Summary of Pathobiologic Presentations

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The results of a large number of investigations on the pathobiology of small cell carcinoma (SCLC) were presented during this meeting. It is difficult to even attempt to summarize adequately and fairly these presentations. Undoubtedly, I will do this inadequately in some instances, and I wish to apologize to those investigators whom I unintentionally slight or whose work I may misrepresent. Unfortunately, some reports were quite preliminary with relatively small groups evaluated. Such presentations contributed little to our understanding and may be misinterpreted or misleading. The biologic studies presented were quite diverse, but many of the papers can be placed into a few major categories, and I will discuss these groups together.

An important area of study continues to be growth factors and growth factor receptors. As more studies are completed, this appears to become a progressively more complex problem.

There were several reports on gastrin-releasing peptide (GRP), although interest in this peptide as a clinically significant growth factor seems to be waning. It was reported that intracellular and secreted GRP was extremely variable in cell cultures, varying from 10- to 100-fold. Of particular interest, it was pointed out that extracellular levels which normally occur in culture were much lower than concentrations used in experiments which demonstrated a growth stimulation effect of GRP. Antibodies to bombesin (BN) had no effect on proliferation in 1 study of cultured cells at low and high density, which was interpreted to indicate that SCLC lines have self-stimulating factors other than GRP.

One such other autocrine growth factor seems to be insulin-like growth factor (IGF). Studies from UK, West Germany, and the US have come to identical conclusions concerning this. SCC lines produce IGF, receptors are present, and antibodies to IGF block the growth stimulatory effect. These are important observations. Tumor growth factor α (TGFα) and transferrin appear to be other autocrine types of growth factors in SCLC.

Now back to gastrin-releasing peptide. Previous studies with radioimmunoassays against the mature GRP molecule rarely detect any elevated levels in patients with SCLC, even though most cell cultures derived from these tumors produce the peptide. However, in a Japanese study it was found that if plasma was passed over immuno-affinity columns, thus concentrating the GRP, elevated levels could be found in most patients with SCC. In an interesting report from Copenhagen, it was shown that a RIA against the prohormone proGRP also indicated elevated levels in many patients. This seems to be a practical assay and thus this provides another candidate as a tumor marker.

Now to the other side of the coin, biologic inhibitors of growth. Little was presented. One report indicated new analogs of substance P which were more inhibitory than others tested previously. Evaluations on 3 SCLC cell lines in serum free medium were described, although, in fact, inhibition of growth has been demonstrated in a total of 8 cell lines. No in vivo studies were reported, and the concentrations used in vitro may be impractical in vivo. In another study it was found that a monoclonal antibody (Mab) to a membrane protein interfered with cell proliferation. Both of these studies suggest the possibility of clinical applications. Of course, there were multiple other reports on Mab defining SCC or subtypes of SCC, but these are too diverse and involved to summarize.

Allelic loss in pulmonary carcinoma was another area of investigation which generated a series of reports at this meeting. In a report from the UK it was demonstrated that deletion of a part of chromosome 3 is a common event in tumors of non-SCLC (NSCLC) as well as in SCLC. This is another important observation. In terms of specific genes it has been found that virtually all SCLC lost heterozygosity at the erb A β locus on chromosome 3P. This locus codes for a thyroid hormone receptor. A similar finding occurred in about 50% of non-SCLC.

In another study of similar type from the US, inactivation and structural alterations of the retinoblastoma gene were described. This gene is located on chromosome 13. The changes were found to be much more common in SCLC than in non-SCLC. Similar inactivation and structural alterations have also been described in breast carcinoma. Allelic loss on chromosome 3P was examined in extrapulmonary SCC and found to be much less common compared with SCLC, even though the tumors look histologically similar.

Deletions actually occur on multiple chromosomes in pulmonary carcinoma, and in another investigation multiple cDNA probes for chromosome 11p were evaluated. Homozygosity was demonstrated in some cell lines, and the group is now examining fresh tumor tissue. In an important study from France it was demonstrated that DNA evaluations of
tumors are feasible on fiberoptic bronchoscopy biopsy specimens. However, the background normal tissue may make the evaluation of allelic loss difficult.

There were relatively few papers on oncogenes in SCLC. Tumors with abnormalities of c-myc or L-myc were found to be more transplantable into nude mice than tumors without such changes. There was a particularly interesting report from the US on the jun oncogene. It was found that this gene was highly expressed constitutively in many SCC and to a lesser extent in NSCLC. This gene apparently codes for a transcription factor, that is, a protein which plays a regulatory role in transcription. The significance of this is unclear but there are multiple possibilities.

