National Toxicology Program
Board of Scientific Counselor's Meeting
October 16 and 17, 1980

Summary Minutes

The National Toxicology Program (NTP) Board of Scientific Counselors met on October 16 and 17 in Conference Room 7, Building 31, National Institutes of Health, Bethesda, Maryland. (Attachment 1: Federal Register Meeting Announcement; Attachment 2: Agenda.)

Review and Discussion of Proposed Changes in the NTP Chemical Nomination and Selection Process: Dr. Rall said that NTP reached thousands of individuals by Federal Register announcements, NTP Technical Bulletins, and personal letters seeking nominations in an attempt to broaden the sources from which nominations are received. He proposed changes in the selection process to include: 1) an initial screening of all nominated chemicals by a small core group of NTP staff. This would not involve a literature search but would be aimed at identifying chemicals which had been tested, were on test, scheduled for testing by NTP or by others, or which had been rejected previously by NTP; 2) involvement of the Technical Information Section in the searching of toxicological and economic data bases; and 3) the solicitation of outside or public advice and comment on nominated chemicals through publication of notices in the Federal Register and mailings to outside organizations. Furthermore, the Chemical Evaluation Committee would form informal subgroups among its membership to consider nominated chemicals for the major testing areas, and to ensure indepth review, primary and secondary reviewers would be assigned to each chemical.

Considerable discussion ensued among the Board and attendees concerning mechanisms for public advice. Considerable support was given for the Board serving as a public advisory group. Dr. Rall agreed but opined that to save time executive summaries should go to the Board and Executive Committee simultaneously. Dr. Horning, however, indicated that for a public advisory group to be effective it should thoughtfully consider and rank nominations prior to Executive Committee action. Dr. Nelson concurred and said that this seemed to be the consensus of the Board, i.e., executive summaries would be forwarded to the Board following review by the Chemical Evaluation Committee. Concurrently, a Federal Register notice would be proposed soliciting advice and comment from outside parties within a given time period (30-60 days). In addition, NTP Technical Bulletins and letters requesting information or comment on the list of chemicals within the same time period would be mailed to interested groups and individuals. Chemicals nominated only for in vitro mutagenicity testing would not be included.

Dr. D. Canter, NTP, presented a revised proposal to the Board in response to the Board's recommendations. The primary change as described by Dr. Canter was to include the Board of Scientific Counselors in the review process. Concurrent with the solicitation of public advice, Executive Summaries would be forwarded to the Board which will meet to review the Summaries and the recommendations of the Chemical Evaluation Committee.

The Board would be augmented by consultants when necessary. Each chemical would be discussed and then assigned a priority for testing. The Board's ratings would then be incorporated into the Executive Summaries and sent to the Executive Committee for review and disposition. A description of the revised chemical nomination and selection process is shown (Attachment 3: Staff Proposal for the National Toxicology Program (NTP) Chemical Nomination and Selection Process). Dr. Horning moved that the revised proposal be accepted. Dr. Harper seconded the motion and it was passed unanimously.

Action Item: Revisions in the NTP chemical nomination and selection process will be implemented and a progress report given to the Board.

Proposed Statements on Human Hazards/Risk Based on the Experimental Results From Carcinogenesis Bioassays: Dr. Huff distributed a working paper for consideration (Attachment 4: Human Risk Assessment for the Carcinogenesis Bioassay Technical Reports).

As a starting point for discussion, the paper included: a) current wordage used in the Forward of the Carcinogenesis Bioassay Technical Reports, b) categories defined by the International Agency for Research on Cancer, c) a degrees-of-evidence classification scheme devised by Griesemer and Cueto, and d) suggestions for future technical reports. Dr. Hitchcock noted that this has been a difficult issue for the reviewers. Dr. Huff said the working paper was drafted in response to requests by the Board and Panel of Expert Reviewers. Dr. Nelson said that proposals had been made to centralize risk assessment guidelines of different agencies, perhaps with NTP as a central point. So we should defer until this issue comes to NTP, at least with regard to quantitative risk assessment. Thus, the current task should be to recommend a hazard warning or caution. Dr. Whittemore commented that risk assessment should be considered in a broader context than just carcinogenesis. Dr. Shepard discussed a classification scheme which is used for quantitative risk estimation with teratogens that involves weighing positive and negative evidence and rating chemicals and drugs on a scale ranging from plus 10 to minus 10.

