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Serum Cardiolipin Antibodies in Cancer Patients with Thromboembolic Events

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Summary: This study was undertaken to investigate a possible association of anticardiolipin antibodies (ACLAs) in cancer patients with thromboembolic events. Twenty-five patients with solid tumors complicated with acute thrombosis, 36 cancer patients without any thrombotic events, and a group of 20 healthy volunteers without thrombosis or malignancy were included. The mean age of the cancer patients with and without thrombosis and healthy subjects were 50 years (range 20–75), 45 years (range 23–66), and 40 years (range 20–68), respectively. Deep venous thrombosis (n = 16) and thrombosis of the central venous port-catheter systems (n = 9) were confirmed by Doppler sonography in all patients. IgG and IgM isotypes of ACLAs were quantitated by enzyme-linked immunosorbent assay with normal levels of <23 GPL and <11 MPL, respectively. Mean values of IgG ACLAs were found similar in cancer patients with acute thrombosis (13.8 ± 4.9 GPL), without thrombosis (12.8 ± 5.4 GPL) or in healthy subjects (14.8 ± 5.5 GPL). Although the mean values of IgM ACLAs were within normal limits in all groups, cancer patients with thrombotic events had higher levels of IgM ACLAs (mean = 10.5 ± 2.2 MPL) than cancer patients without thrombosis (mean = 4.6 ± 2.4 MPL) (p = .01). Healthy subjects also had lower levels of IgM ACLAs (mean = 7.1 ± 3.2 MPL) than cancer patients with thrombosis (p = .16). In addition, a higher percentage of cancer patients with or without thrombosis had IgM and IgG ACLA levels above normal limits compared with healthy controls. In conclusion, our study suggests an association between ACLAs or IgG and particularly IgM isotypes and venous thrombosis in malignancy. Identification of cancer patients who are at higher risk for developing thromboembolic events might lead to a better selection of patients for prophylactic anticoagulant therapy. **Key Words:** Anticardiolipin antibodies—Thromboembolism—Cancer patients.

Hypercoagulability and its clinical consequences as thromboembolic events constitute a major cause of morbidity and mortality in cancer patients (1,2). Hypercoagulability in cancer patients was first described by Trousseau (3) in 1865. In 1938, Sproule (4) reported an increased incidence of venous and arterial thromboses at postmortem examination in a variety of malignancies.

The mechanisms underlying hypercoagulability in cancer patients are complex. It is now clear that the hypercoagulability state in cancer patients represents more than the effects of immobility, inanition, or venous obstruction by malignant masses (1). The coagulation system is a highly regulated cascade of interrelated reactions with a net result of thrombin generation (5). The direct and indirect expression of procoagulant activity by tumor cells, monocytes, and endothelial cells may lead to the activation of the hemostatic system with a wide variety of clinical and laboratory sequelae (6). Although a wide array of laboratory hemostatic abnormalities can be detected in many cancer patients, none of these abnormalities has been determined to have clinical utility in predicting thrombosis (7).

One of the possible contributary causes for thromboembolic disease in cancer is the presence of anticardiolipin antibodies (ACLAs) which may develop secondary to the underlying malignancy (8–10). These autoantibodies that are directed against phospholipids, lead to prolongation of phospholipid-dependent coagulation tests in vitro and are associated with a markedly increased incidence of thrombosis in vivo.

To investigate a possible association between ACLAs and the risk of thrombosis in cancer patients, serum levels of ACLAs in cancer patients with acute thrombotic events were measured and compared with those without thrombotic complications and also with healthy volunteers.

MATERIALS AND METHODS

Twenty-five patients with solid tumors whose deep venous thrombosis (DVT) of the lower extremity or

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thrombosis of the central venous port-catheter system were detected by physical examination and Doppler sonography were enrolled into the study.

The control group consisted of 36 patients with malignancy but without thrombotic events and 20 healthy subjects without malignancy and thrombosis. IgG and IgM isotypes of ACLAs were quantitated by enzyme-linked immunosorbent assay (ELISA) with normal levels of less than 23 GPL and 11 MPL, respectively, in all three groups.

