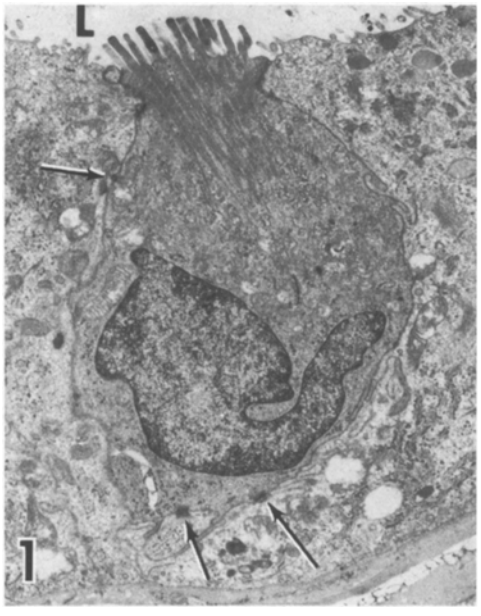

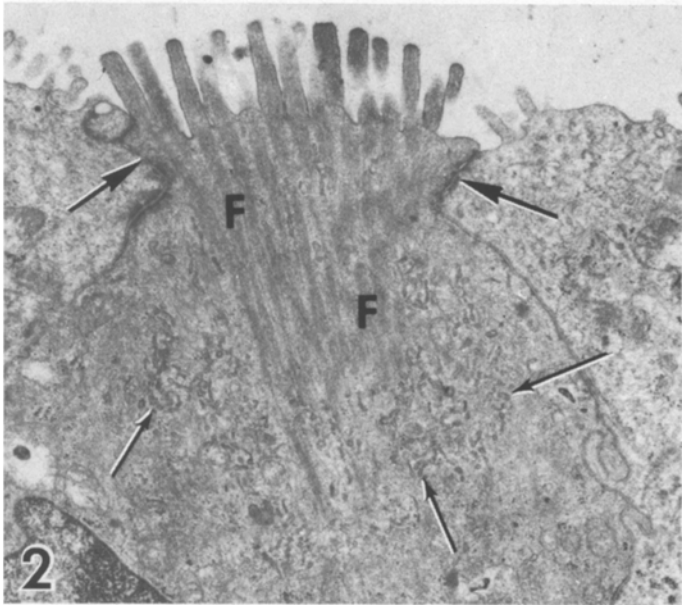

Fig. 1. A caveolated cell taken from the duodenal glands of a white-tailed deer. Note the general pear-shape of this cell type with the narrow apex bordering the lumen (L) of the gland. Scattered desmosomes (arrows) are observed along the lateral cell membranes. × 4000.

Fig. 2. Increased magnification of the apex illustrates bundles of filaments (F) extending from the microvilli and the tight junctions (large arrows) between cells. The caveolae are observed as small tubules and vesicles (small arrows). × 8000.

each microvillus to deep within the supranuclear region (figures 1 and 2). The caveolae appear as small vesicles and/or small irregular tubules coursing through the supranuclear cytoplasm (figure 2). They are, in fact, long tortuous channels that extend from the apical cell membrane between microvilli to deep within the cytoplasm.

The caveolated cells observed in the duodenal glands of the white-tailed deer appear very similar to those reported previously in a variety of entodermal derivatives. Although this cell type has been reported in the mucosa of the stomach, small intestine and colon of a variety of species, it has not been observed previously in the duodenal glands. The caveolated cells of the duodenal glands were observed only infrequently as reported in other regions of the gastrointestinal tract. The function of this unusual cell type is unknown, but it has been suggested that it may function as a chemoreceptor in the respiratory system[8].

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Rosette formation of tumor cells with concanavalin A treated erythrocytes

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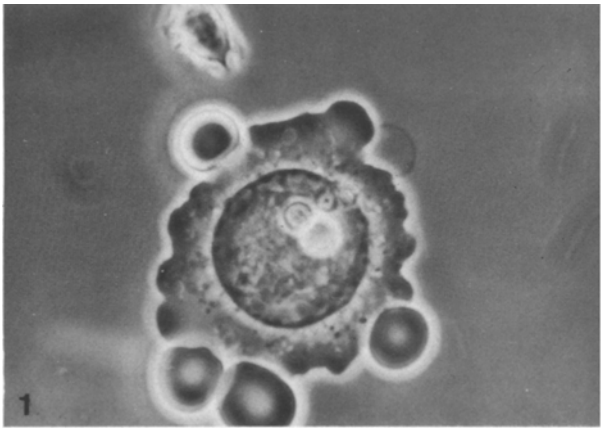
Summary. Trypsinized human erythrocytes were incubated with concanavalin A at 4 °C. After removal of free concanavalin A, the erythrocytes were incubated with Ehrlich ascites tumor cells at 37 °C. The erythrocytes formed rosettes with the tumor cells.

It has become apparent that erythrocytes could be useful as carriers of drugs and other biologically active agents[1-3]. We have reported the effectiveness of daunomycin entrapped erythrocytes against mouse L_{1210} leukemic cells[3]. It is clear, however, that the effectiveness of drug entrapped erythrocytes specifically against tumor cells will demand targeting of the erythrocytes to tumor cells.

