-square for the test of heterogeneity in these studies' estimates of the false positive rate is significant, as shown (see Appendix for further discussion of heterogeneity). In addition, the sensitivity divided by the false positive rate gives the likelihood ratio. This is a useful estimate of the test's power to alter pre-test probability of an outcome. The likelihood ratio for factor V Leiden testing is 1.5, as shown in Table 18-1. This means that individuals found to have a factor V Leiden mutation are one and a half-times more likely to experience a recurrence than would have been known in the absence of testing. This knowledge does not provide strong clinical guidance.

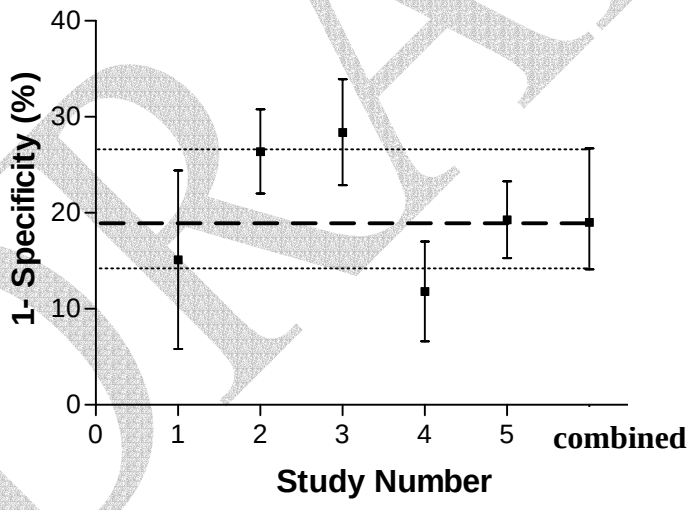

Figure 18-2. Clinical False Positive Rate (1-Specificity) of the Factor V Leiden Mutation Among Individuals Who Will Not Experience a Recurrence. Study numbers are from Table 18-1. The individual and overall mean false positive rates are shown as filled-in squares. Error bars are the 95 percent CI. The combined mean false positive rate is 19 percent (95 percent CI 14-27%). The chi-square for the test for heterogeneity is 36.4 and $p < 0.01$.

Table 18-1. Factor V Leiden -- Clinical Sensitivity, False Positive Rate (1-Specificity), and Likelihood Ratio (LR) for Identifying Recurrent Episodes of Deep Venous Thrombosis

Author	N	N with factor V Leiden	Sensitivity (95 percent CI)	False Positive Rate (95 percent (CI)	LR
1. (Ridker et al., 1995)	77	14	36.4 (10.9-69.2)	15.2 (7.5-26.1)	2.4
2. (Lindmarker et al., 1999)	467	129	35.4 (23.9-48.2)	26.4 (22.1-31.0)	1.3
3. (De Stefano et al., 1999)	395	112	28.3 (20.5-37.3)	28.4 (23.1-34.1)	0.99
4. (Simioni et al., 2000)	224	38	32.7 (20.7-46.7)	11.8 (7.4-17.7)	2.8
5. Unpublished data LETS	474	92	20.0 (11.6-30.8)	19.3 (15.5-23.5)	1.0
Overall	**1637**	**385**	**28** (12.9-34.6)	**19** (14.1-26.7)	**1.5**

Sensitivity and Specificity of Prothrombin G20210A Mutation Testing

Figure 18-3 shows the mean clinical sensitivity and 95 percent confidence interval for the four selected studies of prothrombin G20210A mutation testing. The criteria for selecting these studies were identical to those for the factor V Leiden studies. The asymmetric confidence interval around the estimate for the group of studies is calculated using the DerSimonian and Laird random effects model with an accompanying test of heterogeneity. A total of 1326 individuals was followed for a minimum of 4 years, 95 of whom had a prothrombin G20210A mutation. The overall sensitivity of prothrombin G20210A mutation testing for predicting a recurrence in these studies is 11 percent. The chi-square for the test of heterogeneity in these studies' estimates of the sensitivity is significant. Table 18-2 shows the size of the individual studies, their respective sensitivities and the 95 percent confidence intervals. Details of the studies used for these calculations are in the Appendix. The Appendix also lists studies excluded from the present analyses because study subjects were not fully confirmed to have a first thrombotic event.

