DRAFT STATEMENT OF WORK

PREVENT Cancer Preclinical Drug Development Program: Toxicology and Pharmacology Testing

Independently and not as an agent of the Government, the Contractor shall furnish all the necessary services, qualified personnel, materials, equipment, and facilities, not otherwise provided by the Government, as needed to perform the Statement of Work.

SCOPE

These contracts will support the conduct of regulatory toxicological and pharmacological studies with potential cancer preventive agents to enable or advance human clinical trials. As such, the data from these studies must be suitable for filing with the Food and Drug Administration as part of Investigational New Drug Applications (IND) or New Drug Applications (NDA). Successful Offerors shall perform studies in two or more of the following Technical Task areas: genotoxicity testing; general toxicology in experimental animals; reproductive toxicology studies in rodents and rabbits; and specialized studies as needed.

Specific Task Orders will be submitted to successful Offerors to support the regulatory requirements of specific candidate agent(s), depending on the characteristics of the agent and its position along the drug development pathway. Tasks shall be conducted in accordance with quality oversight that is appropriate to the phase of the specific task and within all applicable and current federal, state, and local laws, codes, ordinances and regulations, as well as all PHS Safety and Health provisions as applicable. The tasks shall also comply with applicable FDA and/or International Conference of Harmonisation (ICH) guidance including, but not limited to, those listed in the Addendum to this Statement of Work.

Task Orders shall range in duration from 1 to 3 years. For proposal purposes, Offerors shall assume that there will be three Task Orders (two for small molecules and 1 for a vaccine) awarded each year, and that there will be only one agent, administered by only one route of administration, per Task Order. Assume that each small molecule will require a 6-month rodent study and a 9-month non-rodent study, and that the vaccine will require a safety study, a cellular immune response and an immunogenicity study.

PROJECT MANAGEMENT

Task Order Initiation Meeting and/or Teleconference: Within 10 calendar days after the effective date of the Task Order or before the start of any tasks, plan for and participate in an Initiation Meeting or Teleconference to review the Task Order requirements and timelines, discuss Task Order technical plans and approaches, and identify any anticipated problems/obstacles to completing the Task Order requirements. The Task Initiation Meeting will be attended by Contractor key personnel (including subcontractor personnel as appropriate), the COR, the designated Subject Matter Experts, the Contracting Officer, and the investigator representatives.

Key personnel or other senior staff coordinator(s) shall be available for ad hoc
meetings/teleconferences with the COR immediately upon a meeting request.

**TASK AREAS**

In performance of work under any Task Area, the Contractor shall comply with all applicable guidelines and Federal Regulations, including, but not limited to, those listed in the Addendum to this Statement of Work, titled “References”. The Technical Requirements have been assembled into the following Task Areas:

- Task Area A – Genotoxicity Testing
- Task Area B – General Toxicology in Experimental Animals
- Task Area C – Reproductive Toxicology Studies in Rodents and Rabbits
- Task Area D – Specialized Studies
- Task Area E – Management Support Activities

1.1 TASK AREA A: GENOTOXICITY TESTING

The Contractor shall determine the homogeneity, concentration, and stability of the test substance/test substance mixture under the conditions of use, as defined in the Task Order Statement of Work (TO SOW).

A. **BACTERIAL REVERSE MUTATION TEST**

The standard set of strains (+S9) shall be used, as cited in the International Conference on Harmonization (ICH) S2B Guidelines to test for G-C and A-T base changes, with appropriate positive and negative control(s). Range-finding studies shall be conducted. In some cases, other strains (e.g. nitroreductase deficient) shall be employed. Endpoints shall assess at least the following based on the cited ICH guidelines:

1. solubility
2. cytotoxicity
3. dose response
4. genotoxicity

B. **AN IN VITRO TEST WITH CYTOGENETIC EVALUATION OF CHROMOSOMAL DAMAGE WITH MAMMALIAN CELLS**

Several mammalian cell systems shall be utilized, including systems that detect gross chromosomal damage such as *in vitro* tests for structural and numerical chromosomal aberrations.

In each Task Order, the Contractor shall propose and justify an appropriate test system that is acceptable under International Conference on Harmonization (ICH) S2B Guidelines.
C. **AN IN VIVO TEST FOR CHROMOSOMAL DAMAGE USING MOUSE OR RAT HEMATOPOIETIC CELLS**

The Contractor shall conduct assays that measure either chromosomal aberrations in bone marrow cells or micronucleated polychromatic erythrocytes in bone marrow or peripheral blood.

The approach to these assays shall consider species, gender, and other parameters that shall be discussed in Task Order proposals.

D. **FOLLOW-UP STRATEGIES FOR POSITIVE TESTS**

The Contractor shall conduct follow-up assays (e.g. RasH2 rat, Big Blue™ (lacI/cII) mouse, p53+- mouse, Comet assay, GreenScreen Human cell GADD45α-GFP, etc.) for agents with positive results in the standard genotoxicity battery.

4.2 **TASK AREA B: GENERAL TOXICOLOGY IN EXPERIMENTAL ANIMALS**

The Contractor shall conduct studies in mice, rats, and dogs. However, on rare occasions the Contractor shall use other species such as rabbits, guinea pigs, hamsters, minipigs, monkeys, or other animals that are deemed more appropriate due to the inherent nature of the chemopreventive agent.

