UPDATE National Toxicology Program
Headquartered at the National Institute of Environmental Health Services NIH-DHHS
JULY 2003

Stokes Named Chief Veterinary Officer for PHS

NIEHS's Capt. Bill Stokes has been named veterinarian chief professional officer for the Public Health Service Commissioned Corps. The appointment, by U.S. Surgeon General Richard H. Carmona, went into effect on May 1.

As the chief veterinary officer, Dr. Stokes will coordinate veterinary professional affairs and oversee recruitment, retention, career development and readiness of the more than 100 PHS veterinary officers, stationed throughout the NIH, the Centers for Disease Control and Prevention, the FDA, and other federal agencies.

Stokes is an internationally recognized authority on the humane care and use of laboratory animals for biomedical research and testing, and has authored or co-authored more than 50 publications and reports. He has served on numerous organizing committees for national and international conferences, and for 15 years was consultant to the accrediting association for laboratory animal care. Before joining the PHS Commissioned Corps, he served in the U.S. Navy and the U.S. Army.

NIEHS Director Ken Olden said, "To have one of our Public Health Service officers selected to serve in such a leadership role in this time of increasing demands on all fronts within public health is indeed a distinction. It confirms what we already knew at NIEHS about Dr. Stokes' high standards of professional excellence."

Dr. Stokes will continue to direct the NIEHS/National Toxicology Program's Interagency Center for the Evaluation of Alternative Toxicological Methods (NICEATM), which has led the federal government's reform of animal testing by introducing the use of scientifically acceptable non-animal testing to replace tests that expose animals to harsh chemicals. As a result of this work, Dr. Stokes received the Russell and Burch Recognition Award from the Humane Society of the United States. Other awards given to Dr. Stokes include the Army and PHS Meritorious Service Medals and Commendation Medals; PHS Outstanding Unit Citation; PHS Unit Commendation; PHS Citation; two Army Achievement Medals; and the Army Expert Field Medical Badge. He is a recipient of the NIH Director's Award and the 1999 Outstanding Veterinarian of the Year Award from the Massachusetts SPCA. Dr. Stokes joined the NIEHS in 1990.

(NIEHS Environmental Factor June 2003)

NTP Launches New Technical Report Series

The new series to be called Genetically Modified Models, or GMM series, was launched May 22 at a meeting at the NIEHS, where the draft findings and conclusions from the first two NTP technical reports in this series, aspartame (GMM-1) and acesulfame potassium (GMM-2), were peer reviewed by the Technical Reports Review Subcommittee, a standing subcommittee of the NTP Board of Scientific Counselors. This new series will contain the results from NTP toxicology and carcinogenicity studies conducted in genetically modified models, such as transgenic mice that have had a key gene related to cancer or other diseases added, “knocked out” or slightly changed.

The actions from this meeting, including the subcommittee’s recommendations on the findings and conclusions from carcinogenicity studies on aspartame and acesulfame potassium in p53 haploinsufficient mice as well as four other NTP studies conducted in traditional rodent models - 2-methylimidazole, propylene glycol mono-*t*-butyl ether, stoddard solvent IIC and triethanolamine - are available on the web (http://ntp-server.niehs.nih.gov/Meetings/2003/May2003Actions.html).

As the final reports from this new series are published, they will be available along with reports from the NTP Technical Report and NTP Toxicity Report series in hardcopy and electronic format, email ehponline@ehp.niehs.nih.gov, visit http://ehp.niehs.nih.gov or write Environmental Health Perspectives, Attn: Order Processing, 1001 Winstead Drive, Suite 355, Cary, NC 27513. Requests for hard copies may also be faxed to (919) 678-8696.

Report on Carcinogens

Prepared by the NTP, the *Report on Carcinogens* (RoC) is an informational scientific and public health document that identifies and discusses agents, substances, mixtures, or exposure circumstances that may pose a carcinogenic hazard to human health. It serves as a meaningful and useful compilation of data on (1) the carcinogenicity, genotoxicity, and biologic mechanisms of the listings in humans and/or animals; (2) the potential for exposure to them, and (3) the regulations promulgated by Federal agencies to limit exposures.