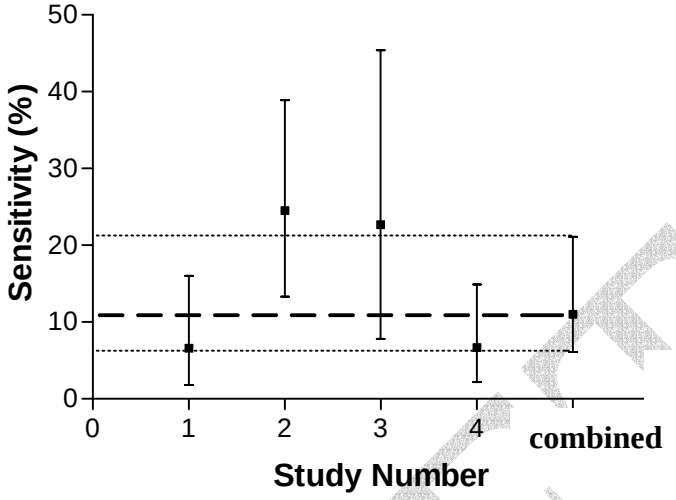

Figure 18-3. Clinical Sensitivity of Prothrombin G20210A Mutation Testing. Study numbers are from Table 18-2. The individual and overall mean sensitivities are shown as filled-in squares. Error Bars are the 95 percent CI. The combined mean sensitivity is 11% (95% CI 6-21%). The chi-square for the test for heterogeneity is 23.3 and p <0.01.

The clinical false positive rate (1-specificity) for prothrombin G20210A mutation testing in the four selected studies is shown in Figure 18-4. The data on which this figure is based are found in Table 18-2. The overall false positive rate is six percent. The chi-square for the test of heterogeneity in these studies' estimates of the false positive rate is significant. Table 18-2 also shows that overall likelihood ratio for prothrombin G20210A testing is 1.8. This is similar to factor V Leiden testing and demonstrates that prothrombin G20210A testing does not provide strong clinical guidance.

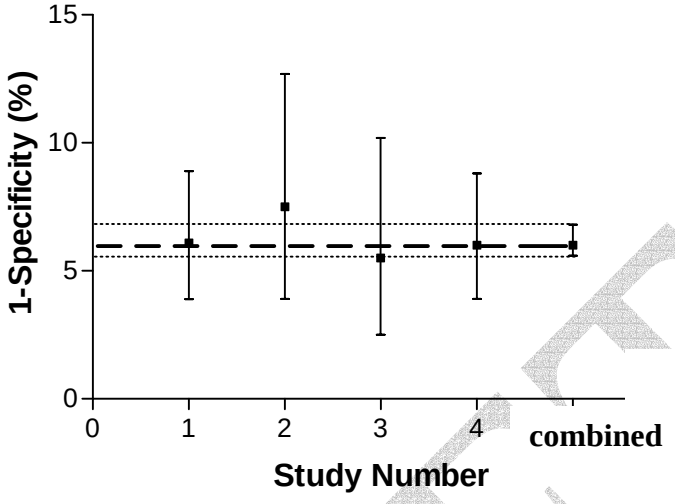

Figure 18-4. Clinical False Positive Rate (1-Specificity) for Prothrombin G20210A Mutation Testing for Individuals Who Will Not Experience a Recurrence. Study numbers are from Table 18-2. The combined mean false positive rate is 6 percent (95 percent CI 5.7-7%). The chi-square for the test for heterogeneity is 5.48 and p <0.01.