The Contractor shall perform work in this Task Area as outlined in each TO SOW. Examples of such study designs, include but are not limited to, the following.

A. **FOURTEEN-DAY DOSE RANGING ORAL TOXICITY IN RODENTS**

The Contractor shall conduct studies as follows:

1. Rats or mice of appropriate age and weight shall be procured by the Contractor from an established, reliable commercial breeder.
2. Animals shall be quarantined for one week prior to study.
3. The appropriate number of mice or rats shall be used for adequate statistical power to allow proper evaluation and interpretation of the data generated from the studies.
4. The test article shall be administered per dose level administered by gavage or other appropriate route determined by the Contracting Officer’s Representative (COR) to obtain a range of toxic effects and mortality rates.
5. Pharmacological or toxic effects shall be recorded as to onset, duration, and disappearance over a two-week period.
6. Food consumption shall be monitored and body weight shall be taken at appropriate times, which will be stated in each TO SOW.
7. Survivors shall be sacrificed at the end of the observation period and gross necropsies performed on all animals.
8. Animals found dead or sacrificed in moribund condition shall be immediately necropsied or refrigerated until necropsied.
9. Tissue samples shall be taken for histopathological examination where abnormalities are noted.
10. A Lethal Dose (LD)\textsubscript{50} or approximate measure shall be determined along with other parameters, including but not limited to dose range, latency, duration, and reversibility of toxic effects.
11. Appropriate records, as outlined in each TO SOW and according to regulatory guidelines, shall be maintained on all observations, animal deaths, and necropsies.

B. TWENTY-EIGHT DAY STUDIES IN RODENTS AND DOGS

1. Route and timing of test agent administration shall be by oral gavage, capsule, food admixture, or other appropriate route once per day for four weeks.
2. As appropriate, the 28-day definitive study will be preceded by a 14-day range finding study to help select appropriate dosages for the 28-day study.
3. In the 28-day study, approximately 160 male and female rats or mice (20/sex/group) and 32 male and female dogs (4/sex/group) shall be chosen to provide control, low, medium, and high dosage groups.
4. If specified in the TO SOW, additional animals shall be included to serve as recovery study animals as well.
5. General appearance, behavior, appetite, elimination, and presence of any signs of toxicity shall be recorded daily.
6. Body weight and food consumption shall be recorded weekly with observations for mortality at least once each morning and each afternoon.
7. Clinical laboratory tests shall be performed as defined in the TO SOW.
8. All survivors shall be sacrificed during the fifth week and subjected to a detailed gross necropsy and histopathologic evaluation of major organs and tissues.
9. Histopathologic studies shall be done on high dose level and control animals and on intermediate dose level animals when there are positive findings at the high dose level.
10. The maximum tolerated and no-effect doses shall be determined and pharmacokinetic evaluations may be incorporated into these studies as defined in the TO SOW.
11. Tissues and/or fluids shall be collected and preserved for potential subsequent drug and metabolite level analyses, genomic, proteomic, metabolomic, and/or other studies.

C. NINETY DAY, ORAL TOXICITY STUDIES IN RODENTS AND DOGS

1. Test material shall be administered daily by oral gavage, capsule, food admixture, or other appropriate route as defined in the TO SOW.
2. Approximately 100 male and 100 female rats or mice (25/dose/sex) and 32 male and female dogs (4/dose/sex) shall be used for control, low, medium, and high dosage groups.
3. Additional animals shall be treated in order to conduct a recovery study as specified in the TO SOW.
4. General appearance, behavior, appetite, elimination, and presence of any signs of
toxicity shall be recorded daily.
5. Body weight and food consumption shall be recorded weekly.
6. Daily observations for mortality and weekly records of appearance, behavior, and
   signs of toxicologic or pharmacologic effects shall be maintained.
7. Hematological, serum chemistry, urinalysis, ophthalmology, cardiology (dogs only),
   and plasma drug measurements shall be made as required by the TO SOW at
   appropriate intervals during the study.
8. All survivors shall be sacrificed after three months and subjected to detailed
   necropsy, including gross and microscopic evaluation.
9. All tissues from the high dose and control groups shall be microscopically evaluated
   and the identified target tissues shall be read in the low and medium dosage groups.
10. The maximum tolerated and no-effect doses shall be determined.
11. Tissues and/or fluids shall be collected for potential subsequent drug and metabolite
    level analyses, genomic, proteomic, metabolomic or other studies.

D. SIX MONTH RODENT AND NINE MONTH DOG CHRONIC TOXICITY STUDIES

1. Test material shall be administered daily by oral gavage, capsule, food admixture, or
   other appropriate route as defined in the TO SOW.
2. Approximately 120 male and 120 female rats or mice (30/dose/sex) and 32 male and
   female dogs (4/dose/sex) shall be used to provide adequate data for control, low,
   medium, and high dosage groups.
3. Body weight and food consumption shall be recorded weekly.
4. Daily observations for mortality and weekly records of appearance, behavior, and
   signs of toxicologic or pharmacologic effects shall be documented in the study file
   and in the study report.
5. Hematological, serum chemistry, urinalysis, ophthalmology, cardiology (dogs only),
   and plasma drug measurements shall be made as required by the TO SOW at
   appropriate intervals during the study.
6. All survivors shall be sacrificed at the end of the respective dosing periods and
   subjected to detailed necropsy, including gross and microscopic evaluation (high
   dose and control with read-down in target tissues).
7. The maximum tolerated and no-effect doses shall be determined.
8. Additional animals shall be treated in order to conduct a recovery study as specified
   in the TO SOW.
9. Tissues and/or fluids shall be collected for potential subsequent drug and metabolite
   level analyses, genomic, proteomic, metabolomic or other studies.

