The recurrent fetal loss was defined by at least three successive abortions. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week. As a miscarriage was considered fetal loss before 28th week of pregnancy, as a stillbirth a termination of pregnancy after 28th week.

Deep venous thrombosis is a serious but rare vascular complication during pregnancy and puerperium and pulmonary embolism is one of the most important causes of maternal mortality (10,15). The risk of thromboembolism in pregnancy (16). The risk is even higher in women with congenital thrombophilia (6,13). In 1993...
Materials and methods

In 1996 we examined 500 blood donors (309 males, 191 females) to get prevalence of APC-R in the East Bohemian region. This prevalence has been found to be 1.6%. We assessed the association of APC-R and fV Leiden with spontaneous fetal loss in three cohorts of women.

1. A prospective group

Thirty women admitted to the hospital for spontaneous or recurrent fetal loss were tested for APC-R and fV Leiden. All females completed the questionnaire about family and personal history of VTE. Characteristics of this group are shown in table 1. All women were also tested for protein C, protein SAT III levels, for the presence of lupus anticoagulant (LA) and antitryptidin antibody (ACA).

2. A retrospective group

In this group we assessed the course of 172 pregnancies in 80 women with APC-R and fV Leiden (72 heterozygous, 8 homozygous) from 57 unrelated families. Diagnosis of APC-R and fV Leiden was made either in the laboratory workup of women with personal history of thrombosis event or in the laboratory work-up of family members of individuals with personal history of thrombosis and with APC-R and fV Leiden. Characteristics of this group are shown in table 2. All women with history of fetal loss were also tested for ACA, LA, protein C, protein S and AT III.

3. A control group

In this group we evaluated the course of 104 pregnancies in 45 women without APC-R and fV Leiden from 42 unrelated families. These women are relatives of individuals with personal history of thrombosis event and with diagnosis of APC-R and fV Leiden. Characteristics of this group are shown in table 3. All women with the history of pregnancy loss were tested for ACA, LA, protein C, protein S and AT III as well.

Methods: Blood samples were collected by venipuncture into plastic tubes containing either 1/10 volume of 3.8% sodium citrate for coagulation assays or 1/10 volume of 0.5 M sodium fluoride (21) for DNA extraction after 15 min (at 2500 G) for prothrombin time (PT), activated partial thromboplastin time (APTT) and AT III assays or after double centrifugation (+10 min at 15 000 G) for protein C, protein SAT-III and LA assays, citrated plasma was either analyzed immediately (PT, APTT, LA) or stored at -70 °C until analyzed (AT III, protein C, protein SAT-III). APC-R was determined by COATEK APC RESISTANCE test (Chromogenix). Low response for APC was defined as SR<2.05 (SR=sensitivity ratio-clot time APTT+APC to APTT without APC). Protein C and protein S were determined by coagulation assays using STACLOT PROTEIN C and STACLOT PROTEIN S kits. AT III was determined by chromogen assays using STACHROM AT III kit. All kits are from STAGO Diagnostics. To detect LA, the following assays were performed: PT, APTT (PTT Automate, Stago D.), PTT with high sensitivity (PTT Automate, STAGO D.), PTT (Tissue Thromboplastin Inhibition Time), dRVVT (diluted Russell’s Viper Venom Time). A solid phase immunoassay technique was used to quantify anticoagulins in the following levels: IgG level >10 U/ml and IgM level >7 U/ml were considered as positive results. PCR method was used for fV Leiden determination.

Statistical analysis: Data were evaluated by software program NCSS 6.0.1 using Fisher’s test for categorial variables.

Results

1. The prospective group

In the group of 30 women admitted to the hospital we obtained the results which are shown in table 4. Stillbirths did not occur in any of these women. LA and protein C, protein S, AT III deficiencies were not detected either. Frequency of APC-R has been found to be 7% (1%-22%), frequency of fV Leiden 3% (0%-17%).

2. The retrospective group

By the assessment of 172 pregnancies in 80 women with fV Leiden we got the results which are shown in table 5. Stillbirths did not occur in any of these women. Protein C, protein S and AT III deficiencies were not found in any of 4 women with abortions. Antiphospholipid syndrome was diagnosed in one woman with recurrent abortions (positivity of LA and ACP). Frequency of women with abortion in this group is 10% (4%-19%). Frequency of abortions is 6% (4%-14%).