Table 18-2. Prothrombin G20210A – Clinical Sensitivity, False Positive Rate (1-Specificity), and Likelihood Ratio (LR) for Identifying Recurrent Episodes of Deep Venous Thrombosis

Author	N	N with Prothrombin G20210A Mutation	Sensitivity (95 percent CI)	False Positive Rate (95 percent CI)	LR
1. (Lindmarker et al., 1999)	456	28	6.6 (1.8-16.0)	6.1 (3.9-8.9)	1.1
2. (Simioni et al., 2000)	210	24	24.5 (13.3-38.9)	7.5 (3.9-12.7)	3.3
3. (Miles et al., 2001)	186	14	22.7 (7.8-45.4)	5.5 (2.5-10.2)	4.1
4. Unpublished data LETS	474	29	6.7 (2.2-14.9)	6.0 (3.9-8.8)	1.1
Summary	**1326**	**95**	**11** (6.2-21.1)	**6** (5.6-6.8)	**1.8**

References

Berlin, J. A., Laird, N. M., Sacks, H. S., and Chalmers, T. C. 1989. A comparison of statistical methods for combining event rates from clinical trials. *Stat Med* **8**(2)**:**141-51.

De Stefano, V., Martinelli, I., Mannucci, P. M., Paciaroni, K., Chiusolo, P., Casorelli, I., Rossi, E., and Leone, G. 1999. The Risk of Recurrent Deep Venous Thrombosis among Heterozygous Carriers of Both Factor V Leiden and the G20210A Prothrombin Mutation. *N Engl J Med* **341**(11)**:**801-806.

De Stefano, V., Martinelli, I., Mannucci, P. M., Paciaroni, K., Rossi, E., Chiusolo, P., Casorelli, I., and Leone, G. 2001. The risk of recurrent venous thromboembolism among heterozygous carriers of the G20210A prothrombin gene mutation. *Br J Haematol* **113**(3)**:**630-5.

Eichinger, S., Pabinger, I., Stumpflen, A., Hirschl, M., Bialonczyk, C., Schneider, B., Mannhalter, C., Minar, E., Lechner, K., and Kyrle, P. A. 1997. The risk of recurrent venous thromboembolism in patients with and without factor V Leiden. *Thromb Haemost* **77**(4)**:**624-8.

Kearon, C., Gent, M., Hirsh, J., Weitz, J., Kovacs, M. J., Anderson, D. R., Turpie, A. G., Green, D., Ginsberg, J. S., Wells, P., MacKinnon, B., and Julian, J. A. 1999. A comparison of three months of anticoagulation with extended anticoagulation for a first episode of idiopathic venous thromboembolism. *N Engl J Med* **340**(12)**:**901-7.

Lindmarker, P., Schulman, S., Sten-Linder, M., Wiman, B., Egberg, N., and Johnsson, H. 1999. The risk of recurrent venous thromboembolism in carriers and non-carriers of the G1691A allele in the coagulation factor V gene and the G20210A allele in the prothrombin gene. DURAC Trial Study Group. Duration of Anticoagulation. *Thromb Haemost* **81**(5)**:**684-9.

Margaglione, M., D'Andrea, G., Colaizzo, D., Cappucci, G., del Popolo, A., Brancaccio, V., Ciampa, A., Grandone, E., and Di Minno, G. 1999. Coexistence of factor V Leiden and Factor II A20210 mutations and recurrent venous thromboembolism. *Thromb Haemost* **82**(6)**:**1583-7.

Miles, J. S., Miletich, J. P., Goldhaber, S. Z., Hennekens, C. H., and Ridker, P. M. 2001. G20210A mutation in the prothrombin gene and the risk of recurrent venous thromboembolism. *J Am Coll Cardiol* **37**(1)**:**215-8.

Ridker, P. M., Miletich, J. P., Stampfer, M. J., Goldhaber, S. Z., Lindpaintner, K., and Hennekens, C. H. 1995. Factor V Leiden and risks of recurrent idiopathic venous thromboembolism. *Circulation* **92**(10)**:**2800-2.

Rintelen, C., Pabinger, I., Knobl, P., Lechner, K., and Mannhalter, C. 1996. Probability of recurrence of thrombosis in patients with and without factor V Leiden. *Thromb Haemost* **75**(2)**:**229-32.