Dr. Horning suggested there should be a small Board working group to help NTP decide on cautionary statements for the bioassay reports. Dr. Harper favored using the IARC guidelines unless these were considered deficient for the NTP needs. Dr. Canter (NTP) asked whether the statements on carcinogenicity in the bioassay reports referred only to evidence in the bioassay. Dr. Rall replied that was the case but thought eventually the reports should be expanded to be more like monographs, and include all available evidence.

Action Item: Dr. Nelson proposed setting up a small subgroup of Board members to deal with categorization of toxicology findings as to hazard. They would also consider potency in relation to risk assessment. Their first charge would be to develop recommendations on the wording of hazard warnings for the bioassay reports. He nominated Drs. Harper (Chairperson), Hitchcock, Horning, and Whittemore as members of the subgroup. Dr. Huff was asked to work with this subgroup as the NTP staff liaison.

Issues Related to Experimental Design of NTP Tests and Statistical Analyses of Carcinogenesis Bioassays: Dr. Whittemore discussed experimental design issues for chemicals selected for NTP testing which had arisen from conversations with Drs. N. Breslow, University of Washington, and R. Shore, NYU, both members of the NTP ad hoc Technical Report Review Panel, and Drs. D. Hoel and J. Haseman, NTP. The issues were subdivided into general design issues, and into those related to short-term tests, in vivo toxicology tests, and carcinogenesis bioassays (Attachment 5: NTP Design Issues).

Dr. Hoel then talked specifically about the statistical analyses used for the carcinogenesis bioassays, and changes agreed on by the group. These changes include: 1) Certain experimental data on individual animals should be in the reports to aid readers in assessing potency, e.g., age of death, cancer positive sites; 2) NTP should survey the state-of-the-art in data analysis and adjust the statistical methodology for such as tumor frequency by dose level, tumor frequency at sacrifice, tumor incidence based on life table analysis; 3) the most sensitive indicator of effect is the trend analysis statistic. This should be used in place of the currently used Fisher exact test. Probability (P) values should be given rather than N.S. (not significant); 4) relative risk is not useful; 5) general survival curves should be given but with improved graphic presentation; 6) overall P values for across sex and across species comparisons should be given in the Conclusions Section; 7) the Data Recording and Statistical Analysis section of the report should be rewritten; and 8) there was no resolution as to when historical control data should be used.

Dr. Hoel stated that for more validated quantitative risk assessment more dose levels were needed so curve fitting could be done. Research in this was underway and new methods developed will be applied to larger data sets.

Action Item: Dr. Moore said that some of the recommendations just made on changes in statistical methodology would be incorporated in a new bioassay report format now under development.

Review of NTP Pathology Quality Assessment: Dr. E. McConnell, pathology program leader, reviewed NTP pathology quality assessment following the prechronic studies (Attachment 6: Pathology Quality Assessment - Acute, Repeated - Dose and Subchronic Toxicity Tests). He said the most well-defined pathology in this aspect of the bioassay process is that for the 90 day studies, with the 14 day repeated dose work-up being less complete. After completion of the 90 day study, the reviewing pathologist would evaluate 10% of all slides (randomly selected) for quality of histotechnique. Furthermore, the Pathology Working Group, composed of NTP pathologists and outside pathologists when needed, would conduct a review of all slides where discrepancies with the original pathologist are noted by the reviewing pathologist. This quality assessment is described in more detail in Attachment 6. Dr. McConnell suggested that the quality assessment procedures might be considered for other NTP areas such as reproductive and developmental toxicology.

