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Effect of low dose cyclophosphamide on the synthesis of acute phase protein and its significance for cancer chemotherapy.

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Abstract

Patients with far advanced colorectal cancers received chemotherapy consisting of low-dose cyclophosphamide (LDCY) 333 mg/m2 every four weeks intravenously and by oral administration of 5'-DFUR (a masked compound of 5-Fluorouracil). Serum levels of immunosuppressive acidic protein (IAP), an acute phase protein, were measured every four weeks for a total of thirty-one LDCY trials of ten patients. LDCY chemotherapy significantly decreased the IAP levels in cancer patients with high IAP levels. These results suggested that LDCY chemotherapy could counteract host responses against tumors and could have decreased immunosuppressive responses in cancer patients.

KEYWORDS: low-dose cyclophosphamide< acute phase protein, immunosuppressive acidic protein

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Brief Note

Effect of Low Dose Cyclophosphamide on the Synthesis of Acute Phase Protein and Its Significance for Cancer Chemotherapy

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Patients with far advanced colorectal cancers received chemotherapy consisting of low-dose cyclophosphamide (LDCY) 333 mg/m^2 every four weeks intravenously and by oral administration of 5'-DFUR (a masked compound of 5-Fluorouracil). Serum levels of immunosuppressive acidic protein (IAP), an acute phase protein, were measured every four weeks for a total of thirty-one LDCY trials of ten patients. LDCY chemotherapy significantly decreased the IAP levels in cancer patients with high IAP levels. These results suggested that LDCY chemotherapy could counteract host responses against tumors and could have decreased immunosuppressive responses in cancer patients.

Key words: low-dose cyclophosphamide, acute phase protein, immunosuppressive acidic protein

More than half of the patients already have disseminated cancer or will suffer relapse after treatment. Treatment of recurrent and metastatic colorectal cancer by chemotherapy is usually disappointing (1-4). Therefore, innovative therapeutic regimens are warranted to offer an alternative to conventional chemotherapy in patients with advanced colorectal cancer. Immunosuppressive acidic protein (IAP), an acute phase protein, is a type of alpha 1-acid glycoprotein, and its level in serum was found to be elevated in various malignancies (5, 6). In malignant diseases, a high or rising level of an acute phase protein such as C-reactive protein (CRP) has been associated with a large tumour volume and dissemination and poor prognosis (7). In addition, patients who failed to respond to immunotherapy had grossly elevated levels of an acute phase protein (8). These reports suggested that patients who were already demonstrating a significant acute phase response were unlikely to respond to anticancer therapy. Cyclophosphamide has been shown in animals and in humans to inhibit suppressor T cells selectively at low doses (15-20 mg/kg in mice and 300-500 mg/m^2 in humans) (9). We present preliminary data showing that chemotherapy combined with low doses of cyclophosphamide (LDCY) decreases host responses against tumors, resulting in decreased serum levels of IAP.

Patients and methods. Ten patients with a mean age of 64 ± 7 years (standard error: SE) were studied. The patients were suffering from histologically confirmed metastasizing colorectal adenocarcinomas. The patients were given low doses of cyclophosphamide (333 mg/m^2) every 4 weeks intravenously with daily 5'-DFUR administration. Serum IAP levels were measured every four weeks for a total of 31 LDCY trials of ten patients, using single radial immunodiffusion assay (5). The cut-off levels of IAP in cancer patients obtained in our laboratory was 580 μg/ml $<$. Data were reported as mean $\pm$ SE and analyzed by paired Student's t test, with a level of $P < 0.05$ considered to be statistically significant.

Results and discussion. The present research clearly demonstrates that combined LDCY chemotherapy decreased the levels of IAP, an acute phase protein and one of the soluble suppressor substances, in patients with 580 $<$ μg/ml level. In contrast, this therapy has no effect on the synthesis of IAP in patients with normal IAP levels (Table 1). These results suggest that

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Table 1 Change in serum IAP levels after LDCY* administration

	IAP levels (μg/ml) at day 0			
	580 < (n = 10)		580 > (n = 21)	
Day 0	749.8 ± 44.3	$P = 0.028$	319.5 ± 19.5	$P = 0.813$
Day 28	561.5 ± 64.3		310.3 ± 30.0	

*LDCY: Low dose cyclophosphamide (333 mg/m^2). IAP: Immunosuppressive acidic propein. Values are mean ± SE. Differences between IAP levels at day 0 and day 28 were determined by Student's *t*-test.

LDCY has an inhibitory effect on the synthesis of acute phase proteins, although its inhibitory mechanism remains unclear.

As described above, high baseline levels of an acute phase protein may be a reflection of either tumor burden or biological aggressiveness and hence enhance tumor cell turnover. The cytokines, IL-1, IL-6 and tumor necrosis factor-alpha (TNF-α) are believed to play a crucial role in the control of acute phase protein synthesis in the liver by gene regulation (10-12).

LDCY has been reported to counteract tumor-induced suppressor functions. Berd *et al.* observed a significant reduction in the assays for Con A-induced suppressor cells after LDCY challanges. In addition, they reported LDCY-induced depletion of CD4 + suppressor-inducer T cells (13-15). CD4 + T cells-induced cytokines usually activate macrophage/monocyte cells which, in turn, produce inflammatory cyokines such as IL-1, IL-6 and TNF-α. Therefore, the LDCY-induced inhibition of IAP synthesis was speculated to be a significant reduction of CD4 + T cells.

One of the reasons why chemotherapy is largely ineffective in colorectal tumors is an extreme host inflammatory response against tumors. Therefore, this LDCY combined therapeutic regimen may perhaps offer an additive host chemotherapy to conventional tumor therapies in patients with extreme host inflammatory responses. It is tempting to speculate that inhibition of host inflammatory responses against tumors could be followed by successful anticancer therapy and long survival. Further careful studies are needed to confirm these findings and to elucidate the possible underlying biochemical mechanisms. Random clinical trials of this regimen are on-going in the First Department of Surgery.

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