The most recent RoC, the 10th Edition, was released to the public on December 11, 2002 and is available on the Internet from the NTP RoC web page at http://ntp-server.niehs.nih.gov/NewHomeRoC/AboutRoC.html or by contacting Dr. C.W. Jameson, Head, Report on Carcinogens (contact information page 3).

11th Edition of the *RoC*

The scientific review of nominations to the 11th RoC is currently ongoing and publication is anticipated in 2004. The nominations to this edition of the report are:

- **1-Amino-2,4-dibromoanthraquinone:** an anthraquinone-derived vat dye that is used in the textile industry.

- **Selected Heterocyclic Amines (three nominations):**
  1. **MeIQ** (2-Amino-3,4-dimethylimidazo[4,5-*f*]quinoline)
  2. **MeIQx** (2-Amino-3,8-dimethylimidazo[4,5-*f*] quinoxaline);
  3. **PhIP** (2-Amino-1-methyl-6-phenylimidazo[4,5-*b*]pyridine)

  MeIQ, MeIQx, and PhIP are heterocyclic amines formed during heating or cooking and are found in cooked meat and fish.

- **Cobalt Sulfate:** used in electroplating and electro-chemical industries, as a coloring agent for ceramics, as a drying agent in inks, paints, varnishes and linoleum and as a mineral supplement in animal feed.

- **Diazoaminobenzene (DAAB):** used to promote adhesion of natural rubber to steel, as a polymer additive and as an intermediate in the production of a number of pesticides, dyes and other industrial chemicals.

- **Diethanolamine (DEA):** used in machine oils and metal cutting fluids and in the preparation of liquid laundry and dishwashing detergents, cosmetics, shampoos and hair conditioners, as well as in textile processing and other industrial uses.

- **Hepatitis B Virus (HBV):** a small DNA-enveloped virus that is transmitted through contact with blood and blood products or other body fluids.

- **Hepatitis C Virus (HCV):** an RNA-enveloped virus mainly transmitted in blood as is HBV above.

- **Human Papillomaviruses, Genital-Mucosal Types (HPVs):** small non-enveloped viruses that infect genital mucous membranes. HPV infections are common throughout the world.

- **X-radiation and gamma radiation:** major exposures of concern for cancer from X- and gamma-radiation are from the past use of atomic weapons and from medical uses of radiation.

- **Neutrons:** exposure normally occurs from a mixed irradiation field in which neutrons are a minor component; however, exceptions are exposure of patients to neutron radiotherapy beams and exposures of aircraft passengers and crew, who are bombarded naturally by the particles.

- **Naphthalene:** used in making many industrial chemicals, and as an ingredient in some mothballs and toilet bowl deodorants.

- **Nitrobenzene:** used in the production of aniline, a major chemical intermediate in the production of dyes.

- **Nitromethane:** used as a stabilizer added to many halogenated solvents and aerosol propellants.

- **Lead and Lead Compounds:** common sources of environmental lead exposure are from paint chips and leaded dusts and soils resulting from aging painted surfaces; major occupational exposures are in the lead smelting and refining industries, battery-manufacturing plants, steel welding or cutting operations, construction, and firing ranges.

- **4,4'-Thiodianiline:** an intermediate in the manufacture of several dyes.

The first set of nominations - 1-amino-2,4-dibromoanthraquinone, MeIQ, MeIQx, PhIP, cobalt sulfate, diethanolamine, naphthalene, nitrobenzene, nitromethane, and 4,4'-thiodianiline - were peer reviewed by the RoC Subcommittee of the NTP Board of Scientific Counselors November 19-20, 2003, at its meeting in Washington, DC Summary minutes from that meeting are available on the NTP web site (http://ntp-server.niehs.nih.gov).