E. CARCINOGENICITY STUDIES IN RODENTS

As defined in the TO SOW, the Contractor shall perform the following:

1. Animals shall be six to seven weeks of age at the time of release from quarantine and
   start of the study.
2. All animals shall be randomized by weight and approximately sixty animals per dose
   per sex and species shall be started in each test group routinely.
3. Additional animals for a recovery study or interim sacrifice at 12 months shall be included as needed.
4. Sentinel animals for serological monitoring and satellite animals for blood collections shall be included as needed.
5. The test material shall be administered daily by oral gavage, capsule, food admixture, or other route.
6. Three dose levels of test agent and control groups shall be used.
7. Body weight and food consumption shall be recorded weekly.
8. Daily observations for mortality and weekly records of appearance, behavior, and signs of toxicologic or pharmacologic effects shall be maintained.
9. Hematological, serum chemistry, urinalysis, and plasma drug measurements shall be made at appropriate intervals during the study.
10. All survivors shall be sacrificed after 18 to 24 months and subjected to detailed necropsy, including gross and microscopic evaluation (high dose and control with read-down in target tissues).
11. The total spontaneous tumor incidence shall be determined and the carcinogenic potential of the test compound evaluated.
12. Tissues and/or fluids shall be collected for subsequent drug and metabolite level analyses, genomic, proteomic, metabolomic or other studies.
13. The Contractor shall also be required to conduct 6-month carcinogenicity studies in p53 +/- mice if specified in the TO SOW for the compound under study.

4.3 TASK AREA C: REPRODUCTIVE TOXICITY STUDIES IN RODENTS AND RABBITS

The Contractor shall conduct studies in this Task Area using mice, rats, and rabbits. However, the Contractor shall use other species when appropriate due to the inherent nature of the chemopreventive agent or a suggestion by the regulatory agencies, and could include guinea pigs, hamsters, monkeys, or other animals.

The Contractor shall perform work in this Task Area as outlined in each TO SOW.

A. SEGMENT II TERATOGENICITY STUDIES

   The following types of studies shall be performed to evaluate the effect of the test article on organogenesis:

1. Dose range-finding study (6 groups of 5 females).
2. Teratogenicity study (4 groups of 20 rabbits and 25 rats per group).
3. Implantations, resorptions, fetal viability and gross malformations, and soft tissue and bone malformations.

B. TWO GENERATION REPRODUCTION STUDIES IN RODENTS

   The Contractor shall perform tests designed to provide information concerning effects of a test article on gonadal function, estrous cycles, mating behavior, conception,
parturition, neonatal morbidity, mortality, lactation, weaning, and the growth and
development of the offspring.

Depending upon the nature of the test article, second filial (F2) teratology evaluations
shall be conducted as needed (F1 and F2 males shall be mated to naive females).

Endpoints include:

1. female fertility index,
2. gestation index,
3. weaning index,
4. sex ratio,
5. viability indices,
6. growth indices,
7. maternal toxicity, and
8. specialized tests, e.g., sperm, neuronal, or immune evaluations.

4.4 TASK AREA D: SPECIALIZED STUDIES

The Contractor shall conduct studies in this Task Area using mice, rats, dogs, rabbits, guinea
pigs, hamsters, minipigs, monkeys, or other appropriate species to evaluate drug-specific
mechanisms of toxicity. Interspecies differences in drug metabolism, pharmacokinetics, and
pharmacodynamics are well recognized and shall be considered in the development of
protocols.

1. Pharmacokinetic studies shall be undertaken as needed to evaluate the absorption,
plasma concentrations, and/or tissue distributions of the test article or metabolites in
different formulations.
2. Deidentified human fluids, tissues, or cells (obtained commercially or from other
studies) shall be employed as needed.

As outlined in Task Orders, the Contractor shall conduct studies as follows:

A. BIOANALYTICAL METHODOLOGY STUDIES

1. The Contractor shall perform bioanalytical method development and validation
studies. Methodologies that shall be considered include High Performance Liquid
Chromatography (HPLC), Gas Chromatography (GC), HPLC-Mass Spectroscopy
(HPLC-MS), HPLC-MS-MS, GC-MS, GC-MS-MS, radioimmunoassay,
imunoaffinity, atomic absorption, bioassay, and other assays.
2. Analytical techniques shall be identified and validated for sample matrices with the
emphasis on selectivity, accuracy, precision, lower limit of quantification, and
stability as approved by the COR.
3. Isomeric composition and stability shall be considered, as necessary.
4. Formulation, plasma, blood, urine, feces, or specific tissues shall be used as
necessary.
5. Applicable guidelines include, but are not limited to: Bioanalytical Method Validation Q2A; Text on Validation of Analytical Procedures; and, Q2B, Validation of Analytical Procedures: Methodology. (See the Addendum to the Statement of Work for additional guideline information.)

B. PHARMACOKINETIC AND PHARMACOKINETIC-PHARMACODYNAMIC STUDIES

The Contractor shall perform the following to determine pharmacokinetic profiles for investigational agents.