Dr. McConnell then described pathology assessment for the two year studies. After the originating (contractor) pathologist has read all the slides and submitted a report, a reviewing pathologist makes a count of all the tissues, looks at the slides for every tumor and every target organ and tissue, and examines all tissues from 10% of the animals. Based on this, the reviewing pathologist either agrees, disagrees, or supplements the originating pathologist's diagnoses. Then the Pathology Working Group examines the worksheets and slides. If there is disagreement with the originating pathologist which cannot be resolved, the technical report will note this with two sets of findings (tables) reflecting and documenting the differences.

At this point, Dr. Whittemore questioned whether the reviewing pathologists should be reading all slides 'blind', i.e., each slide should be coded giving only the animal and histology numbers. After considerable discussion Dr. Moore suggested doing a one-time blind study for comparison with other routine diagnoses used presently. Dr. Whittemore agreed with this proposal. Dr. J. Douglas, NTP, suggested that slides from a completed study be coded and read by a pathologist not associated with the study.

Action Item: Dr. Rall proposed that the pathology group set up a small blind study to be approved by Dr. Hoel. They would report their findings at the next meeting of the Board.

Status Report on Automatic Data Processing Study: Mr. J. Washington, NTP, gave a brief status report on the interim technical review of the Toxicology Data Management System (TDMS) being developed at the National Center for Toxicologic Research. The review had been conducted on site by three non-government experts for the purpose of determining whether TDMS could meet the automatic data processing needs of the NTP's animal bioassay programs. The final report of the consultants should be available in November.

Action Item: Dr. Nelson asked that the consultants meet with the Automatic Data Processing Subcommittee. The Subcommittee then will give a report at the next Board meeting.

Peer Review of Carcinogenesis Bioassay Technical Reports: Dr. Hitchcock summarized the carcinogenesis bioassay technical reports peer review session of October 15. Seven reports were reviewed: two, C.I. Acid Orange 10 and 11-Aminoundecanoic Acid, were deferred for extensive revisions and will be reviewed again at the next review meeting on February 18, 1981. The other five bioassay reports were approved contingent on minor revisions, mainly the stipulation that adenomas and nodules not be combined with carcinomas of the same organ site for statistical treatment. This distinction was to be made clearly in conclusions and summary where applicable.

Testing Needs Study: Dr. R. Tardiff, National Academy of Sciences/ National Research Council (NAS/NRC), described the three-year Testing Needs Study which NAS/NRC is performing under a contract with NTP. The purposes of the study are (1) to determine the magnitude of compounds that are untested or inadequately tested and might be candidates for additional

toxicity testing and the nature of toxicity testing, and (2) to develop a comprehensive and objective framework by which NTP could set priorities among compounds to be tested. A steering committee and three task oriented committees have been formed to effect the study. The committees will be concerned with toxicity data elements, sampling strategies and priority mechanisms.

Toxicology Testing and Test Development Activities: Dr. Moore talked about the NIH/NTP testing and test development activities. There are seven discrete areas: Program Resources, which includes chemical and animal resources, health and safety, and technical information; Program Operations, which is responsible for overall coordination of long-term bioassays; Data Management and Analysis; Cellular and Genetic Toxicology, concerned with short-term testing and test development; Systemic Toxicology, concerned with general toxicology and toxicology of various organ systems; Toxicologic and Carcinogenic Evaluation, concerned with science content and interpretation; and Chemical Pathology. The heads of each of these areas are members of the recently formed NTP/NIH Implementation Committee which meets approximately biweekly.

Dr. Moore then discussed chemical management. This begins with a determination of why the chemical was nominated, and if nominated by a regulatory agency, what is the regulatory need. Nearly all of the NIH/NTP scientific staff serve as chemical managers, although the number of chemicals assigned to a manager varies considerably depending on their other responsibilities. Two senior chemical managers have been identified to orient, train, and supervise or advise chemical managers.