Papers presented indicated that there is increasing interest in studying drug resistance, obviously an important area clinically. In a study of large cell carcinoma with classic multidrug-resistant phenotype there was no detectable P-glycoprotein and decreased EGF receptor gene expression. Thus, this differs from the recently determined characteristics of other drug-resistant cells, and the investigators suggest the possibility that a different mechanism may be involved: I mention this particularly because others have made a similar observation in SCLC. In my opinion these are extremely important provocative leads which should be rapidly pursued.

There were other reports of cell lines of SCLC established before and after chemotherapy as well as xenografts in nude mice from chemoresistant SCLC. These may be useful in further studies of the mechanism of drug resistance. One group found that verapamil increases the sensitivity to doxorubicin in 3/5 newly established SCLC cell lines from patients either before chemotherapy or following relapse. However, sensitivity to doxorubicin in vitro did not seem to relate to the apparent chemical chemosensitivity of the original tumors. This is disappointing.

There was slightly increased interest in studies on metastases at this meeting compared with our last conference in Toronto. There was 1 study seeking the genetic basis of the metastatic phenotype in SCC. Another study correlated the secretion of tissue type plasminogen activator with spontaneous metastases in nude mice. However, the number of tumors studied was small. Then, in another paper from China, an inhibitory effect of a biologic extract on metastases of the Lewis lung carcinoma was described. There were several papers on proteases, which are often studied in relation to the metastatic phenotype. However, these were all related to NSCC.

Now for a summary of some presentations which may have somewhat more clinical relevance. It was shown that serial samples of creatine kinase isoenzyme BB reflected disease status in 80% of patients with SCLC. Both α- and γ-interferon were observed to act as radiosensitizers in cultures of SCLC. A group from Switzerland described a Mab to SCLC which had high tumor localization, long biologic half-life, and resistance to radiolabeling procedures. This was proposed as a promising candidate for selective destruction of SCLC by radioimmunotherapy. Whether this will be clinically useful is as yet undetermined. Another interesting study (in this case from Japan) described a stable mouse-human heterohybridoma which secretes human monoclonal Ab. This reacted with 39/39 pulmonary squamous cell carcinoma but had little reactivity with normal tissue by immunocytochemistry. It was effective in imaging tumors in nude mice and it was suggested that it may have clinical application.

Now in the category of new approaches I wish to mention 2 presentations in particular. One was a study from Denmark in which NMR spectroscopy was utilized to study SCLC xenograft growth in nude mice. The investigators concluded that this approach seems useful in studying alterations in the energy metabolism of individual tumors but indicate that the future role in preclinical drug testing is yet to be defined. The second new approach was the establishment of a hybridoma cell electrophoresis assay by a group in China. The hybridoma cells were used as natural “biobeads” carrying the antibodies in cytoelectrophoresis. The sensitivity, specificity, accuracy, and repeatability of the method suggested that this new technique may be of value in serodiagnosis of pulmonary carcinoma.

In another category there were 5 papers presented comparing SCLC and neuroblastoma. Since tetanus toxin has been used in the differential diagnosis of neuroblastoma, tetanus toxin labeling of 21 cell lines was evaluated using flow cytometry. There was prominent labeling of all SCLC cell lines and much less labeling of NSCLC cell lines. In another study it was found that the opioid antagonist naloxone, which modulates growth of neuroblastoma, also stimulates DNA synthesis in SCLC cell lines.

In a report from The Netherlands it was demonstrated that a panel of Mab to neuroblastoma and a panel of Mab to SCLC recognize an identical protein. This provides evidence for a phenotypic relationship between these tumors. In a similar study from Amsterdam an overlapping panel of Mab to SCLC and neuroblastoma demonstrated a similar tissue distribution pattern of reactivity in 3-4 month-old human fetuses.

In conclusion, it is extremely difficult to judge which papers are more or less significant, particularly over the long run. However, I will go out on a limb and share with you a personal opinion as to what I believe was a particularly important contribution. My nomination for a highly significant report would be the paper from Lyon, France by Trillet and colleagues. This abstract indicates the feasibility of using fiberoptic bronchoscopy biopsy specimens for DNA evaluations. The study was performed on 43 biopsy specimens, and up to 60 μg of DNA could be isolated. This report is very important in my mind. It points the way to applying multiple molecular genetic techniques to relatively early stages of pulmonary cancer. This should be particularly useful in SCC, where primary tumor specimens have rarely been available for study. This biopsy material along with the recently developed polymerase chain reaction, which allows one to amplify predetermined DNA sequences, opens up the opportunity to make an extremely broad range of molecular genetic studies in tumor tissue directly from the patient. I believe this paper was a highly significant contribution, and in looking into my crystal ball I see these techniques as being the basis for 5-10 abstracts on the pathobiology of pulmonary cancer for our next meeting in Australia. At the same time, let me apologize to others who may have presented equally important contributions which I have overlooked.