Simioni, P., Prandoni, P., Lensing, A. W. A., Manfrin, D., Tormene, D., Gavasso, S., Girolami, B., Sardella, C., Prins, M., and Girolami, A. 2000. Risk for subsequent venous thromboembolic complications in carriers of the prothrombin or the factor V gene mutation with a first episode of deep-vein thrombosis. *Blood* **96**(10)**:**3329-3333.

APPENDIX

Two-by-Two Tables

The definitions of clinical sensitivity (Question 18) and clinical specificity (Question 19) for predicting a recurrence of venous thrombosis can be derived using a two-by-two contingency table for data from case/control or cohort studies.

Table 1. A Two-by-Two Contingency Table for Deriving the Four Major Clinical Performance Parameters

	Clinically confirmed Recurrence		
	Yes	No	Totals
Factor V Leiden or Prothrombin G20210A Mutation*			
Yes	A	B	A+B
No	C	D	C+D
Totals	A+C	B+D	A+B+C+D

*This example assumes 100% analytic sensitivity and specificity.

- Clinical sensitivity [A / (A + C)] is the proportion of individuals with a confirmed recurrence of venous thrombosis (A+C) who are correctly identified as being factor V Leiden or prothrombin G20210A mutation carriers (A).
- Clinical specificity [D / (B + D)] is the proportion of individuals without a recurrence of venous thrombosis (B+D) who are correctly identified as not being factor V Leiden or prothrombin G20210A mutation carriers (D).
- Positive predictive value [A / (A + B)] is the proportion of positive tests (A + B) that correctly identifies individuals destined to have a recurrence (A). This can be directly derived only if data in the table are from a population-based cohort study(ies).
- Negative predictive value [D / (C + D)] is the proportion of negative tests (C + D) that correctly identifies individuals destined not to have a recurrence (D). This also can be directly derived only if data in the table are from a population-based cohort study(ies).

Test of Heterogeneity

The DerSimonian and Laird modified Cochran method produces a weighted average of differences in rates between studies (Berlin et al., 1989). This method allows for a between-study estimate of variability providing a test for heterogeneity.

Characteristics of Reviewed Studies

Nine studies were found that had information on the effect of the factor V Leiden or prothrombin G20210A mutation for the rate of recurrence. For the calculations described in Questions 18 and 19, we excluded those studies that did not include adequate documentation of a confirmed first

venous thrombotic event, as specified in the disorder definition (Rintelen et al. 1996, Eiching et al. 1997, Margaglione et al. 1999, Kearon et al. 1999, De Stefano 2001). However, all studies are shown in the tables that follow. Overall characteristics of the studies for factor V Leiden calculations are shown in Table 2, and calculations for prothrombin G20210A mutation are shown in Table 5.

The following calculations are performed: sensitivity, specificity, incidence of recurrence per year, and the relative risk (if not specified in the article by the authors). For each article, the yearly incidence of recurrence among factor V Leiden carriers is calculated by dividing the number of individuals with a recurrent event by the number of person-years of follow-up, once for those with, and once for those without, recurrences of venous thrombosis among individuals with a factor V Leiden mutation. The relative risk is the ratio of the two incidence rates.

Factor V Leiden Mutation

Sensitivity and specificity for all studies

Table 2 gives an overall description of the nine available studies. Table 3 shows the sensitivity, false positive rate (1-specificity) and likelihood ratio for all studies. The proportion of factor V Leiden carriers among those with a recurrence ranges from 18.8 to 54.4 percent (sensitivity), with an overall sensitivity of 28 percent for included studies and 26 percent for excluded studies. Table 3 also shows that 11.8 to 48.4 percent of the individuals without a recurrence had the factor V Leiden mutation (false positive rate), with an overall false positive rate of 19 percent for included studies and 27 percent for excluded studies. Table 3 also shows the individual and combined likelihood ratios. The test of heterogeneity for the studies included and excluded from Table 18-1 is also shown. All calculations except sensitivity in included studies involved heterogeneity.