He described the flow sequence of selected chemicals through the bioassay process after assignment to chemical managers. The chemical manager designs the experimental protocol which is then reviewed by an Experimental Design Group (EDG). Each EDG is composed of toxicologists, a pathologist, a chemist, an expert in carcinogenesis, and as required a biostatistician and experts in specialty areas such as chemical disposition and immunotoxicology. An ad hoc EDG will review proposed inhalation studies. Chemicals proposed for teratology or genetic toxicology studies are reviewed by different mechanisms. After EDG approval and assurance of adequate resources by the Implementation Committee, the chemical is assigned for bioassay to one of the 21 university or commercial laboratories qualified for conducting carcinogenesis bioassays and other toxicologic studies under a basic ordering agreement (BOA). (A BOA is a written understanding which defines the services, costs, and clauses applicable to any future demand order between the government and a contractor.) The 21 laboratories in the BOA 'pool' have been judged qualified to do some or all the testing procedures currently required by the NTP. After the award, there are certain evaluation stages: after the acute, subacute and subchronic (90 day) testing phases. The Chemical Manager in consultation with a Senior Chemical Manager makes recommendations to the Implementation Committee as to whether there should be changes in the protocol originally approved by the EDG. Following completion of the two-year bioassay, the Chemical Manager prepares or assists in preparation of the draft bioassay report which is reviewed by the Implementation Committee. After revision, the draft report is submitted for peer review.

Dr. Moore then discussed how the bioassay process has changed under the NTP to meet the objectives of broadened toxicologic characterization of chemicals. He prefaced this by describing how the prechronic phases of the bioassay were conducted prior to NTP involvement. These testing phases, especially the 90-day studies, are done to determine toxicopathology, species/sex differences, and a maximum tolerated dose (MTD) as a basis for setting appropriate doses for the two-year studies. Under NTP stewardship, other information is now obtained from the prechronic studies, e.g., general toxicology and chemical disposition. Thus, the completion of the 90-day study becomes a major decision point, and the results may indicate special target areas to monitor in the two-year tests. At this point, experimental design modification may be made and/or detailed special studies may be indicated which would be done separately, e.g., reproductive toxicology. Two handouts illustrated the marked increase in special studies between FY 1979 (the last year when chemical management was under the prime contract) and FY 1980 (when direct NTP chemical management was effected), (Attachment 7: Experimental Designs-FY 1979; Attachment 8: Experimental Designs-FY 1980). Dr. Moore discussed some of the tests involved in special studies. Developmental work on renal function assays has shown that existing tests for the most part are not very sensitive. For neurobehavioral and immunologic toxicology, NTP-developed test batteries are being used although validation of these batteries has not been completed. He noted that clinical chemistry is within the capabilities of most of the laboratories under the BOA. He reported that NTP is putting out a RFP for a special support contract to collect, store and analyze the data from the special studies. Finally, he emphasized that there is no standard design for the bioassay but rather, the design is adaptable to special testing needs for each chemical.

There was a brief discussion about the role of the Board as a concept review group. Dr. Moore said that this had been discussed at the Board review of April 7, 1980, and for the record, it was agreed then that discussion of scientific issues by the Board would qualify as a concept review, so that an RFP could be initiated at a later time without further concept review. Dr. Nelson commented that concept review of organ function test development may be most needed.

Action Item: Dr. Moore stated that the agenda for the next meeting should include a discussion concerning what concepts should be submitted to the Board, and mechanisms to use.

Mouse Lung Adenoma Assay: Dr. W. Hartwell, NTP, gave a status report on the evaluation of the Strain A mouse lung adenoma assay for its usefulness as a short-term in vivo test for chemical carcinogenesis (Attachment 9: Strain A Mouse Bioassay). He said that 60 chemicals are being tested in the Strain A mouse at the University of California, San Diego, and more than half of these are being tested at Oak Ridge National Laboratory in Strain A mice from a different source as a measure of interlaboratory reproducibility. Fourteen of the chemicals have been tested previously in the Bioassay Program. The results from 13 of the 14 chemicals have correlated directly with results obtained in bioassays.

Cellular and Genetic Toxicology Test Development: Dr. R. Tennant defined two immediate objectives of the program: (1) to interface selected short-term genetic assays with the bioassay process, and (2) to promote the

development and use of assay systems with better resolution of toxic biological endpoints. He gave an overall description of the current program which includes genetic toxicology, both microbial and mammalian systems; cellular transformation systems; and assay development. He noted that emphasis in assay systems development was on those detecting mutagenesis, chromosomal effects, and cell transformation. For the genetic component of the program, he listed the contracts and their status.