1. Pharmacokinetic studies shall provide the parameters of absorption, blood concentration-time profiles, bioavailability, distribution, and elimination following administration to animals by various routes and schedules. Data on time-dependent tissue concentration of the test agent, determined as part of the toxicology testing, shall contribute to the pharmacokinetic profile.
2. Information on major metabolites shall be included in order to provide as complete a picture as possible of the overall distribution and fate of the test agent.
3. Appropriate modeling shall be applied to determine probable patterns of biodistribution and compartmentalization.
4. Radiolabeled test articles shall be used as needed. The Nuclear Regulatory Commission (NRC) licensure shall be maintained for the life of this contract.
5. Pharmacodynamic profiles (e.g. time dependent biomarker concentration or activity, effect on putative targets) shall also be determined and modeled as needed in conjunction with pharmacokinetics in order to characterize correlation of the response with drug levels and/or total exposures.
6. Determination of free drug fraction (protein unbound drug) shall be evaluated as needed.

C. BIOAVAILABILITY AND BIOEQUIVALENCE STUDIES

The Contractor shall perform bioavailability and/or bioequivalence determinations in experimental animals. Bioavailability and bioequivalence refer to absolute or relative rates and extent of an active pharmaceutical product reaching the systemic circulation or site(s) of action.

1. At least three commonly used indices shall be estimated: the maximum drug concentration in plasma ($C_{max}$), the time needed to reach the maximum concentration in plasma ($t_{max}$), and the area under the plasma-concentration-vs.-time curve (AUC). Possible formulations/routes of administration include: oral, transdermal, subcutaneous, intramuscular, intravenous, buccal, sublingual, respiratory, vaginal, etc.
2. Effects of co-administration of other agents, dietary ingredients, or inactive ingredients shall also be evaluated as needed.
3. Isomeric composition and stability of the agents shall be considered, as necessary.
4. Radiolabeled test articles shall be used as needed and NRC licensure shall be
D. **DRUG METABOLISM/DRUG INTERACTION STUDIES**

The Contractor shall perform studies for characterization and quantification of metabolites and/or detection and characterization of drug-drug interactions.

1. Major and active metabolites shall be identified, characterized, and quantified using bioanalytical technologies such as HPLC, GC, NMR, GC-MS, GC-MS/MS, LC-MS, LC-MS/MS, immunoaffinity, etc.
2. Radiolabeled test articles shall be used as needed and NRC licensure shall be maintained for the life of this contract.
3. Isomeric composition and stability shall be considered as necessary.
4. Drug interaction experiments shall evaluate a potential for drug interactions (pharmacokinetic and/or pharmacodynamic inhibition or induction) during a combination therapy or with a likely-to-be-encountered co-medication.
5. Drug metabolism and interactions studies shall be *in vivo* (different routes of administration possible) and/or *in vitro* (e.g. microsomes, S9 fractions, hepatocytes, liver slices from experimental animals, or deidentified human sources), as deemed necessary.
6. In addition to metabolic considerations, drug transporters shall also be considered, as needed.

E. **SPECIFIC SAFETY STUDIES**

The Contractor shall perform specialized investigations to determine mechanisms of action and toxicity of specified agents.

1. The studies shall be conducted as independent safety pharmacology studies or as a component of a larger study, such as a 90 day toxicity study. Examples of such investigations include cardiovascular toxicity, neurotoxicity, pulmonary toxicity, ototoxicity, endocrine toxicity, neurovascular toxicity, and coagulopathies.
2. Dermal and aerosol formulations/routes of administration shall be investigated, as appropriate.

F. **GENOMICS/PROTEOMICS/METABOLOMICS**

The Contractor shall use genomic, proteomic, metabolomic, and/or other -omic tools to identify and evaluate the potential for toxic reactions.

1. Genomic microarray methodology (cDNA or oligonucleotide microarrays and other related and evolving techniques) and proteomic tools [e.g., surface enhanced laser desorption/ionization time-of-flight (SELDI-TOF); matrix-assisted laser desorption/ionization time-of-flight, (MALDI-TOF); laser capture microdissection; protein, antibody, and tissue microarrays; and other related and evolving techniques] shall be used as needed.
2. These new approaches shall be used if they provide a complementary and more accurate and/or earlier assessment of toxicity risk potential, its mechanisms and targets, and lead to better-targeted animal studies using fewer animals.

G. COMPUTATIONAL (PREDICTIVE) PHARMACOLOGY/TOXICOLOGY

In silico or computational techniques shall be utilized as defined by the TO SOW to estimate solubility, intestinal permeability, metabolism, and predict toxicology (e.g. genotoxicity, hepatotoxicity, etc.) of new candidate chemopreventive agents.

H. SAFETY ASSESSMENT OF VACCINES AND OTHER IMMUNOPREVENTIVE AGENTS

The Contractor shall perform studies necessary to enable human clinical trials with investigational vaccines or other immunomodulatory agents used for cancer prevention (e.g. antibodies). Specifically, these studies shall assess immune activity, potential for local and systemic toxicity, and acute and time-dependent toxicities including potential resolution. Appropriate species, gender, age and endpoints shall be considered by the Contractor in the design of the studies to assure detection of potential safety signals, the expected immune activity and the specificity of the agent. The route of administration, dose levels and dosing schedules to be used shall be specified by the COR in the TO SOW. Generally, these studies will include at least one immunization beyond that planned for clinical use, and a dose equivalent to the highest dose of vaccine planned for the clinical trial. An accelerated schedule of vaccine delivery, relative to the clinical trial, may also be employed.