The second set of nominations – diazoaminobenzene, hepatitis B virus, hepatitis C virus, human papillomaviruses (genital-mucosal types), X-radiation, gamma-radiation, neutrons, and lead and lead compounds - will be peer reviewed by the RoC Subcommittee at an upcoming meeting on October 14-15, 2003, in Washington, DC. When available, additional information about this meeting will be announced in the Federal Register and posted on the NTP web site.

The background documents for the nominations to the 11th Edition of the RoC and public comments received on the nominations are posted on the web in PDF format (http://ntp-server.niehs.nih.gov/NewHomeRoC/AboutRoC.html) along with the criteria for listing and a description of the

RoC review process. A limited number of hardcopies and CDs of the background document are available upon request to Dr. Jameson (see below). The NTP continues to solicit public comment on these nominations. Comments should be directed to Dr. Jameson.

NTP Requests Nominations for Future Evaluation for Listing/Delisting in the RoC

The NTP solicits and encourages the broadest participation from interested individuals or parties in nominating agents, substances, mixtures, or exposure circumstances for listing in or delisting from the *RoC*. Nominations should contain a rationale for the listing or delisting and appropriate supporting background information and relevant data (e.g., journal articles, NTP Technical Reports, IARC listings, exposure surveys, and release inventories) when possible. Nominations should be directed to Dr. Jameson.

Dr. C.W. Jameson, Head, Report on Carcinogens, NIEHS/NIH, 79 Alexander Drive, Rm. 3118, P.O. Box 12233, MD EC-14, Research Triangle Park, NC 27709; T: 919-541-4096; F: 919-541-0144, jameson@niehs.nih.gov

Upcoming Events**

August 12-13, 2003	Scientific Advisory Committee on Alternative Toxicological Methods Meeting NIEHS, Research Triangle Park, NC
September 10-11, 2003	NTP Board of Scientific Counselors Meeting Research Triangle Park, NC
October 14-15, 2003	NTP Board of Scientific Counselors Report on Carcinogens Subcommittee Meeting Washington, DC
November 5-6, 2003	NTP Board of Scientific Counselors Technical Reports Review Subcommittee Meeting Research Triangle Park, NC

** As available information about these meeting will be posted on the NTP website (http://ntp.server.niehs.nih.gov).

NTP Interagency Center for the Evaluation of Alternative Toxicological Methods

ICCVAM Forwards New Test Methods Recommended for Acute Oral Toxicity Hazard Assessments

In accordance with the ICCVAM Authorization Act of 2000, the first test recommendations by the Interagency Coordinating Committee on the Validation of Alternative Methods (ICCVAM) were sent to Federal agency heads on March 21, 2003. They are for *in vitro* methods that can be used to estimate starting doses for acute oral toxicity studies and for a revised test method for determining acute oral toxicity.

The *in vitro* methods are described in two ICCVAM reports - *Report of the International Workshop on In Vitro Methods for Assessing Acute Systemic Toxicity* (NIH No. 01-4499) and *Guidance Document on Using In Vitro Data to Estimate In Vivo Starting Doses for Acute Toxicity* (NIH No. 01-4500).

ICCVAM test recommendations on the revised Up-and-Down Procedure (UDP) are described in the report - *The Revised Up-and-Down Procedure: A Test Method for Determining the Acute Oral Toxicity of Chemicals; Results of an Independent Peer Review Evaluation Organized by the ICCVAM and NICEATM* (NIH No. 02-4501).

ICCVAM recommends that the UDP be used instead of the conventional LD_{50} test to determine the acute oral toxicity hazard of chemicals. When used in place of the conventional LD_{50}, the UDP will reduce and refine the use of animals. Federal testing regulations for which the UDP may be applicable are provided in the UDP report. These test recommendations and the responses from the agencies will be made available to the public.