3. The control group

The results in cohort of women without fV Leiden are shown in table 6. Stillbirths did not occur in any of these women. Protein C, protein SAT III deficiencies were not found in any of 4 women with spontaneous abortions either. Frequency of women with abortion in this group is 9% (2%-21%) and frequency of abortions is 6% (2%-12%). Using Fisher’s exact test we have not found statistical difference (p=0.67) between frequency of women with abortions in the retrospective group and in the control group. We have not proven statistical difference (p=0.63) between frequency of abortions in women in these groups either.

Discussion

Since 1996 several reports have been published about the relationship between APC-R and fV Leiden and fetal loss. These studies assessed either the association of fV Leiden with recurrent fetal loss or the association with miscarriages and stillbirths. EPCOT study (European Prospective Cohort on Thrombophilia) is the largest study which analyzed the risk of fetal loss in women with known thrombophilia. Researchers found that the odds ratios were 3.6 (95% CI 1.4-9.4) for stillbirths and L.3 (95% CI 0.94-1.71) for miscarriage. The odds ratios were 2.0 (0.51-7.7) for stillbirths and 0.9 (0.51-5.5) for miscarriages in women-carriers of fV Leiden (21). On the other in Berkane’s study, fV Leiden was found to be a common risk for stillbirths (3). The most recent prospective study performed in Sweden, comparing 2480 women with APC-R in early pregnancy, elicited no increased recurrence rates. The presence of APC-R was unrelated to adverse pregnancy outcome apart from an 8 fold increased risk of VTE (18).

Three recent case control studies documented a significantly increased prevalence of factor V Leiden mutation in women with recurrent fetal loss (22,8,5). The increased prevalence was not found in study done by Kotwal (17). The discrepancies in the results may be explained by differences in selection criteria, including the ethnic origin of the study populations. Other potential causes for recurrent fetal loss, like chromosomal abnormalities, autoimmune disorders, endocrinologic diseases, infections and anatomic abnormalities should be eliminated as well. fV Leiden is a mild risk factor for thrombosis and is also a mild risk factor for recurrent pregnancy loss (5) but the majority of women who are carriers of fV Leiden will not experience a recurrent fetal loss.

Conclusion

Resistance to activated protein C and fV Leiden were not the risk factors for miscarriage and stillbirth in our study group. They have not been found to be the risk factors for recurrent fetal loss either. Prophylactic anticoagulant treatment is not indicated in women with APC-R and fV Leiden and with the history of fetal loss in their next pregnancies.
Materials and methods
In 1996 we examined 500 blood donors (309 males, 191 females) to get prevalence of APC-R in the East Bohemian region. This prevalence has been found to be 1.6%. We as-
1. A prospective group
Thirty women admitted to the hospital for spontaneous or recurrent fetal loss were tested for APC-R and fV Leiden. All females completed the questionnaire about family and personal history of VTE. Characteristics of this group are shown in table 1. All women were also tested for protein C, protein S, AT III levels, for the presence of lupus anticoagulant (LA) and anticardiolipin antibody (ACA).

2. A retrogressive group
In this group we assessed the course of 172 pregnancies in 80 women with APC-R and fV Leiden (72 heterozygous, 8 homozy-
3. A control group
In this group we evaluated the course of 104 pregnanc-
i.e. 45 women without APC-R and fV Leiden from 42 un-
related families. These women are relatives of individuals with history of thrombotic event or with diagnosis of APC-R and fV Leiden. Characteristics of this group are shown in table 2. All women with history of fetal loss were also tested for ACA, LA, protein C, protein S and AT III as well.

Methods: Blood samples were collected by venipunc-
ture into plastic tubes containing either 1/10 volume of 3.8% sodium citrate for coagulation assay or 1/10 volume of 0.5 M sodium citrate for protein assay. EDTA tubes (10 min. at 2500 G) for prothrombin time (PT), activated partial thromboplastin time (APTT) and AT III assays or after double centrifugation (10 min at 1500 G) for prote-
In this group we obtained the results which are shown in table 4. Stillbirths did not occur in any of these women. LA, protein C, protein S, AT III deficiencies were not detected in any of the 80 women with APC-R and fV Leiden. Characteristics of this group are shown in table 5. Stillbirths did not occur in any of these women. Protein C, protein S and AT III deficiencies were not detected in any of 4 women with abortions. Antiphospholipid syndrome was diagnosed in one woman with recurrent abortions (positi-

The results in cohort of women without fV Leiden are shown in table 6. Stillbirths did not occur in any of these women. Protein C, protein S, AT III deficiencies were not detected in any of 4 women with spontaneous abortions either. Frequency of women with abortion in this group is 9% (2%-21%). and frequency of abortions is 6% (2%-12%). Using Fisher's exact test we have not found statistical difference (p=0.65) between frequency of women with abortions in the retrospective group and in the control group. We have not proven statistical difference (p=0.65) between frequency of abortions in women in these groups either.