Possible endpoints to be evaluated in these studies include, but are not limited to, the following.

1. Mortality, clinical signs, injection site reactions, body weights, diet and water intake, hematology, coagulation, serum biochemistry
2. Cellular and/or humoral immune response evaluation assays, in vitro tissue binding assays (e.g. immunohistochemistry) for tissue cross-reactivity evaluation, and other in vitro studies, as specified in the Task Order
3. Virulence/neurovirulence (e.g. for new live attenuated viral vectors/vaccines)
4. Biodistribution, integration, and/or persistence of encoded antigens in DNA vaccines, as specified in the Task Order
5. Macroscopic and microscopic evaluations of major organs and other relevant tissues, including but not limited to brain, kidney, liver, lung, heart, GI tract, reproductive organs, skin, injection site(s), spleen, thymus, and draining lymph nodes, at study time points specified in the Task Order

In addition, since many cancer vaccine formulations contain adjuvants used in conjunction with vaccine antigens to augment or direct the specific immune response to an antigen, the potential toxicity of the adjuvant alone, and of the investigational clinical vaccine-adjuvant combination, should be assessed in preclinical studies as appropriate.
4.5. Task Area E: Management Support Activities

Scope of Task Area E

Task Area E will support parent contract requirements as stated in the contract. The scope of this Task Order includes management functions performed by the Contractor in support of NCI’s contract entitled “PREVENT Cancer Preclinical Drug Development Program: Toxicology and Pharmacology Testing” which occurs independent of other specific Task Orders to be awarded under the contract.

Requirements for Task Area E include:

1. Kick-off Meeting
   a. Plan and participate in the initial parent contract kick-off meeting to be held either in Rockville, Maryland or in the Contractor’s vicinity. The kick-off meeting shall be attended by the Principal Investigator and other relevant key Contractor technical and business staff, the COR and his/her designees, and the Contracting Officer and/or his/her designees.

5) GENERAL PROCEDURES

A. ANIMAL FACILITY

Laboratories shall be accredited by or registered as follows:

1. The Contractor shall have an approved Animal Welfare Assurance from the Office of Extramural Research (OER), Office of Laboratory Animal Welfare (OLAW) (http://grants.nih.gov/grants/olaw/olaw.htm), Office of the Director, NIH, as required by Section I-43-30 of the Public Health Service Policy on Humane Care and Use of Laboratory Animals. The Contractor shall maintain such assurance for the duration of this contract, and any subcontractors performing work under this contract involving the use of animals shall also obtain and maintain an approved Animal Welfare Assurance certified facility.

2. The Contractor and any subcontractor shall be fully accredited by the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC) International or equivalent and maintain that accreditation for the life of the contract. Information about AAALAC accreditation is available at www.aaalac.org.

4. The Contractor’s Institutional Animal Care and Use Committee (IACUC) shall approve all animal procedures under this contract.

All animals for toxicological studies shall be furnished by the Contractor from established, reputable, known commercial breeders. The Contractor shall quarantine the animals for an appropriate period prior to placing them on test for toxicological studies and their release shall be documented by a veterinarian certified by the American College of Laboratory Animal Medicine (ACLAM).

B. GOOD LABORATORY PRACTICE (GLP) REGULATIONS

The Contractor shall develop experimental protocols and conduct studies using established techniques, study parameters, and statistical methods of data analysis in accordance with current FDA GLP regulations for non-clinical laboratory studies (21 CFR Part 58), revised as of April 1, 2016. ([http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?CFRPart=58 &showFR=1](http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?CFRPart=58 &showFR=1)) and International Conference on Harmonization (ICH) Guidelines ([https://www.fda.gov/regulatoryinformation/guidances/ucm122049.htm](https://www.fda.gov/regulatoryinformation/guidances/ucm122049.htm)). The format and content of the technical report shall conform to current FDA GLP standards. (See Addendum to the Statement of Work)

The Contractor shall possess and maintain knowledge of FDA GLP regulations in order to be able to conduct and report these studies accordingly. The Contractor shall ensure compliance with current GLP regulations on all studies, and provide documentation to the COR of any FDA data audits and inspections (i.e., reports and responses to issues raised) within one (1) week after receipt of the audit or inspection report from the FDA. If it is determined that problems are encountered with the assays or documentation because GLP conditions were not met or maintained, the Contractor shall repeat the assay at no additional cost to the Government.

C. PATHOLOGY

The Contractor shall ensure that examination and reporting of pathologic alterations in organs and tissues in each toxicological study are conducted to demonstrate histopathological evaluations correlated to clinical, hematologic, or clinical chemistry evidence of toxicity to an organ system with direct injury to that system. The Contractor shall accurately report the type and severity of all lesions in the animals in order to increase the efficiency of prediction for toxic manifestations of agents administered to patients in subsequent clinical trials.

