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INTERACTION BETWEEN HEMOSTATIC COMPONENTS AND TUMOR CELLS. N. Esumi (1), S. Todo (1) and S. Imashuku (2). Department of Pediatrics (1) and Children's Medical Research Center (2), Kyoto Prefectural University of Medicine, Kawaramachi, Kamikyoku, Kyoto, Japan.

Involvement of platelets and coagulation systems in the hematogenous metastasis of tumor cells has been suggested from in vivo and in vitro studies, however, there is still controversy about the exact role of hemostasis in metastasis. To date, at least three types of platelet aggregating mechanisms and three types of tumor cell procoagulants have been reported in different tumor cells.

We investigated platelet aggregating activity (PAA), procoagulant activity (PCA) and the relationship between these two activities, using eight human neuroblastoma cell lines, three human leukemia cell lines and human mature lymphocytes. PCA in tumor cells was measured by the single stage recalcification time and the assay with chromogenic substrate S_{2222}. PAA was determined turbidometrically with an aggregometer by adding cell suspensions of tumor cells to platelet rich plasma (PRP). The effects of protease inhibitors, enzymes and thrombin inhibitors on PAA and PCA were also studied.

Neuroblastoma cell suspensions showed high PCAs which were reduced in Factor VII deficient human plasma, indicating a tissue factor-like activity. NCG line possessing the highest PCA also showed a high PAA, which was inhibited by pretreatment of cell suspensions with phospholipase A_2 and abolished in the presence of heparin, hirudin or MD805 in the assay system. Human leukemia cell lines and mature lymphocytes had weak to moderate PCAs without showing PAA, but became active to express PAA after being removed of cell surface sialic acid by neuraminidase. These results suggest that in neuroblastoma, PCA closely linked with PAA may play a role in the hematogenous metastasis. In hemopoietic cells, PAA expressed when cell surface sialic acid is removed does not correlate with PCA, and sialic acid in these cells possibly prevents direct interaction with platelets in the hemostatic homeostasis.

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CANCER CHEMOTHERAPY AND THROMBOSIS. M. Levine, A. Arnold, L. Kelleher, S. Lord, W. Hryniuk, J. Hirsh and M. Gent. Ontario Cancer Foundation, Hamilton Clinic and Department of Medicine, McMaster University, Hamilton, Ontario, Canada.

Malignant disease is recognized as a risk factor for venous thromboembolism. A number of recent reports have suggested that cancer chemotherapy may contribute to this risk, but it was not possible to separate the role of chemotherapy from the effects of the malignant disease. We are conducting a randomized trial to determine the optimal duration of adjuvant chemotherapy in women with Stage II breast carcinoma. These ambulatory patients, with negligible tumour burden, receive either 12 weeks of chemo-hormonal therapy (cyclophosphamide, methotrexate, 5 fluorouracil, vincristine, prednisone, adriamycin and tamoxifen) or 36 weeks of chemotherapy (cyclophosphamide, methotrexate, 5 fluorouracil, vincristine and prednisone). This study has provided us with an opportunity to evaluate the thrombogenic effects of chemotherapy since patients in the 12 week group, while off chemotherapy, can be compared directly to the patients in the other group who are still on chemotherapy. This allows the confounding influence of the malignant process to be circumvented. All patients undergo screening tests for thrombosis (impedance plethysmography and Doppler ultrasound) and routine clinical assessments. Suspected venous thrombosis is confirmed by venography and suspected pulmonary embolism by either pulmonary angiography or high probability ventilation perfusion scanning. There have been 11 episodes of venous thromboembolism to date among 191 patients of whom 164 have completed the first 36 weeks of study. There were 3 episodes in each group during the first 12 weeks. During the subsequent 24 weeks there have been no events in the group whose treatment was stopped and 5 events in the group still on treatment ($p = 0.03$). These findings demonstrate that chemotherapy per se is an important risk factor for venous thromboembolism in patients with malignant disease.