Tab. 1: Characteristics of females with spontaneous or recurrent fetal loss

| Number (No.) of women (w) | 20 |
|---|---|
| Mean age (yrs.) | 24 |
| Age range yrs. | 21-44 |
| No. of pregnancies | 43 |
| Mean age of pregnant w. (yrs.) | 26 |
| Age range of pregnant w. (yrs.) | 21-44 |

Tab. 2: Characteristics of females with APC-R and fV Leiden.

| No. of w. | 80 |
|---|---|
| Mean age (yrs.) | 43 |
| Age range yrs. | 23-70 |
| No. of pregnancies | 172 |
| Mean age of pregnant w. (yrs.) | 25 |
| Age range of pregnant w. (yrs.) | 18-37 |

Tab. 3: Characteristics of females without APC-R and fV Leiden.

| No. of w. | 45 |
|---|---|
| Mean age (yrs.) | 43 |
| Age range yrs. | 24-72 |
| No. of pregnancies | 104 |
| Mean age of pregnant w. (yrs.) | 24 |
| Age range of pregnant w. (yrs.) | 19-38 |

Tab. 4: The results in the prospective group.

| No. of w. with 1 spontaneous abortion | 21 |
| No. of w. with 2 spontaneous abortions | 6 |
| No. of w. with recurrent abortions | 3 |
| Mean week of abortion | 40.5 |
| Week range of spontaneous abortion | 7-18 |
| Week range of spontaneous abortion | 7-18 |

Tab. 5: Frequency of abortions in women with APC-R and fV Leiden.

| No. of w. with 1 spontaneous abortion | 4 |
| No. of w. with 2 spontaneous abortions | 3 |
| No. of w. with recurrent abortions | 1 |
| Mean week of abortion | 40.5 |
| Week range of spontaneous abortion | 7-18 |

Tab. 6: Frequency of abortions in women without APC-
and fV Leiden.

| No. of w. with 1 spontaneous abortion | 3 |
| No. of w. with 2 spontaneous abortions | 0 |
| No. of w. with recurrent abortions | 0 |
| Mean week of abortion | 11 |
| Week range of spontaneous abortion | 5-18 |

Discussion
Since 1996 several research papers have been published about the relationship between fV Leiden and fetal loss. These studies assessed either the association of fV Leiden with recurrent fetal loss or the association with miscarriages and stillbirths. EPCOT study (European Prospective Cohort on Thrombophilia) is the largest study which analyzed the risk of fetal loss in women with known thrombophilia. Researchers found that the odds ratios were 3.6 (95% CI 1.4-9.4) for stillbirths and L and 1.3 (95% CI 0.94-1.71) for miscarriages. The odds ratios were 2.0 (0.5-1.77) for stillbirths and 0.9 (0.5-1.5) for miscarriages in women-carriers of fV Leiden (21). On the other hand in Berkane’s study, fV Leiden was found to be a common risk for stillbirths (1). The most recent prospective study performed in Sweden, compr-

References
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3. The control group
In this group we assessed the course of 172 pregnancies in 80 women with fV Leiden we got the results which are shown in table 4. Stillbirths did not occur in any of these women. LA, protein C, protein S and AT III deficiencies were not detected in any of the 80 women with APC-R and fV Leiden. Characteristics of this group are shown in table 5. Stillbirths did not occur in any of these women. Protein C, protein S and AT III deficiencies were not detected in any of 4 women with spontaneous abortions. Antiphospholipid syndrome was diagnosed in one woman with recurrent abortions (positivity of LA and ACA). Frequency of women with abortion in this group is 10% (4%-19%). Frequency of abortions is 8% (4%-14%).