The Contractor shall ensure that:

1. All lesions are categorized as either drug-related or non-drug-related;
2. Each lesion is listed and coded by the most specific topographical and morphological diagnoses, severity, and distribution using Systemized Nomenclature of Medicine (SNOMED) codes.
Pathology examinations and procedures shall include at least the following, as appropriate and as specified in each Task Order SOW:

**Rats:**

1. A complete necropsy, organ weight determinations, clinical chemistry, hematology, coagulation, and histopathology shall be performed on treated and control animals.
2. Animals shall be observed two times daily (once in the morning and once in the afternoon at least 6 hours apart, including holidays and weekends) for moribundity and mortality; one of these observation periods shall be shortly after dosing to observe potential effects of dosing.
3. Gross motor and behavioral activity, observable changes in appearance, and clinical signs related to the pharmacology and toxicology of the test article shall be recorded with attention to the sign, onset, and duration.
4. Animals whose condition makes it unlikely that they will survive until the next observation, based upon criteria established by the Contractor’s Principal Investigator in concert with the veterinary staff and toxicologist, shall be sacrificed immediately, necropsied, and diagnosed histopathologically (unless the cause of death is grossly related to the gavage procedure). Moribund animals shall be terminated out of sequence with complete necropsy and histopathology as for scheduled necropsies.
5. Antemortem observations shall be recorded for each animal prior to necropsy, including those clinically normal.
6. All significant antemortem observations shall receive comment or confirmation at necropsy. All animals shall have final body weights and required organ weights taken, unless severely autolyzed.
7. A board certified veterinary pathologist shall be available to examine any unusual findings during unscheduled necropsies and shall be present at the terminal sacrifices.
8. The Contractor shall obtain and analyze additional samples during severe or unusual toxicity (e.g. blood for hematology/clinical chemistry/coagulation/plasma drug levels) and/or modify doses of the agents. If severe or unusual toxicity is noted, these additional blood samples shall be obtained at the onset of the observation and the Contracting Officer’s Representative (COR) shall be notified within 24 hours.
   a. Histopathological observations shall be made on all required tissues of animals in the control group and in the highest treated group that has greater than 50% survival.
   b. Tissues from animals in other groups shall be fixed and held for possible further study.
   c. Tissues from organs found to be normal in a higher dose groups need not be examined histologically in lower dose groups.
   d. Tissues from organs which are found to be abnormal in a higher dose group shall be examined in animals from the other groups.
9. Tissues that shall be examined macroscopically in all animals and microscopically in controls and the above-specified animals include, but are not limited to: (*record weight of tissues):

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<tr>
<th>Adrenals (pair)</th>
<th>Jejunum</th>
<th>Skin</th>
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<tr>
<td>Aorta (thoracic)</td>
<td>*Kidneys</td>
<td>Spinal cord (2 levels;</td>
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<td>Bladder (urinary)</td>
<td>*Liver</td>
<td>entire cord if signs</td>
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<td>*Brain (3 levels)</td>
<td>Lymph node (mesenteric)</td>
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<td>Cecum</td>
<td>Mammary gland</td>
<td>*Spleen</td>
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<td>Colon</td>
<td>Ovaries and fallopian tubes</td>
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<td>Corpus and cervix uteri</td>
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<td>*Testes</td>
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<td>Pituitary</td>
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<td>Ileum</td>
<td>Seminal vesicle</td>
<td>Lesions (all gross visible)</td>
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<td>*Tissue masses</td>
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<td>Abnormal lymph nodes</td>
</tr>
</tbody>
</table>

10. Bone marrow smears shall be prepared, inventoried, and properly stored until shipped to the DCP Chemoprevention Repository at the time of Task Order close-out.

11. Teratological examinations of rodents, when needed, shall be similar to that described in the section on Rabbits below.

12. The Draft and Final report of each study shall contain high resolution, illustrative photographs or digital images of drug-induced lesions.

**Dogs:**

1. A complete necropsy, organ weight determinations, and histopathology shall be performed on treated and control animals.

2. Dogs shall be checked for mortality and signs of morbidity twice daily (once in the morning and once in the afternoon at least 6 hours apart, including holidays and weekends); one of these observation periods shall be shortly after dosing to observe potential effects of dosing.

3. Moribund dogs shall be terminated out of sequence with complete necropsy and histopathology as for scheduled necropsies.

4. Antemortem observations shall be recorded for each dog prior to necropsy, including those clinically normal.

5. All significant antemortem observations shall receive comment or confirmation at necropsy.

6. All dogs shall have final body weights and required organ weights taken, unless severely autolyzed.
7. A board certified veterinary pathologist shall be available to examine any unusual findings during unscheduled necropsies and shall be present at the terminal sacrifices.

8. The Contractor shall obtain and analyze additional samples during severe or unusual toxicity (e.g. blood for hematology/clinical chemistry/coagulation/plasma drug levels) and/or modify doses of the agents. If severe or unusual toxicity is noted, these additional plasma samples shall be obtained at the onset of the observation and the COR shall be notified within 24 hours.

   a. Histopathologic observations shall be made on all required tissues of dogs in the control group and in the highest treated group that has greater than 50% survival.
   b. Tissues from dogs in other groups shall be fixed and held for possible further study.
   c. Tissues from organs found to be normal in a higher dose group need not be examined histologically in lower dose groups.
   d. Tissues from organs which are found to be abnormal in a higher dose group shall be examined in dogs from the lower dose groups.

