quantitative and qualitative level.

In general, the contributions elucidated that both in rodent and human cells in vitro the process of spontaneous cell transformation becomes increasingly interesting for the analysis of phenotypic alterations related to differentiation features, cytogenetic changes and the expression of oncogenes in relation to the transition from normal to "premalignant" and finally to malignant cells. In these assays the elimination of the specific carcinogenic factors was considered advantageous for the undisturbed analysis of phenotypic and genotypic alterations and their correlations to the changes from a normal to a tumour cell.

Tumour-Host Interaction - Mechanism of Invasion

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Several of the reports in this seminar dealt with the regulation of proteolysis, a process considered to be critically involved in invasion. This was discussed by A. Vaheri (Helsinki, Finland) in the introductory remarks. He emphasized that invasiveness and metastasis, distinguishing properties of malignant cells, involved penetration through components of the extracellular matrix. Enzymatic degradation of matrix components appears to be involved in these phenomena. In collaboration with S. Barlati's laboratory (Brescia, Italy) it was found that defined gelatin-binding fragments of fibronectin have transformation-promoting activity in experimental conditions. This activity is shared by tissue type plasminogen activator t-PA. Interestingly, t-PA is bound by immobilized fibronectin and immobilized laminin, a process possibly involved in "directional proteolysis" by the cell. Fibronectin fragments detected in body fluids of tumour patients may serve as markers for tumour progression. Pericellular proteolysis appears to be regulated in part through α_2-macroglobulin (α_2M), plasminogen activators and their inhibitors, as detected in cultures of various types of normal and malignant human cells. Proteolytic targets of the pericullular matrices of cells in culture include fibronectin and an M_r 66,000 matrix-associated protein. Interestingly, in cultures of human sarcoma cells u-PA is enriched at cellular adhesion sites where also fibronectin is found.

In previous studies production of α_2M has been confined to normal cells; apart from liver cells in vivo, in culture conditions macrophages and lung fibroblasts synthesize this wide-spectrum protease inhibitor. J. Bízik (Bratislava, Czechoslovakia) now reported that several human sarcoma and melanoma cell lines synthesized and secreted to the culture media a high molecular weight glycoprotein identified by immunological criteria, molecular size and NH_2-terminal sequence analysis as human α_2M. In immunoperoxidase staining some but not all human melanomas in vivo were positive for α_2M. J. Tőzsér (Debrecen, Hungary) reported on plasminogen activator levels in blood of mice bearing leukaemia induced by chemical carcinogens. Increased levels were detected in the blood cell extracts while in plasma the overall levels were unaltered or even decreased. The latter finding suggested that analysis of the molecular forms (size, pI) by zymography might be of interest.

The approach by K. Dingemans (Amsterdam, The Netherlands) was primarily a morphological one: ultrastructural analysis of the invasion of lung parenchyme by bronchogenic squamous cell carcinomas. At the sites of initial contact the tumour cells appeared to have inserted themselves between the alveolar epithelial cells and the tissue retained an essentially intact basal lamine that persisted relatively long. A noticeable feature in the deeper tumour areas was a prominent connective tissue reaction, an interesting and conspicuous process the mechanism of which remains to be defined.

What makes some tumour cells metastatic and how can this be correlated to cellular parameters in culture is being studied by a number of laboratories today. The clonal evolution and selection of tumour cells created obviously considerable problems in any such analysis. P. Veselý (Prague Czechoslovakia) reporting on characteristic patterns of in vitro behaviour of sarcoma cells with different metastatic capacities made an interesting comment about the pH of the study conditions. At the acid pH 6.7 (rather than at pH 7.4) a correlation between the increased locomotory activity and capability of metastasizing was found. K. Lapis (Budapest, Hungary) and collaborators are in the process of a multiparameter analysis of in vivo selected highly metastatic variants of Lewis lung tumour cells. Several cell surface properties (more microvilli, more fibronectin, highly sulphated heparan sulphate, altered lectin-binding sites) were characteristic of the highly metastatic cell line. Perhaps of more interest was the finding that these cells did not attract macrophages to the same extent as the parental tumour cells. The most provocative part in the report was, however, the result that 5-hexyl-2'-deoxy-uridine inhibited the highly metastatic cells in vivo without being cytotoxic.

Destruction of metastatic tumour cells by immunological means was the subject of the report by K. Yokoro (Hiroshima, Japan) using a reticulum cell sarcoma line originating from a C57BL/C3H mouse. Mice receiving spleen cells from specifically immunized donors were protected from metastasis. The observation that after anti-Thy 1 serum treatment the spleen cells lost their protective capacity indicated that specifically activated T-lymphocytes were responsible for metastatic cell destruction. R.Heicappell (Heidelberg, F.R.G.) reported on another approach for experimental prevention of metastatic spread. The highly metastatic murine lymphoma ESb cannot be curatively treated with surgery or chemotherapy alone. Postoperative metastatic spread could, however, be prevented with a combination of surgery and immunotherapy using

autologous irradiated tumour cells modified by Newcastle disease virus infection. Interestingly, this therapy did not work in nude mice, a finding which should help in clarification of the mechanism of the observed remarkable curative effect.

Athymic nude mice provide a convenient host to study the growth of human tumour xenografts but growth kinetics are hard to study because of irregular tumour shape. A. Fiennes (London, U.K.) reported on what appears to be a very useful way to do this rapidly and repeatedly with tumours as small as 50 µl. A snap-setting alginate dental moulding gel allowed accurate (± 5%) impressions of tumour to be taken from live unanesthesized mice.

Not only the pH, as discussed above, but also the oxygen level are variables worthy of study. The report by I. Basic and his colleagues at Houston, U.S.A. dealt with the effect of O_2 on tumour cell growth in culture and their interaction with macrophages. There is evidence that in the tumour area in vivo the level may be as low as 2 to 5% O_2. In culture, lower O_2 levels (2%) greatly enhanced clonogenicity of tumour cells in soft agar but had less effect on growth rate itself. In cocultures with the tumour cells macrophages had in 20% O_2 little effect on tumour cell growth but in 2% eliminated the stimulatory effects of hypoxia. The effects of hypoxia were limited to primary cultures of sarcomas, while the growth of established cell lines was unaffected. This report also serves to illustrate the difficulties we have in trying to understand the mechanisms of tumour growth in vivo. What is true for primary tumour cells is not true for tumour cell lines and only the latter may be analyzed in any great detail for biochemical properties. Moreover, the host response has a decisive role on tumour cell behaviour. Clearly, there seems to be no shortage of important research topics in sight.