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Sjögren’s syndrome (SS) is a chronic autoimmune disease characterized by a progressive lymphocytic and plasmacellular infiltration of the salivary and lacrimal glands. SS is often associated with other autoimmune diseases and can occur alone (primary) or in association with other autoimmune diseases (secondary) (24).

The trapping of leukocytes from the bloodstream and their subsequent rolling along the activated endothelial cell lining of postcapillary venules are the earliest signs of inflammation. DNA and their ligands, which are heavily glycosylated surface molecules of leukocytes e.g. CD15 molecule of granulocytes, could be bound through this interaction. Next step is mediated through the interaction between adhesion molecules of leukocytes and their ligands expressed on the surface of activated endothelial cells and their ligands, which are heavily glycosylated surface molecules of leukocytes e.g. CD15 molecule of granulocytes. Membrane adhesion molecules are shed into the body fluids by the proteolytic cleavage or by alternative splicing on the level of mRNA (15). The elevated levels of soluble adhesion molecules can provide some useful diagnostic or prognostic informations (14).

Neopterin is produced by macrophages after activation with interferon gamma, a cytokine produced by CD4+ helper-inducer lymphocytes. Increased neopterin concentration thus serves as a sensitive marker of many inflammatory diseases, and may provide some useful diagnostic or prognostic informations (14).

**INTRODUCTION**

Sjögren’s syndrome (SS) is a chronic autoimmune disease characterized by progressive lymphocytic and plasma cell infiltration of the salivary and lacrimal glands leading to the serositis and xerophthalmia (sicca complex) (1,9,35). SS is associated with the production of antinuclear autoantibodies and another autoimmune diseases (secondary) (24).

Membrane adhesion molecules are shed into the body fluids by the proteolytic cleavage or by alternative splicing on the level of mRNA (15). The elevated levels of soluble adhesion molecules are found in the serum of patients with various inflammatory diseases, and may provide some useful diagnostic or prognostic informations (14).

**OBJECTIVE**

The objective of our study was to evaluate the serum concentrations of soluble adhesion molecules (sICAM-1, sVCAM-1, sE-selectin) and neopterin in patients with SS and to compare them with healthy blood donors.

**METHODS**

We have consecutively evaluated 26 SS patients (17 women, 9 men, mean age 53.7 ± 12.9) and 26 age-matched healthy blood donors (15 women, 11 men, mean age 44.6 ± 12.5). There were significantly higher (mean ± 1SD) serum concentrations of sICAM-1 (362.0 ± 67.9 ng/ml, p<0.001), sE-selectin (78.7 ± 28.1 ng/ml, p<0.001) and neopterin (17.9 ± 6.4 nmol/l, p<0.001) in primary SS patients in comparison to control group. Sera from patients with secondary SS contained significantly higher levels of sICAM-1 (356.0 ± 62.4 ng/ml, p<0.001), sE-selectin (65.5 ± 27.0 ng/ml, p<0.05), and neopterin (18.8 ± 9.8 nmol/l, p<0.001) in comparison with control group. There were no significant differences between patients with primary and secondary SS in any parameters tested. No statistically significant differences in serum levels of sVCAM-1 were found either in patients with primary or secondary SS compared to control group.

**DISCUSSION**

Granulocytes, monocytes and a subset of memory T cells could be bound through this interaction. Next step is mediated through the interactions between E-selectins on the surface of activated endothelial cells and their ligands, which are heavily glycosylated surface molecules of leukocytes e.g. CD15 molecule of granulocytes. Granulocytes, monocytes and a subset of memory T cells could be bound through this interaction. Next step is mediated through the interactions between E-selectins on the surface of activated endothelial cells and their ligands, which are heavily glycosylated surface molecules of leukocytes e.g. CD15 molecule of granulocytes.

**CONCLUSIONS**

Our results are in agreement with previous studies. We have found significantly higher serum concentrations of sICAM-1, sE-selectin and neopterin in patients with primary and secondary SS compared to healthy blood donors. There were no statistically significant differences in serum levels of sVCAM-1.

**SUMMARY**

Granulocytes, monocytes and a subset of memory T cells could be bound through this interaction. Next step is mediated through the interactions between E-selectins on the surface of activated endothelial cells and their ligands, which are heavily glycosylated surface molecules of leukocytes e.g. CD15 molecule of granulocytes.

**KEYWORDS**

Neopterin; sE-selectin; sICAM-1; Sjögren’s syndrome; sVCAM-1