9. Tissues that shall be examined macroscopically in all animals and microscopically in controls and the above-specified dogs include, but are not limited to: (*record weights of tissues):
<table>
<thead>
<tr>
<th>Adrenals (pair)</th>
<th>Medial lobe with section of gall bladder and left lateral lobe</th>
<th>Spinal cord (2 levels; entire cord if signs indicate cord involvement)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aorta (thoracic)</td>
<td>Lungs and bronchi (left atypical and left diaphragmatic lobes; infuse one lobe with formalin; sample both infused and non-infused tissues)</td>
<td>*Spleen</td>
</tr>
<tr>
<td>Bone (femur with epiphysseal plate of head)</td>
<td>Lymph node (bronchial, mandibular, mesenteric)</td>
<td>Stomach</td>
</tr>
<tr>
<td>Bone marrow (sternum)</td>
<td>Mammary gland (from both sexes)</td>
<td>*Testes</td>
</tr>
<tr>
<td>*Brain (3 levels)</td>
<td>Ovaries and fallopian tubes</td>
<td>*Thymus</td>
</tr>
<tr>
<td>Cecum</td>
<td>Pancreas (head and tail)</td>
<td>*Thyroid/Parathyroid (record combined weight)</td>
</tr>
<tr>
<td>Colon</td>
<td>Pituitary</td>
<td>Tongue</td>
</tr>
<tr>
<td>Corpus and cervix uteri</td>
<td>Prostate</td>
<td>Tonsil</td>
</tr>
<tr>
<td>Duodenum (incl. bile and pancreatic ducts)</td>
<td>Rectum</td>
<td>Trachea</td>
</tr>
<tr>
<td>Epididymis</td>
<td>Salivary gland (mandibular)</td>
<td>Ureter</td>
</tr>
<tr>
<td>Esophagus</td>
<td>Sciatic nerve (longitudinal section from region of proximal femur)</td>
<td>Urinary bladder</td>
</tr>
<tr>
<td>Eyes</td>
<td>Skeletal muscle</td>
<td>Vagina and all gross visible lesions</td>
</tr>
<tr>
<td>Gall bladder</td>
<td>Skin (nonfrictional surface, dorsal thorax; frictional site, elbow)</td>
<td>*Tissue masses</td>
</tr>
<tr>
<td>Heart</td>
<td>*Kidneys (weigh separately)</td>
<td>Abnormal lymph nodes</td>
</tr>
<tr>
<td>Ileum</td>
<td>*Liver (weigh total right)</td>
<td></td>
</tr>
<tr>
<td>Jejunum</td>
<td>*Spleen</td>
<td></td>
</tr>
</tbody>
</table>

9. Bone marrow smears shall be prepared, inventoried, and properly stored until shipment to the Chemoprevention Repository at the time of contract close-out.

10. The Draft and Final reports for each study shall contain high resolution, illustrative photographs or digital images of all drug-induced lesions.

**Rabbits:**

The COR may require rabbits to be used as the non-rodent species in certain instances, including but not limited to, general toxicology studies and developmental toxicology studies. For general toxicology studies, histopathological examination shall be as extensive as that specified for dog studies above. The following shall be followed for developmental/teratology toxicology studies.

The Contractor shall describe criteria for determining the viability of a fetus, early and late resorptions, and for the detection of possible implantations sites.
1. In teratology studies, animals shall be sacrificed on day 20 of presumed gestation.
2. A Caesarian section shall be performed immediately upon sacrifice.
3. The abdominal and thoracic cavities shall be opened and examined.
4. The ovaries of all animals (gravid and apparently nongravid) shall be examined and the number of corpora lutea on each ovary recorded.
5. The uterus of gravid animals shall be examined and weighed.
6. The number and location of fetuses and early and late resorptions shall be recorded.
7. Analyses of the uterus shall include, at least, by absolute number and percent:
   a. Corpora lutea (CL) and implantations per dam
   b. Percent pre- and post- implantation loss
   c. Live and dead fetuses per litter
   d. Resorptions (early and late) per litter
   e. Litter with resorptions, litters with total resorptions
   f. Nonlive\(^1\) per litter, litters with nonlive
   g. Affected\(^2\) per litter, litters with affected
      \(^1\) nonlive = dead + resorbed
      \(^2\) affected = nonlive + externally malformed
8. Fetal observations shall include individual body weight and sex.
9. All fetuses shall be examined externally and findings recorded.
10. Fetuses in the range finding study with gross external alterations shall be preserved in Bouin's solution for possible future examination.
11. Analyses shall include, at least, by absolute number and percent:
    a. Live fetuses per litter, litters with live fetuses
    b. Sex ratio per litter
    c. Mean fetal body weight by litter and by sex within litter
    d. Externally malformed fetuses per litter
    e. Litters with externally malformed fetuses
    f. Externally malformed fetuses by sex per litter
12. A morphological examination of the fetuses shall be performed and include a detailed evaluation of the eyes, palate, head shape, and trunk and extremities.
13. Abnormal findings shall be recorded.
14. In Segment II teratology studies, but not in range finding studies, each fetus shall be further identified by litter and uterine placement, and one-half of the viable and nonviable fetuses shall be fixed in Bouin's solution and viscerally examined using the Wilson's free-hand slicing technique.
15. Remaining fetuses shall be eviscerated and the skeletons stained with Alizarin Red S, examined for alterations, cleared in glycerin, and stored in 99.5% glycerin /0.5% phenol.
16. All abnormal findings shall be recorded.
17. Analyses shall further include all the above and, at least, by absolute number and
percent:

a. Fetuses malformed externally, skeletally, and viscerally by litter
b. Litters with malformed fetuses
c. Malformed fetuses by litter and by sex within litter
d. Types and incidence of individual malformations
e. Fetuses and their malformations by litter and dose
f. Fetuses with variations per litter
g. Litters with fetuses having variations
h. Types and incidences of individual variations
i. Crown to rump length

18. The Draft and Final reports for each study shall contain high resolution, illustrative photographs or digital images of drug-induced malformations.