***In Vitro* Test Methods for Identifying Potential Endocrine Disrupting Chemicals**

On June 3, 2003, a Federal Register notice (Vol. 68, No. 106, pages 33171 – 33172: posted on the NTP web site: http://ntp-server.niehs.nih.gov) announced the availability of the report - *ICCVAM Evaluation of In Vitro Test Methods for Detecting Potential Endocrine Disruptors: Estrogen Receptor and Androgen Receptor Binding and Transcriptional Activation Assays* (NIH No. 03-4503).

The report contains ICCVAM recommendations on minimum procedural standards and reference chemicals for standardization and validation of *in vitro* estrogen and

androgen receptor binding and transcriptional activation assays. This report is the result of an April 2000 request from EPA asking ICCVAM to evaluate the validation status of *in vitro* estrogen receptor (ER) and androgen receptor (AR) binding and transcriptional activation assays proposed as possible components of the EPA Endocrine Disruptor Screening Program (EDSP) Tier 1 screening battery. ICCVAM agreed to evaluate these test methods based on their potential interagency applicability and public health significance.

Test method developers are encouraged to submit *in vitro* test methods for evaluation by ICCVAM that adhere to the minimum procedural standards outlined in the report and that have undergone validation using the recommended substances. Following adequate validation of *in vitro* endocrine disruptor test methods, the NTP Interagency Center for the Evaluation of Alternative Toxicological Methods (NICETAM) will coordinate their scientific peer review on behalf of the ICCVAM. Formal ICCVAM test recommendations will then be forwarded to Federal agencies as required by the ICCVAM Authorization Act of 2000 (P.L. 106-545).

Proposed Minimum Performance Standards for Three Types of *In Vitro* Test Methods for Assessing Dermal Corrosivity Hazard Potential of Chemicals

A Federal Register notice published July 1, 2003 (Vol. 68, No. 126, pages 39104-39105) announces the availability of the ICCVAM Dermal Corrosivity and Irritation Working Group (DCIWG) proposed Minimum Performance Standards (MPS) for three types of *in vitro* methods for assessing the dermal corrosivity hazard potential of chemicals. Public comment is invited. This notice is posted on the NTP (http://ntp-server.niehs.nih.gov) and NICEATM/ICCVAM (http://iccvam.niehs.nih.gov) web sites.

The ICCVAM developed the proposed MPS to communicate criteria that can be used to determine if similar test methods have comparable accuracy and reliability. All written comments received by noon on August 15, 2003, will be posted on the ICCVAM/ NICEATM web site (http://iccvam.niehs.nih.gov) and considered by the DCIWG and ICCVAM during development of the final ICCVAM MPS for these assays. Final ICCVAM MPS will be published as an addendum to previously published ICCVAM reports on these test methods and forwarded to Federal agencies for their consideration. Availability of the final MPS will be announced through the Federal Register and copies of the MPS will be made available electronically on the ICCVAM/NICEATM web site or in hardcopy by contacting NICEATM (contact information below).

Request for High Quality *In Vivo* Ocular and Dermal Irritation Data

ICCVAM and NICEATM are collaborating with the European Centre for the Validation of Alternative Methods (ECVAM) to conduct a validation study on *in vitro* test methods for assessing dermal irritation. NICEATM will formally solicit this information through a forthcoming Federal Register notice and request chemical and protocol information/test data on commercially available chemicals used for dermal or ocular irritancy in rabbits and/or for dermal irritancy in humans using standardized testing methods. The published notice will be posted on the NTP web site.

The notice will invite interested persons and/or organizations to submit data on chemicals tested for skin irritancy in rabbits and/or humans using standardized testing methods. This information will aid ICCVAM in identifying suitable reference chemicals (i.e., those with high quality rabbit and/or human dermal irritation data) for the validation study. Commercially available chemicals that have been tested in both rabbits and humans will be given priority in the selection process. High quality rabbit ocular irritation data will also be requested in order to identify appropriate reference chemicals that can be used in future validation studies of *in vitro* test methods for ocular irritancy.