Patient characteristics are shown in Table 1. The mean age in years of the cancer patients with and without thrombosis and healthy subjects was 50 (range 20–75), 39 (range 33–73), and 40 (range 20–68), respectively. Solid tumor types in the study group included gastric cancer (n = 3), colorectal cancer (n = 4), pancreatic cancer (n = 2), breast cancer (n = 4), ovarian cancer (n = 6), lung cancer (n = 4), and non-Hodgkin's lymphoma (n = 2). Sixteen patients had DVT of the lower extremity and nine patients had thrombosis of port-catheter system (Table 1).

RESULTS

There were no statistically significant differences between mean values of IgG ACLA levels in all three patient populations evaluated. Mean values of IgG ACLAs in cancer patients with thrombosis and without thrombosis and in healthy controls were 13.8 ± 4.9 GPL, 12.8 ± 5.4 GPL, and 14.8 ± 5.5 GPL, respectively (Table 2).

Although the mean values of IgM ACLAs were within normal limits in all patients, cancer patients with thrombosis were found to have statistically higher IgM ACLA levels (mean = 10.5 ± 2.2 MPL) than cancer patients without thrombosis (mean = 4.6 ± 2.4 MPL) (p = .01). Healthy control subjects also had lower levels of IgM ACLAs (mean = 7.1 ± 3.2 MPL) than cancer patients who experienced acute thrombotic episodes (mean = 10.5 ± 2.2 MPL), but without any statistical significance (p = .16) (Table 2).

Further analysis has been performed with reevaluation of all three groups using a percentage of patients with increased serum levels of ACLAs above normal limits, instead of mean values. It was evident that IgG ACLA levels were found to be above normal limits in 4% of cancer patients with thrombotic events and in 8% of cancer patients without thrombotic events compared with only 1% of healthy controls. Twenty-four percent of cancer patients with thrombosis were found to have IgM ACLA levels above normal, while only 3% of cancer patients and 11% of healthy controls had IgM ACLA levels above normal limits (Table 2).

TABLE 1. *Characteristics of cancer patients*

	Patient number		
	Cancer patients with thrombosis	Cancer patients without thrombosis	Total
Sex			
Male	9	21	30
Female	16	15	31
Median age (range)	45 (20–75)	39 (33–73)	
Primary tumor site			
Gastrointestinal	9	17	26
Lung	4	8	12
Breast	4	3	7
Ovary	6	4	10
Non-Hodgkin's disease	2	1	3
Other (head-neck, sarcoma)	—	3	3
Location of thrombosis			
Deep venous system	16	—	16
Port-catheter system	9	—	9
Total	25	36	61

DISCUSSION

Disorders of coagulation are common in patients with cancer. Clinically significant hemostatic abnormalities, such as thrombosis and hemorrhage, may affect as many as 15% of cancer patients (7,11–13). Hemostatic complications are the second most common cause of mortality in cancer patients (7,12,14). An increased incidence of thromboembolic events is reported in cancer patients as compared with either patients hospitalized for non-neoplastic disease or the normal population (11). The spectrum of hemostatic abnormalities ranges from massive thromboembolism to abnormal coagulation parameters in the absence of clinical manifestations (11,13).

The mechanism by which malignant tumors activate the hemostatic system is complex. Multiple variables including abnormalities in blood flow, blood composition, and blood vessel wall were originally suggested by Virchow (15) as the basic events in hypercoagulability. Procoagulants secreted by tumor cells, monocytes, and endothelial cells in response to tumor accompanied with sluggish blood flow contribute to hypercoagulability in malignancy (6). Hypercoagulability in cancer patients may also arise from abnormal increases in platelet numbers, abnormal platelet morphology, and platelet lysis (16).