Annual incidence of recurrent episodes of venous thrombosis among factor V Leiden carriers and overall relative risk

Table 4 shows that the mean annual incidence of recurrence in factor V Leiden carriers is 4.8 percent (range 3.0 to 6.7 percent). The relative risk of recurrence for factor V Leiden carriers compared with non-carriers ranges from 1.0 (95 percent CI 0.7-1.5%) to 4.1 (95 percent CI 1.2-14%) in the studies included in the calculations in Table 18-1.

Table 2. Overall Description of Studies Dealing with the Effect of factor V Leiden Mutation on Recurrence, Including the Five Used for Present Calculations and the Four Excluded

Author	age (range)	Country	mean FU years	Type of study	1st Venous Thrombotic Event (n)*	sex M:F	Recurrence (n)
Papers included in calculations for Table 18-1							
Ridker 1995	… (40-84)	USA	5.7	Prospective	77	77:0	11
Lindmarker 1999	58 (15-70)	Sweden	4	Prospective	467	265:202	65
De Stefano 1999	43 (9-77)	Italy	6	Retrospective	395	171:224	120
Simioni 2000	62 (23-84)	Italy	8.3	Prospective	224	117:107	55
Unpublished Data LETS 2002	44 (14-72)	Netherlands	8.1	Prospective	474	272:202	75
Papers excluded in calculations for Table 18-1							
(Rintelen et al., 1996)	49 (21-73)	Austria	5.0	Retrospective	42 (1st or >1)	10:32	11(>1 prev VT)
(Eichinger et al., 1997)	51.1(not given)	Austria	1.7	Prospective	380 (1st or >1)	174:206	36
(Margaglione et al., 1999)	45 (15-85)	Italy	2.8	Retrospective	465(1st or >1)	228:237	63
(Kearon et al., 1999)	>=60	Canada	0.8	Randomized trial	75 (1st or >1)	Not given	16

*This column only includes individuals with factor V Leiden or control individuals, and thus excludes those participants included with a positive prothrombin G20210A test.

Table 3. Factor V Leiden -- Individual and Collective Clinical Sensitivities, False Positive Rates (1-Specificity), and Likelihood Ratios Found by the Nine Studies in Table 2 for Predicting a Recurrence

Author	N	N with factor V Leiden	Sensitivity (95%CI)	False Positive Rate (1-Specificity) (95%CI)	LR*
Papers included in calculations Table 18-1					
Ridker 1995	77	14	36.4 (10.9-69.2)	15.2 (7.5-26.1)	2.4
Lindmarker 1999	467	129	35.4 (23.9-48.2)	26.4 (22.1-31.0)	1.3
De Stefano 1999	395	112	28.3 (20.5-37.3)	28.4 (23.1-34.1)	0.99
Simioni 2000	224	38	32.7 (20.7-46.7)	11.8 (7.4-17.7)	2.8
Unpublished data LETS	474	92	20.0 (11.6-30.8)	19.3 (15.5-23.5)	1.0
Overall	**1637**	**385**	**28** (12.9-34.6)[1]	**19** (14.1-26.7)[2]	**1.5**
Papers excluded in calculations Table 18-1					
Rintelen 1996	42	21	54.5(23.4-83.2)	48.4 (30.2-66.9)	1.1
Eichinger 1997	380	112	27.8 (14.2-45.2)	29.7 (24.9-34.8)	0.9
Margaglione 1999	465	99	25.4 (15.3-37.9)	20.6 (16.8-24.9)	1.2
Kearon 1999	75	20	18.8 (4.0-45.6)	28.8 (17.8-42.1)	0.6
Overall	**962**	**252**	**26** (18.3-37.8)[3]	**27** (20.1-36.3)[4]	**0.9**

*Likelihood ratio

1- Test of heterogeneity: χ^2=3.43, p>0.05

2- Test of heterogeneity: χ^2=36.4, p<0.01

3- Test of heterogeneity: χ^2=1.66, p>0.05

4- Test of heterogeneity: χ^2=9.03, p<0.01

Table 4. Annual Incidences and Relative Risks of Recurrence Among factor V Leiden Carriers found by the Nine Studies in Table 2