NICEATM will compile the submitted chemical and protocol information/test data on the commercially available chemicals. ICCVAM and the ICCVAM DCIWG will consider all submitted data received by the deadline published in the future Federal Register notice. These groups will review the data and identify chemicals that might be appropriate for use in the upcoming validation study on *in vitro* test methods for dermal irritation. The resulting list of chemicals tested for skin irritancy in rabbits and/or humans and supporting data will be provided to ECVAM for consideration in the upcoming validation of *in vitro* test methods for dermal irritation.

Validation Study on *In Vitro* Methods for Estimating Acute Oral Toxicity Hazard of Chemicals

NICEATM and the ECVAM are conducting a collaborative validation study to evaluate the use of two *in vitro* basal cytotoxicity assays proposed for predicting starting doses for *in vivo* acute oral toxicity assays and lethal concentrations in humans. Expert scientists at an earlier ICCVAM International Workshop recommended the study of *in vitro* methods for assessing acute systemic toxicity. Three laboratories are participating in the evaluation of the neutral red uptake assays using both a mouse cell line (i.e., BALB/c 3T3 fibroblasts) and a primary human cell line (i.e., normal human epithelial keratinocytes). The cytotoxicity results for the 72 coded chemicals, representing a wide range of toxicity, will be used to predict starting doses for *in vivo* acute oral toxicity assays. Phase I testing was completed in May 2003. All labs have Phase II protocols in place and testing of nine coded chemicals began on June 2. With completion of Phase II, final optimized protocols will be prepared and used for Phase III, which will involve testing 60 coded chemicals.

Dr. William Stokes, Director, NICEATM, NIEHS/NIH 79 Alexander Drive, Rm. 3129, P. O. Box 12233, MD EC-17, Research Triangle Park, NC 27709, Phone: 919-541-2384; F: 919-541-0947; stokes@niehs.nih.gov.

NTP Solicits Input on Testing Nominations

The National Toxicology Program (NTP) continuously solicits and accepts nominations for toxicological studies to be undertaken by the program. The nominations are subjected to several levels of review before selections for testing are made and toxicological studies are designed and implemented. As part of this review process, the NTP Interagency Committee for Chemical Evaluation and Coordination (ICCEC) met on June 10, 2003, to review 14 new nominations and make study recommendations. The NTP requests public comment on the nominations and study recommendations and asks for the submission of additional relevant information that it might consider in its continued evaluation of these nominations. A Federal Register notice formally soliciting input is forthcoming and once published will be posted on the NTP web site (http://ntp-server.niehs.nih.gov). Internet links to electronic versions of supporting documents for each nomination and further information on the NTP and the NTP Chemical Nomination and Selection Process can be accessed through the NTP web site.

Substances recommended for testing by the ICCEC

- Acrylamide [79-06-1] and Glycidamide [5694-00-8]: recommended studies - toxicological characterization, toxicokinetics, mechanistic (hemoglobin adducts), carcinogenicity and bioavailability from food and drinking water.
- Antimony trisulfide [1345-04-6]: recommended studies: chronic toxicity/carcinogenicity.
- Cadmium telluride [1306-25-8]: recommended studies - toxicological characterization and chemical disposition (oral and inhalation routes).
- Cedarwood oil, Virginia [8000-27-9]: recommended studies - toxicological characterization and developmental toxicity.
- Chondroitin sulfate [9007-28-7]: recommended studies - chronic toxicity/carcinogenicity and carcinogenicity of chondroitin sulfate and glucosamine combined.
- Dimethylethanolamine [108-01-0]: recommended study – metabolism.
- Drugs positive for QT Interval Prolongation/ Induction of *Torsade* Proarrhythmia [No CAS No.]: recommended studies - initiate a study program to develop *in vitro* and *in vivo* test systems for assessing QT interval prolongation.
- Glucosamine [3416-24-8]: recommended studies - chronic toxicity/carcinogenicity and carcinogenicity of chondroitin sulfate and glucosamine combined.
- Nanoscale materials [No CAS No.]: recommended studies – size- and composition-dependent biological disposition of nanocrystalline fluorescent semiconductor materials, toxicological characterization of high-aspect-ratio carbon nanomaterials, role of particle core and surface composition in the immunotoxicity of the above listed materials, and phototoxicity of representative metal oxide nanoparticles.
- *trans*-Resveratrol [501-36-0] recommended studies - toxicological characterization, carcinogenicity and reproductive toxicity.
- Tetrabromobisphenol A [79-94-7]: recommended studies - toxicological characterization, neurodevelopmental toxicity, and carcinogencity.
- Tetrabromobisphenol A-bis(2,3-dibromopropyl ether) [21850-44-2]: recommended studies - toxicological characterization, *in vivo* genotoxicity, metabolism and carcinogencity.
- Tungsten [7440-33-7]: recommended studies - toxicological characterization and carcinogenicity; studies should focus on a representative soluble tungsten compound.