**Monkeys:**

Depending upon the study design specified in each Task Order, histopathological examination shall be as extensive as that specified for dog studies above. In specialized studies involving monkeys, or other species, specific pharmacokinetic and/or pharmacodynamic endpoints may be required as determined by the COR.

**D. ASSAY OF TEST MATERIAL AND FORMULATIONS**

1. NCI staff shall provide cancer chemoprevention agents (chemical or biological, including vaccines and antibodies) in suitable quantity and quality. Generally, a Certificate of Analysis, Material Safety Data Sheet, and confirmation of identity will be provided with the agent; in some instances, the latter shall be required to be performed by the Contractor as directed in the TO SOW.
2. The Contractor shall determine the purity of the neat compound before and after the in-life study, as well as the homogeneity, concentration, and stability of the formulated material under the conditions of use.
3. The Contractor shall use analytical instrumentation to develop, refine, and adapt the methods (e.g., for plasma or other matrices).
4. The methods shall be validated or cross validated by the Contractor prior to determining homogeneity, concentration, and stability of the formulated material, or prior to determining the concentration of test article in plasma or other matrices.
5. Dose accuracy shall be $\pm 10\%$ of theoretical.
6. Dose concentration shall be adjusted to accommodate animal weight changes.
7. Although most studies shall be conducted using oral gavage to rats and capsules to dogs, other routes such as admixture with food, dissolution into drinking water, dermal, aerosol, etc. may be appropriate.

**E. OTHER CONSIDERATIONS**

1. Protocol Modifications:
a. Additions, modifications, or deletions of protocols shall be approved by the COR prior to implementation.
b. If unexpected adverse events are observed at any stage of evaluation that would jeopardize the progress of a study, the Contractor shall contact the COR as early as possible but within 24 hours to report the findings.
c. If a test fails to provide the information needed, for example as a result of misdosing of animals, errors in formulation, cross contamination of samples, unapproved protocol deviations, loss of data, or non-Good Laboratory Practice (non-GLP) performance, then the protocol shall be repeated at the direction of the COR and at the expense of the Contractor.
d. The rerun may be modified within the original scope of work as agreed by the COR.

2. Quality Assurance Unit:
   a. Studies shall be audited for GLP compliance by a Quality Assurance Unit. Specifically, the quality assurance unit shall be responsible for monitoring each study to assure that the facilities, equipment, personnel, methods, practices, records and controls are in conformance with FDA GLP regulations.
   b. The Final report for each study shall include documentation and state that the study was conducted under GLP.

3. Statistics:
   a. The Contractor’s historical in-house and current referenced data bases shall be used as a basis for the underlying normal distribution and appropriate statistical tests (parametric versus nonparametric).
   b. In addition to analysis of variance (ANOVA) and t-tests, linear regression analysis shall be performed, all using validated computer system(s).
   c. The Final report for each study shall include a description of the statistical analyses used.

4. eCTD and SEND Format:

The Contractor shall prepare study reports with study data generated under this contract in the electronic Common Technical Document (eCTD) format. This requirement is intended to comply with Section 745A(a)(1) of the FD&C Act, which specifies that submissions under NDAs, ANDAs and BLAs must be in eCTD format beginning May 15, 2017, and commercial IND submissions must be in eCTD format by May 15, 2018. This requirement ensures a consistent general framework for organizing study data, including templates for datasets, standard names for variables, and standard ways of doing calculations with common variables. This requirement will also facilitate the exchange of clinical and nonclinical research data between computer systems.
Addendum to the Statement of Work/References


TASK AREA A - GENOTOXICITY TESTING


TASK AREA B - GENERAL TOXICOLOGY IN EXPERIMENTAL ANIMALS


**TASK AREA C - REPRODUCTIVE TOXICITY STUDIES IN RODENTS AND RABBITS**


**TASK AREA D - SPECIALIZED STUDIES**

i. **BIOANALYTICAL METHODOLOGY STUDIES**


ii. **PHARMACOKINETIC AND PHARMACOKINETIC-PHARMACODYNAMIC STUDIES**


iii. BIOAVAILABILITY AND BIOEQUIVALENCE STUDIES


iv. DRUG METABOLISM AND DRUG INTERACTION STUDIES


v. SPECIFIC SAFETY STUDIES


vi. SAFETY ASSESSMENT OF VACCINES AND OTHER IMMUNOPREVENTIVE AGENTS


ICH. Guidance for Industry: S6 Addendum to Preclinical Safety Evaluation of Biotechnology-Derived Pharmaceuticals. 2012 May. Available from:

FDA. Guidance for Industry: Considerations for Plasmid DNA Vaccines for Infectious Disease Indications. 2007 Nov. Available from:

FDA. Guidance for Industry: Content and Format of Chemistry, Manufacturing, and Controls Information and Establishment Description Information for Vaccine or Related Product. 1999 Jan. Available from:

GENERAL PROCEDURES

Good Laboratory Practice for Nonclinical Laboratory Studies, 21 C.F.R. § 58 (revised as of April 1, 2016). Available from:
