

REPORT
Effects of Ecomer on Immunological System in Mice

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REPORT

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Effects of Ecomer on immunological system in mice. Part 1.

Material and methods.

Studies on the effect of Ecomer have been performed in 2-month old, male inbred Balb/c mice. Mice have been of local laboratory breed, weighing 20 g each.

Each mouse received Ecomer in dose of 12,5 mg/mouse/24 hrs, or water- during 7 days. On the day 8-th mice were subjected to narcose by means of chloral hydrate, bled from the retroorbital plexus and sacrificed. Spleens have been dissected under sterile conditions and spleen cell suspensions have been prepared .

The tests performed in the study:

1/ Counting of granulocytes and lymphocytes in heparinized peripheral blood.

2/Measurements of metabolic activity of blood granulocytes using chemiluminescence stimulated by Zymosan, according to the method described by Easmon & Cole (1) in own modification (2). Chemiluminescent activity was measured in scintillation counter (RackBeta 1218, LKB Wallac, Sweden) and expressed as *cpm* per 1000 granulocytes.

In addition to above, *the following complementary tests* have been performed:

1/ Local reaction graft- versus host (test LIA, lymphocyte-induced angiogenesis) according to Sidky and Auerbach(3). Briefly, multiple samples of splenocytes suspension (each sample five hundred thousands cells in 0,05 ml of Parker fluid), cleaned from erythrocytes by centrifugation on Histopaque 1077 gradient, were grafted intradermally into anaesthetised (Balb/c x C3H)F1 6-weeks old male mice. Number of newly formed blood

vessels has been calculated in dissection microscope after 3 days of implantation, using criteria described by authors of the method.

2/ Test of angiogenesis induced by cells isolated from mice cancer – Balb/c sarcoma L1.

Sarcoma cells were kindly delivered by dr.Henryk Skurzak from Warsaw's Oncology Center and then passaged through several generations of locally bred Balb/c mice.

Two hundred thousands sarcoma cells have been implanted (while suspended in 0,05 ml of Parker solution) into regionally shaved, narcotised Balb/c mice and angiogenesis estimated quantitatively after 3 days of implantation, on the inner skin surface, as described previously (4).

Results.

Results of the performed studies are presented in Table 1. Both number of circulating granulocytes ($p < 0,001$) and their chemiluminescence activity, when measured in a scintillation counter increased significantly ($p < 0,05$) after Ecomer treatment.

Results of complementary additional studies are shown in tables 2 and 3. Treatment with Ecomer resulted in an increase of activity of splenic lymphocytes in a local graft-versus-host reaction (table 2).