Substance for Which No Study Is Recommended at this Time

- 4-Phenylcyclohexene [4994-16-5]: low suspicion of hazard based on available human exposure and toxicity information.

Persons submitting comments and information are asked to include their name, affiliation, mailing address, phone, fax, e-mail address and sponsoring organization (if any) with the submission. Written submissions should be sent to Dr. Scott Masten (contact information below) and will be posted electronically on the NTP web site as they are received.

Nominations for new toxicology studies are welcome and should be addressed to Dr. Scott Masten. Information on how to submit testing nominations is available at http://ntp-server.niehs.nih.gov/NomPage/noms.html

Dr. Scott A. Masten, Office of Chemical Nomination and Selection, NIEHS/NTP, P. O. Box 12233, MD A3-07, Research Triangle Park, North Carolina 27709; telephone: (919) 541-5710; FAX: (919) 541-3647; masten@niehs.nih.gov

Toxicogenomics Gordon Research Conference

The inaugural meeting of the 2003 Toxicogenomics Gordon Research Conference was held on June 22-27, 2003, at Bates College in Lewiston, Maine.

Co-chaired by James Selkirk, Raymond Tennant, and Vice Chair Leona Samson,, the meeting was designed to foster discussion among the scientists studying various aspects of microarray, proteomics, and bioinformatics. The key feature of the meeting was its diversity and outstanding collection of world-renown scientists representing academia, industry and government.

The guiding principle at Gordon Conferences is that creative and powerful solutions to research problems are often catalyzed by a broad knowledge of diverse biological research areas and experimental approaches.

The meeting provided both a general educational forum and promoted information and productive communication between investigators studying diversified sub-specialties. Information about the meeting is available at:
http://www.grc.uri.edu/programs/2003/toxico.htm

How to Subscribe to the NTP List-server

The NTP Update is issued approximately 4 times each year. To subscribe to the "list-server" and receive the NTP Update as well as other NTP news and announcements electronically, register online at http://ntp-server.niehs.nih.gov or send e-mail to ntpmail-request@list.niehs.nih.gov with the word "subscribe" as the body of the message, or contact the NTP Liaison and Scientific Review Office.

Additional information about the NTP along with announcements of meetings, publications, study results and its centers is available on the Internet at http://ntp-server.niehs.nih.gov.

The ehpOnline maintains issues of the Report on Carcinogens and the library of NTP Technical Reports and NTP Toxicity Reports and adds new reports as available. The electronic PDF files of completed reports are available free-of-charge and printed reports can be purchased through ehpOnline. To gain access to these reports, go to http://ehp.niehs.nih.gov or call 866-541-3841 or 919-653-2595.

Contact information: NTP Liaison and Scientific Review Office, NIEHS, P.O. Box 12233, MD A3-01, Research Triangle Park, NC 27709; T: 919-541-0530; F: 919-541-0295; liaison@starbase.niehs.nih.gov