Concanavalin A (Con A) preferentially agglutinates Ehrlich ascites tumor cells, rather than other tumor cells[4]. Human erythrocytes are usually not agglutinated by Con A but trypsinized erythrocytes are highly agglutinated. Con A-mediated cell agglutination is temperature dependent. We studied the rosette formation of trypsinized and Con A treated erythrocytes with Ehrlich tumor cells.



Phase contrast microscopy of rosette formation.
Fig. 1. Ehrlich tumor cells (5×10^5 cells/0.2 ml) were incubated with Con A treated erythrocytes (10^7 cells/1 ml).

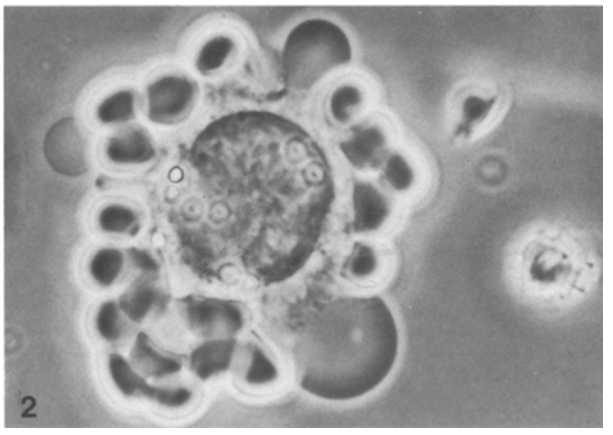


Fig. 2. Ehrlich tumor cells (5×10^5 cells/0.2 ml) were incubated with Con A treated erythrocytes (10^8 cells/1 ml).

Materials and methods. Freshly obtained human blood, with heparin as anticoagulant, was washed 3 times in phosphate buffered saline (PBS) to remove plasma and buffy coat. Erythrocytes were resuspended in PBS at a final concentration of 10^9 cells/ml. The erythrocytes were incubated with trypsin (Sigma Chemical Co. 1 mg/ml) at 37 °C for 30 min with gentle shaking. The suspension was then washed 6 times with PBS to remove trypsin. The erythrocytes, at a concentration of 10^9 cells/ml, were incubated with Con A (Sigma Chemical Co. 50 µg/ml) at 4 °C for 1 h, and the cells were then washed gently twice by centrifugation at $150 \times g$ for 3 min at 4 °C.

Ehrlich ascites tumor cells were collected from a DDY mouse and washed twice with Hanks' balanced salts solution. Rosette formation on mixing the tumor cells with erythrocytes was performed by the following method. In a 10×75 mm glass tube, 0.2 ml of a suspension consisting of 5×10^5 tumor cells was mixed with 1 ml of Con A treated erythrocytes at a concentration of 10^7 or 10^8 cells/ml. The mixture was incubated at 37 °C for 30 min with gentle shaking. The formation of rosettes on mixing the tumor cells with Con A-treated erythrocytes was observed by phase contrast microscope, and rosettes consisting of a minimum of 3 erythrocytes bound to a tumor cell were counted.

Results and discussion. Trypsinized and Con A treated erythrocytes attached to the tumor cells and formed rosettes (figure 1 and 2). 82% of tumor cells formed rosettes with erythrocytes when 5×10^5 tumor cells were incubated with 10^7 erythrocytes and 87% of cells formed rosettes when incubated with 10^8 erythrocytes. A few erythrocytes were agglutinated without rosette formation. At 37 °C a large number of erythrocytes attached to tumor cells, but at 4 °C they did not. α-D-methyl glucoside (Sigma Chemical Co. 10 µg/ml) inhibited rosette formation.

One possible approach for increasing the effectiveness of antitumor drugs would be to find methods of altering their distribution in the body to increase their local concentration at the tumor cell sites. Erythrocytes have been used as a carrier vehicle to enhance the cytotoxic activity of daunomycin against tumor cells[3]. The effectiveness of erythrocytes against tumor cells specifically demands targeting of the erythrocytes to tumor cells. Some success has already been reported in this direction in vivo and in vitro by the use of lectin although the results were not satisfactory[3,5].

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Induction of micronuclei in PHA-stimulated human lymphocyte cultures by therapeutic radiation[1]

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Summary. Micronuclei frequency and percent of chromosome breaks increases significantly in adults whose thymus glands were irradiated in infancy and after irradiation of cancer patients.

Radiation exposure of man produces chromosomal aberrations in human peripheral blood lymphocytes[3]. The micronuclei are formed from chromosomal fragments that are not incorporated into daughter nuclei at mitosis because they lack a centromere. The micronucleus test is a simpler and faster method than chromosome analysis for determining chromosomal fragmentation[4,5]. To determine whether the micronucleus test correlated with chromosome aberrations, we determined the micronuclei frequency in lymphocyte cultures in normal control people, in adults whose thymic glands were irradiated in infancy and in cancer patients before and after radiation treatment.