Dr. E. Zeiger, NTP, then gave a detailed presentation on the genetic toxicology testing and test development efforts. In the NCI component, there has been an interlaboratory comparison using the Salmonella assay in which the mutagenicity of 50 chemicals was tested with good qualitative agreement among the four laboratories in the study. In the NIEHS component, there is a focus on developing information on potent genetic hazards and carcinogenic potential of chemicals, and an objective of looking at endpoints predictive of clinical endpoints.

The Salmonella assay is currently well established in three contract laboratories. Assays are done using coded chemicals and there has been good agreement where chemicals have been assayed in more than one laboratory. Drosophila and in vitro mammalian cytogenetics assays are being evaluated and coded chemicals are being tested in these systems. Generally, chemicals that give positive or equivocal results (and certain negatives) in Salmonella will be tested in Drosophila. Both Salmonella-positive and-negative chemicals will be assayed in the in vitro cytogenetics assay for chromosome aberrations and sister chromatid exchange.

A major effort in the cellular genetic toxicology program is in the development and utilization of systems for handling and analyzing the large volumes of data generated. Included is a collaborative effort with the EPA in Data Systems development, and collaboration with NIEHS biometry staff to develop a family of 'biologically driven' statistical models for results from the Salmonella assay. The data-based management system being used is the PROPHET system which tracks the test status of chemicals and collects and analyzes the data. Data terminals will be on-line in the test laboratories with the provision for automatic plotting of data which can be useful for looking at trends. Dr. Zeiger said that they aim to build correlations between different test systems, using the data based system to do structure-activity-relationship studies and correlations.

Summaries of all test results will be given in the NTP Technical Bulletin, manuscripts are submitted to peer reviewed journals, and new data will be made available, probably through the National Technical Information Service (NTIS).

Dr. Tennant then discussed in some detail the cellular component of the program beginning with a description of contract initiatives for dual laboratory evaluation of the primary candidate mammalian cell transformation systems. The evaluation will require about three years to complete.

He reviewed other major development objectives of the cellular and genetic toxicology program including: (1) development of a contract to serve as a metabolism monitoring (precarcinogen or promutagen activation) resource for the program; (2) evaluation of a multiple-genetic endpoint single cell test system; (3) further development of the mouse heritable translocation and specific locus mutation tests; (4) incorporation of specific genetic endpoints into the 90-day (subchronic) phase of the animal bioassay; and (5) attempts to advance the state-of-the-art in human cell culture systems.

Dr. Tennant briefly described approaches for direct testing in humans. He commented that there has been no systematic attempt to establish background chromosome damage in humans and to estimate the variance of the test. He suggested that criteria for study should include definition of what constitutes exposed vs. nonexposed populations, methodology should be non-invasive, and appropriate protocols and biological endpoints should be defined.

Dr. Mendelsohn commented that there is good baseline data for somatic effects, e.g., bioassay data, but none for heritable genetic effects. He stressed the importance of the NTP cellular and genetic toxicology program addressing problems related to heritable genetic effects in mammals. Dr. Nelson agreed that NTP should put more stress on germ cell studies. Dr. Tennant noted that about one-fourth of the present cellular and genetic toxicology program is devoted to cellular toxicology with about three-fourths invested in genetic toxicology. Dr. Zeiger discussed the multi-locus system where effects on different endpoints, e.g., DNA repair, sister chromatid exchange can be compared in the same cell line, and perhaps even the same animal. There was a discussion about the need for short-term (three to four month) training programs for principal investigators to increase the number of potential contractor laboratories qualified to do short-term genetic toxicology testing in areas such as cytogenetics. The question was raised as to how NTP might support the needed training programs.

The next NTP Board of Scientific Counselors meeting will take place on January 15 and 16, 1981 at N.I.E.H.S., Research Triangle Park, North Carolina. The next carcinogenesis bioassay technical report review meeting will be held on February 18, 1981 at the National Institutes of Health, Bethesda, Maryland.