Author	Factor V Leiden mutation (N)	Factor V Leiden carrier frequency %‡	Factor V Leiden mutation with Recurrence (N)	Incidence of Recurrence in factor V Leiden carriers per year %	Overall RR (95%CI) (with factor V Leiden vs. without factor V Leiden)
Papers included in calculations					
Ridker 1995	14(0)**	18.2	4(0)**	5.0	RR 4.1 [1.2-14]
Lindmarker 1999	129(11)	27.6	23(4)	4.5	RR=1.4 [0.8-2.3]
De Stefano 1999	112(0)	28.4	34(0)	5.0	RR 1.1 [0.7-1.6]
Simioni 2000	38(0)	17.0	18(0)	6.7	RR: 2.4 [1.4-4.1]
Unpublished data LETS	92 (8)	19.4	15 (not given)	3.0	RR: 1.0 [0.7-1.5]
Summary				**4.8**	
Papers excluded in calculations					
Rintelen 1996	21(5)	50.0	6(2)	5.7	RR=1.0 [0.3-3.3]
Eichinger 1997	112(10)	29.5	10(1)	5.2	RR=1.0 [0.5-2.1]
Margaglione 1999	99(not given)	21.3	16 (not given)	5.8	OR: 1.3 [0.7-2.4]
Kearon 1999	20(1)	26.7	3 (1)	19.0	RR: 0.5 [0.1-1.8]

‡ Carrier frequency of the factor V Leiden mutation among individuals with a first venous thrombosis
**Number in ( ) = Homozygotes

Prothrombin G20210A mutation

Clinical Sensitivity and specificity for all studies

Table 5 shows the important characteristics of the seven available studies. Table 6 shows the sensitivity, false positive rate (1-specificity) and likelihood ratio for all studies. The prevalence of prothrombin G20210A carriers among those with a recurrence ranged in different studies from 6.6 to 24.5 percent (clinical sensitivity), with an overall clinical sensitivity of 11 percent for included studies and 12 percent for excluded studies. The prevalence of individuals without the prothrombin G20210A mutation among those with a first but not a second episode of venous thrombosis ranged in these studies from 85 percent to 94.5 percent. This means that 5.5 to 15 percent of the individuals without a recurrence had the prothrombin G20210A mutation (false positive rate), with an overall false positive rate of 6 percent for included studies and 12 percent for excluded studies. Table 3 also shows the individual and combined likelihood ratios. The test of heterogeneity for the studies included and excluded from Table 18-2 is shown. All calculations involved heterogeneity.

Incidence of recurrence in prothrombin G20210A carriers and overall relative risk

Table 7 shows that the mean annual incidence of recurrence in prothrombin G20210A carriers was 4.2 percent (range 2.1 to 6.0 percent) and that the relative risk of recurrence for prothrombin G20210A carriers compared with non-carriers ranged from 0.9 percent (95 percent CI 0.2-2.9%) to 4.9 percent (95 percent CI 1.9-12.9%) in studies included in the calculations included in Table 18-2.

Table 5. Overall Description of Studies Dealing with the Effect of Prothrombin G20210A on Recurrence, Including the Four Used for Present Calculations and the Three Excluded

Author	age (range)	Country	mean follow-up (years)	Type of study	1st venous thrombotic event (n)*	sex M:F	Recurrence (n)
Papers included in calculations							
Lindmarker 1999	58(15-70)	Sweden	4	Prospective follow-up	456	Not given	61
Simioni 2000	62(23-84)	Italy	8.3	Prospective follow-up	210	106:104	49
Miles 2001	(see previous article)	USA	7.3	Prospective follow-up	186	186:0	22**
Unpublished Data - LETS	44 (14-72)	Netherlands	8.1	Prospective follow-up	474	272:202	75
Papers excluded in calculations							
Eichinger 1999	49.8 (not given)	Austria	2.0	Prospective follow-up	492 (1st or >1)	227/265	57(>1 prev VT)
Margaglione 1999	45(15-85)	Italy	2.8	Retrospective follow-up	421 (1st or >1)	197:224	58
(De Stefano et al., 2001)	43.7	Italy	6	Retrospective follow-up	335 (1st or >1)	146:189	115

*This column only includes individuals with the prothrombin G20210A mutation or control individuals, and thus excludes those participants included with a positive factor V Leiden test. However, for the study by Eichinger et al 1999, Miles et al 2001 it was not possible to exclude the factor V Leiden positive individuals from the calculations.

