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6. It is also possible that the colostral IgA of rabbits having IgM specificity Ms1 or Ms2 may also carry these determinants. Accordingly, when does with a known IgM specificity are bred the colostral IgA of these rabbits will be tested for the presence of Ms1 or Ms2 determinants.
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Hemopoietic Colony-Forming Units in Regenerating Mouse Liver: Suppression by Anticoagulants

Abstract. *After hepatic injury induced by carbon tetrachloride, mitotically active hematopoietic cells of nonhepatic origin localize in the liver as judged by an increase in colony-forming nodules in the spleens of lethally irradiated recipient mice on intravenous injection of cells from these livers. The administration of warfarin suppresses the localization of colony-forming units in the regenerating liver by inhibiting the coagulation mechanism of the donor animals.*

While fibrin formation in hemostatic and inflammatory processes has been investigated for decades, the significance of fibrin formation in the growth of primary tumors and in the fixation and extension of metastases has only recently become apparent. Hiramoto (*1*) has shown that extravascular fibrin deposits are found in various spontaneous human tumors. O'Meara (*2*) showed that fibrin forms lattice work on which tumor cells grow and spread. Bale, Spar, and Goodland (*3*) used antibody to fibrin to localize I^{131} in tumor tissue, producing permanent regression of a transplantable rat rumor, and demonstrating the localization of I^{131}-labeled antibody to fibrinogen in spontaneous human tumors (*4*).

Grossi, Agostino, and Cliffton (*5*) have shown that activation of the fibrinolytic system reduced the pulmonary metastases resulting from intravenously administered tumor cells in rats. Anticoagulant therapy as well as fibrinolysis reduces the incidence of pulmonary metastases resulting from intravenous injection of tumor cells or from massaged intact tumors (*6*). Conversely, agents which increase clotting or inhibit fibrinolysis increase metastases (*7*). Alexander and Altemeier (*8*) noted a striking increase in the number of metastases to injured tissues, which Agostino and Cliffton (*9*) attributed to fibrin formation resulting from the inflammatory reaction. These metastases could be greatly reduced by anticoagulant therapy or fibrinolysin. Metastases were produced in liver tissue after carbon tetrachloride feeding, while intravenous injection of tumor cells resulted only in pulmonary metastases in control animals (*10*).

It is our hypothesis that fibrin formed in injured tissues traps normal mitotically active cells which usually are not localized in uninjured tissue and apparently do not play a role in the inflammatory response or subsequent repair processes. One such cell type is the hemopoietic colony-forming cell.

Till and McCulloch (*11*) devised a test system in which cells from bone marrow with the characteristics of stem cells are injected intravenously into lethally irradiated host animals. Gross nodules of proliferating cells, which can be counted macroscopically, are produced in the spleens of the host animals. The spleen colony technique appears to satisfy the requirements of an assay for stem cells, since it detects a cell type which has the capacity to proliferate extensively and to give rise to progeny containing differentiated cells (*12*). Single cells, which have been given the operational name of colony-forming cells (*13*), produce colonies which contain, within a single colony, erythrocytic, granulocytic, and megakaryocytic cells (*14*). The number of colonies in the spleens of irradiated recipients is, over a wide range, proportional to the number of cells or colony-forming units intravenously injected (*15*).

Our work shows that (i) after carbon tetrachloride–induced hepatic injury, colony-forming units, presumably circulating peripheral blood leukocytes, localize in the liver and are trapped in the network of fibrin. Hemopoietic stem-cell elements are found in peripheral blood leukocytes with a frequency 1/30 to 1/50 that in bone marrow cells (*16*). Karyotype studies (*17*) have shown that mitotically active myeloid cells, presumably of bone marrow origin, are present in livers after injection of carbon tetrachloride. (ii) Anticoagulant therapy with warfarin of sufficient dosage to increase the normal prothrombin time 3.4 to 4.5 times inhibits the localization of colony-forming units in the regenerating liver. (iii) Warfarin has no direct effect on colony-forming units; it affects only their localization in the regenerating liver by inhibiting the coagulation mechanism. This inhibition can be reversed by the administration of vitamin K. When bone marrow cells from warfarin-treated mice are intravenously injected into lethally irradiated host mice, spleen nodule counts are the same as those of recipients injected with bone marrow cells from control mice.

The donor and recipient mice used were 12- to 16-week-old genetically homogeneous female F_1 hybrids $(C57L \times A/He)F_1$. All warfarin-treated mice were injected intraperitoneally with the following dosage of warfarin sodium: day 1, 4.8 mg; day 2, 3.36 mg; day 3, 1.92 mg; day 4, 1.20 mg.

All carbon tetrachloride–treated mice were injected subcutaneously on day 2 with 0.2 ml of 40 percent CCl_4 in olive oil. All mice treated with vitamin K (Mephyton) were injected intraperitoneally at the same times as mice treated with warfarin. The dosage (milligrams) of Mephyton was the same as for warfarin. All donor mice were killed on day 5, liver slices were removed, weighed, and homogenized by hand in a glass homogenizer, and the homogenates were suspended in cold TC-199 culture medium to a cell dilution of 10 percent by volume. Recipient mice, which had received 900 rad of whole-body x-irradiation, 250 kv (peak), within the previous 3 hours, were injected intravenously in the tail vein with 0.20 ml of suspended cells containing 20 mg of liver tissue. Approximately 15 minutes before the intravenous injection of liver cells, 10 mg of heparin was injected intraperitoneally in the recipient mice to reduce the mortality from intravascular coagulation produced by the liver-

Table 1. Effect of warfarin on localization of colony-forming units in regenerating liver. All cells were intravenously injected within 3 hours following 900 rad of whole-body x-irradiation of recipients.