Abnormalities in laboratory tests have been reported in up to 90% of cancer patients (13,17). The most common abnormalities are elevations of fibrinogen and clotting factors (V, VIII, IX, XI), elevated fibrin/fibrinogen degradation products, and thrombocytosis, all consistent with low-grade disseminated intravascular coagulopathy (18). Despite the multitude of laboratory abnormalities, at the present time their significance in the development of thrombotic events is uncertain (19). The balance between coagulation, fibrinolysis, and inhibition of the pro-

TABLE 2. *Serum cardiolipin levels in cancer patients with or without thrombosis and in normal healthy subjects*

Volunteers	Cancer patients with thrombosis	Cancer patients without thrombosis	Healthy
*Mean ± SD			
IgG ACLA GPL	13.8 ± 4.9	12.8 ± 5.4	14.8 ± 5.5
IgM ACLA MPL	10.5 ± 2.2	4.6 ± 2.4	7.1 ± 3.2
*Values above normal/total (%)			
IgG ACLA ≥ 23 GPL	1/25 (4%)	3/37 (8%)	2/20 (1%)
IgM ACLA > 11 MPL	6/25 (24%)	1/37 (3%)	2/18 (11%)

* Normal IgG ACLA and IgM ACLA = <23 GPL and <11 MPL, respectively.
ACLA; anticardiolipin antibody.

coagulant and the fibrinolytic system may predispose the patient to localized intravascular thrombosis or a classical disseminated intravascular coagulopathy associated with hemorrhage and thrombosis (18).

Thrombotic events are more common than bleeding complications in solid tumors. It may approach to 40% to 50% in some cancer populations (19). Although a wide array of hemostatic abnormalities can be detected in many patients with cancer, none of the laboratory parameters correlate with the development of a clinical thrombotic episode (18).

Interest in antiphospholipid antibodies began in 1952 with the discovery of the lupus anticoagulant (LA) in 10% of patients with systemic lupus erythematosus (SLE) (20,21). These antibodies comprise a family of immunoglobulins characterized by their pattern of reactivity in a number of laboratory tests. Included in this family are LA and ACLAs. They may occur in a variety of conditions, including autoimmune disorders, infectious diseases, in association with certain drugs, and in malignancy (22). Anticardiolipin antibodies and the LAs are strongly associated with thrombosis and appear to be the most common acquired blood protein defects causing thrombosis. In 1983, Harris et al. (23) developed the ACLA test with IgG, IgM, and IgA idiotypes. Solid phase ELISA is the preferred method for the detection of ACLAs for all idiotypes. Some have advocated assaying more specific anticardiolipin idiotypes by ELISA, such as antiphosphotidylinositol or antiphosohotidylserine (24). Although precise mechanisms whereby ACLAs induce hypercoagulable state remains unclear, interference with endothelial release of prostacyclin, interference with protein C, protein S, and prekallikrein activity, and interaction with platelet membrane phospholipids may lead to thrombosis (25).

The ACLAs may occur in up to 17% of cancer patients, and are proposed as one of the contributing factors in hypercoagulability in malignancy (8,9,26). Although it is not known how these antibodies lead to thrombosis, it has been proposed that there is an interaction in altering prostaglandin release activation of platelets, and changes in prostaglandin metabolism. In addition, it has been proposed that these antibodies may block protein C or alter phospholipid interactions with activated factor V (27). ACLAs should be suspected if the patient has a prolonged partial thromboplastin time that does not correct with an equal mix of normal plasma (6). In a study performed by Zuckerman et al. (8) ACLA positive cancer patients, mainly those with high titres, were found to have a higher rate of thromboembolic events (28%) than ACLA negative cancer patients (14%). Two case reports described widespread thrombosis in patients with ovarian cancer and renal cell cancer who had positive ACLAs (9,10).

In our study, cancer patients with thrombosis had higher mean levels of IgM ACLAs compared to cancer patients without thrombosis and healthy volunteers. However, mean levels of IgG isotypes in all three groups were similar. Further analysis revealed that a higher percentage of cancer patients, with or without thrombotic complications, had IgG and IgM ACLA levels above normal limits. There was a clear association between IgM ACLA levels and venous thromboembolic events in cancer patients. However, our study also showed a possible association of IgG ACLAs with risk of thrombosis in malignancy (Table 2). This study suggests that identification of ACLA-positive patients, especially the IgM isotype, might be of help in selecting cancer patients predisposed to thromboembolic events. Prophylactic measures can then be instituted in these high-risk cancer patients.

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