

CARTA AL EDITOR

Enhancement of E-Rosettes Formation After Treatment of Lymphocytes With Papain

It has been described that peripheral blood human T lymphocytes can bind S.R.B.C. forming non immune rosettes (2, 4). We have found that the treatment of human peripheral lymphocytes with crude papain increases significantly the number of E-Rosettes compared with controls. This enhancement is also apparent in the size and strength of the formed rosettes.

Papain (Merck 1:350) was prepared according to the method described by PORTER (3), using the following buffered system: 0.1 M sodium phosphate pH 7.0, 0.01 M cysteine, 2 mM ethylenediamine-tetra-acetate (EDTA). Lymphocytes were treated at room temperature for 30 min. with 1 ml of enzyme solution containing 0.15 mg of papain. As controls we tested lymphocytes with sodium phosphate, cysteine, EDTA and all together.

We performed the rosette technique mixing 3×10^6 lymphocytes/0.5 ml, washed twice in Hanks solution before and after papain treatment, with 0.5 ml of a suspension of S.R.B.C. (0.05 % in Hanks solution), during 5 min. at 37° C. Centrifugation at 1,200 rpm was followed by incubation at 4° C during 1.5 h. The supernatant was removed and the top layer of the pellet was gently resuspended by shaking. One drop of the cell suspension was mounted on to a glass slide and covered by a coverslip. 300 lymphocytes were counted and all lymphocytes binding more than three S.R.B.C. were considered positive.

The rate of increase of E-Rosette (ta-

Table 1. *Percentage of human peripheral lymphocytes forming E-Rosettes in healthy donors, before and after papain treatment.*

Lymphocytes Treatment	E-ROSETTES %
Lymphocytes controls	57.6 ± 6.5
Papain	89.7 ± 10.1
EDTA and cysteine	70.1 ± 3.5
EDTA	52.8 ± 13.7
Cysteine	51.8 ± 13.6

ble 1) formation with lymphocytes pretreated with papain was 32.1 % (sd 10.1). This result may be comparable with neuraminidase effect described by BENTWICH (1).

With these results we suggest that papain action could unmask membrane receptors with capacity to form E-Rosettes.

References

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