**10% of all non-carriers (n=172) had a recurrence, so the number of recurrences could be 17 or 18, and therefore the total number 22 or 23

Table 6. Prothrombin G20210A Mutation -- Clinical Sensitivity, False Positive Rate (1-Specificity), and Likelihood Ratio for Recurrence

Author	**N**	**N with Prothrombin G20210A Mutation**	**Sensitivity (95%CI)**	False Positive Rate (95%CI)	LR
Papers included in calculations in Table 18-2					
Lindmarker 1999	456	28	6.6 (1.8-16.0)	6.1 (3.9-8.9)	1.1
Simioni 2000	210	24	24.5 (13.3-38.9)	7.5 (3.9-12.7)	3.3
Miles 2001	186	14	22.7 (7.8-45.4)	5.5 (2.5-10.2)	4.1
Unpublished data LETS	474	29	6.7 (2.2-14.9)	6.0 (3.9-8.8)	1.1
Summary	**1326**	**95**	**11** (6.2-21.1)[1]	**6** (5.6-6.8)[2]	**1.8**
Papers excluded in calculations in Table 18-2					
Eichinger 1999	492	42	9.4 (1.1-14.6)	8.1 (6.4-12.0)	1.2
Margaglione 1999	421	55	19.0 (9.9-31.4)	12.1 (9.0-15.4)	1.6
De Sefano 2001	335	52	16.5 (12.5-24.6)	15.0 (10.6-20.4)	1.1
Summary	**1248**	**149**	**12** (5.1-26.9)[3]	**12** (8.7-15.6)[4]	**1.2**

1- Test of heterogeneity: χ^2=23.5, p<0.01
2- Test of heterogeneity: χ^2=35.8, p<0.01
3- Test of heterogeneity: χ^2=5.48, p<0.05
4- Test of heterogeneity: χ^2=18.8, p<0.01

Table 7. Annual Incidences and Relative Risks of Recurrence Among Prothrombin G20210A Carriers Found by the Seven Studies in Table 5

Author	Prothrombin G20210A mutation (N)	Prothrombin G20210A mutation carrier frequency %‡	Heterozygous Prothrombin G20210A mutation with recurrence (N)	Incidence of Recurrence in Prothrombin G20210A mutation carriers per year %	overall RR (95%CI) (with prothrombin G20210A mutation vs. without prothrombin G20210A mutation)
Papers included in calculations					
Lindmarker 1999	28 (0)**	6.1	4 (0)**	3.6	OR: 0.9 [0.2-2.9]
Simioni 2000	24 (0)	11.4	12 (0)	6.0	RR: 2.4 [1.3-4.7]
Miles 2001*	14 (0)	7.5	5 (0)	4.9	RR: 4.9 [1.9-12.9]
Unpublished data LETS	29 (0)	6.1	5 (0)	2.1	RR: 1.1 [0.5-2.5]
Summary				**4.2**	
Papers excluded in calculations					
Eichinger 1999*	42 (1)	8.5	3 (not given)	3.6	RR: 0.7 [0.2-2.1]†
Margaglione 1999	55 (not given)	13.1	11 (not given)	7.1	OR: 1.7 [0.8-3.5]
De Stefano 2001	52 (0)	15.5	19 (0)	5.2^	RR: 1.2 [0.7-1.9]

* In the articles by Eichinger et al 1999 and Miles et al 2001 it was not possible to exclude individuals who carried the factor V Leiden mutation.

^ Mean number of follow-up years was 7 in the prothrombin G20210A mutation carriers.

‡ Carrier frequency of the prothrombin G20210A mutation in patients with a first venous thrombosis event

† RR was adjusted for the factor V Leiden carrier status and age

**Numbers in ( ) = Homozygotes