Mice in recipient group (No.)	Time of killing after injection of cells (days)	Treatment of donors	Nodules per spleen of recipients (Mean ± S.E.)
	Liver cells injected (20 mg)		
13	7	None	0.31 ± .17
14	7	CCl_4	2.64 ± .41
12	7	CCl_4 + warfarin	0.42 ± .23
13	7	Warfarin	.31 ± .14
4	10	Untreated	.50 ± .29
6	10	CCl_4	3.67 ± .16
5	10	CCl_4 + warfarin	0.40 ± .24
6	10	CCl_4 + warfarin + vitamin K	2.83 ± .54
5	10	Warfarin	0.20 ± .20
	6×10^4 *Bone marrow cells*		
4	10	Untreated	11.75 ± .63
6	10	Warfarin	12.33 ± .71

cell injection. Recipient mice injected intravenously with bone marrow cells did not require prior injection with heparin. At the same time that the liver slices were taken, bone marrow cells were taken from washings of the femur with culture medium. Cells were counted and diluted in culture medium to a count of 6×10^4 cells for each intravenous dose of 0.2 ml.

Recipient mice were killed at 7 and 10 days after intravenous injection of cells, spleens were removed intact and placed in formalin–acetic acid, and nodules were counted macroscopically before sectioning.

As measured by the production of macroscopic nodules in the spleens of lethally irradiated mice, the intravenous inoculum of liver cells from mice previously injected with carbon tetrachloride contains approximately eight times as many colony-forming units as control liver cells do (Table 1). The administration of warfarin prevents the increase in colony-forming units in livers injured by CCl_4; concurrent administration of vitamin K restores the increase in the number of colony-forming units in mice treated with warfarin and CCl_4. The effect of warfarin is probably due to its inhibition of fibrin formation in the livers of mice treated with CCl_4, which suppresses the trapping of colony-forming units from the circulating blood leukocytes. Bone marrow cells from warfarin-treated mice contain the same number of colony-forming units as bone marrow cells from control mice.

Our results suggest that extramedullary hemopoiesis, rather than resulting from a transformation and differentiation of primordial cells originally located in extramedullary tissue, occurs when hemopoietic progenitor cells, derived from the bone marrow and circulating in the peripheral blood, localize in extramedullary tissue such as liver. These hemopoietic stem-cell elements, in common with metastatic tumor cells, may require the deposition of fibrin to establish themselves in tissue. The question arises whether the localization and establishment of hemopoietic colony-forming units in "normal" sites of extramedullary hemopoiesis, such as the spleen, can be adversely affected by anticoagulants.

Note added in proof: In cytogenetic studies (*18*) on regenerating liver (after CCl_4 administration) of radiation-chimeric mice, 89 percent of the cells (from 11 chimeras) in mitosis (presumably the cells were parenchymal) were identified as donor type, derived from the injected spleen cells. In view of our results and those of Nowell *et al.* (*17*), it seems likely that at least some of the T6-containing cells in liver observed by Hard and Kullgren (*18*) may in fact be hemopoietic colony-forming units of extra-hepatic origin.

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Chainpur-like Chondrites: Primitive Precursors of Ordinary Chondrites?

Abstract. *Chainpur and similar, apparently primitive, chondritic meteorites may be precursors of ordinary chondrites; a variety of evidence supports this working hypothesis. In general, carbonaceous chondrites seem to be related collaterally to this genetic sequence rather than being direct ancestors of ordinary chondrites. Metamorphic processes may be responsible for fractionations of elements such as indium and iodine, and type-II carbonaceous chondrites seem to be more primitive than types I or IIIA.*

A long-standing problem has been an adequate and self-consistent explanation of the observed properties of chondritic meteorites (*1, 2*). Chondrites comprise about 85 percent of observed meteoritic falls (*3*); they divide into various classes, notably carbonaceous chondrites, enstatite chondrites, and ordinary chondrites. Each of these major classes may be separated into subclasses—for example, the ordinary chondrites are generally subdivided into the Fe-rich group, the Fe-poor group, and Soko-Banjites (*4, 5, 6*). We now consider evidence bearing on the relations among the ordinary and carbonaceous chondritic classes (*7*).

The ordinary chondrites are quite homogeneous in composition in most "nonvolatile" elements, the most prominent exception being the total content of Fe (*2, 4*). The average metal content of the Fe-rich group exceeds that of the Fe-poor group by about 9 or 10 percent; FeO contents of the ferromagnesian silicates of these meteorites vary less, in the opposite sense. Recent comprehensive studies (*6, 8*) suggest that addition of roughly 6 percent metal to Fe-poor chondrites and reduction of some FeO to Fe could have produced Fe-rich chondrites, the silicate phases of these meteorites having essentially the same composition (FeO content excluded). Of course, the process of metal transport coupled with oxidation, or reduction, could have occurred in the other direction, making Fe-poor chondrites from material of Fe-rich–group composition (*6*).

Also, the sequence of events may well have been much more complicated than this simple model suggests. In any case, the materials implicated were probably chemically similar (in a gross sense) to those we observe now but