# VI.
# Summary

The leukocyte adherence inhibition (LAI) test since its early introduction (and up till now) was applied to detect cell-mediated immunoreactivity related to various types of human diseases specially cancer. Our previous studies of human urinary bladder squamous cell carcinoma has been conducted in this issue and expanded to other tumor types as well as shistosomases.

Cancer patients were chosen at early stage of the disease to get the highest reactivity and usage of fetal calf serum (FCS) was avoided because it causes nonspecific adherence inhibition of leukocytes. Following incubation of peripheral blood leukocytes (PBL) from either control subjects or carcinoma patients in serum-free medium, percentage adherent cells was estimated and considered as control level of adherence.

The toxicity of the tumor extracts (SqCC/UB, TrCC/UB, AdnC/UB and SqCC/CX) and other extracts (NorE/UB/82 and Shaem/SEA/83) was examined. The nontoxic concentrations (at which nonspecific reduction of cell adherence was not observed) were determined and found to be of 250 ug/ml for all extracts but 25 ug/ml for Shaem/SEA/83 extract.

Although tumor extracts were prepared by one and the same method, different tumor-antigen preparations varied vastly in their ability to give specific adherence inhibition. All preparations consistently gave a high tumor-specific LAI (20% decreased adherence or more). Only the SqCC/UB2/82 failed to express such level of adherence reduction. Use of this extract was avoided.

Peripheral blood leukocyte samples from SqCC/UB patients showed specific LAI when reacted with SqCC/UB1/80, SqCC/UB1/82 and SqCC/UB3/82, while no reactivity was observed when reacted with TCC/UB1/80, NorE/UB1/82, SqCC/CX1/82 or Shaem/SEA/83 extracts. Peripheral blood leukocytes of other urinary bladder carcinoma patients, cervical carcinoma or even *S.haematobium* patients showed the same way of specificity of LAI in presence of their sensitizing antigens.

Cross reactions between various types of urinary bladder carcinomas was found to be combletely absent, indicating tissue type specificity. Such absence of cross-reactivity was also demonstrated between urinary bladder and cervical squamous cell carcinomas. This indicated organ tissue type specificity. Shaem was also none cross-reacting with SqCC/UB which revealed that *S. haematobium* (as common infection in urinary bladder carcinoma) does not interfere with cellular immune reactions of SqCC patients.

The SqCC/UB1/82 reactive antigen was purified by gel filtration chromatography. The peack C retained all the antigenic activity as detected by the LAI technique at the same 250 ug/ml concentration as the crude extract. Molecular weight of this active fraction was estimated and found to be of 16,227 daltons.

Two versions of the LAI assay (haemocytometer and microplate) were at the same level of sensitivity in detecting CMI in urinary bladder carcinoma patients. The tube modification of the LAI test

failed to show reliable results when tests were performed using PBMC of the same patients.

The difficulties in counting, visually, the adherent cells lead to the introduction of colorimetric modifications of the micro-LAI version. Establishment of what is refered to as the TPC/microcolorimeteric LAI-assay was performed. This assay showed reliable results in detecting specific adherence reduction of leukocytes.

The mechanisms involved in the LAI reaction were detected by the established TPC/microcolorimetric modification of LAI assay. No mediator of the lymphokin family could be detected. Mechanism was confirmed to be dependent on T cells, B cells and monocytes direct interaction with the sensitizing antigen, and on monocyte dependence on B lymphocyte arming. T lymphocytes, regardless that putative mediators, are active whem present in a pure population.