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GENERATION OF ADP BY HUMAN AND MURINE TUMOR CELLS IS SPECIFIC BUT IS UNRELATED TO METASTATIC POTENTIAL. G.A. Jamieson (1) and G. Grignani (2). American Red Cross Laboratories, Rockville, MD 20855, U.S.A. (1) and University of Pavia, Italy (2).

The ability of tumor cells to activate platelets may facilitate the metastatic process. It has been generally assumed that the production of ADP by tumor cells is due to non-specific damage during harvesting in vitro or, in vivo, by frictional interactions with the capillary wall. The present work shows that tumor cell ADP arises not from cell damage but by a specific process under metabolic control. The human 253J urinary carcinoma cell line activated heparinized human platelets by an ADP-dependent mechanism based on inhibition by CP/CPK and the identification of aggregating concentrations (1 uM) of ADP in the cell-free supernatant by HPLC. Tumor cell damage during harvesting was shown not to be a factor since (i) the amount of ADP secreted was unrelated to the appearance of LDH, (ii) was similar when measured in confluent monolayers, in tumor cells after detachment and resuspension or following crossover studied in HBSS and MEM, and (iii) was constant at varying tumor cell concentrations. Metabolic control of ADP generation or transport was indicated by the fact that it was reduced 50% in tumor cells treated with p-chloromercuribenzene sulfonate and was completely abolished in those treated with iodoacetic acid. In order to determine whether this metabolically controlled generation of ADP was related to metastatic potential, we carried out identical experiments with the F1 (low) and F10 (high) metastatic variants of the B16 murine melanoma line. The amounts of ADP produced by the B16 cells were about twice as great as with the human 253J cells but there was no significant difference between the amounts of ADP generated by F1 and F10 variants. These studies demonstrate that ADP production by tumor cells is a discrete process under metabolic control but is not directly related to the metastatic potential of individual tumor cell lines.

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ALPHA-2-ANTIPLASMIN (α_2AP) IN ACUTE NONLYMPHOBLASTIC LEUKAEMIA AS A MARKER OF DISEASE ACTIVITY. E. Cofrancesco, E. Pogliani, M. Salvatore, C. Boschetti, G. Moreo, M. Cortellaro. Istituto di Scienze Mediche, Università di Milano, Italy.

α_2AP (Coatest Antiplasmin Kit, Ortho Diagnostic Systems), antithrombin III (AtIII, Coatest Antithrombin Kit, Ortho Diagnostic Systems) and plasminogen (S2251) were assayed in 21 patients with acute nonlymphoblastic leukaemia (ANLL: $5M_2$, $6M_3$, $5M_4$, $5M_5$) before and after chemotherapy and during bone marrow cellularity recovery. The aim of the work was to investigated disturbances of coagulation and fibrinolysis with special reference to proteases inhibitors and to evaluate the prognostic value of changes in these parameters in ANLL patients. Low α_2AP levels were observed in the initial phase of the disease: 41.5 $\pm$ 24.79 SD in 10 DIC patients versus 64.10 $\pm$ 20.70 SD in patients without DIC ($p < 0.05$). α_2AP normalized only in the 11 patients who achieved haematological remission (71.63 $\pm$ 13.13 SD) and remained low in those who did not respond to chemotherapy (61.88 $\pm$ 17.86 SD, $p < 0.01$). No significant modification of AtIII and plasminogen levels were observed during the course of the illness.
It may be postulated that proteolytic cleavage of α_2AP by granulocyte proteases contributes to the low levels of the inhibitor in ANLL, and suggested that α_2AP may represent a marker of leukaemic disease activity. In fact the mean α_2AP level of all patients at diagnosis plus those who did not respond to chemotherapy was significantly lower than that of patients in haematological remission (55.93 $\pm$ 23.13 versus 71.63 $\pm$ 13.13, $p < 0.05$).